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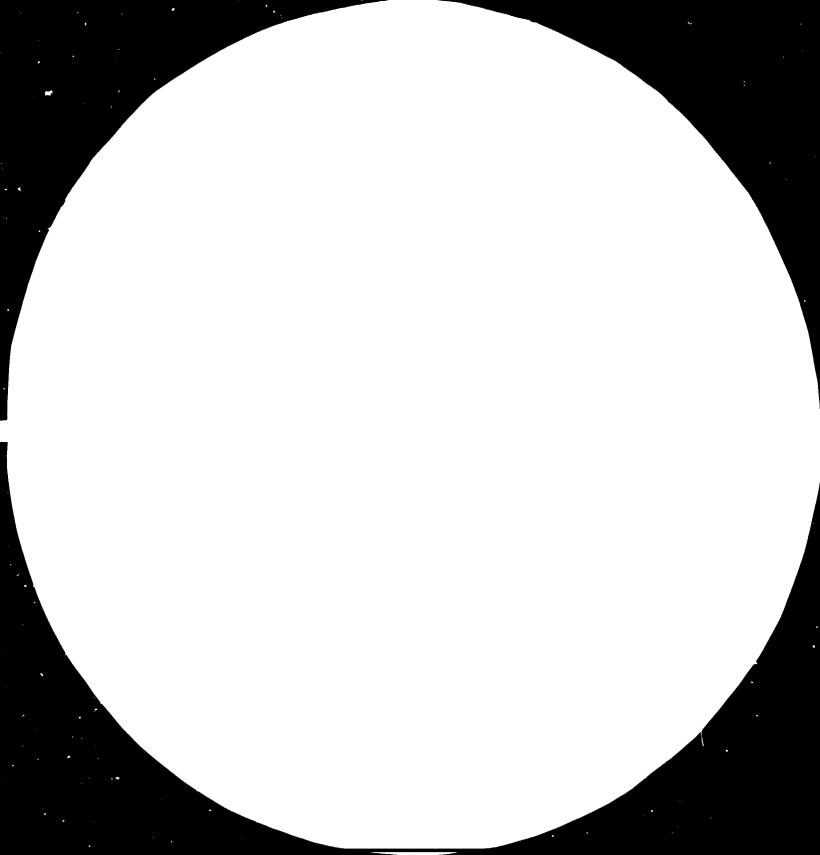
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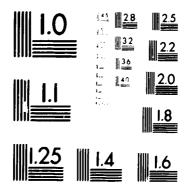
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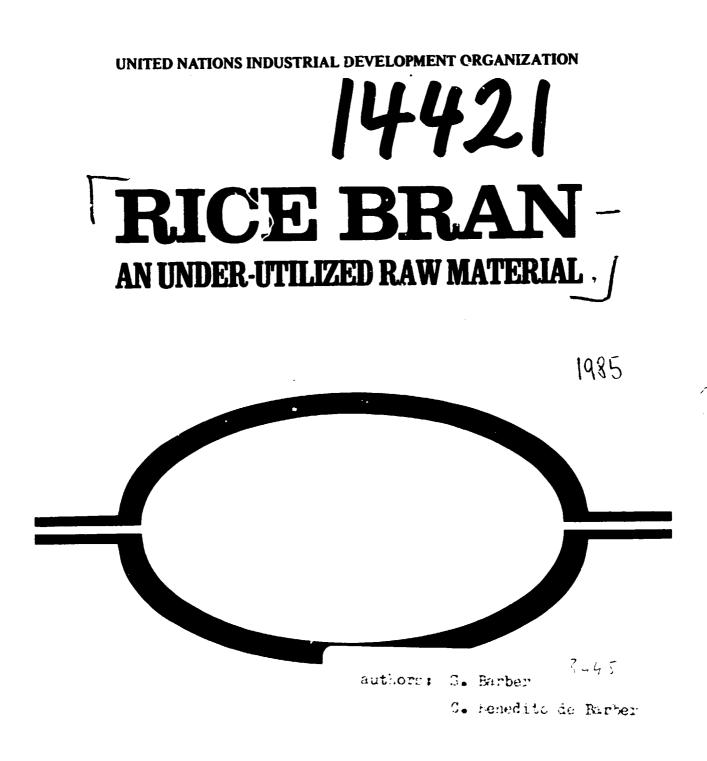
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#### MICROCOPY RESOLUTION TEST CHART NATIONAL BUREAU OF STANDARDS STANDARD REFERENCE MATERIAL 1010a (ANSI and ISO TEST CHART No. 2)





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UNITED NATIONS



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## **RICE BRAN: AN UNDER-UTILIZED RAW MATERIAL**

## LE SON DE RIZ : UNE MATIERE PREMIERE SOUS-UTILISEE

## UNA MATERIA PRIMA SUBUTILIZADA: EL SALVADO DE ARROZ

## ABSTRACT / SOMMAIRE / EXTRACTO

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## ABSTRACT

The annual production of rice bran is approximately 40 million tonnes. Out of this 6-8 million tonnes consists of edible oil, a similar quantity consists of high-quality protein, and there is a high proportion of vitamins, trace elements and other nutrients. In spite of this, rice bran is at present an under-utilized by-product.

There is a great deal of information on rice bran, but it is widely scattered and difficult to obtain, in some cases muddled and in others incorrect and generally fragmentary. A publication which assembles, classifies and critically examines the information available will therefore be useful for teachers, researchers and industrialists concerned with the post-harvesting technology of rice and, in particular, with the problems relating to a by-product as valuable as rice bran.

The United Nations Industrial Development Organization is aware of the importance of this subject and of the need for a comprehensive source of information, and has therefore published the present volume, which simultaneously assembles and analyses the existing information on the subject and presents a rational and systematic discussion of the scientific and technical basis of the industrial processing of bran.

The publication covers in its six chapters the production of bran, the basic principles of stabilization, the characteristics of the by-product, control of the process, the technology for stabilization, and the problems of conservation and storage.

The subject of bran production is dealt with from its beginnings, with an analysis of the rice grain and an examination of the manufacturing process, attention always being focused on the by-product, bran, rather than the primary product, rice. An understanding of the grain as the raw material for the production of bran must include a knowledge of its morphology, microscopic structure and chemical composition. The entire process of bran production is discussed: cleaning, husk removal, husk separation, paddy separation, whitening, polishing, collection of fractions of bran and germ separation. Various types of plant are discussed with the aid of numerous diagrams, covering the technologies, ranging from the simplest to the most sophisticated, that are used throughout the rice belt.

Two chapters deal fully and in detail with the problem of stabilization. In chapter II the basic principles of stabilization are set out, with descriptions of the inactivation of enzymes, the destruction of micro-organisms and the control of other harmful constituents. This information is supplemented by a consideration of stabilization methods, the stability of the constituents of bran, and the effects of the different treatments on the composition and properties of the by-product.

A chapter is devoted to the morphology, anatomy, histology and histochemistry of the discrete particles that make up commercial rice bran. Special attention is paid to the rice germ.

In dealing with the control of rice bran production, the work systematically analyses the methods for measuring the degree of milling and their application to process control in the husk-removing, whitening, polishing and grading stages. Another chapter of the publication is devoted to the storage of rice bran. It describes the different types of deterioration, analyses the causes and factors that determine them, and discusses the changes that take place in the chemical composition and properties of the by-product.

Chapter V is devoted to the specific technology for stabilization. It reviews physical and chemical methods of stabilization, paying particular attention to processes using heat, and it describes and critically examines the processes, with their many variants and diverse conditions, and the corresponding machinery. The chapter ends with an analysis of the criteria for evaluating stabilization methods.

The publication includes a large number of tables, graphs, diagrams and photographs and some photomicrographic material to contribute to the understanding of the information presented. The wealth of carefully chosen bibliographical references reinforce the facts and opinions in the text and provide sources of additional information.

## SOMMAIRE

La production annuelle de son de riz avoisine 40 millions de tonnes. Sur ce total, l'huile comestible représente de 6 à 8 millions de tonnes et les protéines de qualité une quantité équivalente; on trouve aussi une forte proportion de vitamines, d'oligo-éléments et d'autres éléments nutritifs. En dépit de ces atouts, le son de riz est aujourd'hui un sous-produit insuffisamment utilisé.

Il existe une grande quantité d'informations sur le son de riz, mais celles-ci sont éparpillées et difficiles à obtenir; elles sont quelquefois confuses, quelquefois imprécises et presque toujours incomplètes. Il serait donc utile pour les enseignants, les chercheurs et les industriels de disposer d'une publication qui rassemblerait, classerait et analyserait de manière critique toute l'information disponible sur les techniques utilisées une fois le riz récolté et, plus particulièrement, sur l'ensemble des problèmes relatifs à un sous-produit aussi précieux que le son de riz.

L'Organisation des Nations Unies pour le développement industriel, consciente de l'importance du sujet et de la nécessité d'une source exhaustive d'informations, a donc décidé de publier le présent volume, qui rassemble et analyse les données existantes sur le sujet et procède à cette occasion à un examen rationnel et systématique des fondements scientifiques et techniques de la transformation industrielle du son.

Les six chapitres du présent ouvrage examinent la production du son, les principes de base de la stabilisation, les caractéristiques de ce sous-produit, la maîtrise du procédé, les techniques de stabilisation et les problèmes de conservation et de stockage.

La production du son est envisagée en amont : analyse du grain de riz et examen du processus de fabrication, l'attention s'attachant toutefois au sousproduit — le son — et non au produit primaire — le riz. Pour connaître le grain dans sa fonction de matière première pour la production de son, il faut en comprendre la morphologie, la structure microscopique et la composition chimique. L'ouvrage examine dans sa totalité le procédé de fabrication du son : nettoyage, décorticage, séparation du riz d'avec les balles et le paddy, blanchiment, polissage, ramassage des brisures de son et extraction des germes. Différents types de plants sont examinés à l'aide de nombreux diagrammes qui rendent compte des techniques — depuis la plus simple jusqu'à la plus élaborée — utilisées dans la riziculture.

Deux chapitres exhaustifs et détaillés sont consacrés au problème de la stabilisation. Le chapitre II en présente les principes de base, décrivant l'inactivation des enzymes, la destruction des micro-organismes et la maîtrise des autres composants nocifs. Cette information est complétée par l'étude des méthodes de stabilisation, la stabilité des éléments constitutifs du son et les effets des divers traitements sur la composition et les propriétés du sousproduit.

Un autre chapitre porte sur la morphologie, l'anatomie, l'histologie et l'histochimie des particules discrètes qui constituent le son de riz commercial. Le germe de riz retient tout particulièrement l'attention.

En ce qui concerne la maîtrise de la production du son de riz, l'ouvrage analyse systématiquement les méthodes permettant de mesurer le degré d'usinage et l'application de ces méthodes au contrôle des procédés, aux diverses étapes du décorticage, du blanchiment, du polissage et du classement par qualité.

Un autre chapitre est consacré au stockage du sous-produit. Il décrit les différents types de détérioration, en analyse les causes et facteurs déterminants et examine les modifications intervenues dans la composition et les propriétés chimiques du sous-produit.

Le chapitre V est tout entier consacré à la technologie spécifique de stabilisation. Il passe en revue les méthodes physiques et chimiques de stabilisation, en insistant tout particulièrement sur les procédés faisant appel à la chaleur, et il fait une description et une analyse critique de ces procédés avec leurs nombreuses variantes et la diversité des conditions dans lesquelles ils sont employés, ainsi que des machines correspondantes. Le chapitre se termine par une analyse des critères d'évaluation des méthodes de stabilisation.

L'ouvrage comporte un grand nombre de tableaux, graphiques, diagrammes et photos, ainsi que du matériel photomicrographique qui permet de mieux comprendre l'information présentée. D'abondantes références bibliographiques, soigneusement sélectionnées, viennent étayer les faits et opinions consignés dans le texte et représentent une source d'informations complémentaires.

## EXTRACTO

La producción anual de salvado de arroz se aproxima a los 40 millones de toneladas. De ellas, de 6 a 8 millones corresponden a aceite comestible, y otras tantas a proteínas de buena calidad, así como a una elevada proporción de vitaminas, oligoelementos y otros nutrientes. A pesar de ello, el salvado es, en la actualidad, un subproducto insuficientemente utilizado.

Existe mucha información sobre el salvado de arroz, pero está muy dispersa y es dificil de obtener, en algunos casos, confusa, en otros, incorrecta y, en general, fragmentaria. Una publicación que recoja, ordene y analice criticamente la información, será, por tanto, útil para los educadores, investigadores e industriales interesados en la tecnología postcosecha del arroz y, en particular, en la problemática de ese valioso subproducto que es el salvado.

La Organización de las Naciones Unidas para el Desarrollo Industrial, consciente de la importancia del tema y de la necesidad de contar con una fuente global de información, ha editado el presente volumen, en el que al mismo tiempo se reúne y analiza la información existente sobre el tema y se presentan y discuten, de forma sistemática y razonada, las bases científicas y técnicas de la industrialización del salvado.

La publicación incluye, a lo largo de sus seis capitulos, la producción del salvado, los principios fundamentales de la estabilización, las características del subproducto, el control del proceso, la tecnología de la estabilización, y la problemática de su conservación y almacenamiento.

El tema de la producción de salvado se aborda desde su origen, analizándose el grano de arroz y examinándose el proceso de elaboración, con la atención siempre centrada en la obtención del subproducto, el salvado, antes que en la del producto primario, el arroz. El conocimiento del grano como materia prima para la producción de salvado incluye la morfología, la estructura microscópica y la composición química. Se examina todo el proceso de la producción de salvado: limpia, descascarillado, separación de la cascarilla, separación del palay, blanqueo, pulido, recolección de las fracciones de salvado y separación del germen. Se examinan diversos tipos de plantas con ayuda de numerosos diagramas sobre las tecnologías, desde las más simples a las más complejas, utilizadas en todo el cinturón arrocero.

Dos capítulos tratan, con amplitud y detalle, del problema de la estabilización. En el capítulo II se exponen los principios básicos de la estabilización, describiéndose la inactivación de enzimas, la destrucción de microorganismos y la lucha contra otros componentes perjudiciales. Esta información se complementa con un análisis de los métodos de estabilización, la estabilidad de los componentes del salvado y los efectos de los distintos tratamientos sobre la composición y las propiedades del subproducto.

Un capítulo está dedicado a la morfología, anatomía, histología e histoquímica de las partículas discretas que constituyen el salvado comercial. Se concede especial atención al germen de arroz.

Al tratar del control de la producción del salvado de arroz la obra analiza sistemáticamente los métodos de medida del grado de elaboración y su aplicación al control del proceso en las etapas de descascarillado, blanqueo, pulido y clasificación. Otro capítulo de la publicación está dedicado al almacenamiento del salvado. En él se describen los distintos tipos de deterioro, se analizan las causas y factores que los determinan, y se estudian los cambios que se producen en la composición química y en las propiedades del subproducto.

El capítulo V está dedicado a la tecnología específica de la estabilización. Se revisan los métodos físicos y químicos de estabilización, dedicándose especial atención a los procedimientos térmicos y se describen y examinan críticamente los procesos, en sus múltiples variantes y condiciones, así como la maquinaria correspondiente. Cierra este capítulo el análisis de los criterios de evaluación de los métodos de estabilización.

La publicación incluye gran número de tablas, gráficos, esquemas, fotografías y material fotomicrográfico, que contribuyen a la comprensión de la información presentada. Las referencias bibliográficas, cuidadosamente seleccionadas y muy abundantes, respaldan los conceptos y opiniones expresados en el texto y proporcionan fuentes de información adicional.

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RICE BRAN: AN UNDER-UTILIZED RAW MATERIAL

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UNITED NATIONS INDUSTRIAL DEVELOPMENT ORGANIZATION Vienna

14421

## RICE BRAN: AN UNDER-UTILIZED RAW MATERIAL





UNITED NATIONS New York, 1985 The designations employed and the presentation of the material in this publication do not imply the expression of any opinion whatsoever on the part of the Secretariat of the United Nations concerning the legal status of any country, territory, city or area or of its authorities, cr concerning the delimitation of its frontiers or boundaries.

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## Preface

Rice is undoubtedly one of the world's basic human food items. It is by far the most important cereal food in South-East Asia and the Far East and is fast gaining in popularity in Africa and Latin America.

Rice, however, differs markedly in structure from other common cereals, and specialized processing methods are required in order to make it edible. Paddy is thus the raw material for an important branch of the food-processing industry, namely the rice-milling industry. In an efficiently run mill, the production of white rice will go hand in hand with the production of its by-product—rice bran. So far, however, rice bran, with its rich oil, protein, mineral and vitamin content, has not been given the attention it deserves as an extremely valuable secondary raw material for the production of vegetable oil and protein human food or animal feed.

Many national and international research institutes have been working to develop the production and processing of rice bran and thus pave the way for efficient industrial processing operations. As a result of their efforts, and certainly also in view of the fact that the vegetable oil industry in developing countries is suffering from a severe shortage of raw materials, rice bran appears to be well on its way to becoming a recognized raw material for the production of vegetable oil and protein animal feed. Nevertheless, a great deal of work will still have to be done in this area before the food-processing industries can extract the full value from this by-product, thereby enabling it to make an effective contribution to human food supplies, particularly in developing countries.

The present publication has been prepared by Dr. S. Barber and Dr. C. Benedito de Barber, of the Instituto de Agroquímica y Tecnología de Alimentos, Valencia, Spain. It is intended to provide all those interested in rice-bran production and processing operations with detailed and comprehensive information on rice bran as a by-product and as potential raw material. The United Nations Industrial Development Organization (UNIDO) hopes that the publication will provide answers to many of the problems besetting the development of this useful by-product.

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#### **ABBREVIATIONS**

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Besides the common abbreviations, symbols and terms and those accepted by the International System of Units (SI), the following have been used.

#### Technical abbreviations and symbols

CBB	coloured bran balance
CEM	carbon dioxide exchange method
FDNB	fluorodinitrobenzene
FFA	free fatty acids
HgBPB	mercuric chloride - bromophenol blue
KIST	Korean Institute of Science and Technology
KS	Klett-Summerson
NPU	net protein utilization
PAS	periodic acid reagent
PER	protein efficiency ratio
psi	pounds per square inch (1 psi = 0.069 bar)
RHE	relative humidity at equilibrium
rpm	revolutions per minute
sp/g	spores per gram
TBA	tiobarbituric acid
TDT	thermal destruction time

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#### Organizations

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AOCS	American Oil Chemists Society
CFTRI	Central Food Technological Research Institute
IATA	Institute of Agrochemistry and Food Technology
PPRC	Paddy Processing Percent Communication
PPRC	Paddy Processing Research Centre

#### CONTENTS

.

•

.

Π.

2

		Page
Pref	Face	v
Abt	previations	vi
Chap	Dier	
I.	THE RICE GRAIN, MILLING AND THE PRODUCTION OF BRAN	I
	The rice grain: raw material for the production of bran	1
	Anatomy of the rice grain Histology of the rice grain Chemical composition of the rice grain	1 4 13
	The milling process and the production of bran	15
	Processing Mills and the type of equipment used	16 31
II.	BASIC PRINCIPLES OF THE STABILIZATION OF RICE BRAN	50
	Value and end-purpose of stabilization Stabilization and post-stabilization Coexistence of valuable and harmful components	50 50 50
	Destruction of micro-organisms	51
	Thermal destruction	51 57
	Inactivation of enzymes	60
	Fundamental principlesDenaturing (or inactivating) agentsInactivation of enzymes by heatEffects of water activity on enzymatic activityEffects of pH on enzymatic activity and stabilityOther thermal inactivating agents	60 61 69 71 74
	Control of other harmful constituents of bran during stabilization	75
	Loss of valuable constituents of bran during stabilization Means of stabilizing bran and stability of the constituents Effects of various means of stabilization on the valuable constituents	76 76 77
III.	MORPHOLOGY, ANATOMY, HISTOLOGY AND HISTO- CHEMISTRY ()F THE DISCRETE PARTICLES IN COMMER- CIAL RICE BRAN	83
	Introduction	83

ý

		Page
	Simple discrete particles in commercial bran	85
	Fragments of lemma and palea	85
	Fragments of sterile glumes	88
	Fragments of the pedicel	88 89
	Fragments of the pericarp Fragments of the starchy endosperm	89 89
	Germ	92
	Fibres	114
	Compound discrete particles in commercial bran	114
	Fragments of the pericarp with the seed-coat	114
	Fragments of the seed-coat with the aleurone layer	117
	Other types of compound particles	118
IV.	CONTROLLING THE PRODUCTION OF RICE BRAN	123
	Control of processing	123
	Methods for measuring the degree of milling of rice	124
	Control of bran production	127
	Rice bran, rice fractions and related by-products: terminology and	
	definitions	131
	Factors determining the properties of bran	135
	Rice-related factors	137
	Factors related to the milling process	146
V.	TECHNOLOGY FOR THE STABILIZATION OF RICE BRAN	157
	Introduction	157
	Stabilization methods	157
	Chemical stabilization	157
	Physical stabilization	161
	Heat stabilization: a widely favoured technique	163
	Criteria for the evaluation of stabilization methods	202
	Criteria for evaluating the stability of treated bran	203
	Criteria for evaluating the loss of valuable properties in the bran	207
	during stabilization	207
VI.	THE STORAGE OF RICE BRAN	216
	Storage in practice	216
	Factors that influence changes in the bran	216
	The role of water: sorption isotherms	217
	Temperature	221
	Other factors	222
	Causes of changes in the bran	222
	Chemical reactions	223
	Enzymatic reactions	223
	Biological changes	226

and the second

٨

.. .

.

## vlii

	Page
Changes in the composition and properties of bran during storage	230
Colour, taste and odour	
Average chemical composition	232
Composition and characteristics of the chemical constituents of	
the bran	235

#### Tables

Chapt	er I	
1.	Dimensions of rice grains of commercial varieties	3
2.	Anatomical distribution of mass in the rice grain	3
3.	Chemical composition of rice and changes occurring during processing	15
4.	Separation and manual recovery of germ in a germ-removing machine	41
Chap	ter II	
1.	Resistance of various biological units to ionizing radiation	59
2.	Analytical methods for determining changes in oils and fats	78
Chap	ter III	
1.	Types of discrete particles identified in commercial bran, and their	85
•	anatomical composition Anatomical distribution of the rice caryopsis	92
2.	Distribution of organs and structures in histological areas of the rice germ	100
3.	Distribution of organs and structures in instological areas of the free germ	100
Chap	nter IV	
1.	Amount of bran removed from rice during milling, by countries	123
2.	Types of bran and fractions: constituents and English and Spanish	
	terminology	131
3.	Size, shape and weight of commercial brown rice grains from the United	137
	States of America	157
4.		140
	caryposis	140
5.	points on the caryopsis	140
ó.	Removal of the germ during the whitening process	143
0. 7.	Effects of parboiling on the weight of the germ and its oil content	144
8.	Effects of parboiling on the oil content of bran	146
9.	Composition of the bran produced in a huller-type mill and in a cone-type	
	mill	147
10.	Average composition of bran from different types of hulling machines	148
11.	Effect of different types of hulling machines on the oil content of bran from	
	whitening machines	149
12.	Particle-size distribution of different types of bran from whitening machines	150
13.	Average composition of bran from both friction-type and abrasion-type	1.60
	whitening machines	150
14.	Effect of brokens on the oil content of commercial bran from an abrasive	151
	cone mill	121

.

F.

ix

#### Chapter V

.

F.

.

Chaj	Dter V	Page
I. 2.	Testing of chemical compounds for the stabilization of rice bran Different types of heat treatment for the stabilization of bran without the	158
3.	addition of water	164
4.	addition of water Effects produced on the colour of the oil by treating the bran in the manual	167
5.	stabilizer Extrusion cookers: effects of temperature and retention-time on the	170
6.	moisture content and peroxidase activity of rice bran Production capacity, treatment temperature and consumption of energy during the extrusion of rice bran	182
7.	during the extrusion of rice bran	185 189
8.	Recommended conditions for the stabilization of rice bran by direct steam treatment in a fixed bed	191
9.	Stabilization of rice bran by direct steam treatment in a moving bed	191
10.	Stabilization of bran using an industrial prototype of the IATA stabilizer	200
Chap	ter VI	
Ι.	Mould flora capable of producing toxic metabolites in bran	228
2.	Same mycotoxins produced by moulds and their pathological effects	228
3.	Effects of storage on the sensory characteristics of raw rice bran	231
4.	Effects of storage on yield and colour of the oil extracted from the bran by pressing	
5.	Sugar and moisture content of rice bran before and after storage	235
6.	Effect of moisture on the rate of formation of FFA during storage of rice	235
7.	bran at 35° C Changes in mould and microbial count and FFA content during the storage of bran treated for two hours in an autoclave at 121° C and dried for one	237
8.	hour at 60° C Changes in the mould count and FFA content of non-inoculated sterilized bran and bran inoculated with Aspergillus chevalieri during storage at	240
9.	different levels of moisture content	242
	bran	243
10.	Change in relative proportions of FFA in bran during storage	24 <i>5</i> 245
11.	Results of sampling carried out in Spanish rice mills to detect the possible presence of aflatoxins in bran and husk	2.0
	Provence of analoxing in orall and nusk	248

2

#### Figures

#### Chapter I

1.	Rice grain (paddy rice, rough rice)	2
2.	Transverse section of the lemma of the rice grain	4
3.	Histological transverse section of the rice husk	5
4.	Trichome of the rice husk	6
5.	Section of rice bran	7
6.	Histology of the pericarp of the rice grain	é
7.	Section of rice grain, showing the aleurone layer	10
8.	Detail of the outer convering of the rice grain	12

9.	Longitudinal section of the rice germ
10.	Processing in a European rice mill
11.	Scalping machine
12.	Cleaning sieve
13.	Disc husker
14.	Rubber-roll husker
15.	Husk separator with "plansifter"
16.	Double-action husk separator
17.	Paddy separator with compartments
18.	Paddy tray separator with husker
19.	Schematic representation of the whitening process
20.	Whitening machine
21.	Abrasive and friction whitening machines
22.	Huller-type mill
23.	Huller-type mill, modernized
24.	Monobloc unit for husking and whitening rice
25.	Monobloc unit with pre-cleaner, bucket elevator and drive unit
26.	Modernized mill, including a huller-type unit
27.	Diagram of a rice-processing system
28.	Diagram of a rice-processing system, including germ separator
29.	Process for separating the germ from the rice bran
30.	Diagram of a germ separator
31.	Laboratory device for separating the germ from the rice bran
32.	Alternative systems of rice processing and their effect on the type of bran produced

D .....

## Chapter II

~

-

.

1.	Representation, on a linear scale, of changes in the number of surviving micro-organisms after heat treatment
2.	Semilogarithmic plot showing the number of micro-organisms surviving heat treatment as a function of time
3.	Semilogarithmic plot of the survival of micro-organisms in a sample with natural heterogeneous microflora
4.	Thermal destruction time plotted against temperature
5.	Effect of relative humidity on the inactivation of <i>Bacillus subtilis</i> var. niger at 125° C
6.	Plots showing the destruction of micro-organisms by ionizing radiation
7.	Effect of heat on the stability of rice bran lipases
8.	Semilogarithmic plots showing the relationship between thermal inactivation of various enzymes and temperature
9.	Relationship between time and temperature in the inactivation of lipase and peroxidase in milk
10.	Thermal inactivation of a microbiological lipase ( <i>Pseudomonas fluorescens</i> ) and the thermostable fraction of potato peroxidase as a function of
	temperature
11.	Thermal inactivation of rice bran peroxidase: effect of moisture content of the bran
12.	Effect of stabilization conditions (moisture content, time and temperature) on rice bran peroxidase activity
13.	Effect of stabilization conditions (moisture content, time and temperature)
13.	on rice bran lipase activity
14.	Thermal inactivation of rice bran peroxidase: effect of pH
177.	

		Tuge
15.	Effect of initial enzymatic activity on the efficacy of the stabilization process, at 12 per cent relative humidity and a temperature of 110° C	70
16.	Relationship between the regeneration of turnip peroxidase and rate of heating during inactivation	71
17.	Rate of enzymatic hydrolysis of lecithin in a mixture of crushed barley-malt	
	with 2 per cent lecithin, stored at 30° C at different levels of water activity	72
18.	Effect of pH on enzymatic activity	72
19.	Effect of pH on the stability of rice bran lipase	73
<b>20</b> .	Rate of inactivation of soya bean lipoxidase at pH 4 and pH 7	74
21.	Effect of steam treatment on the protein efficiency and trypsin inhibitor	
	activity of raw soya meal	75
22.	Effects of heat on the trypsin inhibitor activity of rice bran as a function of	
	treatment time and moisture content of the bran	76
23.	Paths of loss of nutrients due to oxidative changes in lipids	78
Chap	oter III	
Ι.	Particles in commercial rice bran showing heterogeneous composition	84
2.	Transverse section of the lemma	86
3.	Transverse section of palea	87
4.	Section of a pericarp particle	90
5.	Transverse section of an endosperm particle	91
6.	Histological preparation of commercial rice bran, showing the presence of	
	various fragments of germ (Ge)	93
7.	Transverse section of the rice germ	94
8.	Transverse section through the plumule of the rice germ	95
9.	Histological preparation of commercial rice bran, showing a fragment of	
	plumule	96
10.	Longitudinal section of the rice germ, showing plumule and radicle	97
11.	Transverse section of the radicle of the rice germ	99
12.	Longitudinal section of the epithelium of the rice germ	102
13.	Detail of a longitudinal section of the rice germ, showing the epidermis of	
	the coleorhiza and the outer coverings	103
14.	Detail of longitudinal sections of the rice germ, showing the connecting	
	zone (suspensor) between the aleurone and coleorhiza	104
15.	Longitudinal section of the rice germ	107
16.	Longitudinal section of the rice germ	109
17.	Transverse section of the embryonic axis of the rice germ	110
18.	Outer coverings of the rice germ	111
19.	Longitudinal section of the rice germ, showing the outer coverings	112
20.	Longitudinal section of the rice germ	113
21.	Longitudinal section of the rice germ	115
22.	Section of a compound particle in commercial rice bran	116
23.	Histological preparation of commercial rice bran, showing various compound	
	particles	119

## Chapter IV

۱.	Samples taken from the same lot of rice, milled to different degrees and	
	stained with the May-Grünwald reagent	126
2.	Variation in the surface fat content of rice milled in different whitening	
	machines, expressed as a function of the degree of milling	128
3.	Histograms of the intensity of pigmentation of residual bran in individual	
	rice grains, at different degrees of milling	130

Page

ļ

		Page
4.	Uniformity of the milling of rice at the end of successive stages in the whitening process	132
5.	Histogram representing percentage of sections with denuded bran and	
	aleurone layers at different degrees of polish	138
6.	Projections produced by milling	139
7.	Resistance to abrasion during the milling of different varieties and lots of	
	brown rice	141
8.	Changes in abrasion resistance during the storage period of brown rice	
	milled in a differential mill	142
9.	Parts of the rice caryopsis that are separated as bran during the whitening	
2.	process	143
10.	Oil content of successive fractions of bran extracted from raw and	
	parboiled rice	i45

#### Chapter V

.

R.

1.	Changes in FFA content during the storage of bran treated for two hours at 110° C	166
2.	Manually operated stabilizer	169
3.	Stabilizer using indirect heat from gases produced by the combustion of	
	husk	171
4.	Flow diagram of a stabilization process using indirect heat from gases	
	produced by the combustion of husk	172
5.	Bran inactivator combined with cooler	173
6.	Stabilizer from the Central Food Technological Research Institute (Mysore)	174
7.	Hot air stabilizer combined with husk burner	176
8.	Bran stabilizer	1/8
9.	Pneumatic conveyor dryer	178
10.	Stirred fluidized bed device for the stabilization of rice bran	179
11.	Fluidized bed stabilizer with combined phases	181
12.	Low-cost extrusion cooker: temperature profile	183
13.	Extrusion cooker: moisture losses of rice bran at different temperatures	184
14.	Extrusion cooker: inactivation of peroxidase at different temperatures	187
15.	Direct steam stabilizer, with dryer and cooler	194
16.	Bran stabilization unit being assembled	195
17.	Alternative type of stabilizer	196
18.	Stabilizer developed by the Instituto de Agroquímica y Tecnología de Alimentos (IATA), using direct steam in a fluidized bed, with dryer and cooler	<u>197</u>
19.	IATA stabilizer	198
20.	Stabilization of rice bran using an industrial prototype: retention and	
	nominal dwell-time as a function of the rate of bran-flow	199
21.	Extrusion cooker using direct injection of steam and hot water	201
22.	Experimental extrusion cooker using direct steam injection	202
Chap	oter VI	
1.	Hydrogen bonds between water and various functional groups	217
2.	Typical sorption and desorption isotherms for rice bran (hypothetical)	218
3.	Sorption curve showing the presence of solvent water	219
4.	Desorption isotherms for wheat at different temperatures	220
5.	Hydrolysis of glycerides catalysed by lipase	223
6.	Reaction rates in foodstuffs as a function of water activity	224
7.	Decomposition of hydroperoxides resulting from the oxidation of lipids	225

		Page
8.	Effect of temperature and relative humidity on certain species of mould	226
9.	General limits of temperature and relative humidity for the multiplication	
	of biological agencies	227
10.	Profile of volatile compounds from rice bran shortly after production,	
	stored for 50 days at -20° C and stored at +25° C	233
11.	Effects of storage on the oil content of raw and parboiled rice bran	234
12.	Effects of moisture on the formation of FFA during storage of rice bran at	
	30° C	236
13.	Effects of storage at 25° C and at different levels of relative humidity on	
	the formation of FFA in rice bran	237
14.	Effects of storage at different temperatures on the formation of FFA in rice	
	bran	238
15.	Formation of FFA in neutral rice-bran oil after the addition of bran	239
16.	Variation in FFA during the storage of bran from the first and second	
	cones	240
17.	Changes in FFA content and insect infestation of bran during storage	243
18.	Development of FFA in bran from the first and second whitener cones	
	during storage	244
19.	Development of acidity in rice bran and germ during storage at 30° C	245

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# I. The rice grain, milling and the production of bran

#### The rice grain: raw material for the production of bran

#### Anatomy of the rice grain

The rice grain (paddy) (see figure 1) is composed of the caryopsis (dehusked or brown rice) and associated struc ares—lemma, palea, rachilla, sterile lemmas and arista [1], which together make up the husk. The rachilla is found at the lower extremity and is separated from the pedicel by an abscission layer; on top of the rachilla there are two sterile lemmas ("sterile glumes", "external glumes"), with two flowering glumes, the lemma and the palea, which envelop the caryopsis. The two sterile glumes, which may differ in length and shape, are shorter than the lemma and the palea. The lemma, which is located on the ventral side of the caryopsis, covering the germ, has five vascular bundles. It is longer, wider and of more uniform thickness than the palea (about 0.1 mm), but slightly smaller. The palea, which is on the dorsal side of the caryopsis, has three vascular bundles. At its edges it has a lodicule, ensuring hermetic closing with the lemma [2]. Both the lemma and the palea terminate in an extending tip or apiculus, with a filiform extension—the arista, awn or beard—in the case of the lemma. The external surface of the lemma and palea is covered with numerous trichomes, but hairless varieties have been obtained, with very few or no trichomes [2, 3]. There is a space between the husk and the grain, varying in width from one variety to another, which may partly explain why husking is more difficult in some cases than it is in others [4].

During the threshing process, the grain normally becomes separated from the pedicel at the abscission layer. In some varieties, however, the separation can occur when the pedicel itself is broken [1]. When the grain is husked, the caryopsis is uncovered. It has elongated or rounded oval-shaped fruit, largely composed of seed [5], with ridges on the surface, corresponding to the ribs of the lemma and palea [1]. The external layer of the caryopsis is composed of the pericarp, which envelops and protects the seed, to which it is closely attached. Immediately below the pericarp is the seed-coat, or tegmen;<sup>1</sup> then come the aleurone layer and the starchy endosperm.

The germ is lodged in a hollow, in the lower abdominal region of the grain, adhering to the endosperm. It is covered by the aleurone layer, the seed-coat, the pericarp and, finally, the lemma.

<sup>&</sup>lt;sup>1</sup>The tegmen is often wrongly referred to as the testa. The testa is derived from the internal teguments of the ovule, which is destroyed before the caryopsis matures. The tegmen represents the internal cellular layers of the internal teguments of the ovule [1].

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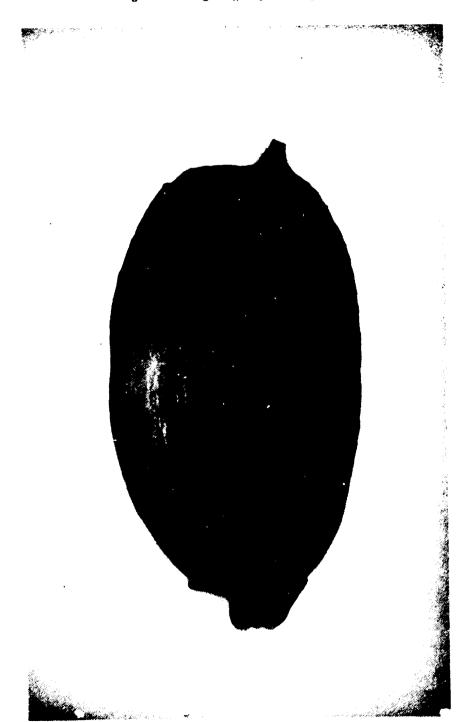


Figure 1. Rice grain (paddy rice, rough rice)

The morphology of the grain, together with other characteristics (geographical distribution, morphology of the plant, sterility of hybrids and serological reaction), has served as a basis for classifying cultivated rice into three subspecies: *indica, japonica* and *javanica*. The grain of the first is thin and rather flattened, that of the second short and rounded, and that of the last wide and thick [1].

There are marked differences in the length and shape (ratio of length to width) of cultivated rice grain, with or without the husk or processed (see table 1). The differences have made it possible to establish grades of rice based on these characteristics, which have been incorporated into the legislation regulating the rice market.

In cultivated rice the proportions of the various anatomical parts of the grain also differ (see table 2).

## TABLE I. DIMENSIONS OF RICE GRAINS OF COMMERCIAL VARIETIES<sup>a</sup>

(Millimetres)

Type of rice		Length	92/id:h	Thickness
Milled:	long grain	6.7-7.0	1.9-2.0	i.5-1.7
	medium grain	5.5-5.8	2.4-2.7	1.7-1.8
	short grain	5.2-5.4	2.7-3.1	1.9-2.0
Brown:	long grain	7.0-7.5	2.0-2.1	1.5-1.8
	medium grain	5.9-5.1	2.5-2.8	1.8-2.0
	short grain	5.4-5.5	2.8-3.0	2.0-2.1
Paddy:	long grain	8.9-9.6	2.3-2.5	1.8-1.9
-	medium grain	7.9-8.2	3.0-3.2	1.9-2.1
	short grain	7.4-7.5	3.1-3.6	2.1-2.3

Source: Webb [6].

<sup>4</sup>Bhattacharya (1974) [7] ...as reported more extreme values: lengths of 10.84 mm for p ddy and 3.99 mm for milled rice; widths of 2.28 nm for paddy and 1.71 mm for milled rice; thicknesses of 1.59 mm for paddy and 1.43 mm for milled rice.

## TABLE 2. ANATOMICAL DISTRIBUTION OF MASS IN THE RICE GRAIN

Anatomical part	Relative mass (percentage)
Husk	17-22 <sup>a</sup>
Pericarp	17-22 <sup>a</sup> 1-2 <sup>b</sup>
Aleurone and seed-coat	4-6 <sup>b</sup> 89-94 <sup>b</sup> 2-3 <sup>b</sup>
Starchy endosperm	89-94 <sup>b</sup>
Germ	2-3 <sup>b</sup>

<sup>a</sup>Most usual value ranges. <sup>b</sup>Juliano [8].

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#### Histology of the rice grain<sup>2</sup>

#### Husk

The histological structure of the two flowering glumes is similar, but there are some differences; for example, the region of the principal rib is rather thicker and the epidermic cells are slightly smaller in the palea than in the lemma. The average thickness is about 120  $\mu$ m [2].

In a cross-section of either of the two flowering glumes, four layers of tissue can be seen: the outer epidermis, the sclerenchyma, the parenchyma and the inner epidermis (see figure 2) [2, 3]. The outer epidermis has a sinuous and ridgy surface formed by radially lengthened epithelial cells with thick walls,

Figure 2. Transverse section of the lemma of the rice grain (Magnification 140×; stained with safranin-phenol)



<sup>2</sup>The dimensions given in this section may be influenced by the specimen preparation techniques, and this fact should be taken into account.

wrinkled, with deep recesses and arranged axially in rows. In histological preparations of samples steeped in nitric acid and potassium chlorate, the epithelial cells of the palea have been found to vary in length from 30 to 80  $\mu$ m and in width from 20 to 100  $\mu$ m, and those of the lemma have varied in length from 60 to 90  $\mu$ m and in width from 50 to 120  $\mu$ m [2]. The trichomes are evenly distributed between these cells. They stick out through the cutin-silica layer and penetrate inside as far as the sclerenchyma (see figure 3). The trichomes (see figure 4) are conical, transparent, colourless, hollow and unicellular, their size depending on the variety of rice and the position of the trichomes in the grain (those in the dorsal region are longer than those on the lateral surfaces). Their walls are thicker at the base and as they age the wall thickens and the cavity decreases in size [2]. The sclerenchyma is formed by two or three layers of cells (measuring 150-600  $\mu$ m in the histological preparations of steeped samples referred to above), with very thick (4-6  $\mu$ m) angular walls. The parenchymatous layer varies in thickness and is composed of from two to four layers of spongy cells [9]. These are of two types, either tangentially elongated, with thick and sinuous walls, or short, with thin walls [2]. They are 20-30  $\mu$ m long and 10-15  $\mu$ m wide [9]. The parenchymatous cells include tubular and cross cells [3, 10]. The vascular bundles lie between the sclerenchymatous and parenchymatous layers. Those of the palea, especially the central vascular bundle, are more differentiated than those of the lemma. The inner epidermis, which is 6-12  $\mu$ m thick [9], is made up of some three layers of isodiametric cells, with very thin walls and empty lumen; these layers are tightly compressed, and therefore difficult to differentiate [2, 10].

## Figure 3. Histological transverse section of the rice husk (Magnification 400×)



Key:

- (1) Cuticle
- (2) Apex of siliceous covering
- (3) Longitudinal siliceous bands
- (4) Outer epidermis
- (5) Branched scierenchymatous cells

Source: Angladette [10].

(6) Sclerenchymatous fibres

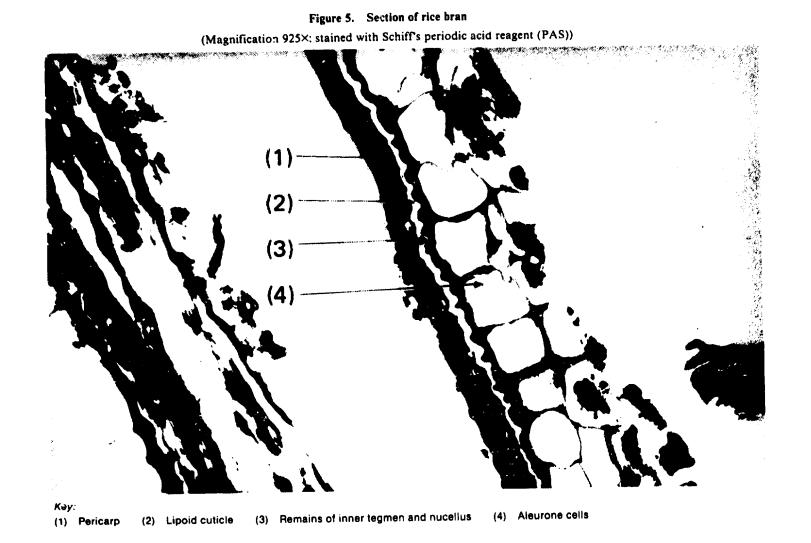
- (7) Tubular cells
- (8) Longitudinal cells
- (9) Inner epidermis
- (10) Attached hair (trichome)

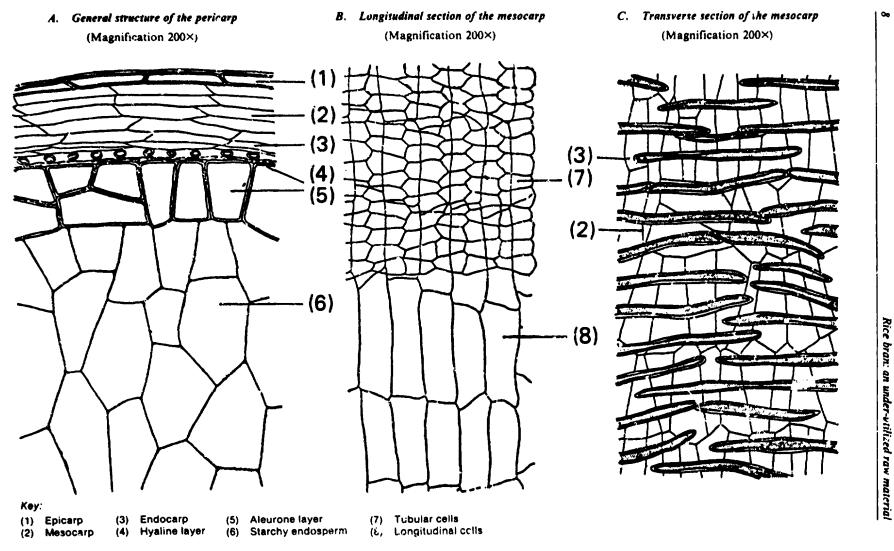
Figure 4. Trichome of the rice husk (Magnification 800×; stained with safranin-phenol)

The sterile glumes have a simpler histological structure than the flowering glumes. They are boat-shaped, pointed at the front and U-shaped at the other end, where they touch the rachilla. The length along the rib can measure from 3.35 to 1.75 mm [9]. In the transverse section the sterile glumes consist of an outer epidermal cellular layer and another inner one, formed by rectangular cells, tangentially elongated, with thick walls. Between these layers are two or more layers of parenchymatous cells; some are elongated, with thick and corrugated walls, while others are short, with thin walls; in the rib region, the surface shows little differentiation [2].

#### Covering structures of the caryopsis

The covering of the caryopsis is made up of three layers: the pericarp, the seed-coat or tegmen and the nucellus (see figure 5). The tissues of the pericarp can be very tightly compressed, making it difficult to distinguish their characteristic component layers. In some varieties they [8, 11] can be seen to consist of several cellular layers (see figure 6). Because of their different morphological characteristics they may be subdivided into epidermal, hypo-





#### Figure 6. Histology of the pericarp of the rice grain

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Source: Angladette [10].

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dermal, transversal and tubular cells. The epidermis consists of a layer of cells, transversally elongated, with ridgy walls, on which there is a cuticle. Sizes have been recorded from  $5 \times 3 \ \mu m$  to  $20 \times 10 \ \mu m$  [9]. The hypodermis is formed by several layers of parenchymatous cells, partially flattened with straight walls and transversely elongated, whose dimensions range between  $5 \times 12 \ \mu m$  and  $15 \times 80 \ \mu m$  (see figure 6). In a longitudinal section, from two to five layers of transverse or crossed cells can be seen, elongated perpendicularly to the major axis of the grain, some 6  $\mu$ m wide and 1-2  $\mu$ m thick; in a transverse section they look like puncta (see figure 6). These cells are very numerous and parenchyma and transverse cells combined produce a tissue that looks somewhat like a grating. The innermost layer is made up of tubular cells. They are elongated and approximately cylindrical in shape, with a diameter of 3-5  $\mu$ m. The pericarp of two varieties of rice (Oryza sativa cv. Caloro and Oryza sativa cv. Labelle) has been measured recently [12] and shown to be about 10  $\mu$ m thick. Immediately below the pericarp is the seed-coat or tegmen, which consists of: (a) the fatty cuticle; (b) the spermoderm on the side nearest to the pericarp; and (c) the perisperm on the side of the aleurone layer. The cuticle can be 1  $\mu$ m or more thick and is sometimes discontinuous [9, 13]. The tegmen is thinner in the vicinity of the germ than it is near the endosperm; in short-grain varieties (Balilla  $\times$  Sollana) thicknesses of 0.8-1.5  $\mu$ m and 1.5-2.3  $\mu$ m, respectively, have been found [14]. The cuticle in the region of the germ is also thinner (about  $0.5 \,\mu\text{m}$ ) [12]. At the back of the seed-coat, and between it and the aleurone layer, is the nucellus, composed of a thick (0.8  $\mu$ m) outer cuticle and a layer of flattened nucellar cells, in the vicinity of the germ [12]. These empty cells, reduced to a mere cellular wall, are arranged in rows, forming a thin but strong membrane [1, 15]. In some cases, the nucellus of the caryopsis of the mature rice has not been detected, while in others it has been found to be present only as a cuticle [12, 14].

#### Aleurone layer

Botanically, the aleurone layer is part of the endosperm in which it originates (see figure 7). It is located beneath the nucellar layer, enveloping the amylaceous endosperm and the germ to which it is attached, except in the immediate area of the scutellum and the coleorhiza (see below, under "Germ"). The thickness and number of layers of cells in the aleurone varies according to the variety of the grain, the region in which it is cultivated, and the conditions under which it is grown [15, 16, 17].

In the aleurone, two different types of cells have been identified: one around the starchy endosperm and another around the germ [12, 14]. The cells of the aleurone layer surrounding the endosperm are largely polygonal; those on the ventral side are almost perfect rectangles and those on the dorsal side irregular hexagons [13, 16]. They are parenchymatous nucleate living cells [5], roughly 15-30  $\mu$ m in size, with walls of uniform thickness (about 2  $\mu$ m) [9, 13, 14]. In the cytoplasm of the cells, the presence of aleurone granules, "fatty bodies",<sup>3</sup> plastids, mitochondria, microbodies, vesiculas and endoplasmic reticules has been noted [12].

<sup>3</sup>Also known as "lipid bodies", which are merged into a larger, single body by mechanical action [12].



Figure 7. Section of rice grain, showing the aleurone layer (Magnification 765×; stained with Sudan black)

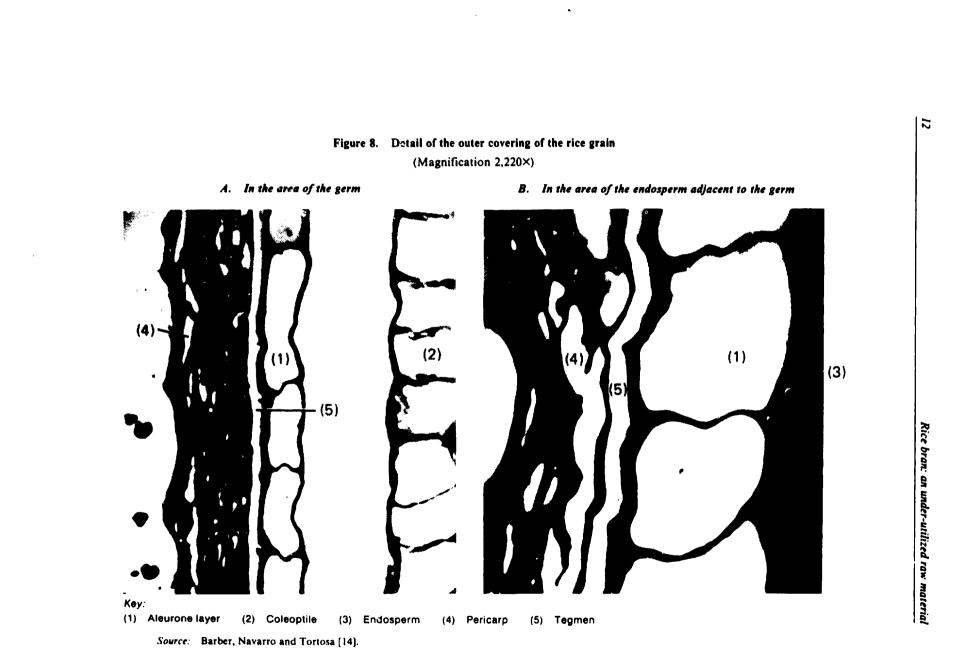
The aleurone granules are spherical, with a 1-3  $\mu$ m diameter [18, 19], and have at least two constituent parts: a particle with a high phytinic content, which is the nucleus of the protein granule; and the surrounding coat, which is composed of proteins and carbohydrates [20]. The nucleus of the granule, which it has been suggested should be called the "phytinic particle" or "phytinic body", has recently been isolated [20]. The phytinic acid accumulates in the aleurone particle [2]] or, more precisely, in the phytinic particle [20]. This has been demonstrated by using micro-autoradiography to trace the distribution of <sup>3</sup>H-myo-inositol, administered to the rice grains while they are maturing. The <sup>3</sup>H appears exclusively in the aleurone granules. The differences between the aleurone granules and protein bodies of the endosperm are particularly marked with respect to: (a) location, as the former are found in the aleusone and the latter in the endosperm; (b) composition, as the aleurone granules have a high proportion of phosphorus (12 per cent) and a low protein content (11.7 per cent), while the protein bodies contain little phosphorus (0.5 per cent) and a high proportion of protein (60 per cent); and (c) structure, since the protein bodies, when viewed through an electron microscope, are seen to be stratified while the aleurone granules have electron-dense bodies [18].

The cells of the aleurone layer surrounding the germ are much more flattened (3-5  $\mu$ m) (see figure 8A) than in the region of the endosperm (15-20  $\mu$ m) (see figure 8B), chiefly in the vicinity of the plumule [14]. The cytoplasm in the aleurone layer surrounding the germ is less dencely packed than it is in the region of the endosperm; the aleurone granules are absent, there are fewer and smaller fatty bodies and numerous vesiculae, the mitochondria and the plastids are dispersed in the cytoplasm and tiny fibres can be seen, some 0.0125  $\mu$ m thick and up to 2  $\mu$ m long. In place of the aleurone granules, other protein structures (possibly protein bodies) have been identified [12].

#### Starchy endosperm

The starchy endosperm is surrounded by the aleurone layer, except in the lower ventral portion, where it is in direct contact with the germ. It is made up of parenchymatous cells, within thin walls (measuring 0.25  $\mu$ m on average). The arrangement of the cells follows a common pattern for all varieties: small cells are arranged, like paving stones, around a central nucleus while larger cells are arranged radially. Their shape also varies according to their position in the grain. The cells of the central portion are rounded and polygonal (measuring from 45 × 50  $\mu$ m to 80 × 105  $\mu$ m). The adjacent cells are larger and isodiametric, somewhat elongated or slightly flattened radially. The cells located in the dorsal or ventral zone next to the nucleus vary in shape, especially radially [13]. It has been pointed out that cultivated rice may have 12-22 cells in the dorsal radius, 10-18 cells in the ventral radius and 10-17 cells in the lateral radius, and 103-256 cells in the longitudinal diameter [8, 22, 23].

In the amylaceous endosperm, the sub-aleurone region and the region of the central endosperm may be distinguished. The sub-aleurone region has a relatively limited number of compound starch granules that are oval in shape. The individual granules are polyhedric and very small (2-4  $\mu$ m). Minute starch granules are also found near the germ [13]. The starch granules are surrounded by protein bodies [24, 25]. These protein bodies can be spherical, with a 1-2  $\mu$ m



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diameter, dense centres and concentric rings or radial faces, spherical but smaller (0.5-0.75  $\mu$ m in diameter), without dense centres but with concentric rings or radial faces, or round, crystalline, and made up of small angular units with a 2-3.5  $\mu$ m diameter [26]. The central endosperm region is largely composed of polyhedric starch granules (5-9  $\mu$ m), bigger than those found in the sub-aleurone layer [13]. The size of the individual granules can vary considerably (0.3 to 13  $\mu$ m) [27]. The compound granules also vary in size (from 12 × 10  $\mu$ m to 20 × 10  $\mu$ m in the Bahia short grain variety) [9]. The individual starch granules are surrounded by protein material located in small sacs. Protein bodies are also found in the central portion of the endosperm; the only ones detected so far are similar to the 1-2  $\mu$ m spherical bodies found in the sub-aleurone layer [26]. There are a large number of protein bodies in the subaleurone region but few in the central portion of the caryopsis [28].

### Germ

The germ is lodged in a hollow in the ventral region and base of the caryopsis, close to where the grain is attached to the ear. It is covered by the aleurone layer on the outer face and by the amylaceous endosperm on the inner face. It is joined to the endosperm by a layer of flattened, protoplasm-deficient cells, with thin cellular walls. The thickness of the layer is variable and it appears as an amorphous mass, in which it is difficult to distinguish the cells. When the germ is separated manually from the caryopsis, the layer remains partly attached to the germ. The flattened cells come from the endosperm and have degenerated during the maturing process of the seed, losing their cellular content. The germ, which varies in size, has a lenticular shape, with a longitudinal outer keel. In the *japonica* variety Balilla  $\times$  Sollana, the measured dimensions of the germ are  $2.12 \times 0.91 \times 1.45$  mm, and those of the grain  $6.03 \times 3.25 \times 2.28$  mm [14].

The germ shows considerable histological differentiation and has a complex anatomical structure (see figure 9). It consists of an L-shaped embryonic axis, composed of the radicle, the plumule, the hypocotyl and the coleoptile; from it grow the scutellum, the epiblast, the coleorhiza and the calyptra.

As the germ is found whole and intact in the bran, of which it is one of the important physical components, its histological structure is described more fully in chapter III below, in which the histology of the bran is discussed in detail.

### Chemical composition of the rice grain

In order to be able to use rice as a food, the paddy rice normally has to be milled in order to separate first the husk (husking operation) and then the bran (whitening operation). Although the only really inedible part is the husk, the separation of the bran increases the edibility of the rice and improves its colour, both of which characteristics are appreciated by the consumer. This change is due to the considerable differences in the chemical composition and physical characteristics of the structure and constituent anatomical layers of the grain. The fact is that husking and whitening produce marked changes in the chemical

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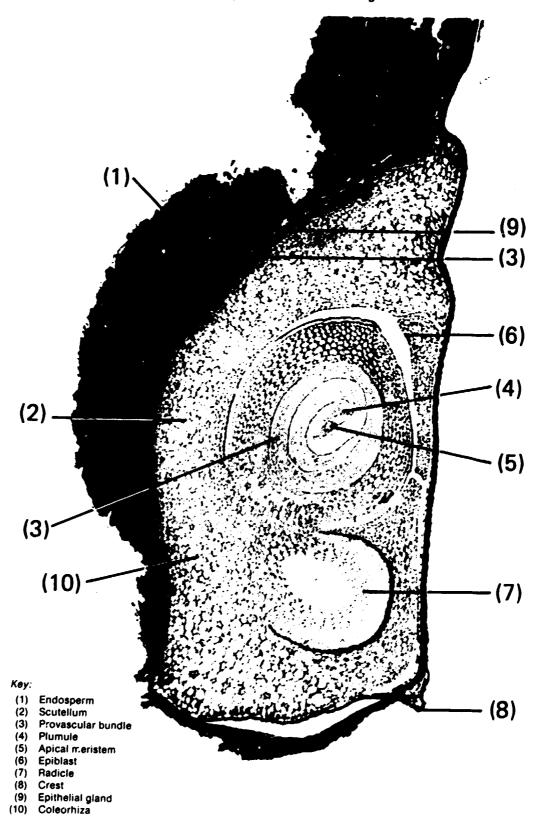


Figure 9. Longitudinal section of the rice germ

composition of rice (see table 3). During the husking process, the most dramatic changes occur in the crude fibre (~90 per cent loss); during the whitening process the main changes are in the fat (~75 per cent loss) and the fibre (~70 per cent loss). Although the extent of these changes varies considerably according to the variety of the rice, its origin, the prior history of the consignment and the milling, they are always important.

TABLE 3.	CHEMICAL COMPOSITION OF RICE AND CHANGES OCCURRING DURING								
PROCESSING									
	(Percentage)								

	1	Relative content in		Change during	
Component	Paddy rice	Brown rice	Milled rice	Husking	Whitening
Proteins	9.24	10.12	8.38	+8.55	-17.19
Fat	2.11	2.38	0.63	+6.16	-73.53
Crude fibre	8.84	0.91	0.29	-89.71	-68.13
Ash	3.35	1.19	0.55	-64.47	-53.78
Nitrogen-free extract	76.44	85.30	90.10	+11.59	+5.62

If the data on the chemical composition of rice are considered in conjunction with the data on the materials remaining when paddy is converted into edible rice, the importance of milling and its by-products is readily apparent.

Detailed descriptions of the chemical composition and chemical constituents of the rice grain have been given by Juliano [8, 29], Barber [30] and Kennedy [31].

### The milling process and the production of bran

No single publication deals exhaustively with the subject of rice milling. The grain, the machines, the working conditions and their effects on both the grain and its by-products, maintenance, management and administration are subjects that must be dealt with one at a time on a scientific and technical basis, taking into account many different fields of study, and focusing on a broader area than simple industrial practice. Excellent contributions have nevertheless been made recently in certain limited areas, for example by Gariboldi [32], Araullo, de Padua and Graham [33] and Spadaro, Matthews and Wadsworth [34].

Paddy rice is invariably processed in order to convert it into grain fit for consumption. Nobody processes rice just to produce bran. For this reason, milling processes concentrate entirely on the grain, on making sure that it loses none of its qualities and on improving its appearance; in short, the goal is to obtain a final product of maximum commercial value. Although this goal does not exclude the possibility of producing quality bran, the basic principles and peculiarities of the process may be ignored, with detrimental effects on 2

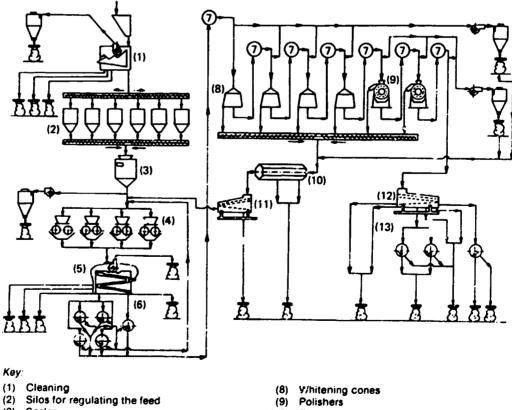
subsequent use of the bran. This is quite apart from the fact that a thorough familiarity with the process of separating the bran must lead to improvements in the processing of rice and in the end product.

In this section, the discussion is deliberately focused on the production of bran as part of the rice-milling process. A brief description of the process is followed by a description of the various types of equipment used and how they work; the characteristics of rice when the bran is separated are then analysed; and lastly, the effects of the different machines and operations of the process of preparation on the quantity and quality of the bran produced are reviewed.

### Processing

The basic stages in the processing of rice are: (a) cleaning; (b) husk removal; (c) whitening; and (d) grading. The flow diagram in figure 10 shows how the process works.

### Figure 10. Processing in a European rice mill



- (3) Scales
- (4) Rubber-roll huskers
- (5) Husk separator
- (5) Separator
- (7) Aspiration

- (10) Bran separator
- (11) Germ separator
- (12) Whole grain separator
- (13) Grading of brokens

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### Cleaning

At the mill, the paddy rice is cleaned so as to remove all foreign matter, including straw, soil, stones, twine, metallic particles and foreign sceds. Although the paddy will have been pre-cleaned before it is dried and stored, it may still contain impurities when it arrives at the mill. Cleaning is based on differences in the size, weight or form of the foreign matter in relation to the grain. The lightest impurities are removed by aspiration or sieving; impurities that are denser than the grain but not of the same size are separated by sieving and those of greater density but the same size, by gravity; grains that are similar in size, form and weight are difficult to separate and therefore pass on with the rice to the following stage.

The machines used at this stage, namely, the scalping machine (see figure 11), the separator sieve (see figure 12), the vibratory grading sieve ("plansifter") the gravity separator or densimetric table<sup>4</sup> and the disc separator [35, 36], may or may not combine suction, sieving and gravity separator. At a later stage the impurities are separated by means of a magnetic separator [33].

### Husk removal

Once the paddy rice is clean, a husker is used to remove the husk from the caryopsis. The machines most often used for this purpose are the disc husker (see figure 13) and the rubber-roll husker (see figure 14).5.6 With the disc husker, the grain of rice is squeezed longitudinally and rubbed lightly to separate the lemma and palea and release the caryopsis. The machine consists of two parallel and coaxial iron discs, the facing surfaces of which are coated with an abrasive material. The gap between the surfaces is slightly smaller than the length of the grains to be husked. The upper disc is fixed, with an aperture in the centre for the rice to enter, while the lower one rotates. The grains are forced to rotate outwards with the lower disc, until they are, for a moment, in a vertical position; they are then squeezed and rubbed between the surfaces, thus removing the husk. The rubber-roll husker (see figure 14) works on the principle that if the sides of the paddy grain are pressed between two elastic surfaces moving at different speeds in the same direction, the glumes will separate [37]. The machine consists of two rollers made out of synthetic elastomer, one in a fixed position and the other adjustable so that the gap between the two is always rather smaller than the thickness of the paddy grains. The rollers rotate in opposite directions at different angular speeds. The grains of paddy are husked when they pass between the rollers.

The discharge from the husker is a mixture of brown rice, husk, bran, fines, powder, chalky and green grains and also unhusked grains—the huskers are regulated so that they husk about 95 per cent of the grains in a single pass. The bran and the powder are removed in a separating sieve with a suitable mesh, or by suction applied in the upper part of a sieve, and they are collected separately (or they can be incorporated into the bran stream of the whitening

<sup>&</sup>lt;sup>4</sup>Also separates stones. It is only used on clean paddy or on brown rice that has been dehusked before it goes into the whitening machine.

<sup>&</sup>lt;sup>5</sup>Centrifuge huskers have a very limited application.

<sup>&</sup>quot;Actually, the "rubber" in a rubber-roll husker is a synthetic elastomer.

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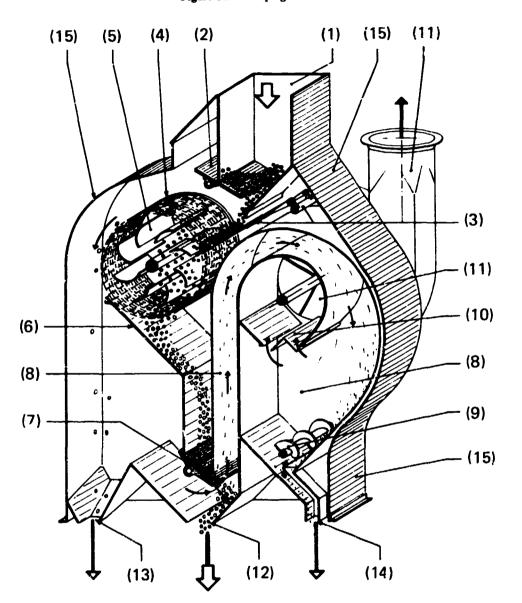


Figure 11. Scalping machine

### Key:

- (1) Feed hopper(2) Feed-regulation gate
- Adjustable rack (3)
- Wire-mesh scalping reel
- Rotating blades Grain-receiving rack
- (4) (5) (6)
- Seal gate (7)
- Aspirating leg and setting chamber (8)

Source: Borasio and Gasiboldi [35].

- (9)
- Worm conveyor Air outlet baffle plates (10) (11)
- Suction fan
- Clean grain outlet (12)
- (13) Outlet for large particles (scalping)
   (14) Outlet for sieve residues (screening)
- (15) Frame and housing

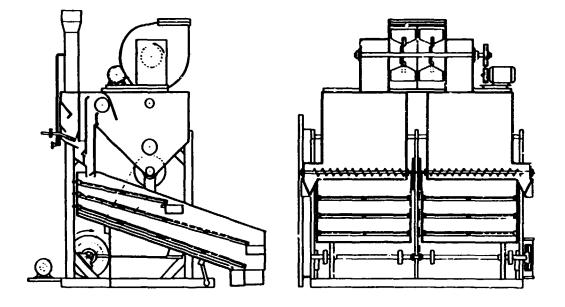


Figure 12. Cleaning sieve

machines). The fines are usually separated in another sieve, before the husks are removed by aspiration, to prevent part of them from being carried away along with the husks. The green and chalky grains are also separated earlier if this can be done without difficulty.

Gariboldi [37] and van Ruiten [38] have described several types of husk separators and the different methods used to separate the bran. In one machine, the separator (see figure 15) is combined with a "plansifter", which is fixed to a solid steel frame placed immediately over the husk aspirator. The "plansifter" is equipped with two automatic unblocking sifters. The first sifter has holes with a small diameter, which will allow bran and powder to pass through them; the second one has larger holes to allow the fines to fall through. The mixture of husk, paddy rice and brown rice is rejected by the sifters. In another machine, the double-action husk separator (see figure 16), the mixture of husk, brown and paddy rice, fines, green and chalky grains, bran and powder is discharged into an oscillating sifter. The bran, powder and fines pass through the sifter and are discharged into two openings (left and right) in the rear part of the aspirator itself. A stream of air, made to pass through the curtain of by-product, separates the bran and the powder as they fall from the fines, which are discharged through an aperture situated at the bottom of the machine.

Both the disc husker and the rubber-oil husker produce what is called "husker bran". This consists basically of fragments of husk and may include a small proportion of pericarp, germ, rachilla, small fragments of endosperm, powder and soil. The disc husker is rougher, and even the most carefully regulated machine will scratch the pericarp and tear away a considerable part of the germ. It usually produces from 1 to 2.5 per cent of bran in relation to

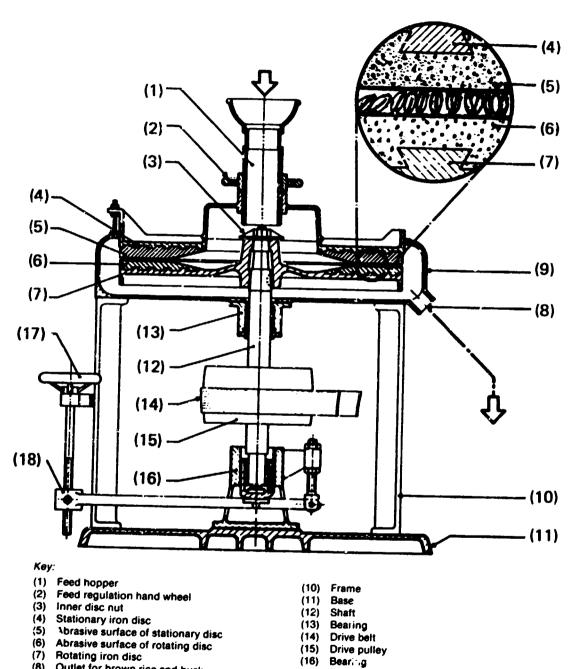


Figure 13. Disc huster

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Source: Borasio and Gariboldi [35].

Outlet for brown rice and husk

Disc housing

(8)

(9)

Disc clearance adjustment hand wheel (17)

(18) Shaft support arm

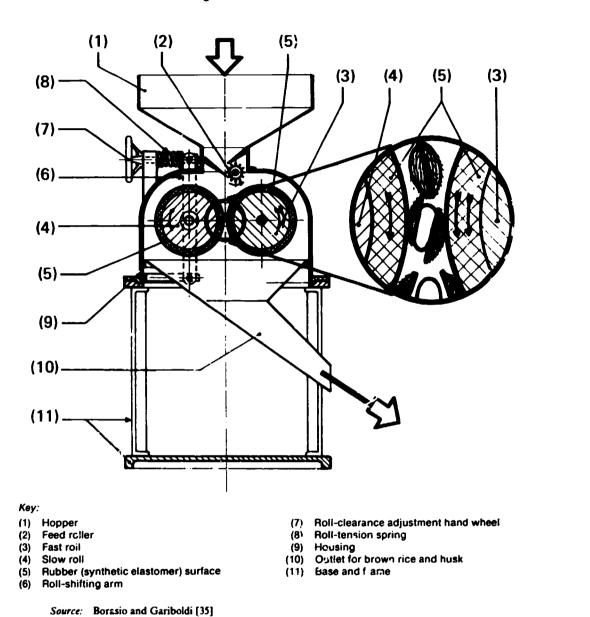
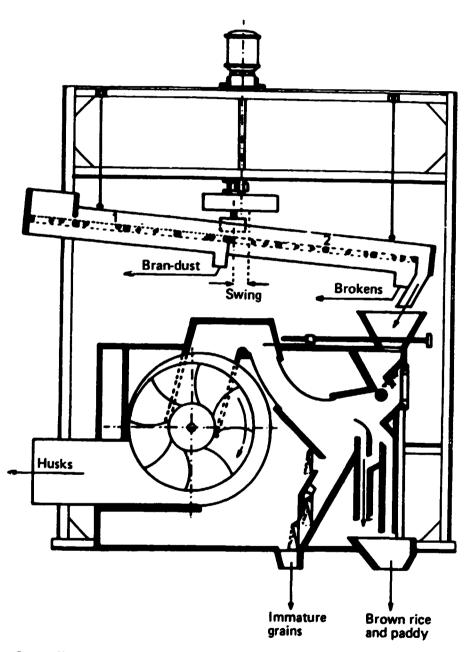


Figure 14. Rubber-roll husker

paddy rice. The rubber-roll husker, if carefully regulated, does hardly any damage to the pericarp and rarely separates the germ from the caryopsis. Consequently, the bran in this case consists of fragments of husk and impurities. Its production represents less than 0.5 per cent of the paddy rice. The very small amount involved is aspirated along with the bulk of the bran or, more usually, discarded with the husk.

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Source: Van Ruiten [38].

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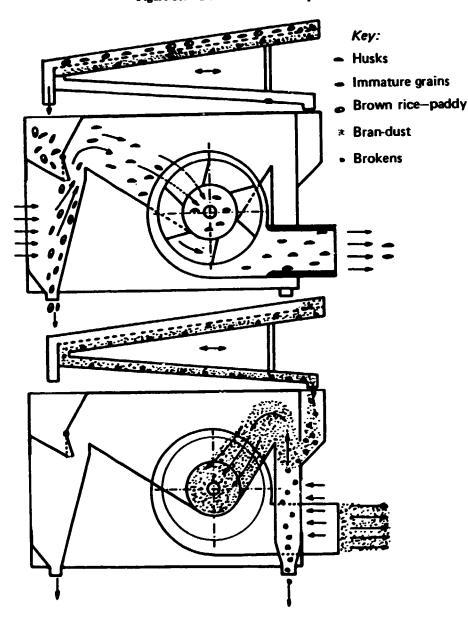


Figure 16. Double-action husk separator

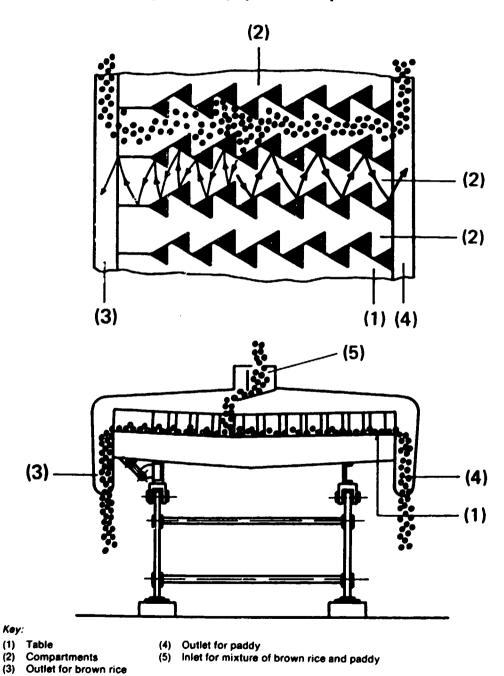
Source: Van Ruiten [38].

# Separation of paddy

Industrial huskers only husk between 80 and 95 per cent of the paddy grains at one time. Therefore, after separating the bran and the powder, the fines and the husk, a mixture of brown and paddy rice is left. Generally, green and chalky grains are present, since, as already noted, it is usually difficult to remove them at an earlier stage. The paddy separator is used to complete the task. 2

The most widely used paddy separators have a compartment-type assembly (see figure 17). Their operation is based on the different behaviour of paddy and brown rice when they move down an inclined plane. The grains of brown rice, being smaller, denser, rounded and smooth, slide more rapidly than the grains of paddy, which are bulkier, lighter and rougher. A more recent type of paddy separator is the tray separator (see figure 18), which also works on the

# Figure 17. Paddy separator with compartments

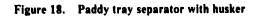


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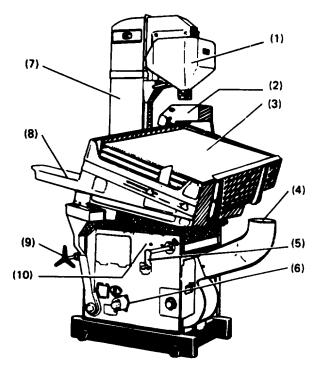
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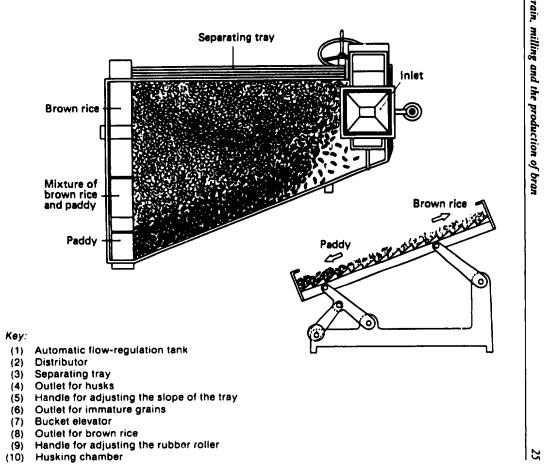
(3)

Source: Gariboldi [37].



A. Separator





B. Husker

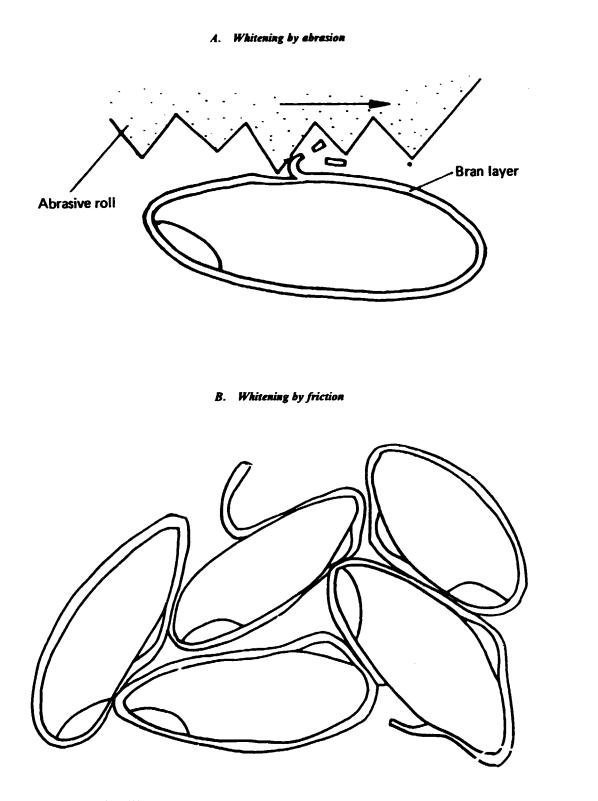
basis of differences in the density and smoothness of the grains. It consists of a series of trays, arranged one on top of another, sloping and with toothed surfaces, driven upwards and forwards towards the highest part. The rice bounces on the tray; brown grains are impelled towards the highest part of the tray, paddy grains towards the lowest. Both separators— with compartments or with trays—can, if properly regulated, yield brown rice that is completely free of paddy.

# Whitening

The brown rice passes from the paddy separator to the whitening machines to eliminate the bran from the grain and to whiten the rice. The machines at present in use operate either by abrasion or by friction. The abrasion machine can have either a vertical shaft, like the European whitening cones, or a horizontal shaft, of Japanese design; the friction machine is like the Japanese "jet polisher", with a horizontal shaft. In abrasion machines, the brown rice is whitened when it passes through the empty space between an abrasive cone and a screen (European mill) or a perforated cylinder (Japanese mill). The abrasive cone-with sharp edges of vitrified particles-acts as a knife that cuts and separates small pieces from the bran layer of the husked germ, like peeling an orange by removing small pieces of peel one by one with a knife without cutting the flesh [39]. Industrial machines also produce some friction, but the abrasive action predominates (see figure 19). The machine (see figure 20) consists of a cast-iron conical rotor, inverted (upright in Spain), covered with abrasive material<sup>7</sup> and mounted on a vertical shaft, which rotates inside a frame or box. There is a screen or perforated sheet in the frame, divided into segments, which is some 10 mm away from the abrasive surface. The screen box holds the rubber brakes, or resistance pieces, which run parallel to the cone, at a distance of some 2-3 mm. The gap between cone and screen and between cone and brakes may be regulated, thus varying the proportion of bran that can be separated from the grain. The brown rice enters the machine through the top; it is thrown outwards by centrifugal force, falling between the cone and the grating so that it rotates in the direction of the cone. The grains strike against the rubber brakes, where they accumulate and, impelled by the cone, are forced to rotate and rub against each other. The grains are thus subjected to the abrasive action of the cone and, to a certain extent, to friction between grains and with the screen (wire cloth or perforated sheet), until they are stopped by the brake. Gradually the grains escape from one brake, only to be trapped by the next one. They continue falling until they reach the outlet. The outer layers of the grain, separated as bran, escape through the holes in the screen, helped by the stream of air which goes through the machine to cool the grain. The bran is collected in a circular collector, from which it is discharged into the general trunk, impelled by rotating vanes. The air stream carries away some bran, which is recovered in a cyclone.

The abrasion whitener with a horizontal shaft (see figure 21A) processes the rice as it passes through the gap between the abrasive central cylinder and the perforated steel cylinder surrounding it. The grain is held by pressure

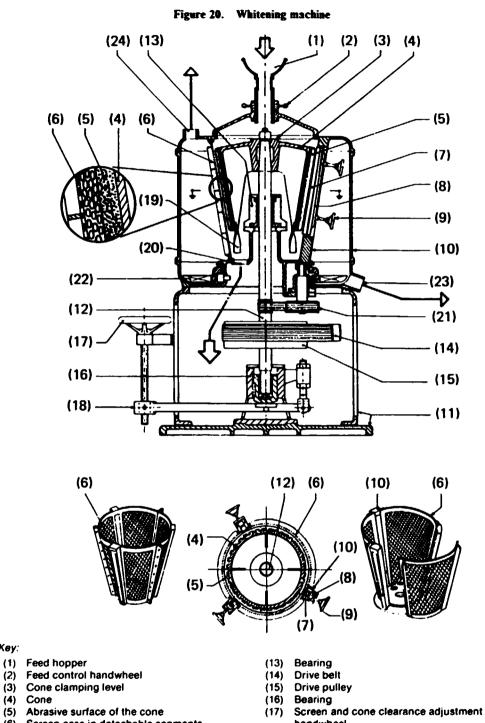
<sup>&</sup>lt;sup>7</sup>For a detailed description of the characteristics of the abrasive materials and instructions for the preparation of the abrasive paste and coating of the cones, see Gariboldi [37].



Source: Koga [39].

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Screen case in detachable segments, with rubber brakes

(?) Brake

Key:

(2)

(3)

(4)

(5)

(6)

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- (8)
- Brake mounting Brake regulation handwheel (9)
- Housing
- (10; (11) Frame
- (12) Cone shaft

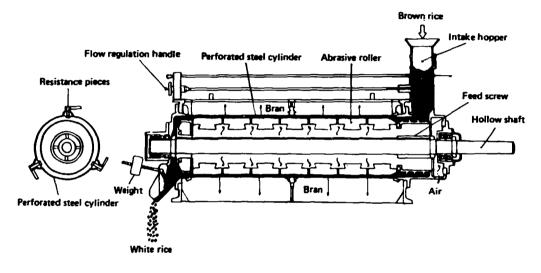
Source: Borasio and Gariboldi [35].

- handwheel
- (18) Shaft supporting arm
- (19) **Rice conveyor**
- (20) **Rice outlet**
- Bran conveyor drive pulley (21)
- Bran conveyor bucket (22)
- (23) Bran outlet
- (24) Air-suction outlet

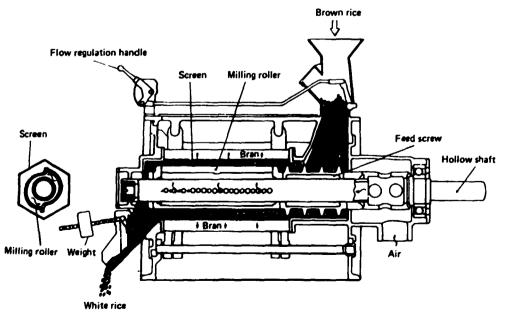
caused by the outlet valve, which regulates the amount of time the grain is retained in the machine and determines the proportion of bran to be separated. Brakes, backing onto the perforated cylinder, change the trajectory and the velocity of the grains in their journey from the intake hopper to the outlet. Air is injected through the hollow part of the central cylinder, and this air passes

# Figure 21. Abrasive and friction whitening machines

#### A. Abrasive whitening machine with horizontal shaft



### B. Friction whitening machine with horizontal shaft



through the grains of rice, carrying the bran with it, and then comes out through the holes of the perforated cylinder. As a rule, two or more units of these machines are used at a time.

The friction whiteners (see figure 21B) work on a different principle. The bran is released and separated when the grains rub against each other under pressure. The friction machine consists of a perforated, partly hollow, shaft; a cylinder of cast steel with friction ridges and inlets for the injection of air, slides on to this shaft. The cylinder is located in a hexagonal chamber with perforated grooves, leaving an empty space for the rice to circulate. A screw in the base of the loading hopper feeds the chamber, while an adjustable outlet valve checks and regulates the flow of the grain, exerting pressure on the mass of rice. The rubbing action thus produced releases the bran from the surface and the air that passes through the mass of rice carries it away through the screen. The internal pressure and the time of r(:ention, which determine the proportion of bran that can be separated from the grain, are regulated by adjusting the discharge valve. These machines are used in many milk to complete the whitening process carried out with one or more abrasion machines.

### Polishing

Although in many mills the whitening process ends at this point, in others it goes on to the polishing stage. Although in some countries whitening machines are sometimes called polishers, strictly speaking polishers are quite different. Their object is to eliminate from the grain that has aiready been whitened the small particles of flour that adhere to the surface, so that the rice acquires a glossy satin-like appearance. At the same time a new fraction of bran is collected. The basic parts of a polisher are similar to those of a whitening machine; the essential difference is that they have no emery coating and no rubber brakes. The two types of polishers most often used-with vertical cones or with horizontal cylinders-are based on the same principle. In the first case, the cone is covered with wood to which strips of leather are nailed. The rice enters the machine in the same way as it enters whitering machines of the same type; on passing through the gap between the cone and the screen, it is impelled by the strips of leather which make it roll around, the grains rubbing against each other and against the grating and the leather itself. The powder that is thus separated is drawn from the machine and is recovered in a cyclone.

### Final steps

In most countries, as far as the production of bran is concerned, polishing is the final stage of rice processing. Although as already noted, each stream or fraction of bran is generally produced separately, the streams are all combined before being marketed. The rice that is discharged from the whitening machines is a mixture of whole and broken grains—the latter of various sizes—which must be separated and graded before marketing. This is a very important part of rice milling, but it is outside the scope of the present study. The subject has been dealt with in detail elsewhere [33].

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### Mills and the type of cquipment used

### Categories of mills

The complexity of the methods used in the processing of rice varies from one country or region to another and according to particular circumstances. Saunders and others [40] have classified installations for rice conversion into categories of different technical and social complexity: (a) subsistence-level; (b) rustic; (c) quasi-commercial; and (d) export. The first category is so called because the rice is produced solely for consumption, generally on an individual or family basis, with no investment in machinery. The second category is represented by the owner of a small mill, consisting of one machine with which he processes the rice brought in sacks by the customers (small farmers or others), from whom he extracts payment, often in the form of rice o. byproducts. This is, according to the authors, the first level at which mechanical improvements of a certain degree of sophistication may be introduced, because the necessary capital may be put up by a group, directly (i.e. through a cooperative) or indirectly (i.e. in the form of shares). The quasi-commercial category is represented by the miller who has very few machines in his mill but who invests capital, has a commercial outlook, and shares the aims and problems of the miller at the next highest level. His field of action is nevertheless very limited, as are his resources. The export or commercial category differs from the preceding one chiefly in the field of action, which is not limited (since it may involve exports) or is expansive (on a national scale), in the quality of rice it requires and in its competitive capacity and technology, which in this case means major capital investments.

# Types of equipment and the way they act on the bran

Leaving aside the manual processing of rice found at the subsistence level,<sup>8</sup> the most simple form of mechanical processing, typical of the rustic category,<sup>9</sup> employs a single machine, involving only two conversions: the processed rice (with a variable quantity of fines) and a by-product consisting of a mixture of ground husk, ground rice, bran and all kinds of impurities, such as straw, soil, stones etc. The machine most commonly used is the "huller" (e.g. kiskisan)<sup>10</sup> (see figure 22). It consists of a cylinder of grooved steel, which rotates on an axle, inside a housing with a screen at the bottom; the rice circulates through the gap between the revolving cylinder and the housing in the direction of the outlet, rotating through the action of the grooves in the cylinder; the first grooves are sloping and exert pressure on the rice, moving it towards the outlet. An adjustable blade controls the amount of rice that passes through and determines the proportion of bran that is separated from the grain.<sup>11</sup> The

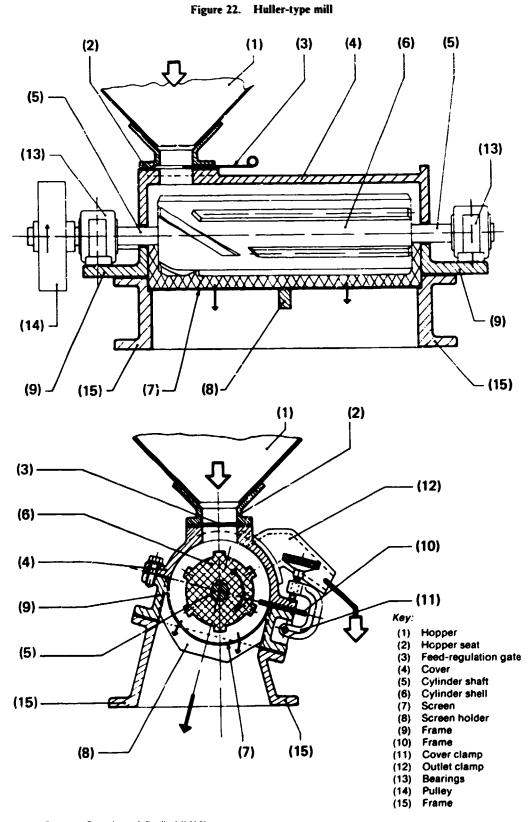
<sup>&</sup>lt;sup>3</sup>In the rural areas of some countries—Bangladesh, India, Indonesia, for example—a fairly substantial amount of rice is processed with a pestle and mortar, operated either by hand or by foot.

<sup>&</sup>lt;sup>9</sup>Less usual at this level are installations with one machine for husking and another for whitening, with a sieve as a third unit to grade or separate.

<sup>&</sup>lt;sup>10</sup>Kiskisan hullers are used for husking and whitening and in a single pass or in two stages and as a whitening machine in conjunction with a disc or rubber-roll husker.

<sup>&</sup>quot;It also influences both feed and discharge.

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ground husk, fine broken grain, bran and impurities pass through the lower sieve and come out through the discharge mouth. The rice and large fragments of husk are discharged together through another outlet. The main trouble with these machines is that much of the rice is broken and the yield is low,<sup>12</sup> the lossof the small and medium grains through the grating being a contributory factor. In addition, since these mills do not have a paddy separator to eliminate the residual unhusked grains from the final white rice, the rice has to undergo more extensive processing in order to ensure that as few paddy grains as possible are left behind. This is particularly true in the case of raw rice, in which the husk is not readily removed. In so far as bran is concerned, the disadvantages are covious, since the bran that is produced has a high silica and fibre content and a very low proportion of oil; in addition it carries along with it the soil and other impurities contained in the paddy.

In spice of all these disadvantages, owners of this type of mill are reluctant to undertake any modernization, particularly in areas where parboiling is practised. Furthermore, the number of such mills is on the increase.<sup>13</sup> Pillaiyar and others [42] have identified several reasons for this: first, the machine is heap, it is easy to install, operate and maintain, even in a rural environment; secondly, some machines have a small capacity, can operate intermittently and can therefore work with the small quantities of paddy—either raw or parboiled—that the customer in the rural area may require; and thirdly, they yield a processed rice of acceptable quality.

In some cases, economic factors have led to a proliferation of mills of this type. In India, for example, the Government imposes a form of taxation by buying a percentage of the rice processed by commercial mills at a price below the market rate. The percentage varies but is generally more than 50 per cent of the total production. The huller mills are considered to be non-commercial and are exempt from this tax, as well as from sales and purchase taxes. The fact that in West Bengal the number of hullers has grown in a few years from roughly 6,000 to more than 22,000 may be attributed to this difference in tax treatment [43]. In the Philippines, there have been cases in which special loans were granted by the World Bank and the Asian Development Bank to install new mills with rubber-roll huskers; then, once the inspection was over, the machines were replaced by husker units [40].

The Indian Ministry of Agriculture and Irrigation has recently introduced a simple procedure for improving single-machine huller mills; an installation of this type is automatically recognized under the Rice Milling Act and Rules as a "modern unit", with all the administrative benefits that accrue to installations of this type [44]. The technical information distributed by the Ministry confirms that the yield is higher, there is less breakage and a bran of superior

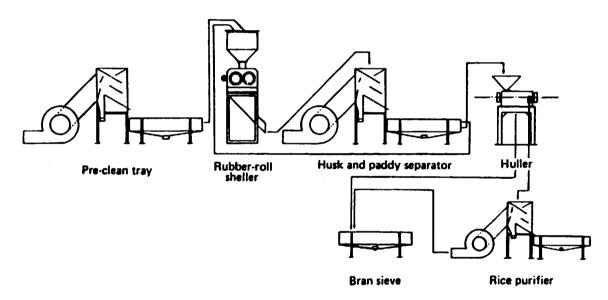
<sup>&</sup>lt;sup>12</sup>The yield of mills with rubber-roll huskers that process paddy rice may be expressed as 2.5 per cent more total processed rice than those with disc huskers and 6.6 per cent more than those with hullers. In the case of parboiled rice, the figures are 0.8 and 1.6 per cent respectively. The differences in whole rice are 6.1 and 15.1 per cent for paddy and 1.3 and 4.1 per cent for parboiled rice, respectively [41].

<sup>&</sup>lt;sup>13</sup>In India, according to data published in 1965, there were more than 30,000 mills of the huller type, with individual or combined units. Statistics for the biennium 1972-1973 raised the figure to 72,000, while in 1977 the number was close to 90,000.

quality is recovered. The machinery consists of a centrifugal husker<sup>14</sup> and a cleaner with paddy separator consisting of a sieve, combining an oscillatory movement with aspiration, which is used first as a cleaner and then as a paddy separator, the work, of course, being done in batches; lastly, the existing huller mill is used to whiten the brown rice.

In the same industrial sector, the following example represents a further step in the modernization process [42]. In a mill consisting of two huller units, capable of processing 1 t/h of parboiled paddy, one of the units was used for husking and the other for whitening; the white rice, together with the fines and a little husk, was cleaned and graded in winnowing tray; the bran thus obtained contained around 6 per cent oil and 15 per cent silica. The mill was modernized by installing: (a) a new winnowing tray for pre-cleaning; (b) a new non-suction rubber-roll husker; (c) the existing winnowing tray, duly modified to work as a paddy separator as well as a husk aspirator and cleaner;<sup>15</sup> (d) one of the existing huller machines, used exclusively for whitening; (e) a new winnowing sieve to separate fines and the loose bran adhering to the white rice; and (f) a simple sieve, also new, to separate the small fines and a little husk from the bran (see figure 23). Apart from achieving a greater yield in rice and reducing the

#### Figure 23. Huller-type mill, modernized



<sup>14</sup>The centrifugal husker consists of two metallic parallel discs, with a small gap in between rotating at a high speed (3,500 rpm). According to the information circulated, its yield is comparable to that of a rubber-roll husker, but rice breakage and mixing of husk and bran are eliminated; the machine is also claimed to leave the caryopsis intact. The paddy enters at a central point and is guided by metallic devices situated between the discs. The grains, projected by centrifugal force, collide with a stationary rubber disc placed against the outer ring, where they lose their husk. Usually machines have a capacity of some 400 kg/h; they can work with small consignments and operate at no more than 1 hp [44]. Better results are obtained with parboiled rice than with raw rice, which may be why Saunders and others [40] report that this type of machinery has not given good results in the Philippines.

<sup>13</sup>The modification presented no great problem, since it was considered satisfactory to have 5 per cent paddy in the husked rice to help in the whitening process.

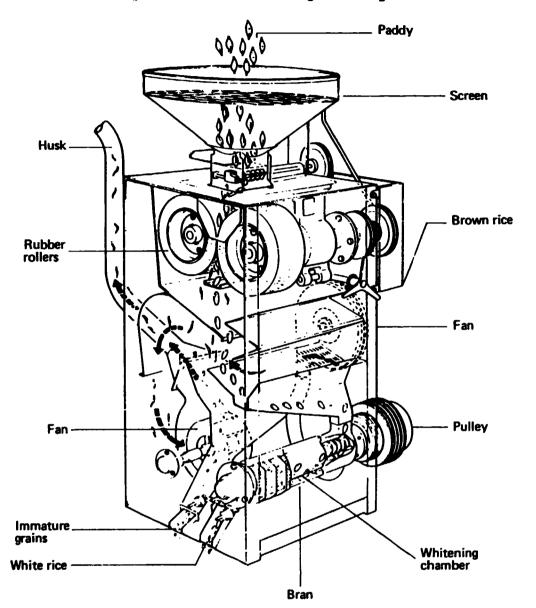
consumption of energy by about 30 per cent, the new machine produced a marked improvement in the quality of bran produced, with an increase in oil content of nearly 20 per cent. This proved to be a low-cost and worth-while investment.

Some manufacturers have combined the husker and whitening machine with some other devices, in a monobloc unit, in order to complete the whole process, from husking and separation of the husk from the husked rice to whitening and separation of the bran in one pass, using only one machine (see figure 24). The machine incorporates a vibratory sieve for the separation of large impurities (straw, rachilla etc.), as well as a magnetic separator, a rubberroll husker, a husk separator that also separates the immature grains, a horizontal whitening machine with cooling air injection, which at the same time extracts the bran. If there are many paddy impurities, the monobloc unit may be combined with a pre-cleaner (see figure 25). The capacity of the machine varies from 0.5 to 1 t/h, according to the model used. Generally, because the rice they process is usually of inferior quality and the whitening is carried out in a single pass, these machines cause more breakage and the bran contains a larger proportion of starchy endosperm. In addition the cleaning and the separation of the rice from the husk are far from perfect, and this also influences the quality of the bran.

With the basic elements described in the preceding section and other accessories, milling units of widely varying degrees of complexity can be constructed (see figures 26-28). Of particular note are the paddy-rice grader and feed-back husker, the stoner, which is used before the whitening machines, the number of whitening stages and the germ separator; the role of all these will be discussed later. The germ separator (see figures 29 and 30) calls for particular comment. Although in most countries commercial bran includes the germ, in others, like Italy and Spain, the practice of separating the germ is widespread. In Spanish mills, the mixture of brans from the different whitening machines (from three to six are used) passes through the revolving sieve and the germ separator. The former, a screen in the form of a hexagonal prism with a mesh of approximately 0.8 mm, separates the bran, the fragments of germ and smaller germ, as well as the very small fines. The germ separator combines a vibratory sieve with aspiration, in order to separate the particles, which differ in size and in density. It comprises a first sieve, with 0.8-mm holes, to separate the residual bran; a second sieve with a 1.5-mm mesh opening to separate the germ, and a third one, with 2-mm perforations, to separate the small fines from the larger fines and particles (including whole grains) that may be present. A suction stream is passed through the germ outlet, in order to remove lightweight impurities (fragments of husk and bran). The final purification of the germ can be done pneumatically. Depending on the types of machines used and the conditions under which they operate, a commercial product is achieved with a pure germ content ranging from 40 per cent to 85 per cent [45].

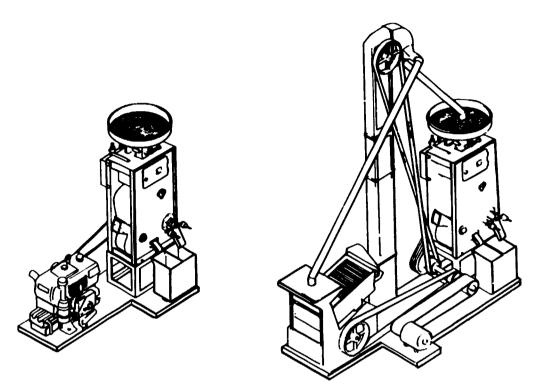
Several types of machinery have been tried out for separating the germ and very small broken grain pneumatically [46].

Another method that has been tried is to remove the germ from the grain before processing [47], as is sometimes done for wheat. After the husk has been removed, the rice is maintained on a fluidized bed, where the friction between the grains and the perforated containing walls results in separation of the germ



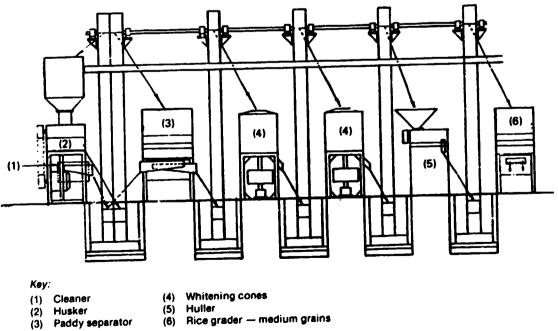
# Figure 24. Monobloc unit for husking and whitening rice

from the caryopsis. It has not been possible to avoid simultaneous separation and breakage. The germ, the bran particles and the small fragments of endosperm, as they are produced, are carried away through the perforations of the chamber by a stream of air. The three fractions produced are separated by later sifting and winnowing. Table 4 supplies data on this device as used with varieties of rice cultivated in India; it also includes data on parboiled rice.



# Figure 25. Monobloc unit with pre-cleaner, bucket elevator and drive unit

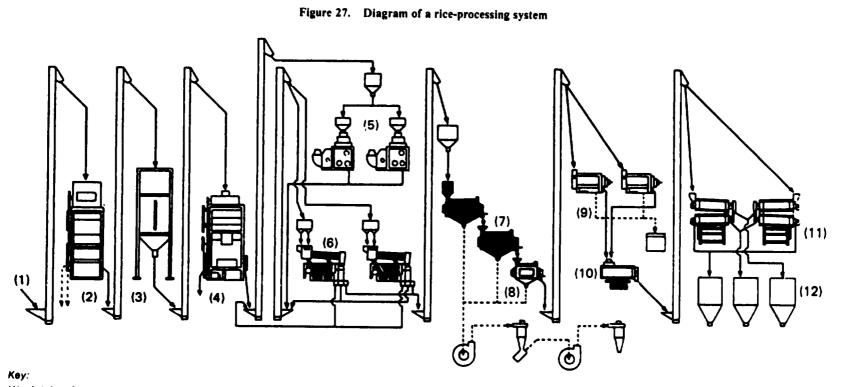
Figure 26. Modernized mill, including a huller-type unit



(1) Cleaner
(2) Husker
(3) Paddy separator

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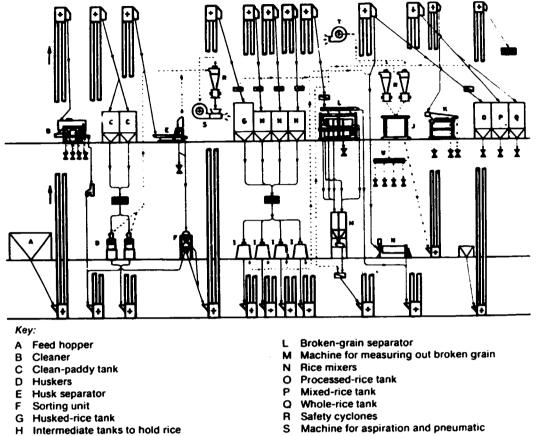
2



- Intake of paddy
   Cleaner
- (3) Tank
- (4) Aspirator with stoner
- (5) Husker(6) Paddy separator (7) Abrasion whitening machine
- (8) Friction whitening machine
- (9) Refining machine
  (10) Gyro sifter
  (11) Rice graders
  (12) Tanks

Rice bran: an under-utilized raw material

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#### Figure 28. Diagram of a rice-processing system, including germ separator

- H Intermediate tanks to hold rice for bleaching machines
- I Bleaching machines
- J Bran separator
- K Germ separator

- transport of husk
- T General aspirator
- U Bran conveyor

The major impurities contained in germ coming from the mill consist of fragments of husk and, in particular, very small fragments of grain. When a higher degree of purity is required, for example in laboratory work, the device illustrated in figure 31 gives satisfactory results. It works on batches of up to 50 grams. The impure germ is sifted in a pair of sieves, with meshes of 1.5 and 0.8 mm (or others more appropriate to the size of the germ of the variety being treated). The part held between both sieves is loaded onto the grating of the purifier, in the lower part of the glass or plastic tube, which constitutes the main body of the device, and air under pressure is blown in through the lower inlet. With a pressure difference of some 20 mm water gauge, practically all the husk is separated. By increasing the pressure to above 100 mm water gauge, the germ can be separated without carrying away the fragments of endosperm, which will remain on the grating.

P

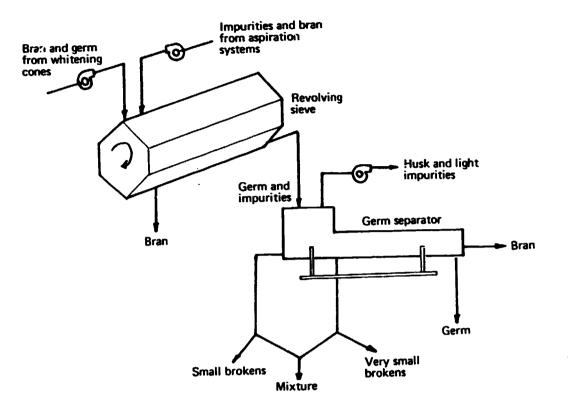
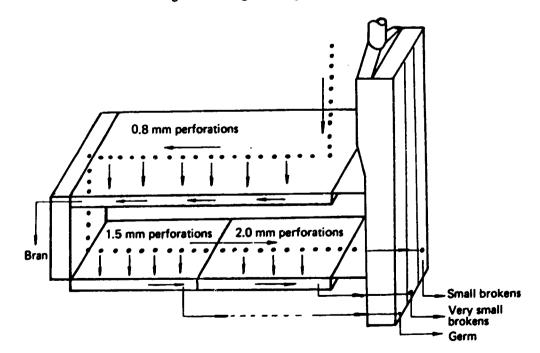


Figure 29. Process for separating the germ from the rice bran





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		Yield	
Variety and type of brown rice	Germ	Bran	Fines
ADT-8			•••
Raw	2.6	3.6	28
Parboiled	1.0	3.0	3
IR-20			22
Raw	1.6	3.1	22
Parboiled	0.5	2.5	3.8

# TABLE 4. SEI ARATION AND MANUAL RECOVERY OF GERM IN A GERM-REMOVING MACHINE<sup>4</sup>

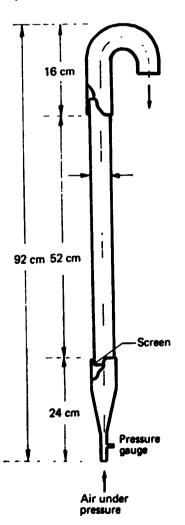
(Grams per 100 grams of husked rice)

Source: Vasan and others [47].

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<sup>4</sup>Operated by the Paddy Processing Research Centre (PPRC), Thiruvarur, India.





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# Brief description of milling installations in selected countries or regions

Saunders and others have prepared a report on post-harvest losses of rice, in which they supply information on milling installations in selected countries [40]. Their descriptions of milling installations in different countries and the type and quality of bran that can be produced in each case are summarized below. Supplementary notes have been added from other sources, including an interesting report on post-harvest technology in four countries in Asia [48].

### Africa

In Liberia there are estimated to be about 150 kiskisan-type units and about 90 installations with rubber-roll husker combined with a friction whitening machine.<sup>16</sup> In some cases, a roller husker is used in the first stage and the kiskisan for whitening. In Liberia, there are only three mills of commercial size. They are equipped with pre-cleaners, rubber-roll huskers, husk aspirators, paddy separators and whitening cones. It is estimated that manual processing accounts for 77 per cent of the production, the small mills for 20 per cent and the large mills for 3 per cent. In Sierra Leone, there are mills at both the rural and the commercial levels. There are several types of rural mill. One consists of a rubber-roll husker combined with a friction whitening machine, single pass, with a capacity of about 200 kg/h, of which there are about 20 in operation.<sup>17</sup> Another mill of the same type, but with a capacity of 500 kg/h, uses a precleaner on occasion. A third type consists of a combined husker and whitener, with a capacity of some 200 kg/h, of which there are about 350 examples in the country. At the commercial level, there are a few mills, representing only 4 per cent of the total milling sector, as against 13 per cent at the rural level. Processing by hand still accounts for 83 per cent of the total. In many countries, a high proportion of the total processing consists of husking by hand pounding, 92 per cent in the Gambia and Nigeria, for example, 62 per cent in Ghana, 46 per cent in the Ivory Coast and 30 per cent in Benin. The mills at the rural level, which are predominantly of the kiskisan type, and those at the commercial level, which are based on a roller husker combined with a friction or kiskisan-type whitening machine, are found in the following proportions: the Gambia and Nigeria 6 per cent and 2 per cent, Ghana 24 per cent and 14 per cent, the Ivory Coast 31 per cent and 23 per cent and Benin 30 per cent and 40 per cent.

### Bangladesh

The large mills in Bangladesh convert only 2-6 per cent of the rice produced; they are located in or near urban areas. In the rural areas, two types of technology—manual and huller-based—are practised: the latter process 20 per cent of the rice production. There are about 7,600 huller mills: their capacity varies from 500 kg/h to 1,300 kg/h. The larger commercial mills use batteries of up to five machines [49].

<sup>16</sup>Other sources indicate 400 kiskisan units and 150-200 with roller huskers and friction whitening machines.

<sup>17</sup>Other sources indicate 50.

### Indonesia

Milling in Indonesia has changed greatly in recent years. It is estimated that in one decade the 80 per cent of production husked by hand pounding has shrunk to 20 per cent.<sup>18</sup> The 700 large and 7,000 small mills, producing up to 7.5 t and 2.5 t a day respectively in 1968, had grown to 1,144 and 28,000 respectively by 1974.<sup>19</sup> Mills built between 1900 and 1925 are still being used in Indonesia.<sup>20</sup> Small Japanese mills are being introduced, for the use of rural co-operatives, at two levels of sophistication: (a) roller-husker machines and one-stage horizontal whitening machines; and (b) roller-husker machines, paddy separators and two-stage whitening machines.

### Japan

The procedure followed in Japan is unusual in that the rice is processed in two separate stages. The paddy rice is husked and stored in its brown form, and is not whitened until just before it is consumed. The husking is done in roller mills or co-operative mills, and the bran is whitened in small family-type machines and in mills, of which there are about 500 with a capacity between 3 and 30 t/h and about 20,000 with a capacity of 0.5 t/h. The machinery is very sophisticated.

#### Malaysia

In Malaysia, there are four types of mills: one-stage hullers and Japanese mills with rollers (in conjunction with husk separators, paddy separators and whitening machines) [48,51], both with a capacity of 0.5 t/h, and traditional mills, one type having a capacity of some 2 t/h and a second type (normally equipped with cleaner, husker, separators, whitening machines and grading machines), with a capacity of 4-8 t/h. According to data for 1969, they were distributed as follows: 45 per cent, 37 per cent, 6 per cent and 12 per cent, respectively, of the total number of units. A very considerable part of the harvest is therefore processed with the primitive huller. Many of the small Japanese mills have been replaced by cone whitening machines with rubber brakes, while commercial mills have replaced stone huskers with rubber rollers.

### Pakistan

In Pakistan, the estimated number of mills is 1,200-1,400. There are two types: hullers with two, three and four units with a capacity of 0.25, 0.35 and 0.5 t/h respectively; and husking mills equpped with two or three whitening machines with a capacity of 0.75 and 1 t/h respectively. Recently, 10 or so automatic plants have been installed, with a capacity of 5-15 t/h [52].

<sup>&</sup>lt;sup>18</sup>Thet Zin and others [50] estimate the proportion of rice husked by hand at 10 per cent for 1974.

<sup>&</sup>lt;sup>19</sup>Data from other sources [48] estimate that in 1974 there were about 20,000 kiskisan-type units, 6,000 to 7,000 Japanese huskers combined with kiskisans as whitening machines, and about 1,000 large-capacity traditional mills. Another source [50] indicates that, in Java in the same year, there were 624 large mills and 6,640 hullers, representing only 60.8 per cent of the total rice production—an unascertained number of small mills account for the rest.

<sup>&</sup>lt;sup>10</sup>The old mills burn the husk to generate steam.

# Philippines

A small proportion (3-5 per cent) of the rice harvested in the Philippines is processed by hand. The greater part (40-55 per cent) is converted in huller mills<sup>21</sup> with a capacity of roughly 500 kg/h. At the commercial level, the cone mill is very popular. It consists generally of a disc husker and two whitening cones. The unhusked rice must be separated and fed back into the husker. The capacity varies between 0.5 and 4 t/h. In some modern designs, the mill has rubber-roll huskers, with disc or roller horizontal abrasion and friction whitening machines for the return feed. Many alternative milling systems are possible with the equipment available (see figure 32).

# Republic of Korea

There are more than 20,000 mills in the Republic of Korea, most of them only equipped to supply one village with rice. In 1971, 784 relatively large mills were reported, 55 per cent of them with a capacity of above 7,000 tonnes a year [54]. Generally, the husking is done in rubber-roll huskers and the whitening with cones or jet-type friction whitening machines. The average mill works at about 120 kg/h.

# Sri Lanka

The government mills in Sri Lanka, which process a third or less of the harvest, use all kinds of installations, from modern Japanese and European types to huller machines. The quota mills, which convert most of the harvest, are usually two-stage machines with rubber-roll huskers, and whitening machines of the vertical cone or huller type. Pre-cleaners and stoners are seldom used. More than 50 per cent of the rice converted in private mills is processed by huller machines in a single stage.

# Thailand

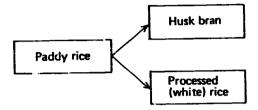
In Thailand, apart from manual processing, which is still practised, huller mills, mills with abrasive discs<sup>22</sup> and "mini-mills" are used at the rural level, while modern or modernized mills are used at the commercial level. The latter usually have disc huskers and two-stage whitening cones, using electric power generated by steam obtained by burning the husk. They include pre-cleaning and grading sieves. In some cases they have rubber-roll huskers as feed-back huskers; in others the roller husker has replaced the classical disc husker. The leather polisher is also common. It is estimated that in 1976 there were about 800 mills with a capacity ranging from 2 to 8.3 t/h of paddy, about 4,000 with a capacity of 0.4 to 1.25 t/h and about 20,000 with a smaller capacity.

<sup>&</sup>lt;sup>21</sup>According to Duff and Estioko [53], using data for 1968, the kiskisan type, with 6,991 mills, accounts for 80.2 per cent of the total number of installations and 53.4 per cent of the industrial capacity, the corresponding figures for the cone type, with 1,728 mills, being 19.8 per cent and 46.6 per cent respectively.

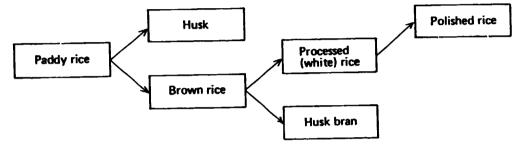
<sup>&</sup>lt;sup>22</sup>These work on a principle similar to that of the husker. The rotor has an abrasive coating and two rubber brakes which project from the housing.

# Figure 32. Alternative systems of rice processing and their effect on the type of bran produced

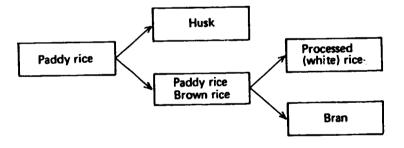
# A. Kiskisan without polisher



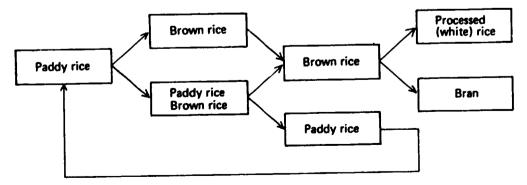
# B. Roller husker and kiskisan with polisher



# C. Cone mill with small-capacity disc husker



# D. Cone mill with large-capacity disc husker



Source: Duff and Estioko [53].

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### Latin America

In Colombia, about 450 mills are operating. Even the smallest, with a capacity of 0.5 t/h, work in several stages. Generally they are composed of a pre-cleaner and rubber-roll and disc husker (when both types are available, the latter is used for the feed-back paddy), a pneumatic husk separator (which independently separates the husker bran and the husk), a paddy separator, cone whitening machines, graders and, in many cases, leather polishers. In Bolivia the mills are built on the old European model and usually have rubber-roll huskers. In Brazil and Peru, they are also based on the old European model. The average capacity is estimated at 1-3 t/h. In Ecuador, there are about 2,000 mills with a capacity of less than 5 t/h. In some cases, they may be very small; one new mill, the biggest in the country, has a capacity of 20 t/h.

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# **II.** Basic principles of the stabilization of rice bran

## Value and end-purpose of stabilization

#### Stabilization and post-stabilization

"Stabilization", in the widest meaning of the word, applies to the entire process, from the time the bran is produced at the mill until it is consumed as feed or used as the raw material for a subsequent process. As a rule, however, the word is used in a much more restricted sense—and will so be used here—to describe a method of treatment designed to limit or prevent the deterioration of the rice bran; the treatment is normally confined to a short, specific, postproduction stage and does not include storage and transport. It should be noted that the broader concept, while less widely used, is actually more appropriate to the industrial sector, since a long-term stabilization process is necessary to ensure the viability of the product. The additional concept of poststabilization must therefore be introduced to complete the cycle. Unfortunately, very little research has been done on post-stabilization technology, and this has to a large extent impeded progress. The future exploitation of the industrial potential of rice bran may be said to depend more today on progress made in post-stabilization technology than on advances in stabilization technology.

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Stabilization and post-stabilization should not be viewed merely as preliminary stages in the extraction of edible oil. Bran not only provides edible oil but is also a potential source of other nutrients and constituents with possible industrial applications, which can only be realized through proper and complete stabilization.

## Coexistence of valuable and harmful components

Harmful components, to be controlled or destroyed, and valuable constituents, to be protected and preserved, may be present simultaneously in the bran. Harmful components include enzymes, micro-organisms, insects, toxins and growth inhibitors, and adulterants and impurities. Oil, proteins, vitamins and other nutrients, on the other hand, are all valuable.

Enzymes (in particular lipases), micro-organisms and insects are the main causes of deterioration in bran. The object of stabilization is to inhibit their activity, or preferably to destroy them, in order to prevent deterioration. It is no use inactivating harmful enzymes if the micro-organisms capable of producing them remain active. The object of post-stabilization is to maintain minimum levels of activity and prevent contamination from outside. There are a number of different means of achieving these objectives, including heat, radiation and chemical compounds. Technical and economic viability at the industrial level is not the only criterion for success. The preservation of valuable constituents is also important. In fact, processes that, in principle at least, do the job they are supposed to do by attacking harmful components may also affect the valuable constituents, thereby reducing the value of the bran as a raw material. Stabilization must therefore be considered as a compromise form of treatment, which reduces the undesirable components to a safe level, with an acceptable degree of risk, while retaining, as far as possible, the quality and quantity of the valuable constituents of the bran.

In order to understand the stabilization process and the scope and significance of the solutions so far proposed, it would seem appropriate to begin by reviewing the fundamental principles determining the stability of both harmful and valuable components. First, the destruction of micro-organisms will be studied, then the inactivation of enzymes and other harmful components, and finally the valuable constituents of the bran.

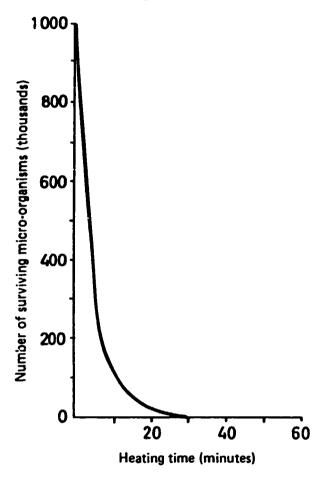
### **Destruction of micro-organisms**

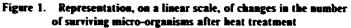
Sterilization implies the destruction of all forms of life. Other less absolute terms are "pasteurization" and "disinfection". Thermal destruction, or death of a micro-organism, signifies the loss of its ability to reproduce under suitable conditions because of temperature changes. Special reference is made in this section to bacteria spores, which are the most heat-resistant microbiological systems. Sterilizing agents may be classified as either physical or chemical. No clear distinction can be made regarding their various actions, however, since physical agents may lead to the formation of chemical products, while chemical agents may cause physical changes, with lethal results in both cases. The most typical agent is heat, which may be dry or moist. Steam under pressure quickly sterilizes permeable materials and surfaces. Dry heat is slower, and requires higher temperatures. The thermal destruction of bacterial cells and spores is an exponential function of time.

#### **Thermal destruction**

#### Curve of the relationship between thermal destruction and time

In order to ascertain the effects of heat treatment on the microbiological population, aliquot parts of the sample are treated at a given temperature for varying periods of time and the number of residual micro-organisms capable of reproducing—that is, of forming colonies—is determined. The relationship between the number of surviving micro-organisms and the duration of treatment at a given temperature is shown by the curve in figure 1. In practice, the initial microbiological population may range from  $10^6$  to  $10^7$  micro-organisms per gram and the final population from  $10^1$  to  $10^3$  micro-organisms per gram. Changes of this magnitude cannot be represented well on a linear scale, especially when the number of living residual micro-organisms approaches





Source: Pflug and Holcomb [1].

zero. This drawback can be overcome by plotting the number of surviving micro-organisms on a logarithmic scale and the duration of treatment on a linear scale, the so-called semilogarithmic plot (see figure 2).

In this way, either one or two straight lines are generally obtained (see figures 2 and 3).<sup>1</sup> The shape of the plot for a given strain of micro-organisms may vary according to the temperature and the test method used. Samples with natural heterogeneous microflora usually give a plot like the one shown in figure 3; as a rule there is a large population of micro-organisms with little heat resistance and a small population of highly heat-resistant micro-organisms.

#### Rahn's thermal destruction model

Assuming that the thermal death of micro-organisms is due to inactivation of a critical molecule in the cell, which is a first-order reaction, the number of micro-organisms  $N_t$  after time t is  $dN_t/dt = -kN_t$ , where k is the reaction rate

Plots can also be parabolic or sigmoidal.

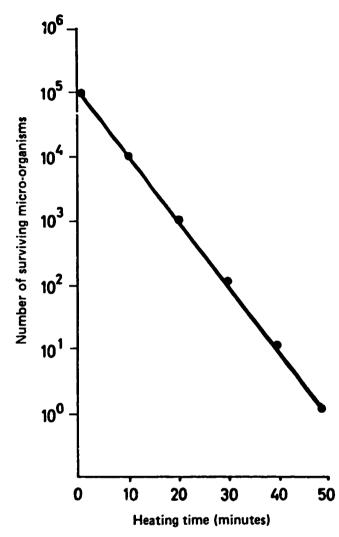
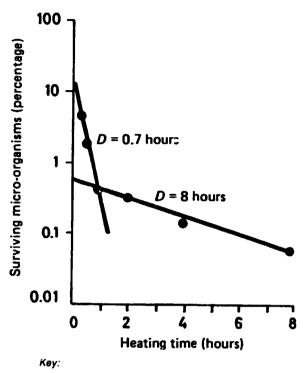


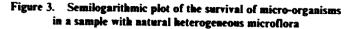
Figure 2. Semilogarithmic plot showing the number of micro-organisms surviving heat treatment as a function of time

Source: Pflug and Holcomb [1].

(assumed constant). The following expression is obtained by integration:  $N_I = N_0 e^{-kI}$ , where N is the initial number of micro-organisms. Symbolizing the time spent at the treatment temperature by U and introducing a new constant D by the substitution  $K = 1/D \log e$ , we have  $\log N_U = -(U/D) + \log N_0$ . The semilogarithmic plot of  $N_U$  against U will be a straight line with a slope of -1/D. The constant D is seen to be the time necessary for the population N to be reduced to N/10, or for the plot to be shifted by one logarithmic cycle—that is to say, the time needed to destroy 90 per cent of the bacterial population existing at the time treatment is begun. Approximately one third of the plots determined experimentally for homogeneous cultures fit Rahns's model.

I





D: Time necessary for population N to be reduced to N/10, or time needed to destroy 90 per cent of the population N.

Source: Pflug and Holcomb [1].

## Temperature coefficient

The temperature coefficient is normally defined as the change in the rate of thermal destruction that occurs with a change in temperature of 10 kelvins:  $Q_{10} \equiv k_{T+10}/k_T$ , where T is the absolute temperature. The range of  $Q_{10}$  values in bacterial thermal destruction processes is given as follows by Pflug and Holcomb [1]:

Dry heat	2.2-4.6
Moist heat	6.8-100

If the logarithm of the time necessary to destroy a given number of microorganisms is plotted against temperature, a straight line is obtained, representing the thermal destruction time (TDT) function  $F_T$  (see figure 4), the equation for which is  $\log F_T^Z = (1/Z)(T_B - T) + \log F_{T_B}^Z$ , where  $T_B$  is the base temperature and Z is the temperature change that produces a ten-fold increase or decrease in  $F_T^2$  It is related to the temperature coefficient  $Q_{10}$  by the equation  $Z = 10/\log Q_{10}$ .

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<sup>&</sup>lt;sup>2</sup>It is also the temperature change needed to change D by a factor of 10. If log D is plotted against T, the straight line obtained is known as the "heat resistance plot" [1].

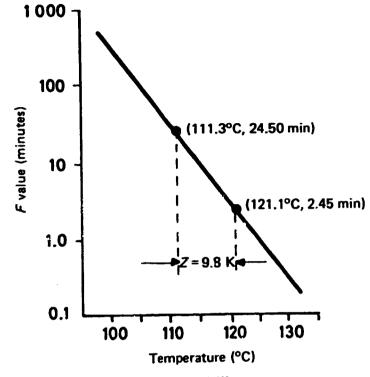


Figure 4. Thermal destruction time plotted against temperature

Source: Townsend, Esty and Baselt [2].

The F and Z values define the behaviour of micro-organisms under u given heat treatment. The TDT at a given temperature can be obtained graphically from the TDT plot or by calculation from the equation above (see Costell and Durán [3]).

The range of Z fc- processes for the thermal destruction of spores are as follows [1]:

Dry heat	15-30 K
Moist heat	5-12 K

Compared with hot air, and on an equimolar basis, saturated steam at 121° C provides at least seven times the heat provided by air at the same temperature. However, not air takes about 2,000 times longer than steam to destroy *Bacillus subtilis* var. *niger* at that temperature.

# Factors affecting the thermal destruction of micro-organisms

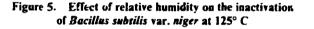
Different forms (breeds and strains) of the same species may produce cells or spores with varying heat resistance in the same medium and under identical treatment conditions. Environmental conditions during the formation of cells and spores also result in similar differences. The conditions of the medium during treatment are of the utmost importance. A pH range of 6.0-8.0 normally indicates the lower heat resistance of the micro-organisms. Variations within this range are not usually very great, but there is a sharp fall in the region of pH 5.5. Ŕ

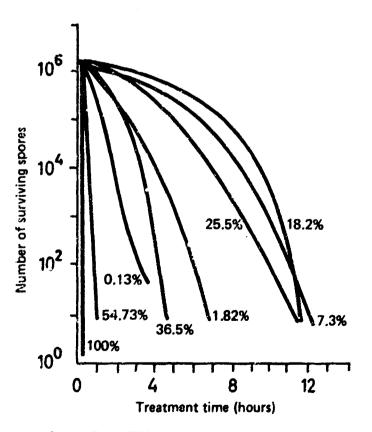
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Materials such as rice and bran harbour some micro-organisms that are easily destroyed by dry heat and some, especially those originating in the soil, that are difficult to destroy. The normal microflora of the soil are variable and heterogeneous, and the plot for micro-organisms surviving heat treatment resembles the plot with two straight lines (see figure 3). Therefore the D value cannot be used, because it is a parameter of a single straight line.

Research based on model systems, using laboratory spores, has shown that there are three primary variables for the mode of action of dry heat—temperature, water content and time—and three secondary variables—open or closed systems, physical and chemical properties of the micro-organism, and the gaseous atmosphere. Temperature is the most important variable in the case of dry heat.

In the 90°-125° C temperature range, spores with an intermediate water content (in equilibrium with a relative humidity content of between 20 and 50 per cent) are more heat-resistant (higher D value) than those with a higher or lower water content (see figure 5); under these conditions, Z is about 21 K whereas it is 8-10 K when relative humidity is 100 per cent (moist heat) [1].







The desruction of micro-organisms by moist heat is characterized by 100 per cent relative humidity, and consequently some water is present in the liquid state. In the case of dry heat, on the other hand, there is no water in the liquid state and the relative humidity value may be anything less than 100 per cent. Since the rate of destruction of dried microbiological cells is a function of their water content, which is in turn determined by the relative humidity of the atmosphere, the rate of destruction varies with relative humidity, which should therefore be specified, along with the treatment temperature, whenever dry-heat rates are quoted. Under conditions of equilibrium, the relative humidity of the atmosphere surrounding the cell is theoretically equivalent to the water activity  $a_w$  within the cell. If the relative humidity for the treatment is known, this should be used and not the water content.

The importance of controlling the moisture content during heat treatment is thus obvious. The main parameters for "closed" systems—where the material is hermetically sealed—are the initial moisture content and the volume of the container or receptacle system. In the case of "open" systems, the material to be treated can lose an unlimited amount of moisture or absorb it, if the system is drier than the atmosphere.

Both the temperature and the water content of the spores during the period of treatment must be known and controlled when studying the destruction of micro-organisms by dry heat, otherwise the results have no significance.

#### Destruction of micro-organisms by ionizing radiation

## Effects of ionizing radiation

At the present time ionizing radiation is too costly a means of destroying micro-organisms in foods. The sensitivity of products to radiation and the lack of suitable toxicological tests are additional obstacles. The method nevertheless warrants at least a brief discussion.

Radiation may be electromagnetic or it may involve particles. Ultraviolet radiation, gamma radiation ( $\gamma$ -rays) and X-rays are of the first type, and  $\alpha$ -rays,  $\beta$ -rays, neutrons, mesons, positrons and neutrinos are of the second type,  $\beta$ -rays (electrons) being the ones most widely used. X-rays and  $\gamma$ -rays have considerable penetrating power, as do electrons, if they are artificially accelerated.

These forms of radiation produce ions, free radicals and excited molecules in the product that absorbs them. Owing, however, to its discrete nature and its ability to penetrate materials, the effects of radiation are precise and localized. Secondary effects, due to the action of electrons with sufficient energy, increase the extent of the areas affected but continue to be localized. These reactions are very rapid: primary ionization in water occurs within  $10^{-18}$  to  $10^{-16}$  seconds and secondary ionization within  $10^{-12}$  to  $10^{-11}$  seconds; molecular products appear within about  $10^{-7}$  seconds.

Natural compounds undergo various chemical changes when they are subjected to ionizing radiation. Irradiated water, for example, may contain appreciable amounts of the free radicals H and OH, which react to form  $H_2$ ,  $H_2O$  and  $H_2O_2$  and react with dissolved substances. In the presence of oxygen

 $\mathcal{O}$ 

appreciable amounts of the hydroperoxide radical  $HO_2$  are produced. This radical and the hydrogen peroxide ( $H_2O_2$ ) produced may act as either oxidizing or reducing agents.

## Plot of the relationship between the destruction of micro-organisms and the radiation dose

Plots representing changes in the logarithm of surviving micro-organisms as a function of the radiation dose can be parabolic, straight-line or sigmoidal (see figure 6). The equation for a straight-line plot is  $N/N_0 = e^{-(d/d_0)}$ , where  $N/N_0$  is the fraction of micro-organisms surviving dose d, and  $d_0$  is the lethal dose, i.e. the dose resulting in the destruction of 63 per cent of the microorganisms. The term  $d_{10}$ , which is equivalent to the dose necessary to reduce the

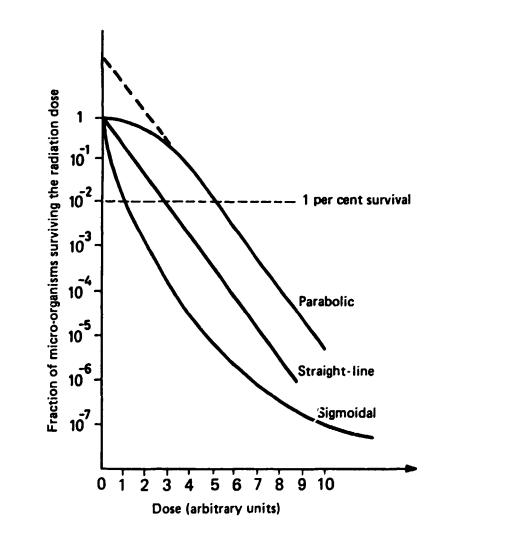


Figure 6. Plots showing the destruction of micro-organisms by ionizing radiation

Source: Silverman and Sinskey [5].

microbiological population by one logarithmic cycle, is used to express the rate of survival. It is easily obtained when the destruction plot is a straight line, but it is more difficult to interpret from curves. A number of equations have been proposed for such cases [5].

# Resistance of micro-organisms to ionizing radiation

The resistance of micro-organisms to ionizing radiation varies significantly. The spores of bacteria, with very few exceptions, are the most resistant, gramnegative rods are the most sensitive, while yeasts and moulds have intermediate resistance levels (see table 1). It should be noted in this context that certain circumstances (such as the physiological state of the micro-organism and the composition of the medium) may give rise to variations in resistivity. Products in which the microflora are not distributed uniformly may require an additional dose.

Species	d <sub>10</sub> (megarads)	Medium	
Anaerobic spore cultures			
Clostridium botulinum			
Type A NCTC 7272	0.12	Water	
Type B 53	0.33	Buffer	
Clostridium welchii (perfringeus)			
Type A	0.12	Water	
Type F	0.20	Water	
Bacillus subtilis	0.06	Saline plus gelatine (5 per cent)	
Vegetative bacteria			
Salmonella typhimurium	0.02	Phosphate buffer	
Pseudomonas sp.	0.003-0.006	Phosphate buffer	
Yeasts			
Saccharomyces cerevisiae	0.05	Saline plus gelatine (0.5 per cent)	
Moulds			
Aspergillus niger	0.047	Saline plus gelatine (0.5 per cent)	
Penicillium notatum	0.02	Saline plus gelatine (0.5 per cent)	

TABLE I. RESISTANCE OF VARIOUS BIOLOGICAL UNITS TO IONIZING RADIATION

#### Process requirements for complete destruction

(megarads)

Inactivation of enzymes Disinfestation of insects	2.0-10.0 0.1-0.5	
Clostridium botulinum	0.37	Canned chicken

Source: Silverman and Sinsky [5].

Note: Coleoptera are more sensitive to gamma irradiation than lepidoptera: 25,000 rads exterminate all phases of the coleoptera Sitophilus oryzae and Tribolium confusum, but over 100,000 rads are required to sterilize the lepidoptera Sitotroga cerealella [6].

A much higher radiation dose is needed to inactivate enzymes and destroy the ability of micro-organisms to reproduce (see table 1) and therefore some products require further heat treatment in order to become completely stable.

Treatment with 4-5 megarads impairs the quality of foods.

#### Inactivation of enzymes

#### Fundamental principles

## Denaturation of enzymatic protein and loss of activity

Enzymes are proteins. When a protein molecule in its secondary, tertiary or quaternary structure undergoes a change, other than the splitting of covalent bonds, it is said to have been denatured. Denaturation consists of the splitting of bonds with a hydrogen bridge, hydrophobic interaction and saline bridges, and unfolding of the protein [7]. One of the main consequences of denaturation is partial or total loss of enzymatic activity.

The sensitivity of a protein to denaturation is determined by the facility with which the denaturing agents affect the integral three-dimensional structure of the molecule. As enzymes vary in structure, their sensitivity to different agents and the changes which they undergo must also vary considerably. That is to say, a particular form of treatment will not affect lipase and lipoxigenase in the same manner. Similarly, denaturation under varying conditions also leads to different types of changes.

#### Reversible and irreversible inactivation

Denaturation of enzymatic protein may be irreversible if the conditions of treatment are sufficiently harsh, or reversible if they are sufficiently mild. In the latter case, an equilibrium constant K may be defined for the reaction E (enzyme) =  $E_d$  (denatured enzyme), which would be obtained from the equation  $K = [E]/[E_d]$ . The change in free energy  $\Delta F$  would be given by the expression  $\Delta F^\circ = -2.3 RT \log K$ , where  $\Delta F^\circ$  is the difference in free energy between a system where all the reagents are found in equimolecular amounts, R is the gas constant and T the absolute temperature once a state of equilibrium has been reached. If K > 1,  $\Delta F^\circ$  is negative and denaturation approaches equilibrium. If K < 1,  $\Delta F^\circ$  is positive and denaturation moves away from a state of equilibrium [8].

#### Activation energy

A definite amount of energy  $E_a$  is required to activate the molecules to enable a chemical or biological reaction to take place, as given by the Arrhenius equation  $k = A \exp(-E_a/RT)$ , where k is the rate of reaction and A (known as the "frequency factor"), as well as  $E_a$  (known as the "activation energy"), is an empirical constant. By converting to logarithmic form and integrating between the limits  $T_1$  and  $T_2$ , the following expression is obtained:

$$\log (k_2/k_1) = \frac{E_a}{2.3 R} \cdot \frac{T_2 - T_1}{T_2 T_1}$$

The activation energy is determined by plotting log k against 1/T, the slope being equal to  $-0.052 E_a$  (joules). The activation energy for the denaturation of proteins is much higher than that for other chemical reactions.

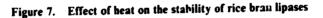
# Denaturing (or inactivating) agents

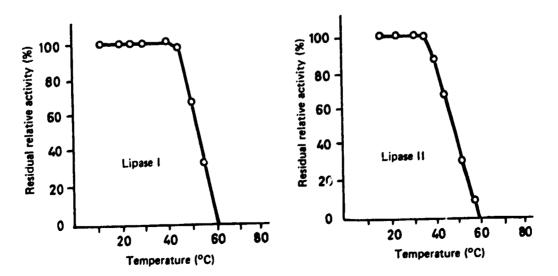
Enzymatic proteins may be denatured by physical and chemical agents. The main physical denaturing agent is heat; other agents include ionizing radiation, ultraviolet radiation, pressure and interfacial tension.

# Inactivation of enzymes by heat

# Thermal stability of enzymes

Aizono and others [9, 10] studied the variation in the stability of lipases I and II, when separated from rice bran and heated (see figure 7). The enzyme was incubated at various temperatures at pH 6.5 for 15 minutes in the presence of CaCl<sub>2</sub>. In the case of lipase I, the residual activity was determined using tributyrin under N<sub>2</sub>, at pH 7.5 and 35° C. For lipase II, the activity was determined in a similar manner, but at 20° C. Lipase I was shown to be stable below 40° C and lipase II below 30° C (see figure 7). Lipase I ceased to be active when treated at 60° C for 15 minutes and lipase II at 57° C for the same period of time. It should be noted that the thermal stability of pure enzymes is different from that found *in situ* in the bran, where they are protected by other constituents. At the same time, enzymes in solution do not exhibit the same stability as in the dehydrated form or with intermediate moisture content levels (see below).





Source: Aizono and others [9, 10].

#### Thermal inactivation plot

The rate of protein denaturation depends largely on temperature. Whereas for most chemical reactions the rate of reaction is doubled for every 10 K increase in temperature, the rate of protein denaturation may increase 600 times over the same interval.

The dependence on temperature (above a certain critical value) is the result of two opposite effects: first the increase in the rate of enzymatic reaction and secondly the increase in the rate of denaturation of the enzymatic protein. The rate of denaturation increases much more rapidly than that of the reaction catalysed by the enzyme. In general, enzymes start to undergo thermal denaturation above 45° C.

An equation analogous to the one used to determine the variation in the rate of thermal destruction of micro-organisms with temperature may also be used here. Figure 8 demonstrates this relationship between thermal inactivation and temperature for enzymes in the potato. The value of Z (that is to say, the temperature increase necessary to reduce D to 10 per cent of its original value) for each enzyme can easily be determined from the plots. It can be seen that the most heat-resistant enzymes have higher Z values—the slope of the straight line is less. This means that, in general, the more heat-resistant enzymes are less dependent on temperature (their activity varies less) than the less heat-resistant enzymes.

The activation energy E, which can be determined from Z, for enzymatic catalysis is 25-65 kJ/mol, while that for enzymatic denaturation varies between 200 and 630 kJ/mol. This means that the enzyme will be relatively stable at lower temperatures, but that denaturation will take place very rapidly at higher temperatures, since a relatively large number of molecules will have sufficient energy to attain the denatured state [12].

The thermal inactivation plot for heterogeneous enzymatic systems may not be a straight line, but may assume other forms, as is the case with thermal destruction curves for micro-organisms.

## Indicators of the efficacy of thermal inactivation

Residual enzymatic activity is an indicator of the efficacy of heat treatment. In systems containing various enzymes of different degrees of stability (as in the case of bran), it is advisable to select as an indicator the most resistant of the enzymes present, or at least of the enzymes that would not subsequently play an undesirable role if not totally inactivated. Peroxidase is one of the enzymes found in plant foodstuffs that shows high resistance to heat. For this reason (and in order to be able to use simple and precise methods of determination), it is used in food technology as an indicator of the effects of heat processes. Inactivation of peroxidase presupposes inactivation of any other enzyme. This criterion is used for many industrial food processes and is not limited to vegetable foodstuffs. In milk, for example, the conditions necessary for the destruction of peroxidase also ensure the destruction of lipase (see

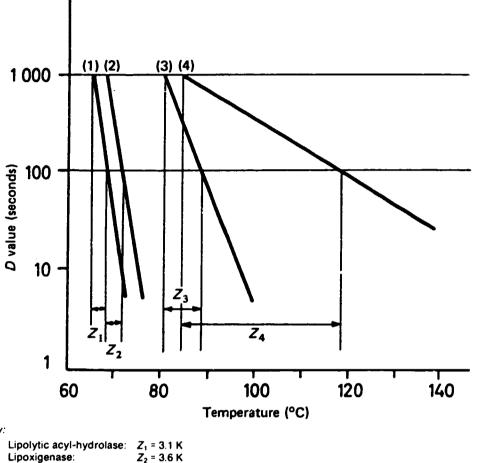


Figure 8. Semilogarithmic plots showing the relationship between thermal inactivation of various enzymes and temperature

Key:

(1)

(2)

(3) Polyphenoloxidase: Z<sub>3</sub> = 7.8 K

(4) Peroxidase: Z₄ = 35 K

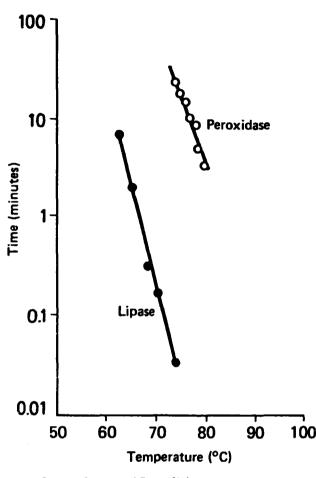
D value = Duration of treatment necessary to reduce enzyme activity to 10 per cent of its original value Z = Temperature increase necessary to reduce D to 10 per cent of its original value

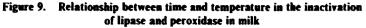
Source: Svensson [11].

figure 9). Peroxidase has been successfully used as an indicator of efficacy in the stabilization of rice bran [13].

It should be noted, however, that the heat resistance of lipases of various origins varies a great deal, and there are some extremely resistant lipases produced by micro-organisms. One of these is produced by the microorganism Pseudomonas fluorescens, which is much more heat resistant than the thermostable fraction of potato peroxidase (see figure 10) [11]. The possible existence of such cases must be taken into consideration when selecting a reliable indicator. As an example, the figure includes the temperature-time range representing F values between 2.7 and 10. At temperatures above  $120^{\circ}$  C the sterilization time is much less than that required for 90 per cent inactivation of )

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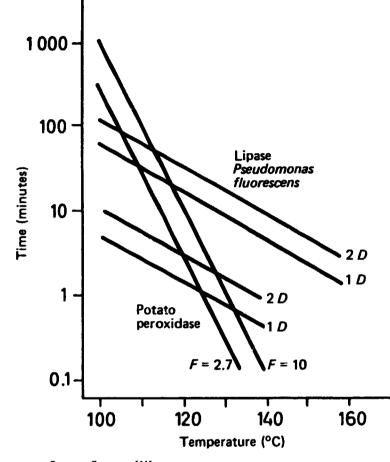
Source: Jenness and Patton [14].

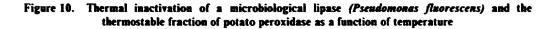
the enzyme (D curve), which leads to insufficient enzymatic inactivation in HT-ST processes, that is to say, in processes involving high temperature and short treatment time. A combination of treatments at a lower temperature for longer periods of time (to inactivate enzymes) and HT-ST treatments (to destroy micro-organisms) may be advisable when highly heat-resistant enzymes are present ( $Z \ge 10$  K).

## Factors affecting thermal inactivation

#### Moisture content

The susceptibility of proteins to thermal denaturation depends on various factors: moisture content, pH, ionic concentration and the type of ions present. The effect of water is extremely important. For example, wheat germ is denatured to approximately the same extent at  $60^{\circ}$  C and 24 per cent moisture content as at  $70^{\circ}$  C and 18 per cent moisture content [5]. Enzymes are also more stable under heat when less water is present. Relevant data have been





Source: Svensson [11].

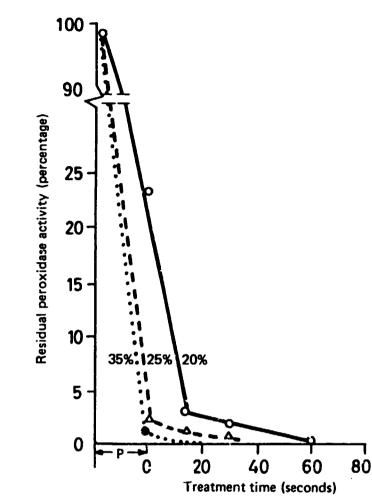
published [13] for rice bran peroxidase in the 20 per cent to 35 per cent moisture-content range (see figure 11). Information has subsequently been obtained for three temperatures (90° C, 100° C and 110° C) in the 6-18 per cent moisture-content range for rice bran peroxidase and lipase (see figures 12 and 13).

## pН

The thermostability of enzymes also depends on the hydrogen ion concentration. The heat resistance of the enzyme is normally greater at pH values near its iso-electric point. The effect of pH on the thermal inactivation of rice bran peroxidase at 121°C and 35 per cent moisture content is shown in figure 14.

## Initial enzymatic activity

Because of the laws governing the thermal inactivation of enzymes and the desirability of eliminating all enzymatic activity, initial enzymatic activity plays an important role in stabilization. It partially determines how much residual ì





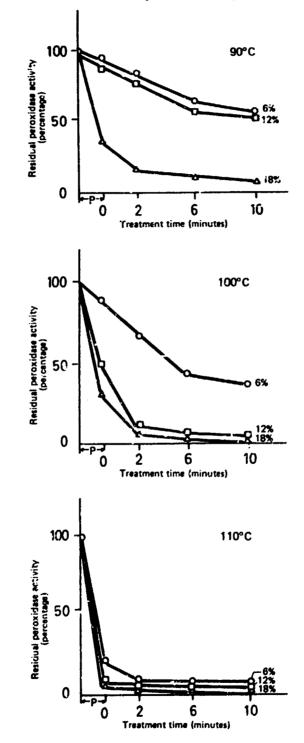
Source: Barber and others [13].

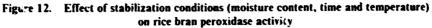
Note: P represents the pre-heating time necessary to reach the required temperature of 95° C. The percentages on the curves are the moisture content of the bran on wet weight basis.

activity there will be after treatment, as shown in figure 15. The combination of time and temperature needed to reach a given degree of inactivation varies with the level of initial enzymatic activity. This means that inactivation that may be suitable for one batch of bran may be inadequate for another.

# Regeneration of enzymatic activity

Some enzymes (peroxidase. lipoxigenase, trypsin) are able to recover their catalytic activity after they have been inactivated by heat. Regeneration takes place during storage after the inactivation treatment has been carried out. The extent of the reactivation depends on the conditions under which the heat treatment was carried out and on the storage temperature. Recovery of catalytic activity is proportional to the rate of inactivation. Processes that combine high temperatures with short treatment times (high rates of inactivation)





Source: Institute of Agrochemistry and Food Technology, Valencia, Spain, unpublished results by V. Cordero and others.

*Note:* P represents the pre-heating time necessary to reach the required temperature of  $95^{\circ}$  C. The percentages on the curves are the moisture content of the bran on wet weight basis.

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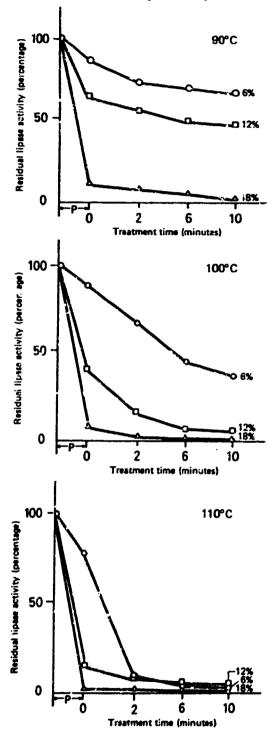
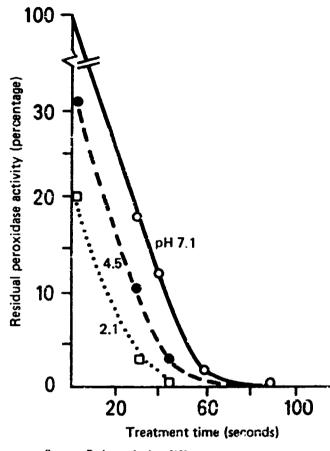


Figure 13. Effect of stabilization conditions (moisture content, time and temperature) on rice bran lipase activity

Source: Instituto de Agroquimía y Tecnología de Alimentos, Valencia, Spain; unpublished results by V. Cordero and others.

Note: P represents the pre-heating time necessary to reach the required temperature of 95° C. The percentages on the curves are the moisture content of the bran on wet weight basis.

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#### Figure 14. Thermal inactivation of rice bran peroxidase: effect of pH

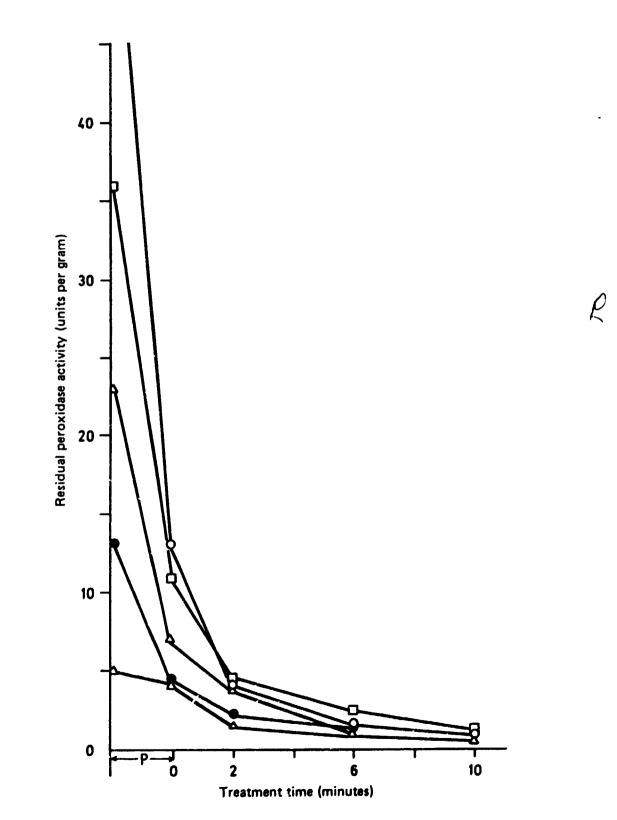
Source: Barber and others [13]. Note: Temperature, 121° C; humidity, 35 per cent, wet weight.

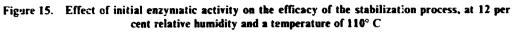
are more conducive to the recovery of enzymatic activity than others using lower temperatures for longer treatment times (lower rates of inactivation) for the same inactivation percentages (see figure 16).

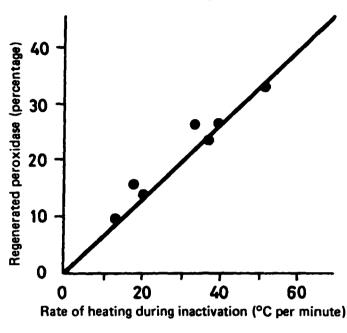
There are various forms of reversible loss of enzymatic activity by heat treatment and subsequent recovery following storage at lower temperatures. These are due to: (a) changes in the secondary and tertiary structures of the enzymatic protein; (b) dissociation of the sub-units of the quaternary structure; and (c) dissociation of the essential co-factors for enzymatic action [12].

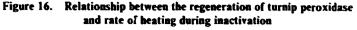
## Effects of water activity on enzymatic activity

The dependence of enzymatic activity on water activity or on relative humidity has already been discussed. In general, enzymatic activity is only significant above the area of monomolecular adsorption. The amount of free water serving as a vehicle for enzymatic processes increases under these conditions. It has already been noted, however, that, if the substrate is sufficiently mobile to combine with the enzyme, the reaction may take place at









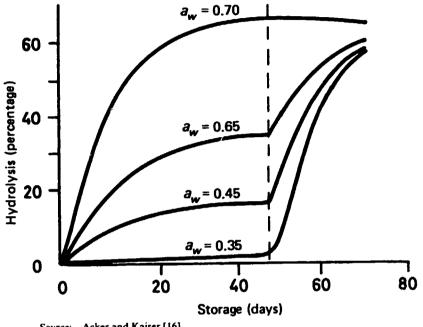
Source: Richardson [15].

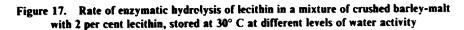
water activity levels below the monomolecular zone. Such is the case with lipolysis of liquid unsaturated triglycerides (oils). In any case, very low values may reduce enzymatic activity to practically zero. When water activity starts to increase again, however, the activity recovers proportionately (see figure 17). It should be noted that solid substrates may be attacked at very low water activity values if they are in close contact with the enzyme [15].

#### Effects of pH on enzymatic activity and stability

The pH value is one of the most important variables in chemical denaturing agents. Enzymes are usually stable in the realtively narrow pH range within which their optimum pH occurs (see figure 18). Extreme pH values reduce the activity of the enzyme; this may first of all lead to reversible and subsequently to irreversible inactivation. Significant ionization changes take place in free amino-acid functional groups, which cause a distortion of the three-dimensional structure of the protein molecule. If no major changes occur, recovery may result in a re-forming of the original structure. Otherwise, irreversible denaturation and inactivation may occur [7, 17, 18, 19].

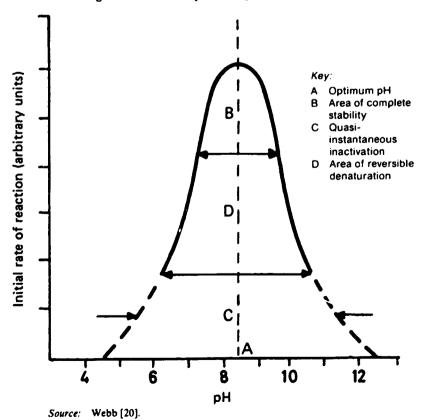
Aizono and others [9, 10] have studied variations in the stability of rice bran lipases I and II with pH (see figure 19). In one case—lipase I—a 66-mM enzyme solution in a buffer with varying pH values was incubated at 8° C for 26 hours and dialysed against 0.5 mM CaCl<sub>2</sub> to remove salts. Moreover, the pH values of the enzyme solutions containing 0.3 mM CaCl<sub>2</sub> were adjusted with 0.1 N NaOH or 0.1 N HCl every three hours throughout the 26-hour



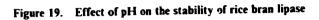


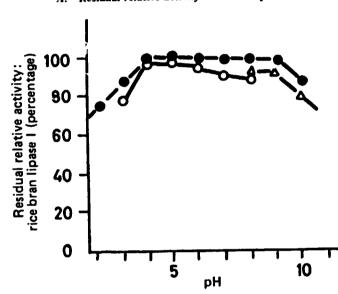
Source: Acker and Kaiser [16].





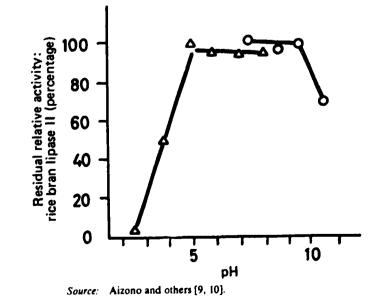
incubation period at 8° C under nitrogen. Residual activity was determined using tributyrin as substrate. In the other case—lipase II—a 34-mM enzyme solution in buffers of varying pH was incubated at 10° C for 5.5 hours. Following incubation, the enzyme solutions were dialysed against 0.5 mM CaCl<sub>2</sub> at pH 6.5. Residual activity was determined at pH 7.5 and 25° C. The results obtained in both cases indicated that lipase I was stable in the pH range between 4 and 9, lipase II was stable between 5 and 9.5 (see figure 19).





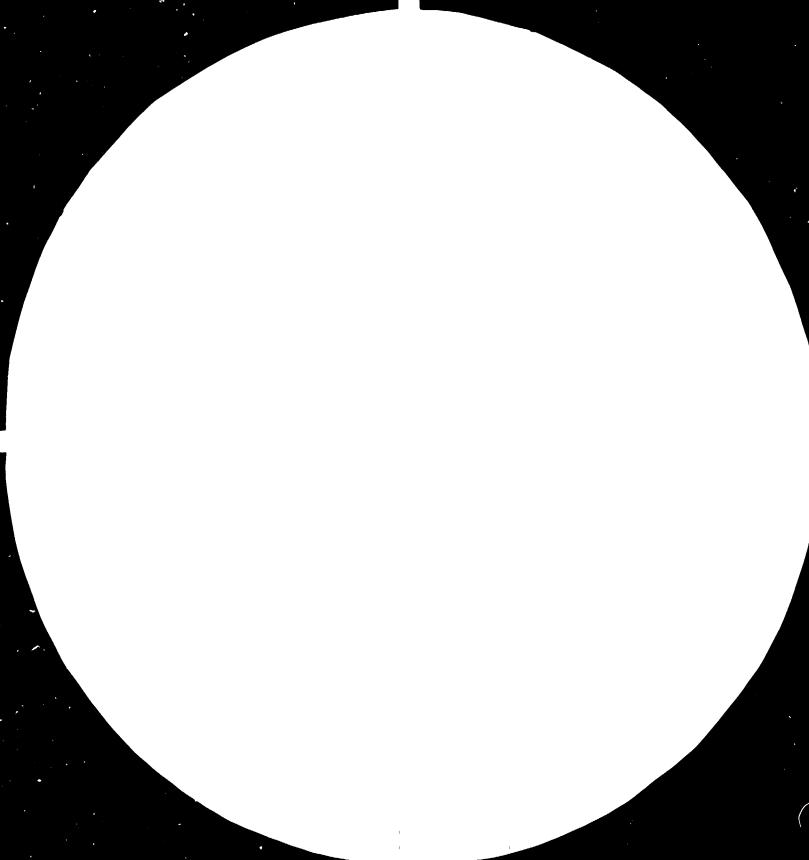
A. Residual relative activity: rice bran lipase I

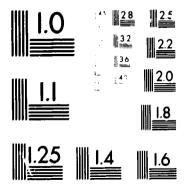
B. Residual relative activity: rice bran lipase II



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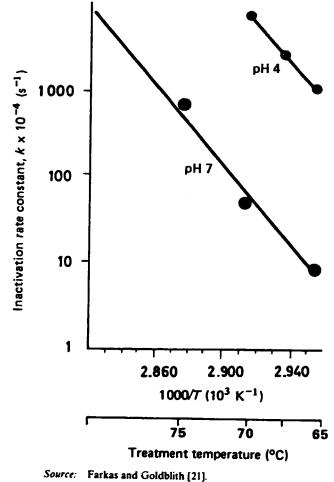




#### MIGROCOPY RESOLUTION TEST CHART NATIONAL BUREAU OF STANDARDS STANDARD REFERENCE MATERIAL 1010a ANSL and ISO TEST CHART No. 21

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A combination of physical agents (e.g. heat) and chemical agents (e.g. pH) is common (see figure 20).





## Other thermal inactivating agents

Electrolytes and ionic strength may act as denaturing agents. Ions from heavy metals  $(Ag^+, Hg^{2+}, Pb^{2+})$  normally have poisonous effects on enzymes. An ion may nevertheless inactivate one enzyme and activate another, or activate one enzyme at one particular concentration and inactivate it at another. High concentrations of electrolytes (brine) therefore inhibit enzymatic action, while low concentrations of Ca<sup>2+</sup> are essential for lipase activity.

The pressure and shearing forces have an inhibiting effect but it is not practical to use them.

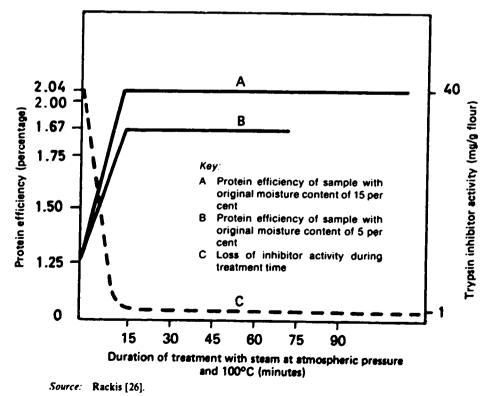
Enzymes may be inactivated by ionizing radiation. The radiation resistance of enzymes varies greatly, the necessary dose for inactivation *in situ* being greater than that required to destroy micro-organisms (see table 1). In general, inactivation by ionizing radiation depends among other things on enzyme concentration, water activity, pH and temperature. Enzymes are more resistant to radiation in the dry state. Inactivation is greater in the presence of water; it involves radiolysis of the water, as well as harmful effects due to the resulting free radicals. Higher temperatures are conducive to inactivation by irradiation. In any case, irradiation poses major economic as well as technical problems.

Proteins tend to become adsorbed in the interfaces, usually reflected in denaturation. The high interfacial tension that may occur on the water-air interface or water-oil interface seems to force the tertiary or secondary structure of the protein to unfold, so that the molecule spreads over the surface. Even lipase, which is believed to be especially well adapted to retain its activity in the water-oil interfaces, may be denaturated by adsorption in the water-triglyceride and water-air interfaces [22].

# Control of other harmful constituents of bran during stabilization

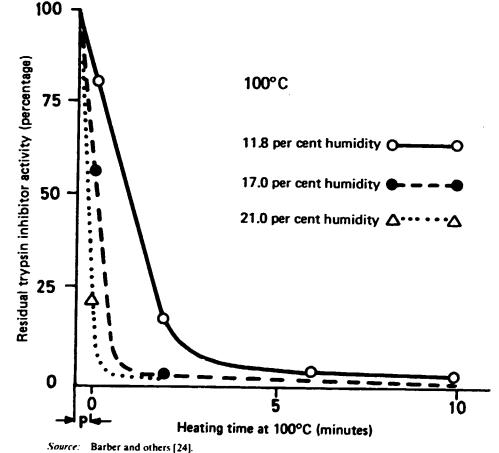
The stabilization process may severely damage insects and some of the antinutritive substances, for example trypsin inhibitors. Insects will not withstand the combination of time and temperature necessary for the destruction of micro-organisms and the inactivation of enzymes. In view of the high risk of infestation in bran, the destruction of insects in all vegetative forms is a very positive result of thermal stabilization. Moreover, trypsin inhibitors are thermolabile, and the right heat treatment will generally result in an increase in the nutritive value of protein [23]. An extensive study has been made of this question in relation to the soya bean. Figure 21 shows the results

Figure 21. Effect of steam treatment on the protein efficiency and trypsin inhibitor activity of raw soya meal



of the steam treatment of raw soya meal with moisture-content levels of 5 and 15 per cent at atmospheric pressure. In both samples over 95 per cent of the inhibitor is destroyed after 15 minutes, and the improvement in the nutritive value of the protein is somewhat greater in the sample with the higher moisture content. A study of the relationship between heat inactivation of trypsin inhibitors in rice bran and treatment time, temperature and moisture content [24] has shown that over 95 per cent of the activity is destroyed in less than five minutes at 100° C; the rate of inactivation increases with the moisture content of the bran (see figure 22). Similar results were obtained for the thermal destruction of haemaglutenins [25].





Note: Prepresents the pre-heating time necessary to reach the required temperature.

# Loss of valuable constituents of bran during stabilization

# Means of stabilizing bran and stability of the constituents

Heat is the most commonly used means of stabilizing bran, and the most promising. Chemical agents and ionizing radiation have also been tested. Bran has been subjected to various conditions, in order to discover the most effective ways of inactivating the enzymes, since the latter causes the bran to deteriorate during storage. Unfortunately, in inactivating the enzymes, stabilization also impairs the valuable constituents (oil, proteins, vitamins and other nutrients), the degree of damage depending on the mode of treatment and the conditions under which it is carried out. Beneficial effects (inactivation of enzymes, destruction of micro-organisms etc.) and harmful effects (reduction in the quality or quantity of valuable constituents) must therefore both be taken into account when selecting the means and conditions of treatment. The stability of the chemical constituents responsible for the functional properties and nutritional value of bran is not covered here, and other specialist works must be consulted. Nevertheless, some indication of possible damage is essential. Although stabilization is mainly considered from the point of view of the subsequent extraction of oil from the bran, the possible use of other constituents—proteins, for example—as nutrients is also examined.

## Effects of various means of stabilization on the valuable constituents

## Oi!

Decolorization is an important stage in the extraction of edible-grade oil from bran. Oil must comply with certain specifications, including colour, in order to be considered edible and marketable. The facility with which crude oil can be decolorized in the refinery is an important indicator of the acceptance or rejection of the crude oil for industrial purposes.

Heat treatment darkens the colour of crude oil extracted from the bran with hexane. The extent of the deterioration depends on the conditions of treatment: the harsher these are, the greater the deterioration. It should, however, be pointed out that in industrial practice a change in the colour of crude oil is less important than a change in the ease with which it can be decolorized. That is to say, the final criterion for evaluating deterioration is not the darkening of the colour of crude oil, but whether or not it can be decolorized, and the ease with which this can be done.

Heat treatment may lead to oxidation, which, besides resulting in a loss of essential fatty acids, has a number of other side effects (see figure 23). The resulting hydroperoxides formed may give rise to the formation of carbonyl compounds or to polymerization. The free radicals and hydroperoxides may react with pigments, aromatic constituents, proteins and vitamins, producing new chemical components and reducing the nutritive value of the sulphur amino-acids and vitamins—such as tocopherols—by oxidation. Carbonyl compounds may reduce the biological value of proteins by combining with the epsilon amino group of lysine.

There are many analytical procedures for determining changes in lipids (see table 2). Again, as far as the extraction of edible-grade oil is concerned, emphasis must be placed not on the colour of the crude oil but on the ease or difficulty with which it can be decolorized. To this end, crude oil must be subjected to a decolorizing test under conditions simulating those used in industry, and its behaviour judged according to yield and final colour.

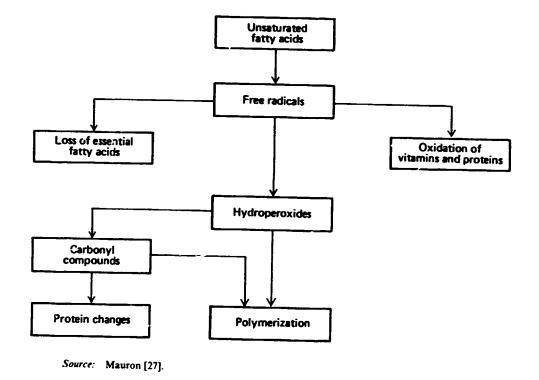


Figure 23. Paths of loss of nutrients due to oxidative changes in lipids

TABLE	2.	ANALYTICAL METHODS FOR DETERMINING	
		CHANGES IN OILS AND FATS	

Change	Methods	
Colour	lodine colour scale Lovibond colour scale Transmission, 360-500 nm	
Conjugation	Extinction at 232 nm Extinction at 268 nm Refractive index	
Polymerization	Iodine index Density Viscosity Gel permeation chromatography	
Oxidation	Peroxide index Epoxide index Aldehyde index Colour in alkali Oxidized fatty acids insoluble in light kerosene Volatile carbonyl compounds Liquid-liquid chromatography	

Source: T. Morton [28].

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#### Proteins

Five types of change in proteins during technological processes have been differentiated [29]. The first type includes changes in tertiary structure of the protein molecule, caused by mild heat treatment. They have no effect on the nutritive value, but do contribute towards changes in physical or chemical properties, for example in solubility, viscosity and the eletrophoretic mobility of the globular proteins. The second type includes the non-enzymatic browning or Maiilard reaction between lysine, through its epsilon amino group, and reducing substances (glucose, lactose, maltose and carbonyl compounds of oxidized fats). The resulting bond is not hydrolysed during digestion and the amino-acid, although present, is not available, so that the biological value is reduced. Other adjacent amino-acids that are not directly involved may also lose their ability to affect the new bond, owing to the specificity of the digestive enzymes. The third type includes harsher forms of heat treatment and implies a loss in the availability of other amino-acids, besides lysine, which may occur in the absence of reducing substances. Cystine may be converted at 115° C into compounds such as methyl mercaptan, dimethyl sulphide and methyl disulphide if the treatment time is sufficiently long. Protein-protein interaction (C-N bonds) may also occur above 110° C over a long period of treatment (measured in hours); new bonds are formed, such as =CHN=, which cannot be hydrolysed by the enzymes of the gastro-intestinal tract, with a consequent loss in nutritive value [30]. The fourth type of change includes deterioration under particularly harsh forms of treatment (toasting of cereals, for example), with the destruction of amino-acids by decomposition or racemization and the formation of crosslinkages producing polyamino-acids [31]. Treatment at 180°-300° C, as in the roasting of coffee and the baking of biscuits, may produce these effects. In one case [32], the baking of a biscuit at 200° C for 15-20 minutes lowered the protein efficiency ratio (PER) from 3.5 to 2.4, while cooking at 130° C for 40-60 minutes lowered it to 0.8. In another case [33], baking at 180° C for 13 minutes reduced the net protein utilization (NPU) by 72 per cent. The fifth type includes changes in an alkaline medium and those due to oxidation (already referred to in the previous section). Peroxides of lipids may react with aminoacid residues of proteins at high temperatures, so that their availability is reduced. Methionine may be affected. Oxidation may lead to the destruction of tryptophan and tyrosine.

#### Vitamins

The main vitamins in bran and related products are in the B group, in particular thiamine, and the tocopherols.

In general, the procedures tested for the stabilization of bran—heat, chemical compounds, ionizing radiation—affect its vitamin values.<sup>3</sup> Thiamine is unstable under heat at neutral and alkaline pH values, but is stable at an acid pH up to 120° C. Thermal destruction of thiamine leads to cleavage of the molecule and the formation of pyrimidine and thiazole rings. Secondary

<sup>&</sup>lt;sup>3</sup>Thiamin and tocopherols, like some other vitamins, exist in a variety of forms in rice byproducts; each has a different vitamin content and level of stability. Retention of the total vitamin value, following physical or chemical treatment, depends on the relative concentrations of the various forms present.

products are derived (including elemental sulphur, hydrogen sulphide and thiophene), which all have a peculiar smell. No data are available for bran, but thiamine losses of 5 per cent at pH 6.0-6.4, 25 per cent at pH 6.9 and 55 per cent at pH 7.5 have been recorded for various bakery products [34]. The retention of thiamine depends on time of exposure, temperature and moisture content. Thiamine losses of around 25 per cent at a moisture content of 20-30 per cent and over 70 per cent at a moisture content of i0 per cent have been recorded [35]. Losses may be significant if the cooking is done by extrusion; figures of 10 per cent and 40 per cent have been recorded under given standard conditions at 149° C, and these rose to 50 per cent and 80 per cent at 193° C.

Thiamine is destroyed by the action of  $SO_2$  (sulphite,  $HSO_3$ ); the molecule splits into two, in rather the same way as it does when exposed to heat. In this case the rate of destruction depends on the pH value. Deterioration is slow at a pH of 3, more rapid at a pH of 5 and instantaneous at a pH of 6 [29].

Tocopherols are destroyed under oxidizing conditions such as exposure to air and light, and their destruction is accelerated by heat. About 50 per cent of the tocopherols in flour are lost during the baking of bread [36]. Vitamin E activity also falls considerably if auto-oxidation of the lipids is significant.

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# III. Morphology, anatomy, histology and histochemistry of the discrete particles in commercial rice bran

## Introduction

Commercial bran is derived from milled rice, being composed of a mixture of quite distinct fragments (see figure 1). In view of the diversity of the machines used to produce them and of the conditions under which they are processed, it is not surprising that commercial brans should display very marked differences, even when they are derived from a single variety and are included in the same consignment of rice.

The anatomical layers of the grain that are separated during milling constitute the raw material from which bran is obtained. A knowledge of them is necessary for a full understanding of the preparation of rice and the nature of commercial bran, but because of the complexity of the transformations undergone by these anatomical layers during milling, and their effects, this knowledge is not enough to identify and determine the essential characteristics and properties of the bran, which are unique. Commercial bran and bran layers in the grain are two entirely distinct products, which must be studied individually.

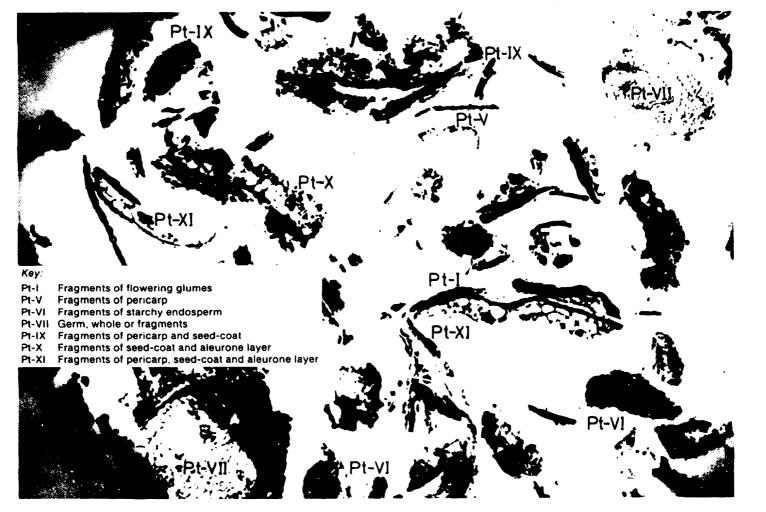
Most works on histology and histochemistry refer to bran in the form in which it appears in the caryopsis, not as a mixture of individual discrete particles. Very little research has been done on the mechanism by which bran particles are formed during milling, despite the fact that it is essential to know something about the subject not only in order to produce top-quality bran but also in order to improve the milling process.

In commercial bran produced from raw rice, more than 15 different types of discrete particles have been identified, in addition to a few fragments of covering material from the rubber-roll husking machines and of the abrasive coating of the whitening machines, fragments of other seeds and other materials. In the following description<sup>1</sup> they are divided into two major groups: simple particles, consisting of tissues or cells corresponding to a single anatomical structure, and compound particles, consisting of several of these<sup>2</sup> (see table 1) [1, 2].

<sup>&</sup>lt;sup>1</sup>Incorporating part of the research work for the doctoral thesis of L. Navarro Lucas and A. J. Pineda, carried out in the authors' laboratory.

<sup>&</sup>lt;sup>2</sup>The dimensions given for histological and histochemical preparations may be influenced by the preparatory techniques used, and should therefore be considered with reserve.

Figure 1. Particles in commercial rice bran showing heterogeneous composition



Source: Barber, Pineda and Benedito de Barber [1].

	Types of particle													
Anatomical structure	Simple					Compound								
	1	2	3	4	5	6	7	1	2	3	4	5	6	7
Flowering glumes														
Glumes	X													
Trichomes only	×												·	
Sterile glumes		X												
Pedicel			X											
Pericarp				×				×		×	X			X
Starchy endosperm					Х						X	X	X	
Germ						X							X	X
Fibres							X							
Seed-coat								×	×	X	х			×
Aleurone									×	X	××	X		X

TABLE I.	TYPES OF DISCRETE PARTICLES IDENTIFIED IN COMMERCIAL BRAN,	
	AND THEIR ANATOMICAL COMPOSITION	

Source: Pineda [2].

## Simple discrete particles in commercial bran<sup>3</sup>

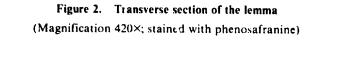
The simple particles are fragments of the flowering glumes, sterile glumes, pedicel, pericarp, starchy endosperm, germ (also whole) and fibres.

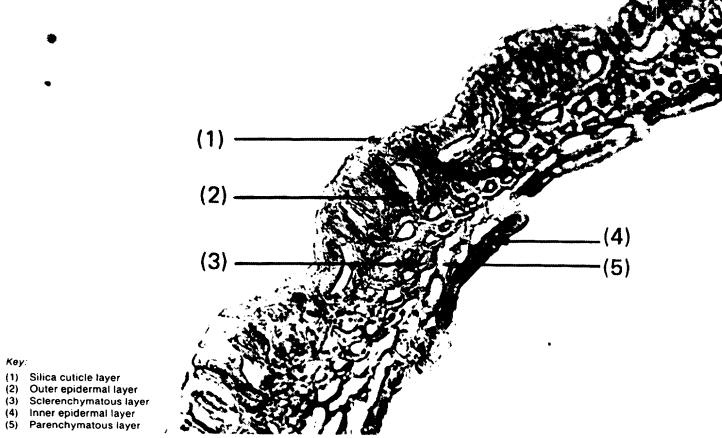
## Fragments of lemma and palea

The fragments of lemnia and palea found in commercial bran (see figures 2 and 3) are in the form of irregular polygons. The larger fragments measure about  $1.5 \times 2.5$  mm (although they may vary considerably, depending upon how fine the meshes of the whitener screen and subsequent sieves are) and are very rare; smaller fragments are more numerous, especially those measuring 25-40  $\mu$ m; the transverse section (see figure 2) is about 80-120  $\mu$ m thick. One of the faces is smooth and the other rough and wrinkled, with undulating folds and covered with trichomes.

The trichomes, both whole and broken, are inserted obliquely, at an angle of  $30^{\circ}-45^{\circ}$ ; they are more numerous along the nerves and sparser on either side of them. In addition, fragments consisting only of the nerve, 0.3-0.5 cm long and some 10  $\mu$ m thick, and other material derived from the awn of the lemma, have also been identified. Frequently, these particles of lemma and palea are found adhering to small fragments of starchy endosperm in the folds and between the trichome.

<sup>&</sup>lt;sup>1</sup>The commercial brans studied [1, 2] come from the *Japonica*, Bahia and Bahia × Sollana varieties of rice, cultivated in Spain. The processing programme of the industrial mill used consisted of: scales, cleaners (with air), rubber-roll husker, husk extractor, paddy separator, return husker (rubber-roll), whitening cones with abrasive coating (four successive units), graders and germ separator.





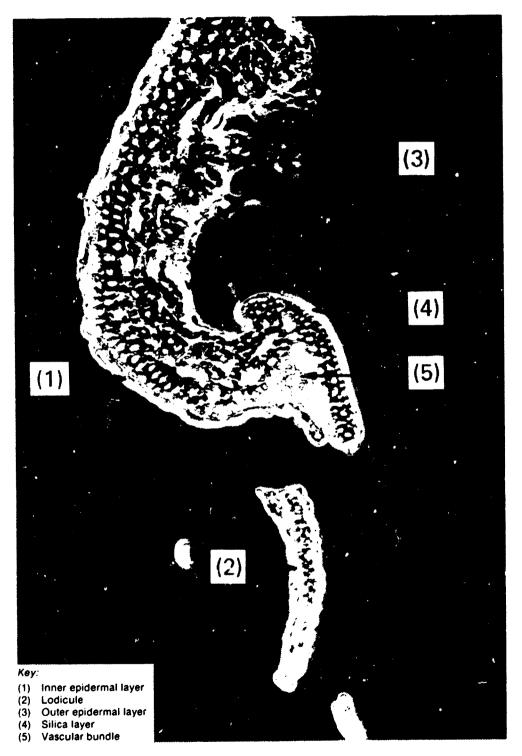
Source: Barber, Pineda and Benedito de Barber [1].

Key:

(4)

Figure 3. Transverse section of palea

(Magnification 270×)



Source: Barber, Pineda and Benedito de Barber [1].

The carbohydrates are contained in the cellular walls [1]<sup>4</sup> and are mainly in the form of cellulose, with smaller amounts of hemicelluloses, especially pentosans; they contain no starch [3]. Lignin is found in abundance in all the histological layers, but especially between the walls of the cells of the sclerenchymatous layer and outer epidermis and the vascular bundles [1, 4]. It is also found encrusted between the fibrils of the cellulose of the cell walls—the cells being mainly those of the outer epidermis and sclerenchymatous layer [2, 4]—and chemically combined with the hemicelluloses [7]. No proteins or fat globules have been detected. Wax and cutin are localized in the epidermal layer [1]. Silica has been found on the cuticle of the cutin, on the outer epidermis (where it forms a layer varying in thickness from 2 to  $6 \mu m$ ), in the inner epidermis (1  $\mu m$  thickness) and in the walls of the sclerenchymatous cells, vascular bundles and epidermis [3, 8].

Whole or fragmented trichomes that have become detached from the glumes are common components of commercial bran. In commercial samples of bran of the Bahia and Balilla × Sollana varieties, trichomes or trichome fragments 100-700  $\mu$ m long, with a maximum diameter of 40  $\mu$ m, have been found. Carbohydrates, lignin and lipids have been detected in the trichomes [1].

#### Fragments of sterile glumes

Fragments of sterile glumes, which generally come from cracks along the nerves, are not very numerous. They are yellowish-white and their outer surface is relatively smooth. Trichomes are present, of course, although not in great numbers, being much more characteristic of the flowering glumes (see also chapter I above). The fragments of sterile glumes are not of uniform size, the smallest being some 2 mm long. They vary in thickness (60-120  $\mu$ m), being at their largest where they are closest to the vascular bundle. Although a very rare occurrence, sterile glumes that are almost whole have also been found. In the particles analysed, three layers of the histological structure proper to the sterile glumes have been detected: outer epidermis, inner epidermis and intermediate parenchyma. The first is formed of a layer of elongated cells, 50-70  $\mu$ m long by 5-10  $\mu$ m wide, arranged tangentially; the outer side of the cell wall, 3  $\mu$ m thick, is covered by a thick cuticle (5  $\mu$ m). The inner epidermis is similar. The intermediate lamina has from two to four layers of cells of variable length (20-70  $\mu$ m) and more uniform width (some 7  $\mu$ m), with thinner walls than the epidermal cells [2].

The chemical components of the sterile glumes, and their distribution, are similar to those of the flowering glumes.

### Fragments of the pedicel

Pedicel fragments are normally very rare in commercial bran. They are cylindrical, elongated and of variable size (with a maximum length of 3 mm). Their surface is striated, and the cell walls contain carbohydrates and lignin [2].

<sup>&</sup>lt;sup>4</sup>Starch, cellulose, hemicelluloses, pectic substances, glycoproteins and glycolipids, which give a positive reaction with Schiff's periodic acid reagent (PAS) [5, 6].

#### Fragments of the pericarp

Most of the pericarp fragments identified in commercial bran (see figure 4) are elongated, varying in length (not more than 800  $\mu$ m) and of relatively uniform thickness (6-8  $\mu$ m). In the particles investigated, the tissue constituents were frequently compressed, forming a dense film, the constituent cell layers not being differentiated [2]. The histological characteristics are described in chapter I of the present publication.

The following have been identified in the pericarp: cellulose [9] and hemicellulose [1, 9], lignin [1], proteins [1, 9], lipids, cutin and waxes [1, 10], phytin [1, 4], anthocyanins [11] and silica [1]; lipase activity has also been identified [10]. The PAS reaction for carbohydrates [5] is positive in the cell walls of the pericarp, with the exception of the transverse cells [1]. The lumen of the epidermal and tubular cells stains likewise. It has been noted that such cells are empty [4]. Nc starch has been detected in the pericarp [1, 4]; cellulose and the hemicelluloses appear to be the main carbohydrates [9]. Lignin is found in the cell walls of the pericarp, where it is easily identified with Schiff reagent; on the other hand, the Schiff reagent does not show the presence of lignin in the tubular cells [1], even when they are known to be lignified [4]. Proteins, as indicated by the Hg-BPB reagent [12] can be found in the hypodermis and in the transverse cells [1]. Their localization in the cell walls of the pericarp has been noted by various authors [9, 13]. A film of lipoidal material, about 1  $\mu$ m thick, has been found on the epidermis, using Sudan black [1]. Cutin has been detected in the pericarp [14]. The lumen of the pericarp cells likewise stains with Sudan black, since these are empty cells with no fat globules.

#### Fragments of the starchy endosperm

Fragments of the starchy endosperm are found in great numbers in commercial bran (see figure 5). They vary in size from a few micrometres to more than 1 mm, the larger ones depending upon the mesh size of the screen of the whitening machine and subsequent sieves, if any. The cell fragments are among the smallest particles. The cells of the starchy endosperm particles have an irregular pentagonal or hexagonal section. The former are usually smaller  $(30 \times 25 \,\mu\text{m}, \text{ as against } 60 \times 40 \,\mu\text{m})$ . The walls of the cells along the torn surface are generally broken. The cracks seen in many of the fragments are thought to be the membrane covering the starch granules and crossing the juxtaposed cell walls.

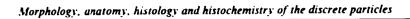
In addition to starch and proteins, cellulose, hemicellulose and lipids have also been identified in fragments of the starchy endosperm of commercial bran. While starch is found in the lumen of the cells, cellulose and hemicelluloses are found in the cell walls [1]. The proteins in the starchy endosperm exist not only as protein bodies [15, 16] but also as cementing material between the protein bodies and the compound starch granules [9, 17] and as a lipoprotein membrane surrounding the starch granules. In the endosperm, the protein bodies measure  $1-5 \mu m$  in diameter [15, 16, 17, 18]. The fats are located between the compound starch granules and in the membrane surrounding both simple and compound granules. The fats in some particles of the starchy endosperm are barely stained by Sudan black [1]; these particles probably come from the central portion of the endosperm, where their presence has not been detected histochemically [19].

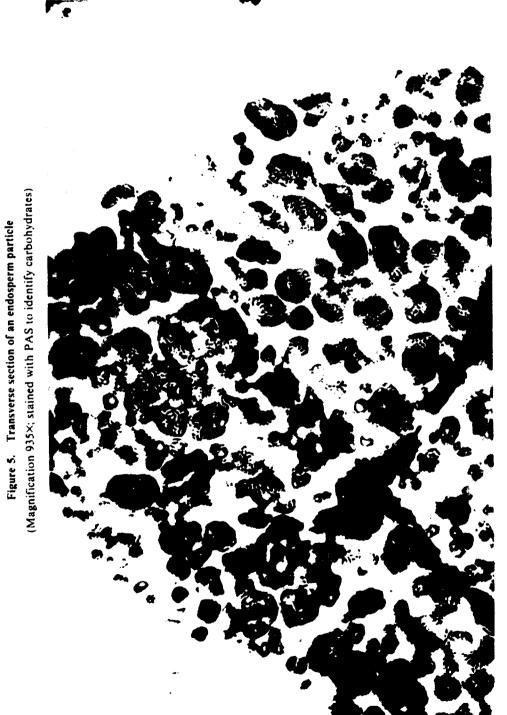


**Figure 4.** Section of a pericarp particle (Magnification 925×; stained with mercuric chloride-bromophenol blue (Hg-BPB) to identify proteins)

Source: Barber, Pineda and Benedito de Barber [1].

Rice bran: an under-utilized raw material





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Source: Barber, Pineda and Benedito de Barber [1].

#### Germ

Commercial bran, obtained from paddy and not from parboiled rice, always contains whole germ, as well as germ fragments (see figure 6). Because of its high tissue differentiation, these particles are readily distinguished from the other components of the bran. The most commonly found fragments of germ are whole plumule, coleoptile, coleoptile and plumule combined, radicle combined with calyptra and coleorhiza and scutellum combined with epiblast. Whole germ, or fractions containing scutellum, usually have adhering to them endosperm from the layer of crushed cells.

As has already been noted in chapter I above, commercial germ consists of: (a) an embryonic axis; (b) the tissue surrounding the embryonic axis; and (c) outer coverings which correspond to other anatomical parts of the caryopsis [20] (see figure 7). The three fractions represent 20 per cent, 71.78 per cent and 8.32 Fer cent by weight, respectively [21]. The distribution by weight of the various anatomical parts of the caryopsis of rice are shown in table 2.

Anatomical part	Percentage of caryopsis	Percentage of germ		
Pericarp and aleurone	7.0			
Starchy endosperm	90.7			
Germ	2.3			
Plumule	0.34	12.91ª		
Radicle	0.18	7.09		
Scutellum	1.4	52.1		
Coleorhiza	0.18	8.33		
Epiblast	0.26	11.25		

TABLE	2.	ANATOMICAL	DISTRIBUTION	OF	THE	RICE

Source: Hinton [21] and Hinton and Shaw [22]. <sup>a</sup>With the coleoptile.

# Histology of the main parts of the germ

## The embryonic axis<sup>5</sup>

The embryonic axis is L-shaped and is situated in the central part of the germ. It consists of plumule,<sup>6</sup> coleoptile, radicle and hypocotil. The plumule is sited on the larger side of the L that forms the embryonic axis. It is in the shape of an igloo, with a circular transverse section, some 0.30 mm in diameter and with a height of about 0.40 mm (Balilla  $\times$  Sollana short grain variety) (see figures 7-10). Only two or three leaves have been found on the plumule, surrounded by the epidermis and by conducting bundles.<sup>7</sup> In the centre of the

'Nerves of the mature leaf.

<sup>&</sup>lt;sup>3</sup>The part of the germ which develops during germination, giving rise to the new plant.

<sup>&</sup>lt;sup>6</sup>The cauline apex of the embryo; it includes the primordial leaves. During germination it gives rise to the primary leaves and to the stem.



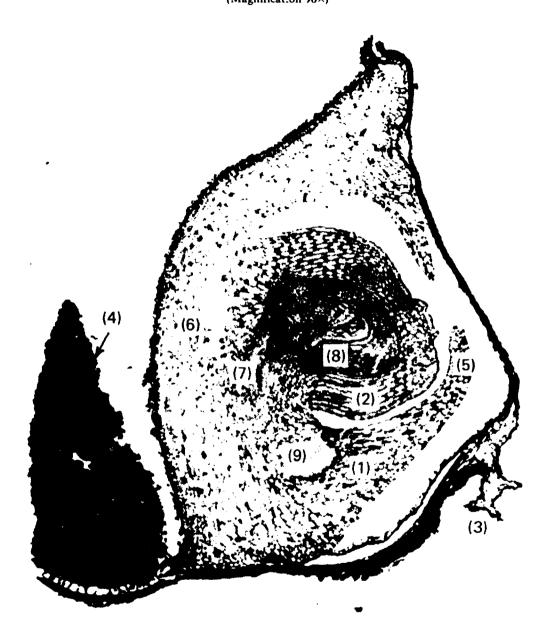
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Figure 6. Histological preparation of commercial rice bran, showing the presence of various fragments of germ (Ge) (Magnification 35×; stained with PAS to identify carbohydrates)

Source: Pineda [2].

2





#### Key:

Coleorhiza	(6)	Scutellum
Coleoptile	(7)	Hypocotyl
Crest	(8)	Plumule
Endosperm	(9)	Radicle
Epiblast		
	Coleoptile Crest Endosperm	Coleoptile (7) Crest (8) Endosperm (9)



Figure 8. Transverse section through the plumule of the vice germ

(Magnification 170×; stained with PAS)

Source: Pineda [2].

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Figure 9. Histological preparation of commercial rice bran, showing a fragment of plumule (Magnification 210×; stained with PAS to identify carbohydrates)

Source: Pineda [2].

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Figure 10. Longitudinal section of the rice germ, showing plumule and radicle (Magnification 135×; stained with PAS)

Source: Pineda [2].

spiral formed by the leaves the apical meristem can be seen (see figure 8). The cells of the embryonic leaves have thin walls; their transverse profile is polyhedral and approximately isodiametric (7  $\mu$ m), while the longitudinal profile is extended. The cells of the epidermis are differentiated from the remainder in that they are arranged in a palisade. Externally they are covered with a thin cuticle (0.1-0.2  $\mu$ m). The cells of the conductor bundles (3-6) are polygonal, measuring some 4  $\mu$ m in transverse section, and rectangular, measuring some 4 × 11  $\mu$ m in longitudinal section. The apical meristem contains isodiametric cells (4  $\mu$ m).

The coleoptile<sup>8</sup> is the outer cover of the plumule (see figures 7 and 8). It has a pore at the apex. It varies in thickness (0.11-0.18 mm), depending on where it is located, and consists of some 9-12 layers of cells, which are irregular polygons in transverse section and rectangular in longitudinal section. These are arranged longitudinally (see figure 8). The cells of the other layer, which are prismatic, form an inner epidermis; this and the outer epidermis are covered by a cuticle of 1.1-1.3  $\mu$ m and about 0.5  $\mu$ m respectively.

The hypocotyl<sup>9</sup> connects the plumule with the radicle<sup>10</sup> (see figure 7). It consists of compound provascular bundles of large, aligned cells, surrounded by parenchymatous cells.

The radicle is cylindrical and about 0.45 mm long (see figures 7 and 11). It is situated on the short arm of the L forming the embryonic axis (see figure 6). The transverse section (about 0.35 mm) shows a variety of tissues arranged in radial symmetry (see figures 7 and 11), including: (a) a cuticle 6-10  $\mu$ m thick (1.5  $\mu$ m in the apical meristem); (b) epidermis and subepidermis of prismatic cells, measuring about 11 × 23 and 7 × 4  $\mu$ m respectively, arranged in a palisade; (c) exodermis of two layers of cells of different shape and size; (d) cortical cylinder, about 70  $\mu$ m thick, with from five to seven layers of oval and round cells (measuring 10-20  $\mu$ m); (e) endodermis of one layer of prismatic cells measuring 6 × 5  $\mu$ m; (f) pericycle of one or two layers of cells; and (g) a central cylinder, in which the protophloem, the protoxylem and the metaxylem can be discerned among the numerous parenchymatous cells. At the apex of the radicle is the apical meristem.

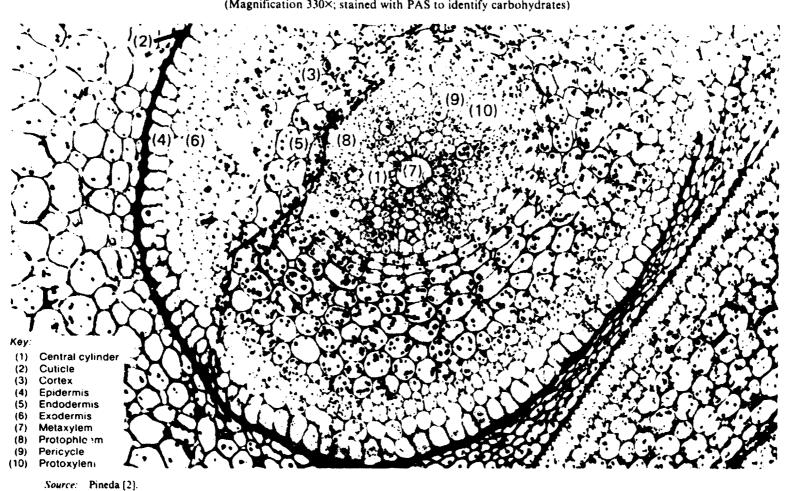
The outermost layer of the meristem is the calyptrogen, which produces the calyptra. The dermatogen, which gives rise to the epidermis, the periblem, from which the cortical cylinder is formed, and, finally, the plerome, which gives rise to the central cylinder, are also present. The apex of the radicle is covered by the calyptra.

Table 3 indicates the distribution of organs and structures in various histological areas of the embryonic axis [23]. The cells of the scutellum contain many round particles, around 2-3  $\mu$ m in diameter, covered by a membrane, which are similar in appearance and composition to the aleurone grains [24].

\*The primary stem.

<sup>&</sup>lt;sup>8</sup>Some authors consider the coleoptile to be a young modified leaf, while others affirm that it is a cotyledon, or even that it forms part of the latter in conjunction with the scutellum [21]. During the first few days after germination it develops along with the plumule, but is subsequently reabsorbed.

<sup>&</sup>quot;The primary root that during germination develops into the main root of the plant.



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Figure 11. Transverse section of the radicle of the rice germ (Magnification 330×; stained with PAS to identify carbohydrates)

8

Morphology, anatomy, histology and histochemistry of the discrete particles

Histological area	Golgi apparatus	Protein bodies	Bundles	Fatty bodies	Endo- plasmic reticulum	Vacuoles	Cutich
Scutellum proper							
Epithelium (an epidermis)	+	+a	_	+b	_	+	_
Farenchyma	_	+a	_	+	_	- -	
Provascular tissue	+	_	+	+ <i>h</i>	+۲	+	_
Epidermis	-	+ <sup>a</sup>	_	+b. d	-	-	+
Ventral zone of scutellum							
Epidermis	_	+2	_	+b. d			+
Parenchyma	-	+2		+	_	_	-
Lateral zone of scutellum							
Epidermis	_	+°	_	+b. d	_		+
Parenchyma	_	, +a	_	+	_	_	-
Coleoptile		•		•			
Epidermis	_	+a	+	+ <sup>b</sup>			
Parenchyma	_	+a	+	+		-	+
Provascular tissue	+	_ _	+	+ +b	- + <sup>c</sup>	+	_
Plumule	•		•	•	т	Ŧ	_
	,						
Apical meristem Embryonic leaf	+	_	+	+b	-	+	+
Epidermis		+a		+b			
Parenchyma	+	+"	+	+0	-	+	+
Provascular tissue	+	_	+ +	+0	+° +°	+	
	т		Ŧ	<b>T</b>	+.	+	
Mesocotyl Parenchyma		. 0					
Provascular tissue	-	+ª	_	+ +b	-	-	-
	+	_	+	+"	+ <sup>c</sup>	+	_
Radicle							
Calyptra	+	+	-	+	-	+	-
Apex	+	+	—	+ b	-	+	~-
Epidermis	-	+	+	+b	-	-	-
Hypodermis	-	-	+	.⊧b	-	~	-
Cortex (parenchyma) Endodermis	_	+a	+	+ + <i>b</i>		-	-
Vascular cylinder	-	-	+	+"		—	-
Pericycle	_	_		+ ^			
Metaxylem	_	_	+ +	+" + <sup>b</sup>	 + ¢	_	-
Procambium	_	_	+	+» +b	+°	+ +	_
Colecrhiza						•	
Epidermis close to the radicle		+	_	.Lb	_	-	
Parenchyma	_	+a	-	+		_	
External epidermis	-	+a	-	+b. d	_	_	+
Epiblast							÷
Epidermis		+	+	+b, d	_	-	+
Parenchyma		+a	+	+			-

# TABLE 3. DISTRIBUTION OF ORGANS AND STRUCTURES IN HISTOLOGICAL AREAS OF THE RICE GERM

Source: Bechtel and Pomeranz [23].

<sup>a</sup>With inclusion of electron-dense material.

<sup>b</sup>Generally peripheral.

Whorls.

R.

dWith dense material at the surface of the section.

#### The tissues surrounding the embryonic axis

Although they are not clearly separated, it is possible to distinguish: (a) the scutellum; (b) the coleorhiza; and (c) the epiblast.<sup>11</sup> The scutellum is connected with the coleorhiza in the lower part of the radicle and with the epiblast in the upper cuter part of the coleoptile.

The scutellum<sup>12</sup> is the largest part of the germ (see table 2). It lies between the embryonic axis and the endosperm of the grain (see figures 7 and 8). The larger part of the scutellum consists of parenchymatous cells, of irregular polyhedral profile, some  $14 \times 18 \mu m$  in longitudinal section. On the outside, except where it is joined to the epiblast and the coleorhiza, it has an epidermis of prismatic cells, covered by a thin cuticle; adjacent to the endosperm, the epidermis is modified to form a layer of cylindrical absorption cells, known as epithelium, with some invaginations known as epithelial glands. In transverse section, the cells of the epithelium appear in disorder, but in longitudinal section they are palisaded (see figure 12). A conductor bundle extends from the upper part of the scutellum to the base of the plumule.

The coleorhiza is a protective tissue for the radicle, around which it is wrapped (see figure 7). It is connected to the scutellum at the lower and rear part of the germ and with the epiblast at the top of the lower outside part of the plumule. It consists of parenchymatous cells of irregular polygonal profile. Externally it has an epidermis of prismatic cells, broadened and of different size, covered by a thin cuticle (see figure 13). Between the coleorhiza and the aleurone layer an empty space has been observed, but in the region of the crest of the germ, both parts are joined by means of a "suspensor"<sup>13</sup> (see figure 14).

The epiblast is a protective cover for the plumule, wrapped around it on the outside and laterally, but without coming into contact with it (see figures 7 and 10). The upper junction with the scutellum is incomplete and at the lower junction the epiblast is fused with the coleorhiza. It is formed of polygonal, isodiametric parenchymatous cells with flattened walls, some 6  $\mu$ m in transverse section and elongated from 7 to 12  $\mu$ m in longitudinal section, and is covered by a monocellular epidermis with a thin outer cuticle.

Table 3 records the organella and structures found in the scutellum, coleorhiza and epiblast [23].

#### Germ conductor system

The germ conductor system consists of the provascular bundle extending through the hypocotyl, from the radicle to the base of the plumule. In the cotyledon node it divides into two branches, the provascular bundle and the branch that enters the plumule, where it branches again to give rise to the provascular bundles of the coleoptile and of the primary leaves.

<sup>13</sup>It has been pointed out that in the wheat germ it facilitates the entry of water into the grain [27].

<sup>&</sup>lt;sup>11</sup>Some authors regard the epiblast not as an independent morphological unit but as part of the scutellum [25].

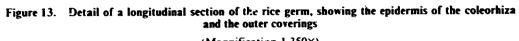
<sup>&</sup>lt;sup>12</sup>During germination, it acts as an organ for absorbing and conducting nutrient materials from the endosperm to the embryonic axis. Some authors regard it as a single cotyledon, while others claim that the cotyledon is formed by the scutellum and the coleoptile [26] and the epiblast [25].



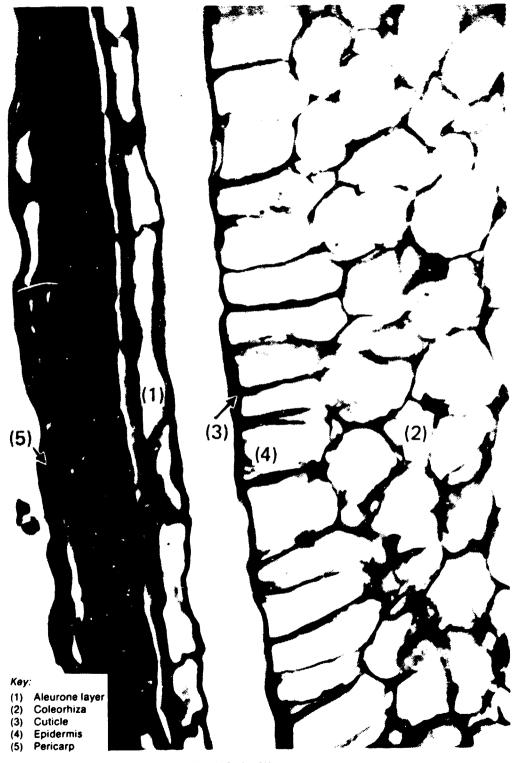
Figure 12. Longitudinal section of the epithelium of the rice germ (Magnification 1,350×)

Source: Barber, Navarro and Tortosa [20].

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(Magnification 1.350×)



Source: Barber, Pineda and Benedito de Barber [1].

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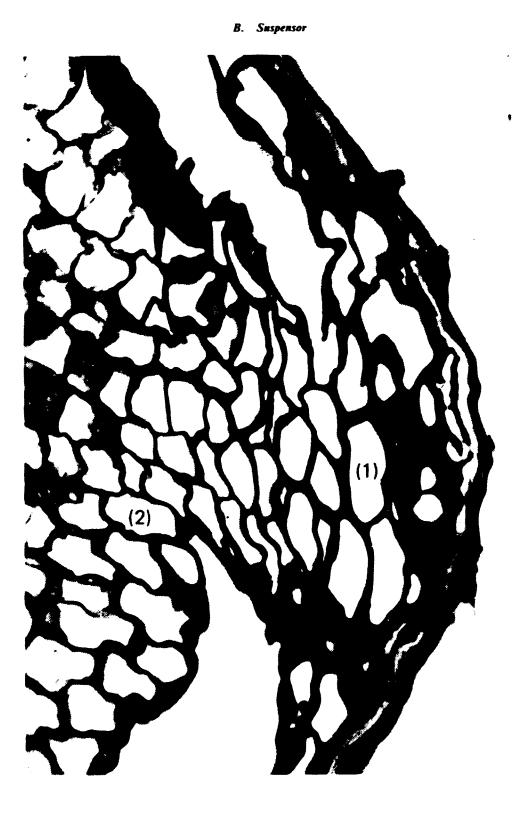
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Figure 14. Detail of longitudinal sections of the rice germ, showing the connecting zone (suspensor) between the aleurone and coleorhiza (Magnification 1,350×)

A. Expansion of the aleurone layer



Source: Barber, Navarro and Tortosa [20].



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## Covering of the germ

The germ is surrounded by the aleurone layer, the tegmen or seed-coat and pericarp on the outside, and by the crushed cells of the endosperm on the inside.

The monocellular aleurone layer undergoes profound changes in the vicinity of the germ. Its cells are more flattened  $(3-5 \,\mu\text{m})$  than they are in the region of the endosperm (14-18  $\mu$ m). The seed-coat<sup>14</sup> appears as a continuous sheath, with no differentiation in the cell walls. It is thinner in the region of the germ (0.8-1.5  $\mu$ m) than in that of the endosperm (1.5 to 2.3  $\mu$ m). In the vicinity of the germ the pericarp is present as a mass of undifferentiated cells, with no protoplasm. In the region of the coleorhiza it forms a fold or crest, increasing the thickness of the pericarp (see figure 7). The sponginess of the pericarp is accentuated around the crest. As has already been seen in chapter I above, the germ is in contact with the endosperm through the layer of crushed cells (see figure 12).

## Histochemistry of the rice germ<sup>15</sup>

Sugars, starch, hemicelluloses, cellulose, lignin, lipids, proteins, basic and sulphur-containing amino-acids, tryptophane and mineral ashes have all been identified by histochemical methods in rice germ [32, 2, 37], and phosphorus and magnesium and potassium have been found by electron microprobe X-ray analysis [21]. The reaction with ferric chloride-hydroxylamine, which is specific for pectic substances [28, 29, 30], and the alkaline hydrolysis method [36], followed by staining with PAS [5], gave negative results [32]. The reaction with phenosafranine reagent for silica [33] has also been negative.

The sugars are found in most of the germ cells; they appear to be absent in the pericarp and aleurone layer cells [4]. Glucose and sucrose have been found in the scutellum [34]. Carbohydrates<sup>i6</sup> are mainly localized in the cell walls and sometimes in the cytoplasm (see figure 15). The range of distribution is common over the various histological regions, although there are some differences. In the embryonic axis, the intense staining of the calyptra, the cuticle covering the epidermis and, to a lesser extent, the cuticle surrounding the coleoptyle, stands out. In general, carbohydrates are not present in the cytoplasm of the cells, but in some preparations of the embryonic axis granules of starch have been detected. They are round and vary in size from  $0.5-5 \mu m$ . They are unevenly distributed, being more numerous in the plumule and the coleoptyle than in the radicle. In addition, starch has been identified in the tissues surrounding the embryonic axis, where the granules are more numerous, in the scutellum and in the coleorhiza. In the tissues surrounding the embryonic axis, the staining of the cell walls with PAS is more intense in the epiblast and

<sup>&</sup>lt;sup>14</sup>This is a semipermeable membrane. Water has been shown to penetrate the endosperm of cereals more rapidly through the area around the germ, possibly because the tegmen is less thick in this area.

<sup>&</sup>lt;sup>13</sup>In the text, reference is made to the intensity of staining of certain histochemical preparations. Although this does not specifically indicate the concentration of the chemical component, it is nevertheless informative.

<sup>&</sup>lt;sup>14</sup>Including carbohydrates that give a positive reaction to periodic acid—PAS [5, 6]: starch, cellulose, hemicelluloses, pectic substances, glycoproteins and glycolipids. The sugars and some lipids also give the PAS reaction [6], but most of these are eliminated during the preparation of the sections.

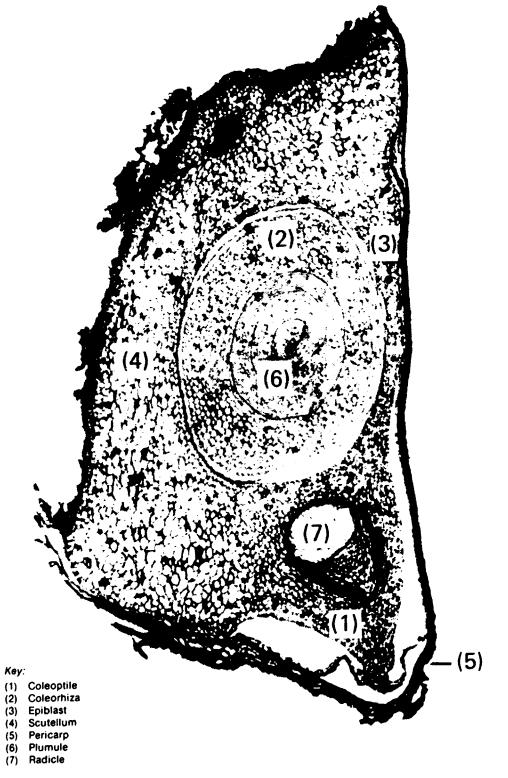


Figure 15. Longitudinal section of the rice germ (Magnification 88×; stained with PAS to identify carbohydrates)



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coleorhiza than in the scutellum. No starch has been detected in the outer coverings of the germ and in the crushed cell layer; carbohydrates are found in the cell wall but not in the cytoplasm [2, 32]. In the cellular protoplasm of some preparations of germ, the presence has been noted of scattered material that stains with PAS [2].

Cellulose is found in the cell walls of all tissues of the germ. Basically it is found in the outer coverings, particularly the pericarp [9, 32, 35].

Using the Schiff reagent [36], lignin has been detected in the outermost layer of the pericarp (outer covering of the germ), but this result has not been confirmed [32] by using phloroglucinol and chloride sulphite stains [36].

Lipids are present in all tissues of the germ (see figure 15 and table 2) [23, 37]. In general, the lipids are found in the cytoplasm, in the form of globules [4, 9, 23, 37, 38, 39], also called fatty bodies [23], of various sizes (0.1-1  $\mu$ m). The fatty bodies are not surrounded by a membrane, even though they have a thin electron-dense margin [23]. The distribution of the fatty bodies in the cells may be used as the basis for classifying them into categories [23]. In some cases the fatty bodies are dispersed in the cytoplasm (e.g. parenchymatous cells of the coleoptyle, cortex and epiblast), in others they are peripheral (e.g. epidermal cells of the scutellum and of the apical meristem). In the tissues of the embryonic axis the lipids are not distributed uniformly (see figures 16 and 17). In the coleoptile the fat globules are more numerous than in the plumule, the hypocotyl or the radicle. The provascular bundles of the coleoptile show little positive reaction to Sudan black staining. There are also differences in the radicle. In the tissues surrounding the embryonic axis, the scutellum is more pientiful than the coleorhiza and epiblast. In the outer coats of the germ, lipids abound in the aleurone and in the cytoplasms, where they are found in the form of globules (see figure 18). When the seed-coat is stained with Sudan black, they are seen to be present in a continuous layer. The pericarp and the layer of flattened cells show very little reaction to this reagent [37] (see figure 19).

The proteins are found in most of the germ tissues (see table 2) [23, 37]. They are present in the cytoplasm, in the form of scattered discrete granules, and in the cell nucleus [37]. Protein bodies have been detected in practically all but the provascular tissues [23]. The cells of the scutellum contain protein particles 2-3  $\mu$ m in diameter, which, because of their proteins and phosphorus, magnesium and potassium content, are similar to the aleurone granules in the aleurone layer [24]. The ultramicroscopic characteristics of the protein bodies vary depending on the type of tissue involved. On the basis of these differences and the distribution of the fatty bodies, three categories of parenchymatous cells have been identified [23]: (a) cells with electron-dense inclusions in the protein bodies and numerous fatty bodies dispersed through the cytoplasm (e.g. the parenchymatous cells of the scutellum); (b) cells with protein bodies with or without electron-dense inclusions and peripheral fatty bodies (e.g. epidermal cells, except the apical meristem); and (c) cells without protein bodies and peripheral fatty bodies (e.g. cells of the provascular system of the plumule and radicle).

In the embryonic axis, staining with mercuric chloride-bromophenol blue [12] is more intense in the coleoptile and in the radicle than in the plumule (see figure 20). The nucleus stains more strongly than the cytoplasm. In the tissues surrounding the embryonic axis, the proteins are present mainly in the form of

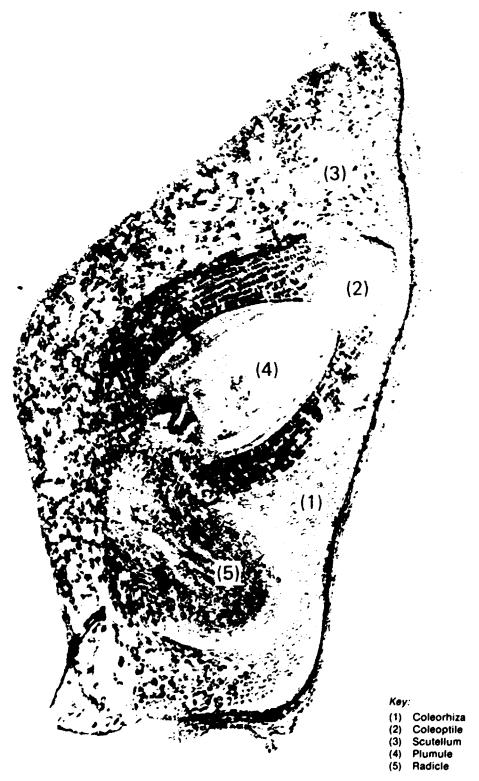


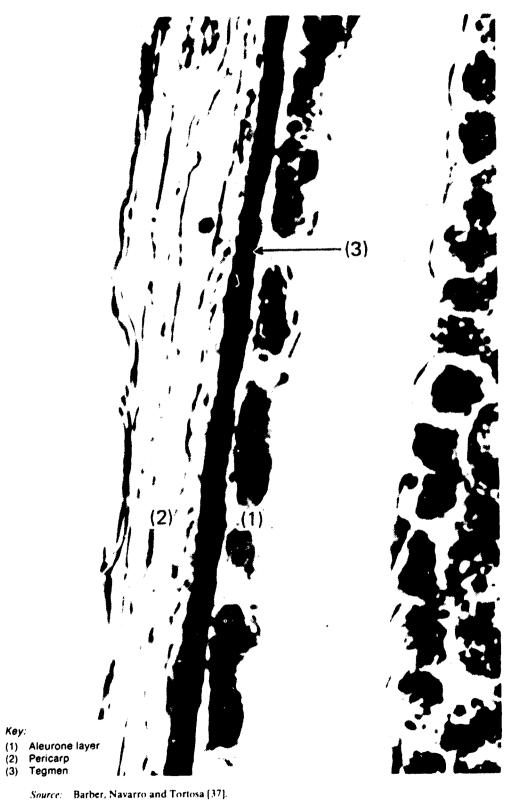
Figure 16. Longitudinal section of the rice germ (Magnification 90×; stained with Sudan black to identify lipids)

Source: Navarro [40].



Figure 17. Transverse section of the embryonic axis of the rice germ (Magnification 170×; stained with Sudan black to identify lipids)

Source: Barber, Navarro and Tortosa [37].



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Figure 18. Outer coverings of the rice germ (Magnification 1,350×; stained with Sudan black to identify lipids)

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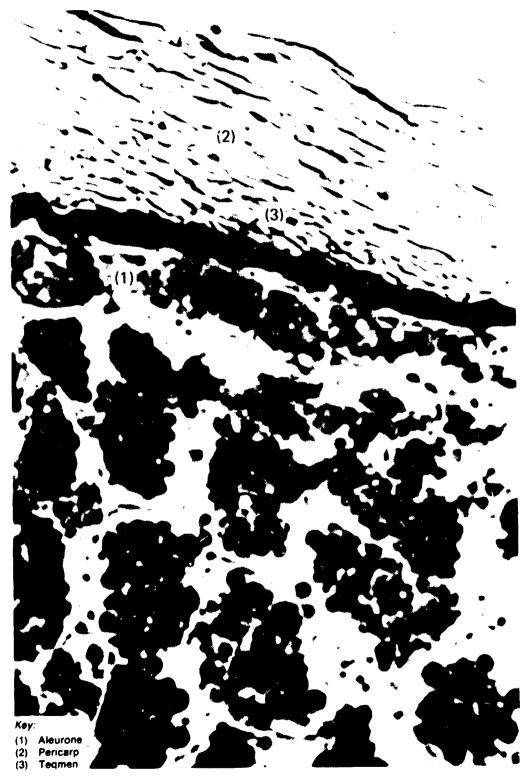


Figure 19. Longitudinal section of the rice germ, showing the outer coverings (Magnification 1,350×; stained with Sudan black to identify lipids)

Source: Barber, Navarro and Tortosa [37].



Figure 20. Longitudinal section of the rice germ

(Magnification 40×; stained with mercuric chloride-bromophenol blue to identify proteins)

Source: Barber, Navarro and Tortosa [37].

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discrete granules measuring 0.5-2  $\mu$ m. The staining is more intense in the outer epidermis of the epiblast and coleorhiza. The provascular bundles show only a very faint stain, and cuticles do not stair. at all [37]. Unlike the aleurone surrounding the endosperm, which contains proteins in the form of discrete granules [41, 42], the aleurone cells in the outer covering of the germ, containing dispersed proteins but no discrete granules, appear stained [37, 41].

It has been found [37] that all the germ tissues that contain proteins can be stained with naphthol yellow S [43], which stains lysine, hydroxylysine, histidine, arginine and terminal amino groups, with 2,2'-dihydroxy-6,6'dinaphthyl-disulphide (DDD) and diazo blue B [44] which stain the sulphurcontaining amino-acids, and with DMAB-nitrite [45], which is specific for tryptophan.

Inorganic materials (residual ash on ignition) are present in almost all the germ tissues (see figure 21). In general they are found in the cytoplasm and, in the case of the pericarp, in the cell walls. In the embryonic axis they are more numerous in the plumule and in the radicle than in the coleoptile, in the plumule they are distributed uniformly and in the radicle they are concentrated in the central cylinder; they are present in the cuticle covering the epidermis. In the scutellum, inorganic materials are abundant, particularly close to the endosperm [37]. The protein particles of the scutellum contain a high proportion of magnesium and potassium salts of phytic acid [24].

#### Fibres

The fibres originate in the nodes at the base of the particle. Although rare, they are conspicuous owing to their great length, which in some cases exceeds 1 cm. They are banded and twisted and exhibit no defined cellular structure; in the fragments identified, two thin walls have been observed with an intermediate and empty cavity some 30  $\mu$ m wide. Like the fibres in the nerves of the glumes, they give a positive reaction to the PAS stain for carbohydrates. It has not been possible to identify proteins, lipids or silica, since they do not stain with the corresponding histochemical reagents [2].

## Compound discrete particles in commercial bran

The following compound particles have been detected in commercial bran: (a) fragments of the pericarp with the seed coat; (b) fragments of the seed-coat with the aleurone layer; (c) fragments of the pericarp, seed-coat and aleurone layer; (d) fragments of the pericarp, seed-coat, aleurone layer and starchy endosperm; (e) fragments of the starchy endosperm and aleurone layer; (f) fragments of the germ and starchy endosperm; and (g) fragments of the germ, aleurone, seed-coat and pericarp.

## Fragments of the pericarp with the seed-coat

This is one of the more common types of particle in commercial bran (see figure 22), forming what some authors refer to as "true bran" [46].



Figure 21. Longitudinal section of the rice germ

(Magnification 90%; micro-incinerated and photographed against a dark field)

Source: Navarro [40].

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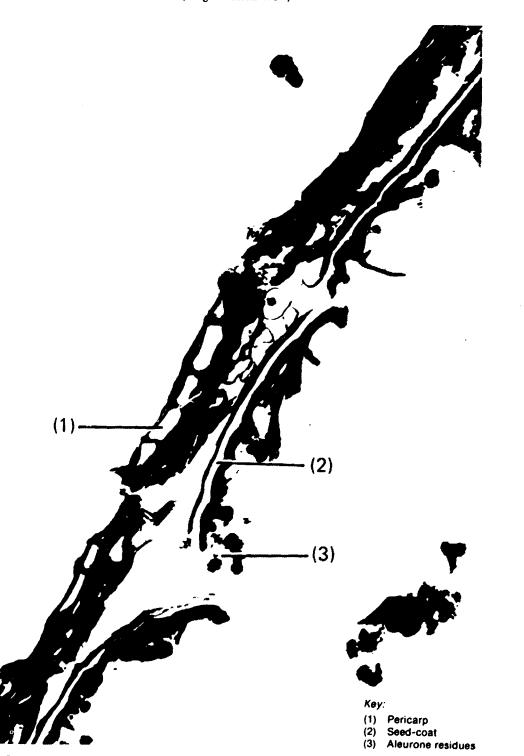


Figure 22. Section of a compound particle in commercial rice bran (Magnification 470×)

Source: Barber, Pineda and Benedito de Barber [1].

The fragments are in the form of twisted or coiled flakes with a smooth and lustrous outer face (free surface of the pericarp) and a rough interior. They are generally elongated and of different sizes (mostly measuring around 300  $\mu$ m). The tissues are usually compressed, ranging in thickness from 10 to 15  $\mu$ m; sometimes larger particles are found, particularly when the tissues are more spongy. This sponginess is particularly noticeable in the vicinity of the crest. As with the particles of the pericarp alone, the cells of the spongy parenchyma of the hypodermis are difficult to distinguish, but in some particles of flattened structure they can be readily discerned. The cross or transverse cells are also visible [2]. The histological characteristics of the pericarp have been described in chapter I above. In particles of commercial bran, the seed-coat and cuticle are combined, together with residues of the nucellus; some sections of the seed-coat may be interrupted. The pericarp and seed-coat are tightly joined along their whole length. In some particles, the seed-coat is accompanied only by the cuticle [2].

The histochemical characteristics of the pericarp have already been described. The seed-coat, with the exception of the cuticle, stains with the PAS reagent for carbohydrates and reacts positively with the Hg-BPB reagent for proteins. The lumen does not stain [1]. Cellulose is the main component of the cell walls [9]. The cuticle stains intensely with Sudan black [1]. It has been suggested by Little and Dawson [9] and Esau [47] that its main component is suberin. The remains of the internal tegument and the nucellus stain very little [1]; the cell walls of the internal tegument are cutinized [14]. The tegument and nucellus contain lignin. No silica has been detected in the pericarp or in the seed-coat [2].

## Fragments of the seed-coat with the aleurone layer

Fragments of the seed-coat with the aleurone layer are not very common in commercial bran. Only particles with a single layer of aleurone cells have been found. Usually neither the seed-coat nor its constituent layers becomes detached during milling. Although botanically the aleurone layer is part of the endosperm, the fragments of the aleurone that separate during processing are more often found with the seed-coat than with the endosperm (see table 1). The size of the fragments varies between 100 and 300  $\mu$ m in length and 15 and 20  $\mu$ m in width. The layer formed by the seed-coat measures 6  $\mu$ m and that formed by the aleurone up to 10  $\mu$ m [2]. The histology of both layers has been described in chapter I above.

The histochemistry of the seed-coat and seed-coat layer and the portion of the aleurone of the endosperm, carbohydrates, proteins, lipids, lignin, vitamins and inorganic materials have been identified. The carbohydrates are localized in the cell walls and in the cytoplasm. In the latter, grains of starch have been found measuring up to almost 5  $\mu$ m.

Although some authors claim that no starch is present in the aleurone [4, 11], it has been mentioned that starch granules may be found in the aleurone cells of mature grains [19]. The walls of the aleurone cells give a positive reaction for cellulose and for hemicelluloses or pectins when they are stained with zinc iodochloride and ruthenium red respectively [9]. Proteins, stained

with the Hg-BPB reagent, are present in granular form and in dispersed form in the protoplasm. Proteins are also present in the form of a thin film around the cell wall [23]. Staining with ferric ferricyanide for proteins also gives positive results for the aleurone cell walls [9]. The cellular nucleus is difficult to identify [2, 17]. The distribution of aleurone granules is not uniform [9]: the lumen in the dorsal region of the caryopsis is largely composed of aleurone granules, whilst other cells have a large number of starch granules and, in particular, fat globules [23]. It has been known for some time that fat globules are present in the aleurone cells [4]. A thin fatty film surrounds the aleurone granules [9].

### Other types of compound particles

The information on compound particles of types 3 to 7 given in table 1 may be summarized as follows (see also figure 23). Particles of type 3 consist of the pericarp, seed-coat and aleurone. It has been shown that separation from the grain during processing takes place more easily from the ventral side than from the dorsal side [14]. The size of the particles varies between  $130 \times 50 \,\mu\text{m}$  and  $560 \times 105 \,\mu\text{m}$ . Most of them have one or two layers of aleurone cells, although few have from five to seven layers [2]. This is probably because the *Japonica* varieties have one or two layers of cells in the ventral side of the aleurone and from five to seven layers on the dorsal side [14] and also because of the difference in resistance to abrasion of the two sides of the grain. Although in some particles the surface aleurone cells are broken, the surface and interior cells generally remain intact. There are particles in which the cuticle of the seed-coat appears to be broken.

The type-4 particles are not very numerous; they consist of the pericarp, seed-coat, aleurone and starchy endosperm and are usually larger than other particles, ranging in size from  $400 \times 260 \,\mu\text{m}$  to  $735 \times 200 \,\mu\text{m}$ . They are of irregular shape. The external cells of the starchy endosperm are, for the most part, broken. The type-5 particles, which are not very numerous, consist of starchy endosperm and aleurone. Usually they are round and relatively small (less than  $95 \times 60 \,\mu$ m). The type-6 particles consist of starchy endosperm and germ. In addition to whole germ, to which a number of flattened endosperm cells are attached, they also include fragments of germ with different layers of adhering endosperm cells. As a rule, however, the germ is the major component. The particles are usually large (ranging from  $690 \times 320 \,\mu m$  to  $410 \times 225 \,\mu$ m). The type-7 particles consist of germ, aleurone, seed-coat and pericarp and are rare in commercial bran from which the germ has been removed. They are abundant in bran that contains the germ since the latter detaches itself from the caryopsis with the various coverings mentioned. Particles which have the same composition but contain only a fragment of germ may also be found, the proportion depending on the type of rice and the milling process. The latter type of particle is seldom found in commercial bran of the Japonica varieties from Spanish mills. When they do occur, they consist mainly of epiblast or coleorhiza, together with fragments of the germ-coat.

The histological and histochemical characteristics of the tissues present in the various types of particle referred to here are the same as those described in earlier sections.



# Figure 23. Histological preparation of commercial rice bran, showing various compound particles (Magnification 220×; stained with PAS to identify carbohydrates)

Source: Barber, Pineda and Benedito de Barber [1].



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# IV. Controlling the production of rice bran

**Control of processing** 

Under industrial conditions the quantity of bran recovered from the rice during processing varies, in the main, from 4 to 11 per cent by weight for rough rice and from 5 to 13.5 per cent by weight for brown rice (see table 1). The proportion varies from country to country and from market to market, as well

TABLE	1.	AMOUNT	OF	BRAN	REMOVED	FROM	RICE	DURING	MILLING,	BY
					COUNTRI	ES				

Country	Grams of bran per 100 grams of rough rice	Source
Colombia	8	Ospina [2]
	8-10	Jaramillo [3]
Costa Rica	9	Vargas and Murillo [4]
Egypt	8	Saunders and others [5]
Spain	7-13	Rivero [6]
•	7.4-11.1	Barber and Benedito de Barber [7]
Philippines	7	Reddy, Gariboldi and Joko [8]
Guatemala	10	Elias and Bressant [9]
India	4-6	Brown [10]
	4.5-8.5	Chakrabarty, Bhattacharya and Vaidyanathan [11]
	4	Reddy, Gariboldi and Joko [8]
	3-4 <sup>a</sup>	Raghvendra Rao, Narayana and Desikachar [12]
Indonesia	4	Reddy, Gariboldi and Joko [8]
Iran (Islamic Republic of)	7-10	Kachru and Eghtedari [13]
Japan	7	Reddy, Gariboldi and Joko [8]
Republic of Korea	7	Reddy, Gariboldi and Joko [8]
	8.7-10.8	Kwon and Jo [14]
Liberia	4-6	Saunders and others [5]
Malaysia	4-5	Arnott and Lim [15]
-	5.45-13	Van [16]
Mozambique	10.5	Berberan [17]
Pakistan	4	Reddy, Gariboldi and Joko [8]
	6-7	Magsood Ali, Abdul Haq and Hameed Khan [18]
	10	Khan [19]
Sri Lanka	4	Reddy, Gariboldi and Joko [8]
_	3.5	Private communication from industry
Thailand <sup>b</sup>	7	Reddy, Gariboldi and Joko [8]
United States of America	9.7	Hunnell and Nowlin [20]
Venezuela	11.5	Guerra and Jaffe [21]

<sup>a</sup>Usually when rice is milled for the Government.

<sup>b</sup>A study carried out in more than 80 mills in Thailand gave the following results: (a) bran from brown rice--2.86 per cent on the paddy rice, varying from 1.95 to 3.87 per cent; (b) white bran-7.19 per cent on the paddy rice, varying from 5.08 to 9.16 per cent [1].

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as in other ways.<sup>1</sup> Parboiled rice requires less processing than crude rice and the amount of bran does not usually exceed 6 per cent. The proportion of bran produced at the mill from rough rice does not usually represent the true proportion of bran separated from the caryopsis, since the bran may contain crushed husk (e.g. huller mills or inefficient paddy separators) or fragments of endosperm (especially when grain breakage is high). Nevertheless, the proportion of bran produced from rough rice is important to the miller for costbenefit calculation. But the processing also has to be assessed in terms of its effect on the grain itself and on the quality of the by-products. Only in this way can the milling process be understood and evaluated, possible improvements be decided upon and the end-products—white rice and bran—given due recognition. Despite the many alternatives available, it has usually been the practice to judge the effects of milling by the quantity of residual bran in the milled grain, using direct visual observation. This is undoubtedly a very inadequate way of assessing the performance of machinery and the behaviour of the grain. Other things have to be monitored. Direct observation of the bran, for example, just at the whitener outlet, provides some very useful information.<sup>2</sup> It should be made clear that the milling process does not always work in the same way, or in the way the miller would like it to. The amount of bran removed is not always the same; the layers of cell tissue removed can be from different depths and the area of the grain from which it is taken may also vary in size and location. The end-product is a rather heterogeneous mixture of grain particles.

### Methods for measuring the degree of milling of rice

The essential purpose of milling is to release the starchy endosperm from the other fractions enclosing it—including the germ<sup>3</sup>—in order to improve the appearance and eating qualities of the grain. In countries where rice is a staple food, the operation must be carried out with as little loss of nutrients as possible. A longer milling time means that there will be more bran and tailings, which are worth less than the full grain, so a means must be found of eliminating unnecessary by-products while still meeting the primary requirements of appearance and palatability. Another important, though less obvious, objective is that the removal of the outer layers of the caryopsis should be homogeneous both in each single grain and among individual kernels; this not only affects the appearance of the end-product but has a direct bearing on its keeping qualities [22]. Fortunately, none of these requirements conflicts with the one final objective, which has so far been largely ignored, namely the production of high-quality bran.

<sup>2</sup>It is sufficient to press a small amount of bran between the palms of the hands in order to reveal defects that would not otherwise be noticed—for example, broken and even whole grains that have passed through damaged screens, fragments of foreign seeds that are present because of inadequate cleaning, particles of emery caused by wear etc.

<sup>1</sup>A special process leaves the white milled rice grain with the germ attached to increase its nutritive value.

<sup>&</sup>lt;sup>1</sup>A study carried out in Thailand [1], in which the yields of the different fractions from 80 mills throughout the country were compared, showed that the main causes of variation in yield were: (a) the quality of the paddy rice; (b) the size, type and general condition of the mill; (c) the degree of processing or the pattern of processing; and (d) other factors such as environmental conditions and the method of production control used.

Many methods have been devised to determine the degree of milling of rice, and several attempts have been made to bring the information on the topic up to date [23, 24, 25]. The various methods in use can be grouped as follows: (a) procedures for ascertaining the quantity of separated or residual bran; and (b) procedures for evaluating the effects of milling on the basis of changes in the chemical composition or optical properties of the end-product.

### Methods for estimating removed bran or residual bran

Both in the mill and in the laboratory it is standard practice to measure the percentage by weight of rice (rough rice or brown rice) that has been separated as bran; brown rice is usually used, in order to avoid errors caused by the varying husk content of rough rice. There are, however, a number of snags. For example, a representative sample of paddy or brown rice must be available, and the processing conditions have to be identical if the results are to be reproducible and readily comparable. Unless these conditions are me: equal quantities of bran will not necessarily show the same degree of whiteness, nutritive value, eating qualities or keeping qualities.

There are several alternative methods for estimating the residual bran. These include: (a) visual inspection of the bran, either directly or by means of optical instruments; (b) visual assessment of the residual bran by prior staining of the rice grains to distinguish the bran from the starchy endosperm; and (c) colorimetry, using bound stains or developed pigment colours in situ or after extraction from the grain.

The most usual method is to make simple visual comparisons, sometimes with the aid of a magnifying glass, against standard samples. Unfortunately, differential staining is only used in the laboratory. The following stains have been tried: indigo carmine and fuchsin [26]; Congo red and methylene blue [27]; Sudan III [28]; iodine [29]; eosin and methylene blue as the May-Grünwald reagent [30]; and an alkaline alcohol solution [27, 31]. The results can only be interpreted subjectively and a comparison of samples of different types of rice is difficult. They are, however, much more reliable than those obtained when undyed grains are inspected, since staining shows up differences that could not otherwise be detected.

The eosin and methylene blue reagent (May-Grünwald reagent) [30] stain the outer layer of the bran green, the inner layer of the bran blue and the starchy endosperm pink. Standard reference patterns for different degrees of milling are needed, and the evaluation is subjective, but it does make the anatomical layers of the grain stand out from one another very vividly and is very useful. Staining with the May-Grünwald reagent has been used as a preliminary step in the preparation of the grain in the development of an objective method for measuring the degree of milling [24]. The stained areas—green or blue for the bran and pink for the starchy endosperm—are measured with a planimeter on magnified plane images. The degree of milling is expressed as the proportion of the area of the grain still covered by bran, or coloured bran balance (CBB) index. The values of the CBB index vary from 100 for brown rice to 0 for fully milled rice (see figure 1). Samples of wellmilled rice yield values of around 5. The method provides an objective and very precise measure of the degree of milling, and is particularly significant in that

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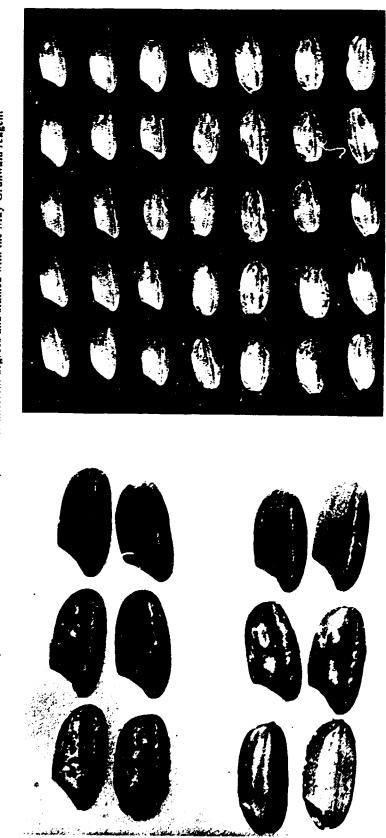


Figure 1. Samples taken from the same lot of rice, milled to different degrees and stained with the May-Grünwald reagent

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for the first time an evaluation of the homogeneity of the milling effects on the rice kernel is possible. It has been used in various laboratories, but although its advantages in terms of accuracy and evaluation of homogeneity are acknow-ledged, there is general agreement that it is too sophisticated and time-consuming and needs to be improved.

The solvent extraction of bound stains or developed pigment colours [27, 31] depends on the availability of representative samples of the original paddy or brown rices and, despite its simplicity, is possibly still too complex a process for routine use by millers.

# Methods based on the effects of bran removal on the chemical composition or optical properties of the milled rice grain

The anatomical layers of rice differ in their chemical composition: most constituents show a gradient of decreasing concentration (or initially increasing, then decreasing, concentration) from the outer to the inner layers of the carvopsis. There are numerous methods of evaluation that use the progressive decrease in the concentration of one constituent as the milling proceeds. The various constituents considered in this connection are: fat [27, 32, 33]; ash [34] and soluble minerals [35]; silica and crude fibre [12]; proteins [36]; phytin [37] and phytinic phosphorus [38]; and thiamine [37]. All these methods require careful execution and probably more time than a mill can spare on a routine basis. Some of them, for example those using fat [39] and soluble minerals [35], have been greatly simplified. Nevertheless, all of them have one basic disadvantage; to be of general validity, the concentration of the constituent employed or the ratio of its concentration in brown rice to that in milled rice would have to be relatively constant for all varieties of rice and in all lots. This is not, however, the case. Furthermore, the processes that are capable of altering the distribution of constituents within the grain (parboiling, for example) limit their own validity. Lastly, the same measurement is obtained over a relatively wide range of the milling process (see figure 2).

Methods based on the grain's optical properties make use of reflectance and transmittance in the visible part of the spectrum [26, 38, 40, 41, 42]. Because of the effect of colour and of the crystalline structure of the rice itself, which differs from one variety to another, on the readings obtained, these methods cannot be widely used. To avoid this problem it has been proposed that the readings should be taken on two different wavelengths: 600 nm (far red) and 850 nm (near infra-red) [44]. The readings are, however, influenced by the humidity, the presence of abnormal grains and the age of the processed rice.

### Control of bran production

The methods described above provide a means of monitoring industrial milling step by step in order to produce high quality rice and bran with optimum yields. It might also be useful to carry out laboratory tests to evaluate the behaviour of a particular variety or lot of rice during the milling process, in order to predict the quantity of bran that would be produced in industrial processing and the ease with which the germ could be separated, or to study the properties of the bran thus obtained. In this case the following

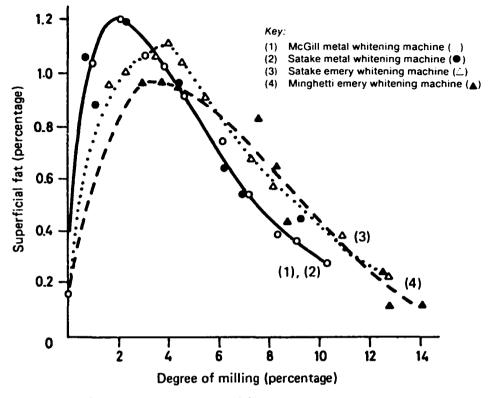


Figure 2. Variation in the surface fat content of rice milled in different whitening machines, expressed as a function of the degree of milling

Source: Shams-ud-Din and Bhattacharya [43].

factors would have to be taken into account: (a) the previous history of the rice lot; (b) the type of experimental mill to be used; and (c) the conditions under which the milling is carried out. The sample of rice to be tested, which has to be representative of the lot as a whole, must be completely freed of foreign matter, following standard procedures [45]. The moisture content of the rice must also be appropriate. If it is too low (less than 12 per cent, wet basis), the rice will tend to break too much and the bran will be adversely affected. The samples must be tested at the moisture content recommended for industrial milling, roughly 13 per cent. Once the sample has been prepared, a suitable experimental mill must be chosen, which might simulate better the conditions under which the rice in question will be processed. In this connection it should be remembered that various designs and working methods are available [45], differing in: (a) the number of stages in processing, namely husking and whitening in a single step, using a single machine, or in two different steps, using two devices; (b) the method of feeding the rice, either in batches, in a continuous stream, or using a combination of a continuous-feed sheller with a batch whitener; and (c) the method of milling, using either an abrasion-type whitener or a friction whitener. There are many laboratory mills using rubberroll shellers, but few simulate the conditions produced by under-runner disc huller.

### Control of hulling

It has been mentioned elsewhere that the husk is easily removed from the caryopsis by compressing the grain between its end-points (under-runner disc sheller) or on its sides by imparting a sort of shearing motion (rubber-roll sheller). But despite the ease with which the lemma and the palea can be opened and separated, the risk of damage to the caryopsis is high. In industrial practice, the outer layers of the grain are abraded or damaged and the germ, to a greater or lesser extent, is torn out. The outcome is that, if the brown rice is not whitened immediately (for example, if it has to be transported), it deteriorates. As a consequence of damage of the outer layer tissues, the enzyme lipase acts on the lipids, hydrolysing them, while oxygen, in conjunction with other enzymes such as lipoxygenase, oxidizes them. Apart from the loss of quality in the basic product, the bran produced when brown rice is milled will have a very high acidity level, even immediately after milling. Removal of the germ during hulling also has an adverse effect on the quality of the bran. Abrasion and germ separation at the hulling stage can represent a weight loss of between 1.5 and 2 per cent of the grain.

Staining with the May-Grünwald reagent as described in detail by FAO [30] provides a simple and rapid means of detecting the effects of hulling [7], so that the settings and operating conditions of the machine may be corrected while it is running. The caryopsis shows as green, if it has not been damaged at all, or as blue where the outer layers of the bran have been damaged. The germ also shows as blue if it has become fragmented. A pink colour is visible where the starchy endosperm has been laid bare by removal of all the layers of bran or germ. The caryopsis is always damaged when disc shellers are used, but machines can be adjusted to reduce damage to a minimum, while maintaining an industrially acceptable level of efficiency. With rubber-roll shellers, damage can be avoided and the brown rice will have excellent keeping qualities.4 However, this type of huller, because of wear on the rollers, is frequently operated under less than optimal conditions. In any event, the caryopsis sustains a proportionate amount of damage. Staining with indigo carmine or fuchsin [26] has also given good results in assessing damage. Indigo colours the bran red, while the germ is coloured yellow when it is undamaged but red if even only a little of it has been separated from the bran. Using this method, it was possible to detect 10 per cent of the grains with damaged germ in a sample from the Bluebonnet variety and 27 per cent or more in the Zenith variety when a stone disc huller was used; practically no change was seen when a McGill rubber-roll sheller was used.

### Control of whitening

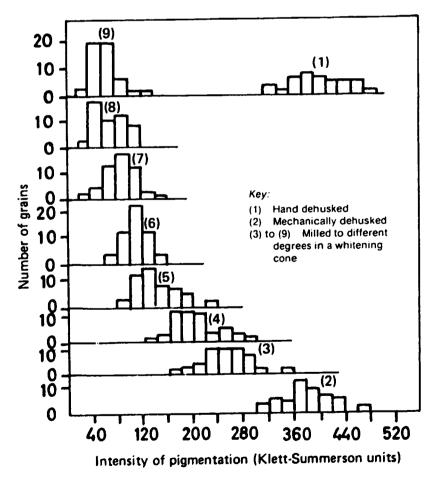
Srirangarajan and others [47] have studied the variations in the amount of residual bran in individual grains of rice at various successive stages in the whitening process, using a commercial abrasion cone. They used a variety with a red pericarp. From each individual grain of seven samples of 50 grains taken from each of the seven stages into which the whitening process was subdivided,

<sup>&</sup>lt;sup>4</sup>Houston, McComb and Kester [46] have shown that brown rice from rubber-roll shellers keeps better than brown rice from disc shellers. Before the introduction of rubber-roll machines in India, rice would not keep even for two months.

they made an extract using 3 ml of boiling 2-per-cent sodium bicarbonate for 20 minutes; the extract from each grain, made up to 3 ml and filtered, was measured by a colorimeter and the readings normalized for an average weight of 25 mg to make them comparable. The data (see figure 3) revealed a wide disparity in the amount of bran recovered from each individual grain at every stage in the whitening process.

Tests, based on the CBB index, of varieties with a normal (uncoloured) pericarp also showed a clear lack of uniformity in the degree of milling among grains of the same batch at the same stage in the whitening process. This means, of course, that a high percentage of grains of the same degree of milling will be found in samples taken from different stages in that process [24] (see figure 4). Two important conclusions may be drawn from the results of these tests. First, even in the first two cones (out of a total of five), there will be a high percentage of grains that have lost over three quarters of their bran content, while in successive cones there will also be a loss of starchy endosperm, so that a less oil-rich constituent is incorporated into the by-product. Secondly, this lack of uniformity at the same stage of whitening varies

Figure 3. Histograms of the intensity of pigmentation of residual bran in individual rice grains, at different degrees of milling



Source: A. N. Srirangarajan and others [47].

markedly from one mill to another, depending on the conditions under which the grains are processed, the variety of rice involved and other circumstances. In general, the degree to which the bran is contaminated by starchy endosperm is directly related to the lack of uniformity at each stage of the whitening process.

### Rice bran, rice bran fractions and related by-products: terminology and definitions

Bran is a by-product resulting from the processing of rice. Even though the process, which consists in removing certain outer layers of the caryopsis, is basically the same in all cases, it can be carried out in a number of ways, some of which have a vital bearing on the characteristics of the end-product. The bran thus produced may have quite different properties; therefore their particular identity should be differentiated and maintained. The variety of products is, however, sometimes accompanied by a profusion of terms used to designate one single product which creates great confusion, particularly at international level, and has serious implications for anyone consulting a specialized bibliography. For instance, the data published on "commercial bran" from Spanish mills cannot always be properly interpreted outside Spain, since the germ is separated from the industrial product before it leaves the mill. In any case, no mill, no matter how perfect, can separate pre-fixed cell layers of the grain. This makes it difficult to establish rigid definitions for the various milling products, and means that more or less arbitrary classifications must be used. Table 2 summarizes the most important or usual designations for bran, bran fractions and related by-products, all of which are defined in the following paragraphs.

Product	Husk	Pericarp. tegmen. seed coat	Aleurone	Starchy endo- sperm	Germ	Impurities
Hulling bran—from disc huller						
(salvado de descascarilladora						
de piedra) <sup>a</sup>	+	+	ο	tr	+	×
Bran (salvado)	×	+	+	+	+	×
Bran with germ removed						
(salvado desgerminado)	×	+	+	+	0	×
Germ (germen)	×	tr		o	+	×
White bran (harina cilindro) <sup>b</sup>	×	tr	+	+	tr	×
Polish (harina de pulidora)	×		+	+		×
Bran from huller-type mill						
(salvado de molino "huller") <sup>c</sup>	+	0	0	0	0	×
True bran (salvado verdadero)		+		2	-	

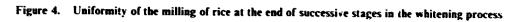
 TABLE 2.
 TYPES OF BRAN AND FRACTIONS: CONSTITUENTS AND ENGLISH AND SPANISH TERMINOLOGY

*Note:* + Major constituent, o Minor constituent, tr Trace,  $\times$  Proportion varies depending on the type of rice used, the milling procedure etc.

<sup>a</sup>Bran from rubber-roll hullers consists principally of husk and impurities.

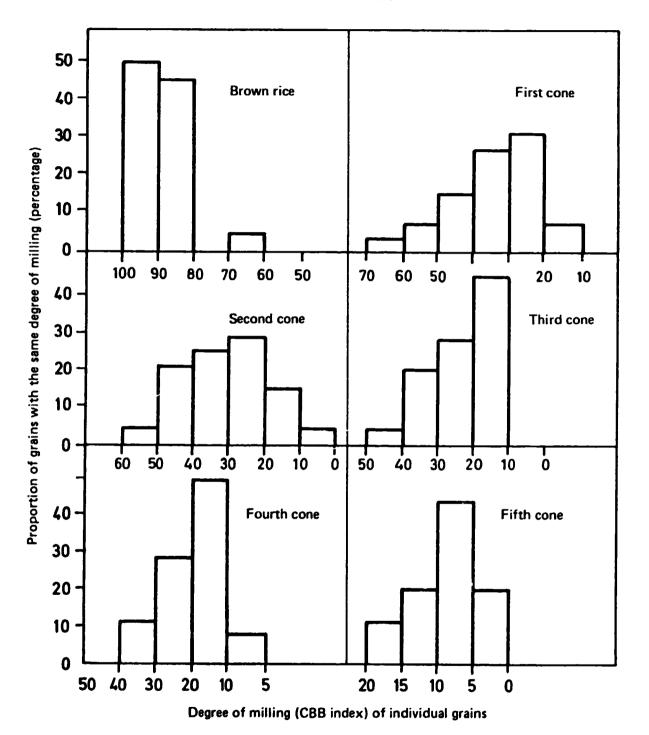
<sup>b</sup>Also called polish or polishings from whiteners.

<sup>c</sup>One stage huller-mill.



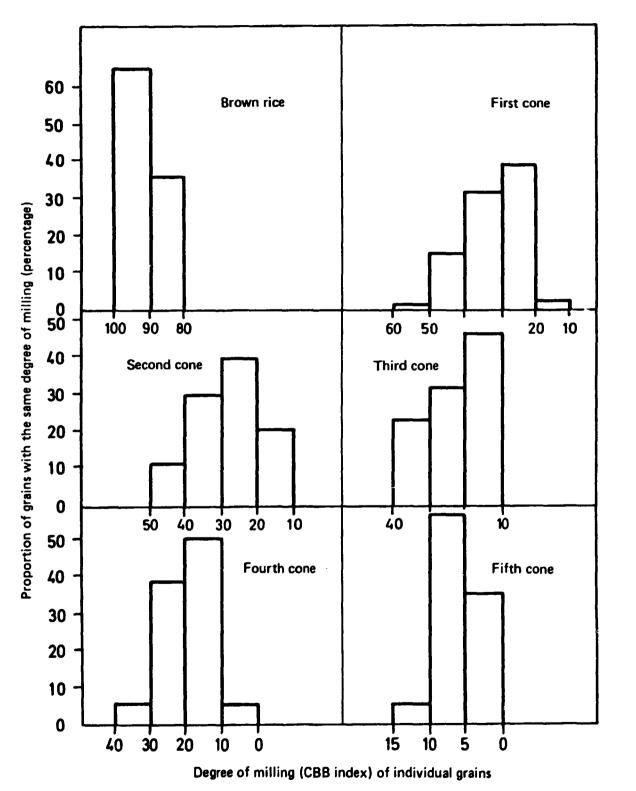
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A. Industrial mill A

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### B. Industrial mill B

### Bran from the hullers<sup>5</sup>

Bran from the huller, called husking bran or hulling bran, is the bran obtained when removing the husk from paddy rice. It can be produced from hullers with abrasive discs or rubber rollers. When the former are used, the bran is largely made up of fragments of husk, pericarp and germ; it also includes a small proportion of tegmen, aleurone and traces of starchy endosperm, and contains a great many impurities. The bran produced from rubber-roll huilers consists essentially of fragments of husk and impurities, with a very small proportion of pericarp. These bran fractions are usually mixed with the bran from whiteners (see later) to make them commercially acceptable. Other names used include "dehusking bran" (salvado de descascarado) [48]. In certain parts of Mexico this is called simply "bran" (salvado), to distinguish it from the by-product obtained form the whitening process, which is called "polishings" or "polisher flour" (puliduras or harina de pulido).

### Bran

Bran is the by-product obtained by removing the outer layers from brown rice to whiten the kernel. It consists mostly of pericarp, tegmen, aleurone, whole germ and crushed germ and starchy endosperm, in the form of dust and small fragments; it includes varying amounts of scraps of husk and impurities. When the term is used to distinguish "bran" from "white bran", it refers to the by-product obtained in the first (undefined) stages of whitening.<sup>6</sup> In practice, the demand for white bran is usually much lower than the actual production and in many countries it is not even marketed. As a result, white bran and bran are usually mixed in the proportions in which they were produced, being offered commercially as a single by-product under the general term "bran". Other names used in Spanish are: *afrecho* (another synonym for bran) [48]; *puliduras* and *pulido de arroz* in Mexico; *polvillo* in Peru; *polvo de arroz* in Cuba; *harina de pulimento* in Colombia; and *semolina de arroz* in Costa Rica; in the Philippines the terms *darak* and *tikitiki* are used.

#### Bran with germ removed

Bran with the germ removed is a bran from which the whole germ and some of the fragments of non-powdery endosperm have been extracted by sieving and suction. It always contains some of the smaller whole germ and some crushed germ.

### Germ

This by-product is the germ of the rice, detached and removed in the milling process as part of the bran from which it is separated later. It is mostly whole, although it can include some crushed germ; it also contains fragments of starchy endosperm (generally between 15 and 30 per cent by weight) and some husk and impurities. Other terms used are "embryo" and, in Spain, *morret*.

<sup>&</sup>lt;sup>5</sup>Machines used to remove the husk from the paddy grain are known by different names: hullers, huskers, dehuskers, shellers, hulling mills.

<sup>\*</sup>Defined by FAO as "a by-product of rice milling, consisting of the outer bran layers of the grain together with part of the germ".

### White bran<sup>7</sup>

White bran is the by-product obtained in the last stages of whitening. It is a whitish, floury material, soft to the touch, somewhat fibrous, mostly consisting of starchy endosperm and aleurone, with the remains of pericarp, tegmen and crushed germ. It contains very little, if any, husk or impurities. It is usually sold mixed with bran (see above). Other names used are *cilindro* (in Spanish) and polish or polishings (from whiteners).

White bran from various whitening machines can be differentiated by indicating the number of the whitening stage—fourth cone, for example. The term in English is "rice polish" or "polishings", though the latter term also includes the by-product from the brushing machine (see below).

#### Polishings<sup>8</sup>

Polishings are produced exclusively by a polishing device made of leather or some similar material (see above). For all practical purposes, it contains only starchy endosperm and aleurone; husk residue and impurities are usually absent.

### Bran from the huller-mill

Bran from a single-stage huller mill consists of a mixture of husk crushed with particles from all the outer layers (including the germ) and starchy endosperm. It contains a large number of small finings and impurities. It is known as *kiskis* in the Philippines [50].

Using graduated screens, this product can be sieved to produce two fractions: coarse bran and fine bran. In the former, the larger fragments of husk predominate. In any case, the mix of the components varies not only according to the mill and type of rice used but also to the fineness and efficiency of the final sieving.

### True bran

"True bran" is a term used by certain authors to decribe the bran fraction consisting of the pericarp layers, tegmen and seed covering. It is not obtainable industrially.

### Other fractions

The cooling air of the whitening machines carries away particles of bran that can be recovered, generally using a cyclone. The composition of the flour from the cyclone is very variable. It depends on the streams involved and the number and type of machines producing them.

### Factors determining the properties of bran

No whitening machine is able to separate consecutively the distinct structural layers of the outer part of the caryopsis in all the grains at once. One

<sup>&#</sup>x27;Defined by FAO as "a by-product of rice milling, consisting of the inner bran layers together with part of the germ and a small proportion of the starchy interior".

<sup>\*</sup>The machine used is also called a "brushing" machine [49]. In certain countries, whitening machines are called "polishers", and this leads to some confusion in the naming of by-products.

reason is that whitening is an operation in which the grains do not come into uniform contact with the abrading surface and are not all subjected to the same amount of rubbing action. Other reasons lie in the grain itself. From one variety to another and even within a single lot, the rice grains differ substantially from each other in their anatomical, physical and chemical characteristics. Some of these overlap, and in the combined result it is hard to determine the influence of the various constituents. It therefore seems advisable to make a detailed and methodical analysis of all the factors that affect the properties of industrially produced bran. To this end, they are classified into: (a) rice-related factors; and (b) factors related to the milling process. Both groups can be further subdivided as follows:

### A. Rice-related factors

- 1. Anatomo-morphological
  - (a) Size and shape of the grain
  - (b) Anatomical layers
- 2. Mechanical
  - (a) Resistance of the grain to abrasion and friction
  - (b) Resistance of the grain to breakage
  - (c) Ease of separation of the germ
- 3. Chemical
  - (a) Average chemical composition
  - (b) Distribution of constituents
  - (c) Defective grains
- 4. Soundness
  - (a) Physical contaminants
  - (b) Chemical contaminants
  - (c) Biological contaminants

### B. Process-related factors

- 1. Preliminary operations
  - (a) Cleaning
  - (b) Grading
- 2. Milling stages
  - Single-stage versus multi-stage
- 3. Machinery and operational conditions
  - (a) Hullers
  - (b) Paddy separators
  - (c) Destoners
  - (d) Whitening machines
  - (e) Grading machines

The groups and subgroups are discussed in detail below.

### **Rice-related** factors

### Anatomo-morphological features of the grain

It is possible to distinguish: (a) the size and shape of the grain; and (b) the dimensions of the anatomical layers.

### Size and shape

The size (length), shape (ratio of length to width) and weight of a grain of brown rice varies substantially from one commercial variety to another (see table 3). The maximum and minimum values—length 5.4-7.5 mm, ratio of length to width 1.8 to 3.6 and weight 16-24 mg for rice varieties from the United States [51]—reveal a wide range both in the sphericity index and the ratio of surface area to weight, bearing in mind the fact that the varieties with the longest grains weigh the least. Assuming that all the other properties are equal (thickness of the bran layer, size of the germ etc.), the least spherical grains and those with the greatest surface to weight ratio should produce the greatest quantity of bran. It seems that no study has been made to date of the effects of these factors. A wide range of grain lengths for paddy rice will reduce the efficiency of the huller and if there is no paddy separator, or if there is one but it does not work efficiently, a considerable proportion of rough rice will be present when the whitening process begins.

TABLE 3. SIZE, SHAPE AND WEIGHT OF COMMERCIAL BROWN RICE GRAINS FROM THE UNITED STATES OF AMERICA

Type of grain	Average length (millimetres)	Average ratio of length to width	Average weight (milligrams)
	7.0-7.5	3.4-3.6	16-20
Long Medium	5.9-6.1	2.2-2.4	18-22
Short	5.4-5.5	1.8-2.0	22-24

Source: Webb [51].

A grain of rice has been described as a conical-elliptical cylinder, or as a paraboloid of revolution, with ridges and grooves on its surface, and this is why the separation of bran is not uniform across the whole surface. An investigation under the microscope into the effects of whitening (using McGill No. 3) on different parts of the grain [52] (see figure 5) has shown that: (a) the most prominent ridge is subjected to a greater abrasive effect, with a correspondingly higher loss of bran, than other points on the surface; (b) less bran is lost from the grooves; (c) the abrasive effect is less in the dorsal area than in the ventral portion of the grain; and (d) the central portion of the grain is altered less than the terminal portions.

Differences between varieties or lots in the morphological properties cited above are translated into differences in the bran produced. Thus, for example, the presence of deep grooves forces the miller to prolong the milling process in order to obtain rice of the customary standard of whiteness. As a result, much more starchy endosperm is incorporated into the bran, because of the deeper abrasion and the additional fines that are produced.

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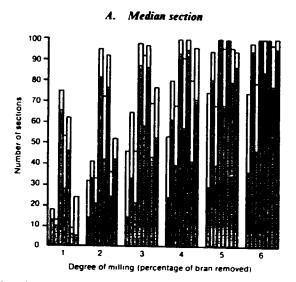
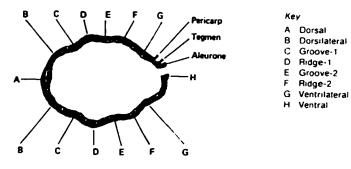
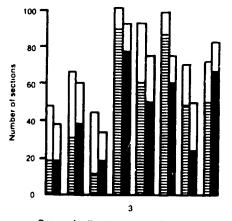


Figure 5. Histogram representing percentage of sections with denuded bran and aleurone layers at different degrees of polish

*Note:* The eight bars from left to right in each group pertain respectively to the portions labelled  $\Lambda$  to H in the transverse section of the rice grain shown below:



B. Distal and median sections compared



Degree of milling (percentage of bran removed)

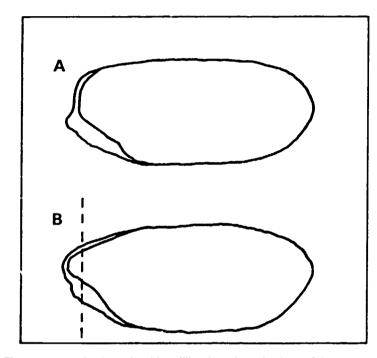
*Note:* In the distal sections the bran is represented as  $\Box$  and the aleurone as  $\blacksquare$ ; in the median sections the bran is represented as  $\Box$  and the aleurone as  $\blacksquare$ 

Source: Srinivas and Desikachar [52].

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Grains in which the lower end of the starchy endosperm protrudes are typical examples of the distinctive, though still normal, shapes that may be found (see figure 6). The elongated projection, which must be considered as a defect, breaks easily during the whitening process and this tends to increase the fractions of small broken germ and bran.

Figure 6. Projections produced by milling



*Note:* The percentage of ends produced by milling depends on the shape of the caryopsis. Shape A resists breakage better than shape B.

### Anatomical layers

As has been shown elsewhere, the number of cell layers in the bran varies. Varieties with a short oblong grain generally have more leurone cell layers than those with a long grain. Highland varieties have more layers in the dorsal and ventral portions of the grain than lowland varieties. High temperatures during ripening also increase the number of aleurone layers, although only in the dorsal portion. In a study made of 40 varieties, including the *indica* and *japonica* sub-species and hybrids [53], the ranges of variation in thickness of the outer bran layer and the aleurone layer in various portions of the grain—dorsal, lateral, central and terminal—have been evaluated (see tables 4 and 5). The ranges of variation are wide; the minimum values are generally a half or even a third of the maximum values. The variations imply substantial differences in the bran obtained and in the oil it may contain, although these have not been evaluated.

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	Outer layer	Aleurone layer		
Statistic	Dorsal portion	Lateral portion	Dorsal portion	Lateral portion
Maxin am	69.0	37.0	89.0	56.0
Minimum	28.0	3.0	30.0	17.0
Mean	45.9	19.1	67.3	32.3
Standard error	1.9	0.9	2.1	1.8
Coefficient of variation (%)	25.5	27.2	19.3	34.7

### TABLE 4. STATISTICS OF MEASUREMENTS OF THE THICKNESS OF THE BRAN LAYER OF THE RICE CARYOPSIS (Micrometres)

Source: Manoharkumar and others [53].

Note: Summary data from a study made on 40 pure strains of paddy representing indica and japonica sub-species and their hybrid selections.

TABLE 5.	STATISTICS	OF	MEASUREMENTS	OF	THE	THICKNESS	OF	THE	BRAN
	LAYER	AT	DIFFERENT POINT	IO 81	N THE	CARYOPSIS			

	(М	(icrometres)	1			
	Germ	portion	Centra	l portion	Opposite end to the germ	
Statistic	Dorsal	Lateral	Dorsal	Lateral	Dorsal	Latera
Maximum	167.0	63.0	150.0	71.0	146.0	67.0
Minimum	67.0	25.0	75.0	25.0	84.0	29.0
Mean	120.0	47.4	112.8	52.2	108.0	49.0
Standard error	2.85	1.36	2.77	1.61	2.75	1.52
Coefficient of variation (%)	15.0	18.2	15.3	19.3	15.5	18.8

Source: Manoharkumar [53].

Note: Summary data from a study made on 40 pure strains of paddy representing indica and japonica sub-species and their hybrid selections.

The size of the germ is another factor to be taken into account. Germ is an important part of the bran, especially when the bran comes from lightly processed brown rice to only a little milling, in which case it can account for more than 25 per cent of the end-product. The size of the germ in different varieties can vary considerably.

### Mechanical properties of the grain

The mechanical properties of the grain include: (a) its resistance to abrasion and friction; (b) its resistance to breakage; and (c) the ease with which the germ can be separated.

### Resistance of the grain to abrasion and friction

The bran becomes detached most easily in places where the aleurone layer is thinnest [54]. During whitening, the bran comes away earlier in the lateral portion of the grain—where the layer is least thick—than in the dorsal portion [53]. Findings have been published on differences in resistance to abrasion, not only between one variety and another (see figure 7) [7] but also between lots of the same variety (see figure 8) [7, 55].

Reports on variations in the resistance of the grain to abrasion according to moisture content [57] have shown that the lots with the lowest moisture content (6-10 per cent) were much more resistant. The fact that grains of parboiled rice have a high resistance to abrasion is particularly well established [56].

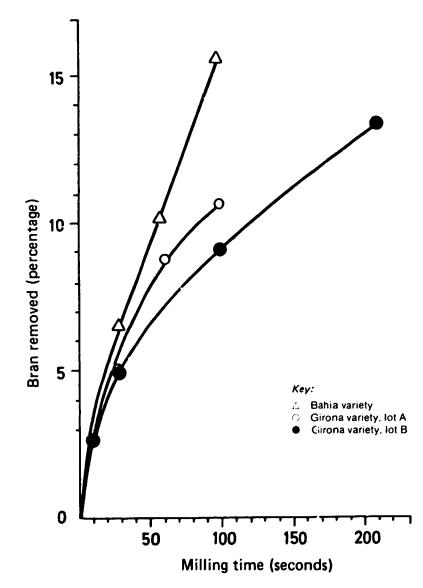


Figure 7. Resistance to abrasion during the milling of different varieties and lots of brown rice

 $\ell$ 

Source: Barber and Benedito de Barber [7].

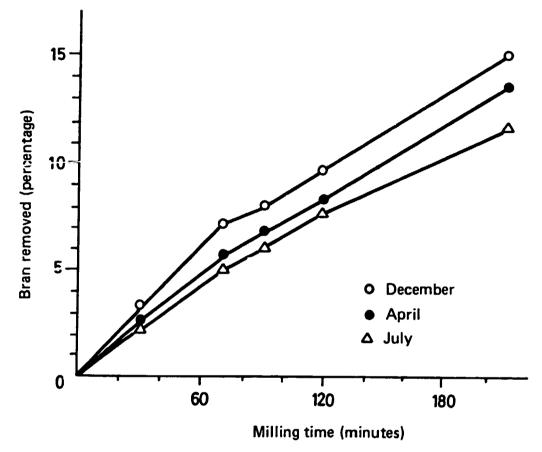


Figure 8. Changes in abrasion resistance during the storage period of brown rice milled in a differential mill<sup>a</sup>

Source: Barber [55].

<sup>*a*</sup>The rice was stored in hermetically sealed containers, at a temperature of  $35^{\circ}$  C. It had a moisture content of 13.2 per cent.

### Resistance of the grain to breakage

Koga [58] has depicted graphically what happens to the broken rice during the whitening process (see figure 9). The brokens are abraded more easily than the whole grain, especially on the surface of rupture, and the flour that is removed from the starchy endosperm goes into the bran. In varieties and lots with a high yield of brokens, this factor greatly affects the quality of the bran.

### Ease of bran removal

It has been pointed out [59, 60] that the ease with which the germ can be removed from its normal position in the caryopsis probably depends on two factors: first, the flimsiness of the scutellar epithelium, which is made up of empty, flattened cells; and, secondly, the poor cohesion of the starchy endosperm in this region, due to its powdery consistency and lack of welldefined walls of cellular tissue. In consequence, the germ is not separated

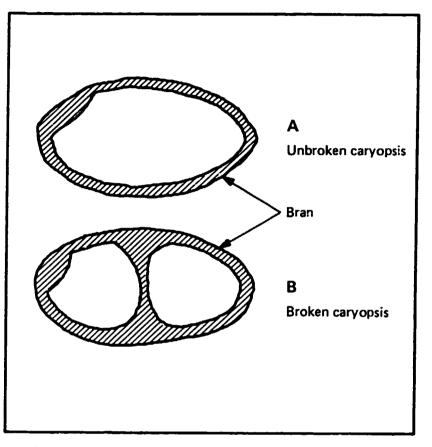


Figure 9. Parts of the rice caryopsis that are separated as bran during the whitening process

through continued abrasion but is usually torn out suddenly under the force of a well-placed impact. Yet, either because of differences in the degree of cohesion or because the concavity of the endosperm surrounding the germ protects it in different ways, varieties of rice differ considerably in the ease with which the germ can be removed during the whitening process (see table 6) [61].

Variety	Туре	Percentage of grain. with residual germ
Kinmaze	Glassy	5.6
	Pearl	5.6
Fukuminosi	Glassy	8.3
	Pearl	6.6
Ginmasari	Glassy	40.6
	Pearl	32.3

 
 TABLE 6.
 REMOVAL OF THE GERM DURING THE WHITEN-ING PROCESS

Source: Kanda, Ikehashi and Ito [61].

Source: Koga [58].

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Since the germ forms an important part of the bran, such variations may well mean that there will be major differences in the make-up of the bran.

In parboiled rice the situation is different. Treatment gelatinizes the starch, and so the endosperm and the germ remain effectively bonded together. The germ does not "jump" out, and its separation from the rest of the caryopsis is brought about by continued abrasion [62]. Unless milling is very prolonged, parboiled rice always contains some residual germ.

Vasan and others [63] have studied the changes in the weight and oil content of the germ caused by parboiling (see table 7). During the process the germ loses a great deal of weight but its oil-content increases; it does in fact retain most of its original oil. If, however, germ removal from parboiled rice is total (which is difficult to achieve before 5 per cent of the bran calculated on the brown rice has been eliminated), its contribution to the oil-content of the bran is the same as in the case of raw rice.

 
 TABLE 7.
 EFFECTS OF PARBOILING ON THE WEIGHT OF THE GERM AND ITS OIL CONTENT

Samples (brown rice grain)	Germ (perceniage)	Oil in the germ (percentage)	
Raw rice	3.4	35.7	
Parboiled rice	2.1	45.8	

Source: Vasan and others [63].

### Chemical characteristics of the grain

One factor that is responsible for considerable variations in the characteristics of the bran is the chemical composition of the grain. It is probable that its importance will increase in proportion to the advance of crop improvement programmes that may include, amongst their objectives, the development of rice varieties with higher-quality bran. Certain rice varieties have already been obtained which, instead of containing between 1.2 and 2.0 per cent oil (the usual proportion), contain approximately 4 per cent [64].

The distribution of chemical constituents in the grain is a second factor to be considered in relation to the properties of the bran. In the caryopsis there is a gradient of decreasing concentration—except for starch, which increases from the outer layers towards the centre of the grain. The fat distribution is different:<sup>9</sup> the concentration increases from one outer layer to the next inner one, reaches a maximum, and then falls off towards the middle of the grain. Although the shape of the distribution curve is generally the same for all varieties of rice, there are quantitative differences that have a bearing on the composition of the bran [22]. Parboiling produces quantitative changes in the oil concentration by regions but does not affect the general pattern of distribution (see figure 10) [63, 65]. The increase in the fat content of bran has been well documented, as have some cases of differences between brans resulting from different degrees of milling (see table 8).

<sup>&</sup>lt;sup>9</sup>Protein distribution also.

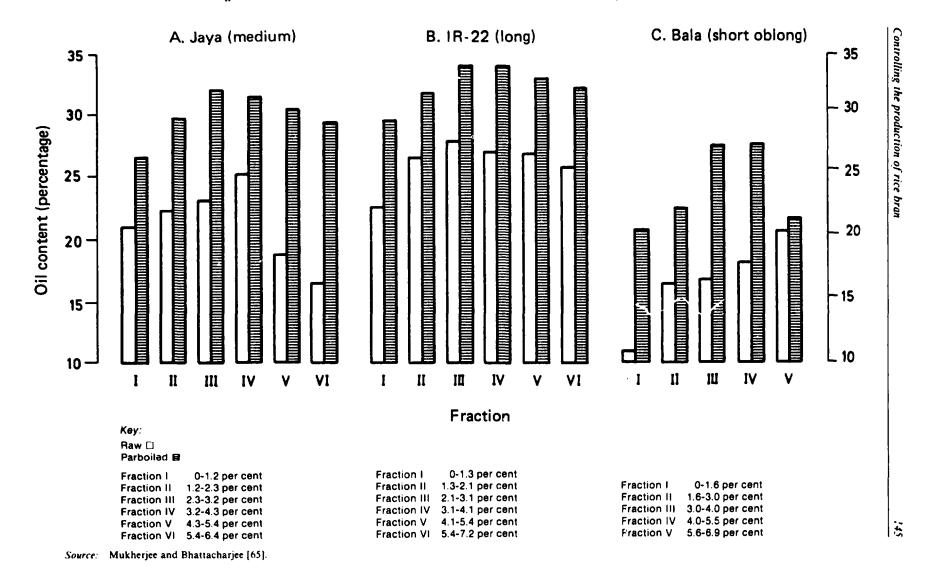


Figure 10. Oil content of successive fractions of bran extracted from raw and parboiled rice

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Amount of bran after polishing (grams of bran per 100) grams	IR-8		GEB-24		CO-33	
of brown rice)	Ran	Parboiled	Ran	Parboiled	Raw	Parboiled
2	24.1	32.2	30.2	32.9	26.4	30.6
5	25.4	34.2	30.2	34.0	27.2	36.0
8	22.5	30.1	26.9	29.1	27.3	34.0

TABLE 8. EFFECTS OF PARBCILING ON THE OIL CONTENT OF BRAN

Source: Vasan and others [63].

### Sanitary condition of the grain

Physical and chemical contaminants, micro-organisms and insects all affect the sanitary condition of the grain. It is customary for rice to be stored for long periods, with the accompanying risk of deterioration caused by the action of insects and micro-organisms. The use of chemical compounds to protect the harvest is widespread. Although most of the contaminants are removed along with the husk, the bran is not necessarily free of them. Some insecticides —malathion, for example—are known to degrade in time, but the degradation takes several weeks (months in the case of malathion) and metabolites (mainly dimethyl-phosphorothionate) are produced in the process [66].

The usual microbial flora of rice include micro-organisms capable of producing mycotoxins. It has been shown that a high proportion (60 to 80 per cent by weight) of the toxins in the grain pass into the bran during milling. The resulting bran contains 10 times as much toxin as milled rice [67]. Given that, under favourable conditions, rice is sometimes exposed to the rapid growth of micro-organisms, with the consequent production of toxic metabolites, the presence of these toxins in bran is not at all improbable.

### Factors related to the milling process

As has already been shown, there are three factors related to the efficiency of the milling process: (a) preliminary operations; (b) number of steps involved; and (c) machinery used and its operating condition during milling.

### **Preliminary** operations

Paddy rice can contain stones, sand, soil, metallic objects, straw, weed seeds etc., which greatly reduce its value. Most, and possibly all, of this foreign matter ought to be removed before the husk is removed. In addition to improving the quality of the bran, there would be the additional advantages of a more attractive milled rice product, and more efficient operation of the hulling and whitening machines and reduced wear and tear on them. The hardest and most durable foreign matter causes excessive wear on the most delicate parts of the equipment and degrades the actual conditions of processing—for example, it can damage the screens of the whitening machine and lead to the inclusion of brokens and even whole grains in the bran. Many of the operations carried out immediately after the rice is harvested (e.g. sun drying and threshing with livestock) contribute in large measure to the presence of foreign matter in paddy rice, so much so that any improvement in the operations can go a long way towards solving the problem. Nevertheless, paddy rice, whatever its provenance, can not reach the hulling stage in good condition unless it has been subjected to a thorough and effective cleaning. This cleaning is also made easier if the size of the grains of rough rice is uniform; otherwise, not only is the efficiency of the operation reduced but there is a risk that grain will be lost in the process.

Where this foreign matter finally ends up depends on the nature of the particles themselves and on the pattern of milling. It has been pointed out elsewhere that at present the single-step huller-type mill, without a cleaning stage, is the prevailing mill in many rice-growing countries, particularly in Asia. In this case, most of the inert impurities (sand and mud) always pass into the bran-although crushed rice husk is the major impurity. The efforts of some Governments to promote even limited modernization of these installations, if successful, must contribute towards a substantial improvement in the quality of the bran. In more recently built installations, and in older ones that have been modernized, cleaning forms part of the process. In many cases, however, the cleaning capability of the machinery is not put to full use. Soil, sand and husk fragments, even when only present in small quantities, greatly affect the food value of the bran. The effect of chewing a bran-based product containing a small grain of sand is surprisingly unpleasant. In anticipation of this sort of problem, the cleaning of paddy rice will have to meet more rigorous standards if quality bran is to be produced.

The pre-cleaning of paddy rice has been dealt with in detail by Gariboldi [68] and van Ruiten [69].

### Number of steps in the milling process

The huller-type mill, used to carry out the simultaneous hulling and whitening of paddy rice in a single stage, produces a by-product of poor quality, as it is a mixture of bran (15-25 per cent) and crushed husk (75-85 per cent) [70]. Since crushed husk is included, as well as many small brokens, the oil content is low (about 12-15 per cent) (see table 9).

## TABLE 9. COMPOSITION OF THE BRAN PRODUCED IN A HULLER-TYPE MILL AND IN A CONE-TYPE MILL

(Percentage, wet weight)

	(	Cone-type mill		Huller-type mill			
Component	Variesy Anuradhapura	Variety Hambantota	Variety Batticaloa	Variety Anuradhapura	Variety Hambantota	Variety Batticaloa	
Protein	12.68	12.67	12.29	8.53	8.97	9.27	
Fat	15.31	14.94	13.59	5.85	7.50	6.12	
Fibre	8.69	9.47	8.68	18.85	21.24	17.32	
Ash	10.09	10.95	10.97	22.40	18.82	17.81	
Nitrogen-free							
extract	42.17	41.72	44.74	33.90	32.00	38.10	

Source: Siriwardene [71].

Attempts have been made to improve the bran obtained in this way by subsequent sigving, separating it into a fine fraction, containing more oil, and a coarse fraction, containing more husk. Although well-established in some places as a commercial practice, this further process does not really solve the problem. No husk that has been reduced to small particles, soil and mud can be separated in this way; the bran is still silica-rich and low in oil (below 10 per cent), for which reason it is still a low-quality product in the two alternative uses to which it can be put directly—for the extraction of oil and as fodder for livestock.

A large-scale inclusion of husk in the bran can only be avoided by using two-stage or multiple-stage processes. Only in this way can a bran be recovered with the high oil content and low silica needed for a quality product.

There are a number of advantages to be gained from using a multi-stage whitening process. First, since the pressure on the grains is less, there is also less risk of deep abrasion, which rubs away portions of the starchy endosperm together with scraps of the bran layers; secondly, since fewer brokens are produced, less flour and fewer small fragments of the starchy endosperm are included in the bran. The result is a bran with a higher oil content.

### Types of machines and processing conditions

The hulling machine also plays a part in the industrial production of bran. It has been shown elsewhere that the two types of hullers in common use differ in their effects on the rice, in that the stone-disc huller damages the bran layers and takes some of the germ out, while the rubber-roll type leaves the caryopsis practically intact. With the former machine, between 1.5 per cent and 2 per cent of bran is produced, in the latter less than 0.5 per cent. The difference lies not only in the quantity of bran produced but also in its composition (see table 10).

Component	Disc-type huller	Rubber-roll hulle
Moisture	9.2-13.5	11.7
Protein	8.1-11.6	3.8
Fat	6.5-10.4	0.8
Fibre	14.8-22.6	41.5
Ash	11.2-20.4	13.2
Nitrogen-free extract	31.0-40.3	28.9

 	 COMPOSITION PES OF HULLING	 	
	(Parcentega)		

Source: Borasio [72], Leonzio [73], Primo and others [74].

The bran from the stone-disc machine consists of fragments of husk, bran (basically pericarp), some germ and rachilla, and small endosperm fragments, together with inorganic impurities, while bran recovered from roller machines contains scarcely any germ and bran. This explains why the bran from the rubber-roll machine is poorer in fat and richer in fibre. The above descriptions apply only to machines in which the adjustment is perfect. Unfortunately this is not usually the case, particularly with stone-disc machines, for which a precise adjustment of the gap between the two abrasive surfaces is most critical. It will be recalled that the disc machine presses on the grain at both ends of its longer axis (length), while the roller machine presses on the ends of the minor axis (thickness). The length of the grains varies more than the thickness; furthermore, the surface of the discs is not elastic like that of the rollers. If the lengths are very different, the longest grains break and the shortest ones emerge unaffected. If the gap between the discs is not correctly adjusted or if, as a result of wear, it is not uniform, or if the machine is fed grain too fast, the resultant abrasive action on the grains leads to the elimination of larger quantities of bran.

When rice is dehusked on a rubber-roll machine, the bran separated in the subsequent whitening process is richer in oil than that obtained with a stonedisc hulling machine (see table 11).

TABLE 11.	EFFECT (	OF DIFFERENT	Г TYPES OF	HULLING	
MACHINES	ON THE OI	L CONTENT OF	F BRAN FROM	M WHITEN-	
ING MACHINES					

Type of huller	Oil content (percentage)
Abrasive disc	15-17
Rubber-roll	18-20

Source: Aggarwal and Sarda [75].

The efficiency of the hulling machine also affects the properties of the bran. No machine dehusks 100 per cent of the paddy rice in a single pass. Moreover, if the hulling machine is worn or badly adjusted, or if the size of the rice grain varies, the percentage of the grains that remain unaffected on passing through the machine is high. Unless the machine used to separate the paddy rice is operated with the greatest care (which would in this case imply a very low yield), the brown rice going into the whitening machines will be adversely affected.

The average size of the grains of rough rice rejected by the paddy separator is considerably smaller than that of the rice that goes to the hullers, and this is why some grains come out unprocessed. The installation of a "return" hulling machine, which only processes the grain rejected by the paddy separator and is specifically adjusted to deal with the average size of these grains, is one good solution, which makes it easier to ensure that no rough rice is fed into the whitening machines.

As has already been pointed out, paddy separators can operate very close to 100 per cent of their efficiency, with industrially acceptable yields. This fact should be taken into account when a high-quality bran is wanted.

Destoners should also be mentioned. They can be placed before the hulling machines or before the whitening machines. Stones and clods of earth of roughly the same size as the rice grains have to be eliminated before whitening, since that is when they are likely to disintegrate and become permanently mixed in with the bran.

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Whitening machines have a decisive influence on the properties of the bran. Of particular interest in this connection are the type of machine, the number of passes and the grade of emery used and the size of the mesh in the screen. The main variables in operating conditions are the homogeneity and the degree of milling, and the possible use of dust to polish the rice.

Whitening machines that operate by friction or rubbing produce a different bran from that obtained from abrasion machines. Bran produced by the former is coarser and somewhat more crimped or curly (see table 12); usually it contains more fat (see table 13).

		Fraction of bran retained by sieve (percentage)			
Sieve			Abrasion type machine <sup>b</sup>		h
US sieve Mesh size		Friction-type	Bran without germ		
	(micrometres)	machine	Rangec	Mean <sup>C</sup>	Bran with germ
20	841	4.4-16.0	0.2-0.4	0.3	18.3
30	595	12.0-28.0	1.8-3.8	2.9	9.7
50	<b>29</b> 7	42.0-62.0	14.9-21.1	19.1	20.3
80	177	2.0-16.0	22.9-29.6	25.0	18.1
100	149	0.5-2.0	3.2-5.6	4.1	4.4
>100	<149	_	43.0-53.8	48.6	29.2

# TABLE 12. PARTICLE-SIZE DISTRIBUTION OF DIFFERENT TYPES OF BRAN FROM WHITENING MACHINES

<sup>a</sup>Data adapted from Mihara [76].

<sup>b</sup>Data from Spanish mills.

<sup>c</sup>Data from four mills.

Component	Friction-type machine <sup>a</sup>	Abrasion-type machine <sup>b</sup>	
Moisture	11-13	12-14	
Protein	14-16	13-16	
Fat	18-21	14-16 <sup>c</sup>	
Fibre	8-10	9-10	
Ash	9-12	8-9	
Nitrogen-free extract	33-36	45-50	

 TABLE 13.
 AVERAGE COMPOSITION OF BRAN FROM BOTH

 FRICTION-TYPE AND ABRASION-TYPE WHITENING MACHINES

<sup>a</sup>Data on Japanese mills, from Mihara [76].

<sup>b</sup>Data on Spanish mills, from Primo and others [74].

<sup>c</sup>Levels of 19 per cent are not infrequent.

Single-stage whitening produces bran with less fat and more nitrogen-free extract than bran produced in several stages. First, the separation of the outer layers of the grain is more irregular, and penetration is deeper; secondly, the percentage of brokens is higher and a greater proportion of starchy endosperm is incorporated into the bran. The mesh-size of the screen in the whitening machine does not vary much from one machine to another, but wear caused by excessive use and, sometimes, incorrect adjustment, lead to adverse results. The presence of brokens and even of whole grains in the bran is usually held to be the result of a damage in the screen. If the defect is not made good, the bran quality will be observed to be progressively impaired, with a corresponding effect on the yield of milled rice. Table 14 shows these effects in a cone mill. The  $841-\mu$ m fraction contains a large number of very small brokens, as a result of the makeshift use of a damaged screen. Its influence on the oil content of the bran as a whole is easily visible.

Sieve				
US sieve No.	Mesh size (micrometres)	Fraction of bran retained (percentage, dry weight)	Oil content of fraction (percentage, dry weight)	
20	841	16.78 <sup>a</sup>	5.96	
30	595	13.24 <sup><i>a</i></sup>	12.77	
50	297	28.22	)	
80	177	10.31		
100	149	11.04	> 18.34	
>100	<149	20.41	J	
Total		100.00	15.35	

 
 TABLE 14.
 EFFECT OF BROKENS ON THE OIL CONTENT OF COMMERCIAL BRAN FROM AN ABRASIVE CONE MILL

<sup>a</sup>Usually less than 5 per cent. The high percentage reflects the presence of small brokens.

The number of stages in the whitening process and the types of whitening machine used can affect the temperature of recently produced bran. A multistage whitening process, using machines with air injection or air suction, yields bran at a lower temperature, which retards deterioration caused by enzymes.

The degree of milling has a particularly marked effect on the properties of the bran, but the ultimate reason for its influence is the distribution of chemical constituents in the rice grain, dealt with earlier.

In some countries, calcium carbonate is used in the whitening process. This material, having a particle size of less than 74  $\mu$ m, acts as an abrasive and helps to give an attractive finish to the white rice. The proportion in which it is used varies widely (0.5-2 per cent on the paddy rice) according to the class of rice and the machine used. But calcium carbonate not only facilitates the whitening; it also modifies the composition of the bran. Its use at a concentration of 0.5 per cent in brown rice represents a carbonate concentration in the bran of 10 per cent if the degree of milling is 5 per cent. This affects two properties of the bran: the measurable acidity and the ratio of calcium to phosphorus; in the first case, the carbonate acts as a neutralizing agent and in the second it has a directly beneficial effect, since the ratio of calcium to phosphorus is roughly 1:10, when for animal feed purposes it must be closer to 1:1.

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# V. Technology for the stabilization of rice bran

# Introduction

In chapter II above, it has been clearly established that the basic objectives of stabilization are: (a) to destroy enzymes, micro-organisms and insects (the chief causes of deterioration of the bran), reducing their activity to safe levels; and (b) to preserve to the maximum the valuable constituents of the bran—chiefly oil, proteins, vitamins and other nutrients. It has also been recognized that the known means of attaining the first objective are inimical to the second, and so a compromise solution is the only viable alternative, provided that the risk of deterioration is within the bounds permissible in industrial use.

In general, however, research and development work on stabilization processes so far has been largely directed towards the partial goal of halting the development of FFA. The substantial amount of work done so far now needs to be reviewed and an assessment made of the results achieved in the light of the new evaluation criteria that have been developed. In the following sections, therefore, the information available is classified on the basis of the means used to check enzymatic activity and other causes of deterioration. First, processes using chemical agents are reviewed. Subsequently the physical processes are dealt with, first analysing those based on cooling, inert atmospheres and irradiation. Finally, an in-depth study is made of the processes that use heat, the processing conditions and equipment being analysed separately.

# Stabilization methods

The stabilization methods investigated thus far may be classified into two major groups: chemical and physical. The latter may be subdivided into several types, such as cold storage, storage in an inert atmosphere, irradiation and heat stabilization.

## Chemical stabilization

Desikachar [1] has pointed out that the conditions required to inactivate isolated and purified lipase differ from those needed to achieve the same result *in situ* in the bran. This applies not only to bran but to most of the enzymes found in vegetable tissues. The effects of a number of very diverse chemicals have been studied with a view to ascertaining their possible use for stabilizing rice bran (see table 1).

157

Chemical compound	Formula	Treatment	Effectiveness	Source
Acetic acid	СН,СООН	Conditions not specified	_	Desikachar [1]
Hydrogen chloride (gas)	HCI	15 minutes stream plus 15 hours	_	Gómez Fabra and Primo [3]
Humic acid		0.01 per cent aqueous solution	+	Gómez Fabra and Primo [3]
Acrylonitile	CH <sub>2</sub> CHCN	Conditions not specified	_	Desikachar [1]
Ethyl alcohol	CH <sub>3</sub> CH <sub>2</sub> OH	Steam at 100° C for 5 minutes	-	Gómez Fabra and Primo [3]
Methyl alcohol	CH,OH	Conditions not specified	-	Desikachar [1]
Ammonia (gas)	NH,	Treat with 15 minutes stream and hold for 50 hours	-	Gómez Fabra and Primo [3]
Sulphur dioxide (gas)	SO <sub>2</sub>	(a) Treat for 15 hours in cold SO <sub>2</sub> atmosphere and aerate	-	Gómez Fabra and Primo [3]
		(b) Hold in SO <sub>2</sub> atmosphere	+	Gómez Fabra and Primo [3]
		(c) Treat for 1.5 hours in SO, stream, hold for 6 hours and aerate	-	Adachi and Futsuhara [4]
		<ul> <li>(d) Treat with 5-10 per cent SO<sub>2</sub> for 5-10 hours and dry in the sun</li> </ul>	+	Loeb and Morris [5]
		(e) Conditions not specified	-	Desikachar [1]
Methyl bromide	CH <sub>3</sub> Br	Conditions not specified	-	Desikachar [1]
2-Chloroethanol <sup>a</sup>	CH <sub>2</sub> CICH <sub>2</sub> OH	0.31 per cent		Loeb, Morris and Dollear [6]
Chloropicrin <sup>b</sup>	CCl <sub>1</sub> NO <sub>2</sub>	Conditions not specified	_	Desikachar [1]
Ethylene dibromide	CH <sub>2</sub> BrCH <sub>2</sub> Br	Conditions not specified	-	Desikachar [1]
1,3-Dimethyl-4,6-bis (chloromethyl) benzene	1,3(CH <sub>3</sub> ) <sub>2</sub> -4,6- -(CH <sub>2</sub> Cl) <sub>2</sub> C <sub>6</sub> H <sub>2</sub>	0.03 per cent	-	Loeb, Morris and Dollear [6]
Carbon disulphide	CS,	Conditions not specified	-	Desikachar [1]

# TABLE I. TESTING OF CHEMICAL COMPOUNDS FOR THE STABILIZATION OF RICE BRAN

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Formaldehyde Furfural Hexane	CH <u>.</u> O C <sub>4</sub> H,OCHO CH,(CH <sub>2</sub> ) <sub>4</sub> CH,	Conditions not specified Conditions not specified Conditions not specified (a) Soak in 4 per cent aqueous	-	Desikachar [1] Desikachar [1] Applied Scientific Research Corporation of Thailand [7] Hermans and Ratanapunvorakul [8]
Bleaching powder <sup>c</sup>		solution and dry in the sun (b) 3-10 per cent in water and dry in the sun		Loeb and Morris [5] Panduranga Rao, Ahmed and Rao [9]
Potassium iodide	KI	5 and 10 per cent on weight of bran		Loeb, Morris and Dollear [6]
Propylene glycol dipropionate Propionaldehyde Sodium bromide Sodium cyanide Sodium chloride Sodium chloride Sodium fluoride Sodium fluoride	CH,CH(C,H,COO)- -CH <sub>2</sub> (C <sub>2</sub> H,COO) CH <sub>3</sub> CH <sub>2</sub> CHO NaBr NaCN NaCI Na <sub>2</sub> CrO₄ NaF NaF	Conditions not specified 5, 10 and 20 per cent on weight of bran 0.03 per cent 5 and 10 per cent on weight of bran 0.1 per cent on weight of paddy, 72 hours 5, 10 and 20 per cent on weight of bran Hold in 5 per cent aqueous solution (10 per cent on weight of bran) and dry in stove at 105° C		Desikachar [1] Panduranga Rao, Ahmed and Rao [9] Loeb, Morris and Dollear [6] Hermans and Ratanapunvorakul [8] Chowdhary and Mukherjee [10] Panduranga Rao, Ahmed and Rao [9] Filho, Germany and Melo [11] Azeemoddin and others [12]
Sodium metabisulphite Topanol OC (antioxidant) Topanol OF (antioxidant)	Na <u>2</u> S2O5	2 per cent on weight of bran 0.5-1.0 per cent 0.5-1.0 per cent	+  	Azeemoldin and others [12] Arnott and Lim [13] Arnott and Lim [13]

<sup>a</sup>Or ethylene chlorhydrin, 2-chloroethyl alcohol.

bOr trichloronitromethane.

CAlso called calcium hypochlorite (Merck).

Technology for the stabilization of rice bran

159

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In spite of some apparently encouraging results, chemical methods have not been satisfactory. The products tested do not have the desirable capacity for inactivating lipase. To this circumstance, which in any case makes the use of chemical methods impractical, must be added other considerations of a toxicological, nutritive and economic character. Of all the chemicals tested, SO, has shown the greatest capacity for inactivating lipase. According to a 1919 Japanese patent [2], the lipase from rice bran is destroyed by treating the byproduct with 5-15 per cent of SO<sub>2</sub> for 5-10 hours in a closed receptacle and by drying it in the sun. Treatment of bran or correctial germ in an atmosphere of SO<sub>2</sub>, at ambient temperatures for  $7\frac{1}{2}$  hours [1] or 15 hours [3], followed by subsequent aeration in order to eliminate the gas, does not, however, give satisfactory results. The by-product must be exposed to sulphur dioxide for days in order to halt the formation of FFA [3].

Recently, sodium metabisulphite has been used as a source of sulphur dioxide.<sup>1</sup> Mixed directly with the bran, in the proportion-regarded as optimum—of 2 per cent by weight, it is said to inhibit the development of FFA. In laboratory tests, the mixture of bran and metabisulphite, in sealed glass bottles, showed satisfactory storage characteristics. A batch of 25 kg, packed in jute bags and stored for 30 days, was also satisfactory: over that period the fatty acid content of the oil in the fresh bran only increased from 2.2 per cent to 3.5 per cent. According to the authors [12], the treatment does not reduce the oil yield, and the crude oil can be subjected to the usual refining processes in order to obtain edible oil. Other authors [3] have detected a marked reduction in the iodine index of the corresponding oils in batches of bran and of germ kept in an SO<sub>2</sub> atmosphere for 70 days at ambient temperature. In the treated bran, the iodine index fell from 108.8 to 75.8 and in the germ from 104.8 to 71.5. Sulphur dioxide has been used in the treatment of cereals and cereal products, such as the soaking of maize in the manufacture of starch, protection of wheat flour against micro-organisms, prevention of the growth of moulds and bacteria in the silage of maize etc., although in some cases its use in foodstuffs is prohibited. The sulphur dioxide reacts with several useful components of the bran. It destroys the thiamine molecule by splitting it into two parts, one of which is a sulphonic acid. In the form of a sulphite it reacts with disulphides like cysteine to produce thiols and sulphonates. In wheat flour this reaction reduces the bread-making strength; in bran proteins these effects do not appear to have been evaluated. The sulphur dioxide reacts slowly with sugars, forming the so-called bisulphite combinations; the reaction is reversible,  $SO_2$  being released under favourable conditions. Treatments with  $SO_2$  can lead to corrosion of the equipment, and this must be avoided. In addition, the product treated with SO<sub>2</sub> requires additional treatment to eliminate the residual gas, which presents its own problems. If this treatment is not carried out, the continuous release of SO<sub>2</sub> creates a toxic and unpleasant atmosphere, which can give rise to many problems. The use of hermetic containers or receptacles also seems to have a number of disadvantages.

<sup>&</sup>lt;sup>1</sup>The forms usually employed in food technology are: gaseous sulphur dioxide (SO<sub>2</sub>) and sodium and potassium sulphites (SO<sub>2</sub><sup>2</sup>), bisulphites (HSO<sub>3</sub>) or metabisulphites (S<sub>2</sub>O<sub>4</sub><sup>2</sup>).

# **Physical stabilization**

Physical stabilization methods include: (a) storage at low temperatures; (b) storage in an inert atmosphere; (c) irradiation; and (d) heat stabilization. The first three methods are discussed briefly below. Heat stabilization, however, is of considerable practical interest, and a vast amount of information is available on the subject; it is therefore dealt with in a separate section.

## Cold storage

It has already been noted that the storage of bran at temperatures close to  $0^{\circ}$  C (ranging from  $+3^{\circ}$  C to  $+5^{\circ}$  C) [6, 14, 15] and  $-3.3^{\circ}$  C [16] reduces the speed of hydrolysis but does not halt it. When the bran again reaches the ambient temperature, the original enzymatic activity is resumed. It must be pointed out that the data available on the effects of cold on the enzymatic activity of bran refer to small samples and laboratory tests. The situation is very different when industrial batches are being handled; for example, when hundreds of sacks are involved. Given the low heat conductivity of bran unless a highly effective heat transfer system is used, a long time is required for the by-product to reach the standard refrigerated storage temperature, during which deterioration can reach relatively high levels. These results, and considerations of an economic order, make it impractical to use refrigeration as an industrial solution to the problem of stabilizing rice bran.

At the laboratory level it is interesting to note that by storing bran in a vacuum flask at  $-70^{\circ}$  C, it is possible to maintain the level of FFA without any deterioration whatever for 15 days [17]. Roseman and others [18] reported having kept samples of bran for eight months without any change in the percentage of FFA, after having dried the bran to 2 per cent moisture content in an oven at 85° C with forced ventilation for three hours, and storing it in hermetic containers at  $-22^{\circ}$  C. Both the samples kept at  $-70^{\circ}$  C and those stored at  $-22^{\circ}$  C showed enzymatic activity when they were returned to ambient temperature and humidity.

#### Storage in an inert atmosphere

Little information is available on the behaviour of bran in inert atmospheres. Suppression of oxygen can reduce the growth of moulds, the proliferation of insects and oxidizing reactions, but it cannot be expected to destroy lipolytic activity. Indeed, there is evidence that bran, packed in an atmosphere of nitrogen and stored at ambient temperature (20°-25° C), develops FFA—in one case, around 40 per cent in 23 days [6].

The packing of bran in a carbon dioxide atmosphere has not given satisfactory results either [6]. With storage at ambient temperature, values of 40 per cent of FFA have been recorded in three weeks. On the other hand,  $CO_2$ has been tested with success for storing milled rice and preserving it from insects and moulds. In general, insects cannot survive concentrations of oxygen in the intergranular air of below 2 per cent by volume. Neither can they withstand levels of  $CO_2$  above 35 per cent in atmospheres rich in  $O_2$  (15-2! per cent) [19]. The carbon dioxide exchange method (CEM) combines these

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principles with the capacity of the rice to absorb large quantities of CO<sub>2</sub> reversibly (at 20° C, processed grain absorbs some 200  $\mu$ l per gram in 24 hours) [20, 21, 22]. In the process, the grain is packed in a CO<sub>2</sub> atmosphere in thermosealable plastic of low permeability, which is sealed hermetically. The rice absorbs the CO<sub>2</sub>, and the pack acts as a vacuum pack with internal protection provided by the CO<sub>2</sub>. Bran has a lower capacity for CO<sub>2</sub> absorption than processed rice (109  $\mu$ l per gram at 25° C in 24 hours) [23], but this is sufficient for the CEM process. In addition, the absorption of the gas is due not only to capillary condensation in the porous zones but also to absorption by the nitrogenous compounds, particularly the proteins. The enzymatic protein does not, however, seem to be affected. The interaction between protein and CO<sub>2</sub> depends more on physical absorption than on chemical reaction or chemisorption.

#### Irradiation

Irradiation has been tested for its potential as a means of stabilizing rice bran. The information available dates from the beginning of the 1960s. In one case [17], the bran was treated with gamma rays. The tests were carried out in equipment with 500-curie Co<sup>60</sup> sources, submitting the samples of bran (of 100 grams) to treatments of 225,000 rads an hour. The doses tested, which varied from  $2 \times 10^6$  to  $12 \times 10^6$  rads, proved ineffective in preventing an increase in the FFA content during subsequent storage at 20° C. The irradiation doses  $(2 \times 10^6 \text{ rads})$  did not, of itself, produce any increase in acidity. Totally different results were arrived at in another case [24], in which irradiation with gamma rays was carried out using a 1,000-curie Co<sup>60</sup> source and submitting the samples of bran to treatments of 102,300 rads an hour for various periods, with resulting doses varying from  $1 \times 10^5$  to  $1.2 \times 10^6$  rads. The effects of the treatments were evaluated by mixing the treated bran with neutral vegetable oil (cottonseed oil), in the proportion of 1 to 10 by weight, incubating the mixture at 35° C and determining the FFA content as a function of the incubation time. The fact that the level of FFA remained constant and equal to that of the initial bran throughout the six-week incubation period led the authors to deduce that highest dose  $(1.2 \times 10^6 \text{ rads})$  completely inactivated the lipase. Irradiation at the doses tested produced no changes in the content of fatty acids or in the idine index of the oil.

Brown rice and bran have been irradiated with gas plasma with a view to checking the development of FFA from the bran oil, but the results were not satisfactory [13, 25, 26]. Brown rice (samples of 30 g), irradiated for 15 minutes at 30 mA intensity and at 2 mm of mercury, packed in kraft paper bags and stored at 23° C and 50 per cent relative humidity, deteriorated less than rice that had not been treated, but the FFA content of the bran separated from it increased from 3.1 per cent to 6.3 per cent in six days. Bran (samples of 10 g), previously dried to 2 per cent moisture in an oven for three hours at 85° C and irradiated for seven minutes at 50 mA or five minutes at 100 mA intensity, at 2 mm of mercury, also deteriorated in a few days when stored at 23° C in atmospheres of 75 per cent and 92 per cent relative humidity. Irradiation at 100 mA and 2 mm of mercury for five minutes produced marked changes in the lipids of the bran. The yield of oil extractable with petroleum ether fell by almost half, the iodine index fell sharply from 101 to 70, and the average molecular weight almost doubled (from 765 to 1,243).

The irradiation technique also has serious technical and economic limitations. It is so sophisticated, the installations so large and the investment costs so high that it is not compatible with the technical and economic resources available for the production and marketing of bran.

## Heat stabilization: a widely favoured technique

In the light of present knowledge, heat treatment offers the most likely solution to the problem of stabilization. Browne demonstrated as far back as 1903 that by heating the bran immediately after milling to 99° C, hydrolytic changes were considerably retarded [27]. At the present time there are several alternative processes and types of equipment for stabilizing rice bran. The progress achieved since Browne carried out his studies is not, however, very great, considering the amount of time that has elapsed, particularly if one takes into account the notable efforts made in the last 30 years. Ways must still be found of meeting the various economic and functional requirements for adopting this technology on a large scale.

Heat stabilization processes may be divided into two major groups: (a) those which do not add water (in the form of liquid or steam) to the bran at any stage; and (b) those which do add water (in the form of liquid or steam) to the bran at one stage or another. This classification seems more convenient than the distinction frequently made between treatment with "moist" heat and treatment with "dry" heat, inasmuch as it is difficult to define precisely the terms "moist" and "dry" in connection with the treatment of bran and to apply them accurately to the various processes known. Furthermore, the addition of water to the bran—common to a number of methods—creates a differential characteristic by reducing the thermo-resistance of the enzymes and microorganisms.

# Heat stabilization without the addition of water

Heat can be imparted to the bran in a number of ways, for example, by maintaining the by-product in a hot atmosphere (stove or muffle); by putting it into direct contact with a stream of hot air; by transmitting heat to it through the walls of an open receptacle (a screw conveyor, for example) or one which is closed hermetically; by subjecting it to friction (as in the case of extrusion cookers); by using infra-red radiation etc. All these methods cause the temperature to rise to given levels for specific periods.

Table 2 summarizes the conditions recommended by various authors for the stabilization of bran by heat treatment without the addition of water. It is immediately obvious that there are some very disparate figures, which are difficult to reconcile. Examples of this are processes A-2 and A-5 (105° C for three hours as against 80° C for half an hour) and B-1 and B-2 (350° C for 10 minutes as against 200° C for 10 minutes). Regrettably, the information that has been published on the processes studied is incomplete (partly because it was not possible to check all the parameters involved). Almost all the studies use the increase in FFA during storage as the sole criterion for measuring the ٦

Processing conditions		onditions			
Group	Temperature (°C)	Time	Type of treatment and final moisture content (percentage)	Remarks	Source
A-1	110	2 h	3.7	With no moisture absorption	Loeb, Morris and Dollear [6], Pe [28]
A-2	105	3 h	In oven, continuous agitation: $\simeq 0$	With no moisture absorption	West and Cruz [29], Arnott and Lim [13]
A-3	105	1 h		-	Jaganmohan and others [30]
<b>A-4</b>	98 ± 2	30 min	30 min hermetic cooling: 0.75	In airtight containers	Instituto de Investigaciones Tecnológicas (Colombia) [31]
4-5	80 ± 2	30 min	30 min hermetic cooling: 2.2	In airtight containers	Instituto de Investigaciones Tecnológicas (Colombia) [31]
B-1	350	10 min	Heated in muffle, on aluminium tray: 1.95	Without protection from moisture	Sidhom, El-Tabey Shehata and Mohasseb [32]
B-2	200	10 min	Drying on trays in 6-8 mm layers	Dried. The bran charred at 160° C for 25 min and at 200° C for 15 min	Arnott and Lim [13]
<b>B-</b> 3	110	20 min	-	Residual lipase activity equal to zero	Srimani and others [33, 34]
B-4	120	15 min	Infra-red: 1.0	_	Gómez and Primo [3]
3-5	120	5 min	_	Low residual lipase activity	Ramkrishniah, Sawarkar and Sen [35]
B-6	(P+T+C) <sup>a</sup>	30 min	6-7	Gradual increase in FFA	Viraktamath and Desikachar [36] and Ramanathan, Krishna and Sen [37]
3-7	$(P+T+C)^a$	45 min	7-8	Gradual increase in FFA	Pillaiyar, Yusuff and Narayanasamy [38]
B-8	110-120	l s per cycle	Number of cycles 1-3: moisture not specified	Increase in FFA	Khem Chand and Gupta [39, 40]
B-9	155	18 s in extruder		Residual peroxidase activity 3.1 per cent in Bonnot extrusion cooker	Cheigh [41]
	130	18 s in extruder		Residual peroxidase activity 20.8 per cent in MFM-KIST	
B-10	132	Not specified (in extruder)	6	Residual peroxidase activity 30 per cent No increase in FFA	Lin and Cater [42]

# TABLE 2. DIFFERENT TYPES OF HEAT TREATMENT FOR THE STABILIZATION OF BRAN WITHOUT THE ADDITION OF WATER

 $^{a}P =$  pre-heating from ambient temperature to 110 -115° C (about 15-20 minutes); T = treatment at 110°-115° C for 5 minutes; C = cooling from 110°-115° C to atmospheric pressure and discharge of the product.

# Rice bran: an under-utilized raw material

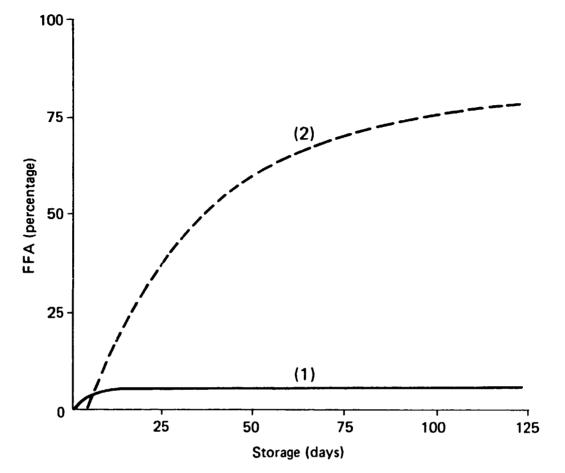
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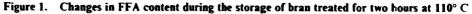
efficacy of stabilization. This index depends also on factors unrelated to the efficacy of the heat treatment, such as subsequent contamination with microorganisms producing lipase and the absorption of moisture during storage. In addition, much of the published work does not indicate the precise storage conditions (temperature, moisture content and relative humidity, or whether the bran is stored outdoors or in permeable containers), or the characteristics of the bran immediately after treatment (residual lipase activity, microbial flora producing lipases, moisture content). If one takes into account the fact that storage of the bran depends on all these parameters, the difficulty of making a comparative study of the data recommended (summarized in table 2) and the effectiveness of the treatments indicated by the various authors is evident. There are nevertheless a few points that may be relevant to determining the effectiveness of the various processing conditions listed in table 2.

Here the studies carried out by West and Cruz [29] on the effects of the heat treatment of bran in an electric oven, testing different time and temperature conditions, are of basic importance. Heat treatment involves a drastic reduction in the amount of moisture of the bran and a consequent reduction in enzymatic activity. In order to determine the effects of heat treatment, bearing in mind the difference between the moisture produced and actual enzymatic inactivation, they designed and carried out the following experiment. Part of the sample was placed in hermetic containers and another part in sterile glass bottles plugged with sterile cotton wool. Under these conditions, the bran could reabsorb moisture while remaining protected from external microbial contamination. If the enzymes had really been inactivated by the treatment, the FFA content should not increase when the lost moisture was reabsorbed. If, on the other hand, the enzymes had not actually been inactivated and their activity had merely been checked because of the lack of water, the bran would show no change in FFA content while it was kept in an airtight receptacle but would recover its enzymatic activity as soon as the moisture returned to normal levels. The results of this experiment indicated that enzymatic activity was not completely destroyed by treatments lasting from three to four hours at temperatures between 100° C and 120° C, since the FFA content increased when moisture (5.5-6.5 per cent) was reabsorbed. If, however, the treated bran did not reabsorb moisture (i.e. if the degree of absorption was less than 1 per cent), there was no increase in the FFA content. Bearing in mind the fact that temperatures above 105° C over prolonged periods of treatment darken the colour of the bran, the authors therefore recommended that the bran should be treated at 105° C for three hours and then stored in airtight containers.

Loeb, Morris and Dollear [6] arrived at the same conclusions, using a procedure similar to that followed by West and Cruz in what must also be considered a basic study. The authors observed that crude bran treated at 110° C for two hours, with a resultant moisture content of 3.7 per cent could be kept for more than two months at 25° C, with no increase in the FFA content, if it was placed in an airtight container, but that there was a rapid increase of acidity if the original moisture content (12.9 per cent) was restored through the addition of water (see figure 1).

Heat stabilization processes that do not add water can reduce or destroy enzymatic activity not only because they reduce the amount of moisture present





Key:

(1) Sample stored with the moisture resulting from the treatment (3.7 per cent)

(2) Sample stored after adding water to restore the moisture content of the untreated bran *Source:* Loeb, Morris and Dollear [6].

but also because they really do inactivate the enzyme; without the addition of water, however, the enzymes become more resistant, and more stringent conditions are required if this method is to have any significant effect.

Provided the moisture of the treated bran is maintained at a sufficiently low level, the stabilization treatment will apparently be satisfactory. The levels of moisture recommended for preventing an increase in FFA content vary between 3 per cent and 6 per cent [6, 28, 29, 43, 44]. This probably accounts for the disparity in the conditions listed in table 2 and the difficulty in reconciling them.

The conditions required for the group B processes listed in table 2 are different from those required for group A. Higher temperatures are used for shorter periods. The reduction in treatment time is not only achieved by using higher temperatures, as in process B-1, which employs a temperature of  $350^{\circ}$  C for 10 minutes [32], but also: (a) by increasing the speed of heat transmission

(whereby the bran reaches the standard temperature earlier), as in process B-4, which uses infrared rays at 120° C for 15 minutes [3], and (b) by increasing the drying speed [45], as in processes B-5, using a temperature of 120° C for five minutes and B-8, using a temperature of 120° C for three cycles at 1 second per cycle [39, 40].

In experiments conducted by Srimani and others [33, 34, 46], the bran was treated at  $100^{\circ}$  C,  $110^{\circ}$  C and  $120^{\circ}$  C for 10, 15 and 20 minutes, in a small container fitted with a slow stirrer, the whole submerged in a thermostated oil bath. The effectiveness of the treatments was evaluated by determining the residual lipase activity. The results (see table 3) show that the bran has to be heated for at least 20 minutes in order to destroy the lipase. These results seem to contradict the conclusions of West and Cruz [29] and Loeb, Morris and Dollear [6], who, after treating the bran for three and two hours respectively at  $110^{\circ}$  C, still detected lipolytic activity.

Treatme	nr		
Temperature (degrees Celsius)	Time (minutes)	Residual lipas 2 activity <sup>a</sup>	
		1.04 (original bran)	
100	10	0.90	
100	15	0.34	
100	20	0.14	
110	16	0.97	
110	15	0.20	
110	20	0	
120	5	0.97	
120	10	0.41	
120	15	0.14	

TABLE 3. CONDITIONS FOR INACTIVATING THE LIPASE IN BRAN BY HEAT TREATMENT WITHOUT THE ADDITION OF WATER

Source: Srimani, Chattopadhyay and Bose [46].

<sup>4</sup>Micro-equivalents of acid produced per minute per cubic centimetre of enzyme solution.

It should be pointed out that when using high temperatures there is a risk that the bran will be scorched and that as a result the oil will take on a permanent dark colour. This may occur when the treatment is carried out for 1 second at 250° C [45], for 15 minutes at 200° C or for 25 minutes at 160° C [13] and for several hours at 105° C [29]. It has been pointed out that prolonged heating at high temperatures reduces the quality of the oil (colour removal becomes difficult), and since bran scorches at temperatures over 120° C and the oil gives off a burnt smell, a maximum temperature of 110° C has been recommended [33, 34]. Another disadvantage of heating at high temperatures for a prolonged period is that the bran oil begins to change as a result of oxidization. If the treated bran is not cooled down before it is stored, there is a risk of spontaneous combustion. 2

It must also be pointed out that, provided enzymatic denaturation results from the process, high temperatures applied for very short periods, particularly to bran with a low moisture content, provide a very favourable environment for regeneration.

# Types of stabilizers in use

In the study made by West and Cruz in 1933 [29], it is reported that the Philippines Bureau of Science had had a cooker installed for stabilizing bran. The apparatus was in the shape of a drum 100 cm in diameter and 40 cm high. The base and walls were surrounded by a steam jacket, with an agitator located near the bottom to move the bran round. The same study describes a simple and rudimentary domestic procedure for stabilizing bran that will keep it in good condition for a week or more. The procedure consists in heating the freshly obtained bran in an oven over a low flame, stirring continually for an hour and afterwards putting the product into a hermetically sealed jar or glass bottle. In the 50 years that have elapsed since then, a variety of types of apparatus have been developed, specially designed to stabilize bran by heat processes that do not add water, and other devices already in use for other purposes have also been tested. All the types can be classified into four groups of basic apparatus, namely: conveyor or open drum; closed rotary drum; fluidized bed; and extrusion cooker (or "extruder").

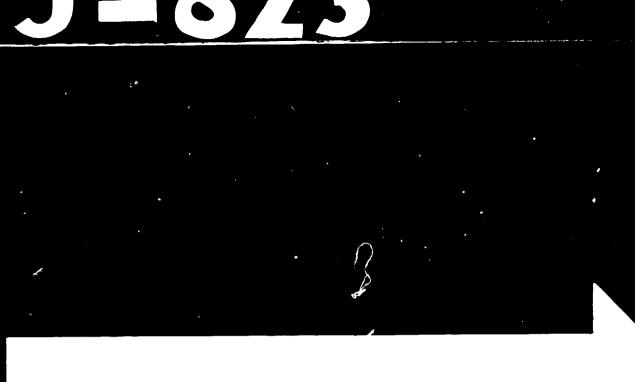
#### Stabilizers based on a conveyor or open drum

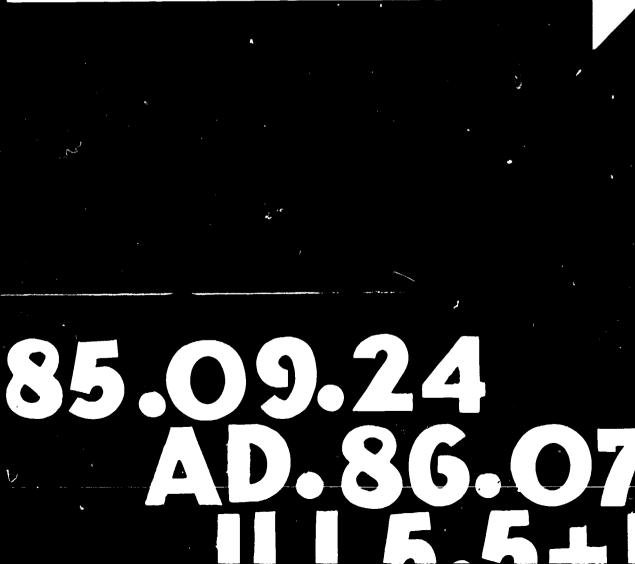
Pe [28] reports on a plant installed in a mill in Burma that has a capacity of 5 tonnes a day. It consists of a dryer, fitted with a screw or scoop conveyor with a steam jacket, in which the bran is circulated in a thin layer for 90-120 minutes and comes out with a temperature of not more than  $110^{\circ}$  C and a moisture content of about 4 per cent. The Pe paper does not indicate the holding time or the time required to reach the working temperature. When it comes out, the bran is cooled down to about 80° C and put into waterproof sacks. The storage life of the product thus treated is reported to be at least three or four months if there is no insect infestation. At the date of publication (1971) the stabilizing unit described by Pe had not yet been put into use commercially.<sup>2</sup>

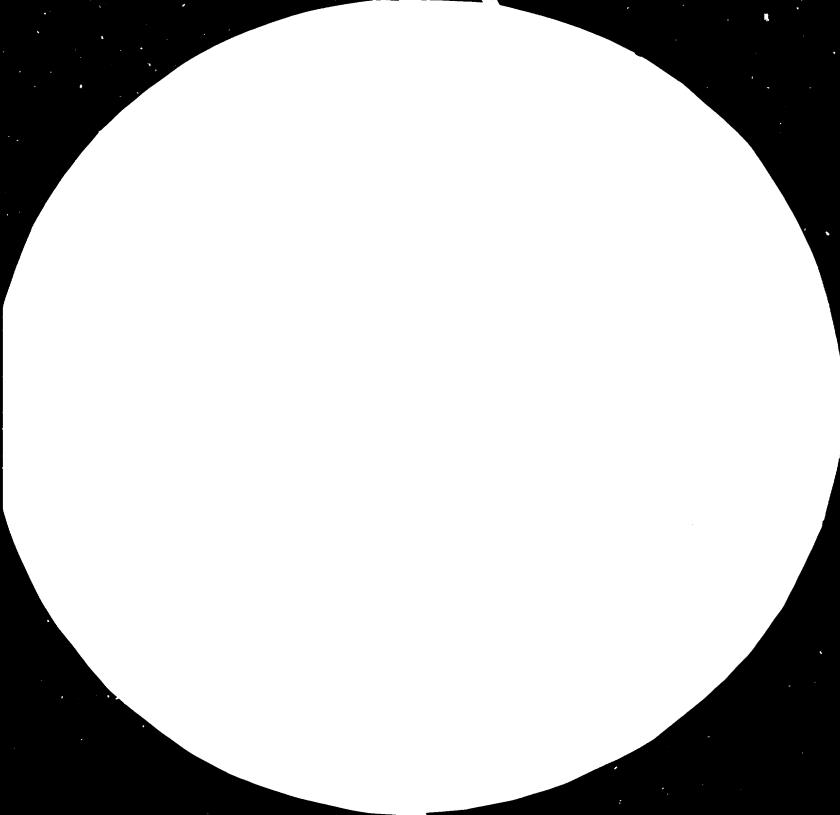
A stabilizer consisting of a screw conveyor, with mixing scoops, fitted with a jacket through which oil is circulated as a heating fluid is reported to have been designed in India [47]. The use of oil makes it easy to reach temperatures higher than 100° C and to circulate the heating fluid with a consequent saving in fuel. This particular model must, however, present some problems because it has not been introduced into industry.

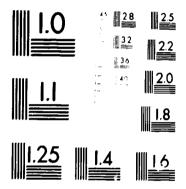
An experimental continuous stabilizer with a capacity of 8-16 kg/h, in which the gases produced by the combustion of husk are used for indirect heating of the bran, has also been reported [48]. It consists of a worm-gear conveyor, operated by a l-hp motor. The conveyor is housed in an underground masonry trench, which leads from the husk burner to the chimney. The combustion gases circulate through the free space between the trench and the walls of the conveyor, so heating the bran.

<sup>&</sup>lt;sup>2</sup>A large number of industrial-size stabilizing units, somewhat different from the type described here, have recently been installed in Burma.







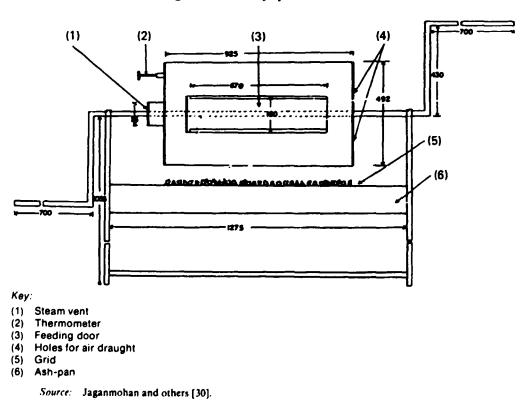


#### MICROCOPY RESOLUTION TEST CHART NATIONAL BUREAU OF STANDARDS STANDARD REFERENCE MATERIAL 10104 (ANSU and ISO TEST CHART No. 2)

Stabilization tests have also been carried out on a forage-dehydrating unit, consisting of a rotary cylinder, in which the bran and the hot air circulate in parallel. With treatment temperatures of  $110^{\circ}$  C and  $120^{\circ}$  C and a retention time of about eight minutes, it is possible to reduce the initial moisture from 9.8 per cent to 1.5 per cent. However, the treatment does not prevent the formation of FFA when the bran is stored in jute sacks and under ambient conditions (average temperature of 29° C and average relative humidity of 73 per cent).

Kuppuswami [49] has reported on a stabilizer to be used in small rural mills, which has been designed, constructed and tested in India. The unit, with a capacity of only 5 kg, consists of a metal cylinder, fitted with a central axle which serves as a support and is used to make the drum rotate by hand. The axle has arms with scoops, which, as they rotate, move the bran round. The cylinder is placed over two or three small ovens that can burn rice husk. According to Kuppuswami, satisfactory levels of stabilization are achieved by maintaining temperatures close to 100° C for sufficiently long periods of time.

A similar idea has been pursued on a larger scale [30] with a simple portable manual stabilizer, batch-operating, with a 75-kg capacity. The stabilizer consists of a drum of galvanized iron sheet with a central axle, supported on two stands, fitted with a handle to make the drum rotate manually (see figure 2). The drum has a rectangular inlet for loading and unloading, and a



#### Figure 2. Manually operated stabilizer

place for a thermometer to check the temperature of the bran. On one of the sides a vent allows the escape of steam and volatiles produced during treatment. On a grid placed a few centimetres below the drum, rice husk, wood or any suitable agro-industrial waste is burnt to heat the bran contained in the drum. In order to carry out the operation, the bran, once loaded into the drum, is heated for between half an hour and an hour at 90°-105° C; it is then unloaded and cooled to the ambient temperature, when it is ready for bagging and storage. The bran starts out with a moisture content of 10 per cent, which is reduced to 5 per cent. The reduction of the treatment conditions to less stringent levels (0.5 hour at 90° C) does not, apparently, reduce its efficiency. The increase in the percentage of FFA after a week of storage (conditions not specified) is insignificant, but the lipase activity is not halted. Refining losses in the oil extracted from the original bran and from the treated bran stored for a week are comparable, as is the colour of the corresponding oils once these are neutralized and bleached (see table 4).

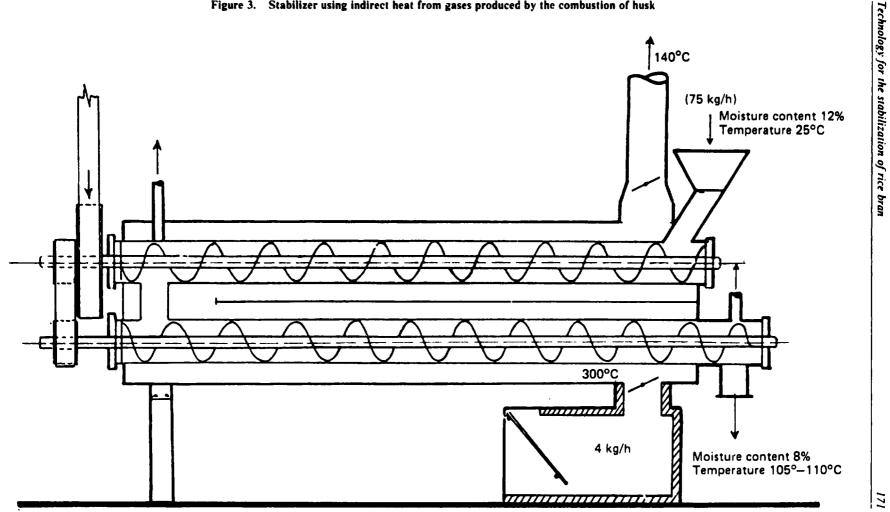
	Lovibond colour <sup>b</sup> (2.54 cm cell)		
Bran	Yellow	Red	Yellow + 5 red
Original batch No. I	9.5	0.9	14.0
Batch No. 1 treated for one hour at 105° C, and stored			
for seven days	11	1.5	18.5
Original batch No. 2	3.5	0.4	5.5
Batch No. 2 treated for one hour at 90° C, and stored for seven days	3.0	0.5	5.5

# TABLE 4. EFFECTS PRODUCED ON THE COLOUR OF THE OIL BY TREATING THE BRAN IN THE MANUAL STABILIZER<sup>a</sup>

<sup>a</sup>See Jaganmohan and others [30].

 $^{h}$ Oil neutralized and bleached according to methods prescribed by the American Oil Chemists Society.

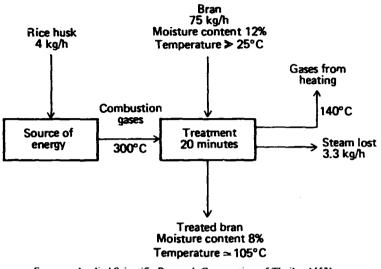
The idea of combining the stabilizing unit with a source of energy based on the combustion of rice husk is very attractive. Indeed, it has been recommended by UNIDO [50, 51]. Figure 3 shows the layout of a stabilizer designed for that purpose [52], which did not, however, reach the construction stage. The design includes two screw conveyors, 22.8 cm in diameter, with a total length of 3.8 m, with heating jacket. The maximum total power is 4.5 hp. The combustion gases circulate through the jacket in the opposite direction from the bran circulating through the conveyors. Vents make it possible to remove the steam given off by the bran. Given an imput of 75 kg/h of bran, at 25° C and with a 12 per cent moisture content, the treated product would come out at  $105^{\circ}-110^{\circ}$  C with an 8 per cent moisture content, with a retention time of 20 minutes. The source of energy would consume about 4 kg/h of husk. The flow diagram is summed up in figure 4. The treated bran must be cooled down to 70° C before being put into hessian sacks.



## Figure 3. Stabilizer using indirect heat from gases produced by the combustion of husk

Source: Applied Scientific Research Corporation of Thailand [52].

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# Figure 4. Flow diagram of a stabilization process using indirect heat from gases produced by the combustion of husk

Source: Applied Scientific Research Corporation of Thailand [52].

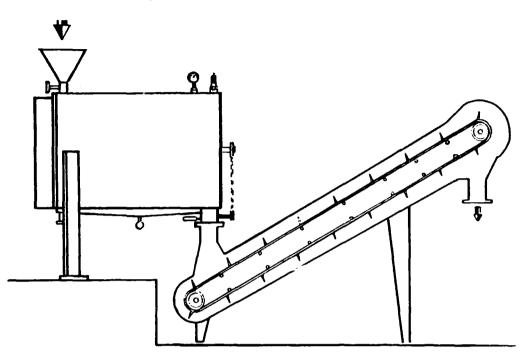
Salgado de Oliveira [52] has designed a stabilizing unit incorporating a band cooler (see figure 5).<sup>3</sup> The inactivator body is a horizontal metallic drum, with a steam jacket; the axle that passes through it, which is fixed to the drum, is driven by a suitable motor. The unit is designed to work continously. The drum has a belt conveyor inside, which mixes and at the same time circulates the bran towards the outlet end. The steam given off by the bran when heated (the heating flow temperature is 105°-110° C) passes out freely through the outlet valve. The treated bran is discharged directly over the cooler, which works by natural convection or, better still, with a small fan.

## Stabilizers based on a closed drum

Viraktamath and Desikachar [36] have published the results of their stabilization experiments using a rotating drum, heated by electrical resistors. The apparatus was provided with a hermetically sealed lid and devices for controlling temperature and pressure, as well as with a safety valve and steam vent. The drum was loaded with 20-25 kg of bran, the lid was then closed, leaving the moisture vent open, and the temperature was raised. The vent was kept open for two minutes after the steam started to push through in order to vent the air; it was then closed and the heating process continued until the thermometer showed a bran temperature of  $110^{\circ}-115^{\circ}$  C. This temperature was maintained for five minutes, and then the vent was opened until the excess pressure was reduced to zero. When the lid had been raised, the bran was taken out and allowed to cool. The FFA content of bran, which was packed in cloth sacks and stored at 37° C and 70 per cent relative humidity, only increased the FFA content from 4 per cent to 10 per cent in 120 days. This was the only evaluation criterion used.

<sup>&</sup>lt;sup>1</sup>There are no reports of the unit having been constructed.



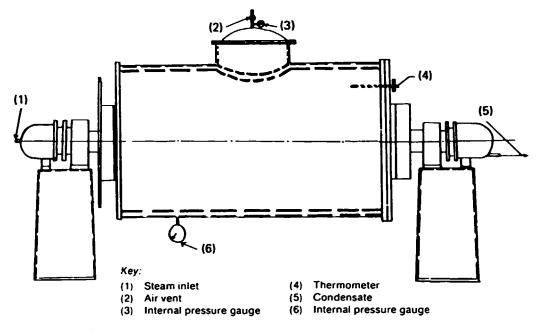


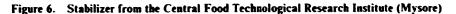
Source: Salgado de Oliveira [53].

The basic idea of heating the bran in a sealed container has since given rise to a number of alternative solutions. One of these (see figure 6) [37] consists of a metallic horizontal cylinder, with a steam jacket and two hollow concentric axles, for heating with steam under pressure. The inlet for loading and unloading the bran has a lid suitable for hermetic sealing, fitted with a steam vent. The apparatus includes temperature and pressure indicators and safety valves. The cylinder revolves at 8 rpm, driven by a 5-hp motor. Operation is by loads of 200-250 kg.4 The operative function is identical to that described for the preceding stabilizer. The treatment is somewhat less drastic (five minutes at 100° C instead of at 110°-115° C), but the complete operation, excluding the subsequent cooling, still requires about 30 minutes. The bran loses from 4 to 5 percentage points of its moisture. The published data indicate that after 70 days storage (in unspecified conditions) the FFA level of the stabilized bran was 4 per cent, that of the original bran being 2.8 per cent. The moisture content of the stabilized bran when storage commenced was 4.3 per cent, and this increased to 5.5 per cent in the course of one month.

Lakshminarayan [54] briefly describes a similar unit, for which he gives the following data: (a) capacity,  $0.8 \text{ m}^3$ , accommodating about 240 kg of bran; (b) thickness of the drum plate, 6 mm; (c) speed of rotation, 10 rpm; (d) motor power, 5 np; (e) pressure in the inside drum, 5 psi for crude bran and 2.5 psi for parboiled bran; (f) consumption of steam per load, 30 kg at 40 psi; and (g) time required for the total operation, 45 minutes.

<sup>&</sup>lt;sup>4</sup>The authors have technical information available on units with a 500-kg capacity [37].





Pillaiyar, Yusuff and Narayanasmy [38] have designed, constructed and carried out trials with a stabilizer also based on a sealed, horizontal, rotating (8-10 rpm) drum, with a heating jacket, which uses hot air as the indirect heating fluid (see figure 7). A central duct,<sup>5</sup> passing horizontally through the inner drum, to which small metallic plates are soldered, contributes to better heat transmission. The small plates facilitate the homogeneous mixing of the product, and prevent the formation of lumps, a common occurrence in bran. Instead of using ball-bearings, which wear out at high temperatures, they use guide rollers. The apparatus has an opening for loading and unloading, with a lid that can be sealed hermetically: it is fitted with devices for temperature and pressure control, as well as steam vents and safety valve. It also has a butterfly valve to regulate the entry of hot air. The apparatus, which can work with any source of hot air, has been tested with success on the basis of a husk burner manufactured in India and a fan of 12.75 m<sup>3</sup>/min with a 1-hp motor.<sup>6</sup> The stabilizer takes about 150 kg of bran, leaving from one eighth to one quarter of the volume of the inner drum free to facilitate mixing during rotation. The operation involves loading the product, closing the lid hermetically, beginning the rotation, heating with the air vent open until the bran reaches 90° C, then closing the valve and continuing heating up to 105° C, maintaining this temperature for five minutes and then, opening the vent until the internal excess pressure is reduced to zero, opening the loading mouth and unloading and allowing the bran to cool before bagging. With a load of 150 kg of bran,

Source: Ramanathan, Krishna and Sen [37].

<sup>&</sup>lt;sup>3</sup>Other possible models may have three such tubes.

<sup>\*</sup>Instead of the fan, a chimney 9 m tall and 20 cm in diameter supplies the flow of air needed for the burner.

and with an air inlet temperature of 240°-260° C, it takes 45-50 minutes to reach 105° C, running the apparatus continuously. The moisture loss during treatment ranges from 2.5 to 4.0 percentage points. The storage capability of batches of treated bran, with a final moisture content of around 6 per cent, is satisfactory; the FFA content (the only characteristic measured) barely increased from three to four months' storage in unspecified conditions.

The stabilizer shown in figure 8 has a novel feature in that it incorporates a vacuum pump to speed up the elimination of moisture from the bran. There are no reports on its use in industry.

# Fluidized bed stabilizers using a stream of warm air

The fact that enzymatic activity decreases considerably when the moisture content of the bran is reduced to very low levels (1-3 per cent) suggests that it might be possible to stabilize the product by drying it. In order to achieve such low levels of moisture without using lengthy treatment times or high temperatures that might damage the bran, some investigators have explored the possibility of drying the bran in a fluidized bed. This does not mean that, under different treatment conditions, the degree of stabilization achieved is not due partly to the reduction of moisture content and partly to actual inactivation by enzymatic denaturation.

Srinivasa Rao and others, for example, have carried out a pilot plant study of the stabilization of bran in a pneumatic conveyor dryer [45]. The outstanding characteristic of this equipment is the extraordinarily short time the bran is in contact with the hot air that serves as a drying medium. The equipment used is shown in figure 9. It consists of a centrifugal fan operating at 170 m<sup>3</sup>/h that drives the air through a hole at 457 mm water gauge pressure, a 12-kW electrical heater and an injector. The bran is introduced into the stream of hot air through the injector. The bran and air circulate through a thermally insulated column 75 mm in diameter and 7.75 m tall. The dried bran is separated in a cyclone. The performance data on this type of dryer are as follows:

Capacity	54.4 kg of moist product per hour
Evaporation capacity	3.63 kg of water per hour
Initial moisture content of bran (dry basis)	10.1 per cent
Final moisture content of bran (dry basis)	2.76 per cent
Temperature of air at inlet	196° C
Temperature of air at outlet	111° C
Air flow at inlet temperature	189 m³/h
Ratio of air to bran	2.62
Temperature of bran at inlet	27° C
Temperature of bran at outlet	75° C
Number of transfer units	0.981
Height of column	7.75 m
Height of transfer units	7.93 m
Diameter of column	75 mm
Type of energy used	Electricity
Energy consumption	3.321 kWh/kg of evaporated water

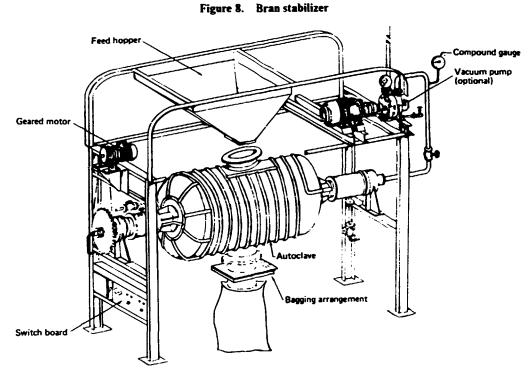
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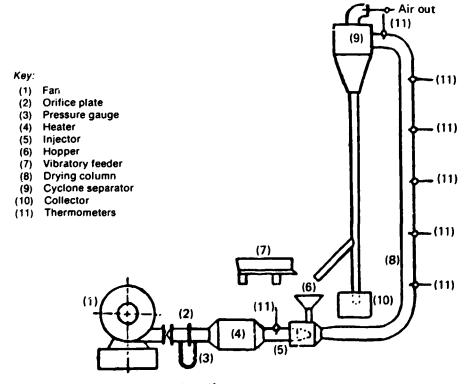
Figure 7. Hot air stabilizer combined with husk burner

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Source: Servotech. Eng., PVT, Ltd.

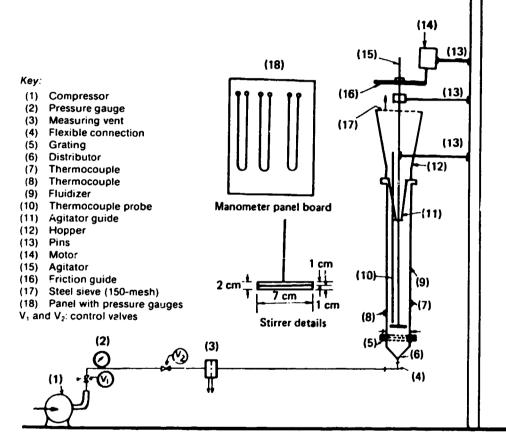




Source: Srinivasa Rao and others (45).

Bran has some characteristics that make it unsuitable for fluidization; for example, it has a tendency to form "channels" during the operation. The immediate consequence of this is that the temperature in the bed is not uniform, and the effects of the treatment, therefore, are also uneven. In order to overcome this disadvantage, Ramkrishniah, Sawarkar and Sen [35] used a slow-speed agitator, which breaks up the channels and improves fluidization. The equipment used is shown in figure 10. It consists essentially of a stainless

#### Figure 10. Stirred fluidized bed device for the stabilization of rice bran



Source: Ramkrishniah, Sawarkar and Sen [35].

stee column 7.5 cm in diameter and 50 cm tall, fitted with an electric tubular heat exchanger, a 150-mesh screen which serves as a distributor at the foot of the column, a blade agitator 7 cm long and 0.5 cm thick, located 1 cm above the distributor, and a small hopper. For an agitator speed of 60 rpm, the air speed required for the fluidization of 250 g of bran was 3.85 m<sup>3</sup>/h. The equipment was tested at different times (2-10 minutes) and temperatures (105° C and 120° C). The effectiveness of the treatment, on the basis of the increase in FFA (the only criterion used) during storage (under unspecified conditions), is apparently satisfactory.

The low conductivity of the bran and the mixed size and shape of the particles make it difficult to heat a batch uniformly. In order to solve this problem, which seems to be common to most of the conventional stabilization methods involving heat treatment, Kem Chand and Gupta and Bal, Savarkar and Bhati [39, 40, 55] have used a method for transferring heat in a fluidized bed combining dense and dilute phases. The particles in the dense phase (made up of sand) are such that they cannot be transported in the stream of air but remain in a state of constant circulation in the zone of heat transfer. The particles of the dilute phase (made up of bran), being lighter, are carried in the stream of air and pass through the dense phase, where they are heated uniformly. This system has the following advantages: (a) the coefficient of heat transfer is 3.5-7 times better than it is in a fixed bed column; (b) the particles have a greater opportunity to reach a uniform temperature; (c) the treatment time is shorter, and therefore the column is shorter too; and (d) the capacity of the equipment can be easily increased without any change in its dimensions, simply by increasing the amount of air and the number of calories supplied. The stabilizer in figure 11 was designed and manufactured in accordance with these principles. It consists of a column 5 cm in diameter and 1.8 m tall, fitted with electrical resistances for heating.<sup>7</sup> A fan supplies air at a maximum pressure of 5 cm water gauge, the flow being regulated by a butterfly valve as required; the flow of air is made uniform by means of a Venturi tube in which the reduction is from 5 cm to 2.5 cm. A cyclone separates the bran as it leaves the column. Sand 0.5-1 mm in diameter is loaded from the top, and the air speed is adjusted in order to keep the dense zone stable. The bran is loaded at the base of the column, through a small hopper in the suction zone of the fan. The bran is retained in the column for 30-70 seconds. According to the authors, the maximum rates of heat transfer are obtained by using a column 5 cm in diameter and 150 cm tall, sand particles 0.4-0.5 mm in diameter and an air speed of 3 m/s; working continuously, the stabilizer can raise the temperature of the bran from 25° C to 120° C in a single pass, with a capacity of 0.72 kg of bran per minute; according to the authors no sand is entrained to damage the quality of the bran; the FFA content of bran treated at 150° C and stored under unspecified conditions for a cycle of four weeks increased from 3.3 per cent to 5 per cent. This was the only parameter evaluated. The authors recommend three passes at 150° C for more effective stabilization.

#### Extrusion cookers

The extrusion cooker (or extruder) transforms the mechanical energy of a pressure screw into heat by means of the friction and shearing that occur when a granular product is compressed and forced to pass through a hole. It consists essentially of a screw, a body, a die, a feed device, a supply of energy and a drive unit with the corresponding control mechanisms.

Williams and Baer [56] used a machine of this type, of United States manufacture, to stabilize bran, although water was added. Subsequently, Viraktamath and Desikachar [36] used an "expeller" press of Japanese manufacture for the same purpose and, more recently, a number of researchers (Lin and Cater [42]; Pablo and Sangalang [57]; Mukhopadhyay [58], Bhumiratana [59]; Harper and others [60]; Sayre [61] and Enochian and others [62]) have explored the possibilities of the Brady Crop Cooker, manufactured in

<sup>&</sup>lt;sup>7</sup>Twelve i,000-watt heaters were used.

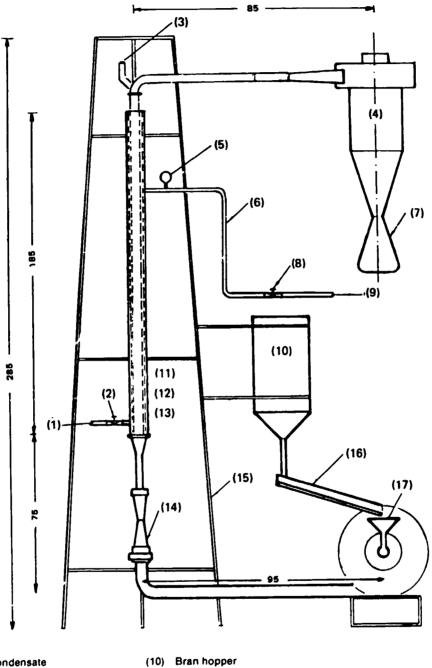


Figure 11. Fluidized bed stabilizer with combined phases<sup>a</sup>

# Key:

1

- (1) Condensate
- Valve (2)
- Sand-feeding arrangement (3) Cyclone separator
- (4) (5) (6) Pressure gauge
- Flexible steam pipe Sterilized bag
- (7)
- (8) Stop valve
- Steam from boiler (9)

Asbestos lining

- (11)
- (12) Steam pipe
- Galvanized iron pipe (13)
- Venturi pipe (14)
- (15) Frame
- (16) Vibratory feeder(17) Feed opening on suction side
- Source: Kem Chand and Gupta [39, 40] and Bal, Savarkar and Bhati [55]. <sup>a</sup>Not to scale; dimensions in centimetres.

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the United States of America, while Cheigh and there from the Korean Institute of Science and Technology (KIST), have experimented with the MFM-KIST extruder, manufactured by the Institute, using an original design by the Meals for Millions Foundation (MFM) of the United States [41, 63] Cheigh also tested a laboratory Bonnot extruder [41]. Although there are some other more or less similar types of extruders, they do not seem to have been used for the stabilization of bran.

Extruders have been widely used in recent years for the preparation of pre-cooked foods based on mixtures of cereals and legumes or oil seeds. Some of the functions they perform are equally suitable for use with bran, for example, heat inactivation of enzymes and enzyme inhibitors with a physiological action, destruction of micro-organisms and insects in order to achieve a hygienic product and prolonged storage of the extrudates obtained, because of their low moisture content. The extruders operate at a temperature ranging between 80° C and 155° C, although the most suitable temperatures seem to be around 110° C [36]. It must be pointed out, however, that the retention-time is very brief and that the product completes the greater part of its journey along the screw before reaching the maximum temperature, exposure to the highest temperatures taking place only along a very short section of the extruder (see figure 12). Despite this, the effect on bran enzymes can be considerable. Cheigh [41] succeeded in reducing the peroxidase activity of the bran to 3.1 per cent of the original value, with a retention-time of 18 seconds and a temperature (at the die) of 155° C in a laboratory extruder, and to 20.8 per cent, for the same length of time, at 130° C in an MFM-KIST extruder (see below) with a 100 kg/h capacity (table 5).

As a result of the reduction in pressure when the bran leaves the extruder, coupled with the high temperature of the bran, moisture is lost as the water suddenly evaporates. Lin and Cater [42] have evaluated the loss in terms of the temperatures of the bran on leaving the extrusion cooker (see figure 13). Cheigh [41] has published data on a laboratory Bonnot extruder (see table 6).

The Brady 206 Crop Cooker has an extruder screw 15 cm in diameter and 1 m long,<sup>8</sup> connected directly to the axle of the agricultural tractor that drives

Type of extrusion cooker	Oulei iemperature (degraes Celsius)	Time (seconds)	Moisture content (percentage) <sup>a</sup>	Residual peroxidase activity (percentage) <sup>b</sup>
Bonnot	121	18	9.2	45.1
Bonnot	138	14	8.5	23.1
Bonnot	155	18	7.0	3.1
MFM-KIST	130	18	8.1	20.8

# TABLE 5. EXTRUSION COOKERS: EFFLCTS OF TEMPERATURE AND RETENTION-TIME ON THE MOISTURE CONTENT AND PEROXIDASE ACTIVITY OF RICE BRAN

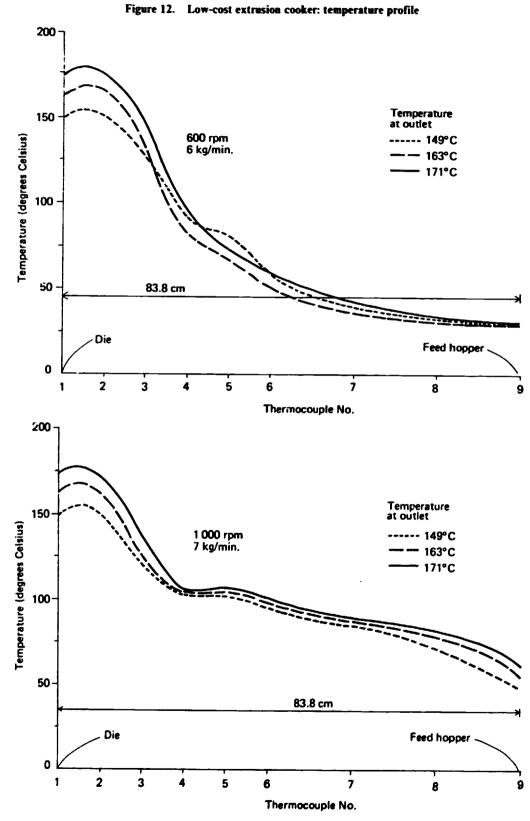
Source: Cheigh [41].

<sup>a</sup>Original moisture content 11.1 per cent.

<sup>b</sup>Original peroxidase activity 100 per cent.

<sup>4</sup>The extruder screw measures 12.7 cm in diameter and 86.4 cm in length. The screw has a constant pitch of 3.81 cm and fits into a jacket fitted with three "breaker bars" [64].

# Technology for the stabilization of rice bran



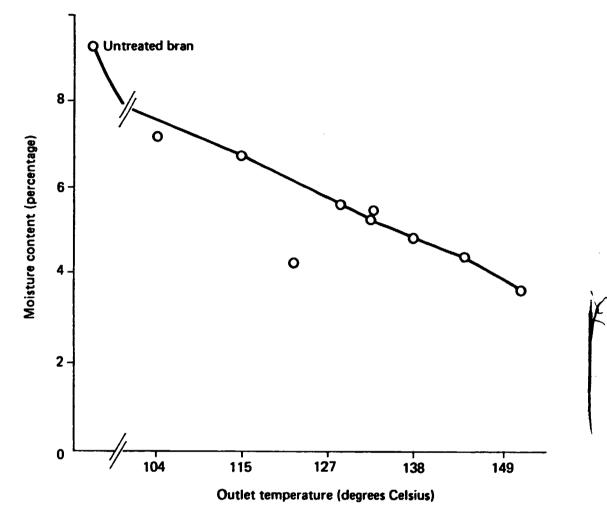
Source: Harper and others [60].

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Source: Lin and Cater [42].

it. The machine includes a hydraulic oil system with hydraulic pump, oil tank, control valve and hydraulic motor. This motor turns a small worm which meters the product from the feed hopper to the inlet of the extruder. Using the control valve, the metering rate can be varied from 0 to 50 rpm, regulating the volume of product fed to the machine; the valve helps to control the treatment temperature and the load capacity, depending on the power of the tractor. The product is forced from one end to another of the extruder screw by the rotor, which turns at 500-1,200 rpm, depending on the power of the tractor. The temperature of the product rises noticeably as it approaches the outlet end. This consists of a cone 16 cm in diameter, which fits into a fixed housing, both of which are replaceable. The gap between the rotating cone and the fixed housing can be varied in order to modify the extrusion temperature. The shape of the extrudate is not determined by the periphery of the rotating cone. Some

Production (kilograms per hovr)	Outlet temperature (degrees Celsius)	Specific consumption of energy (kilowatt hours per kilogram)
110	146	0.040
145	140	0.038
167	133	0.037
188	131	0.038

#### TABLE 6. PRODUCTION CAPACITY, TREATMENT TEMPERA-TURE AND CONSUMPTION OF ENERGY DURING THE EX-TRUSION OF RICE BRAN

Note: Bran was extruded through a 4-mm die.

of the units work with a 100-hp electric motor and others are fitted with diesel units but the majority, as mentioned earlier, are coupled to 65 to 130-hp tractors [65]. There are now some 30-hp models.

The parts that wear out most quickly are the cone and the housing. Bran is known to be a particularly abrasive by-product, especially in its dry state, but its actual effect on the life of these two parts has not apparently been determined. With other products, including cereal grains, legumes and seeds, depending on the contents and type of impurities, the parts can cope with weights varying from about 10 tonnes to over 500 tonnes before wearing out. The presence of dust, dirt and sand accelerates the rate of wear. The rotor is also subject to some wear and tear [65]. In some similar machines it is advisable to install a magnetic separator to eliminate metallic impurities before they enter the extruder screw.

In the Brady 206 model, two control devices regulate the temperature of the extrusion cooker. The first is the cone adjustment handle, which regulates the discharge opening and influences the dwell time and the input of energy. The second control regulates the feed of the product to the extruder [64].

The type of extrusion cooker described above is simple to operate. Once it is going, all that needs to be done is to feed the bran into the machine and to regulate the outlet. In order to ensure that the working conditions are correct, a certain amount of the product must be put through the machine for preheating. To stop the machine, the outlet cone is opened as far as possible, allowing the temperature to fall without shutting off the bran feed. There is no need to strip down the machine for cleaning, but care must be taken to see that no bran is left inside [64].

Considerable attention is being given to the improvement of the Brady 206 extruder. Jackson [66] has pointed out that some characteristics of the machine need to be modified in order to facilitate its maintenance in hygienic conditions, to avoid blocking up the area between the feed thread and the extruder screw, and to reduce wear on the latter. The Brady 206 was originally designed to work intermittently on farms. For it to be able to work continuously under industrial conditions, Harper and others [60] have recommended several modifications, for example to the lubrication and transmission systems. Smith, Kellerby and Tribelhorn have advocated the use of stronger materials for the screw-thread and the cone that regulates the discharge [67]. Mukhopadhyay [58] has reported having stabilized parboiled rice bran with satisfactory results, using the Brady 206 at outlet temperatures of 104°-149° C. The only criterion was the rate of formation of FFA, and this remained at under 0.5 per cent when the bran was stored for a month at an unspecified temperature. During that time the moisture content ranged from about 3.5 to 5.5 per cent. Bhumiratana [59] reports having arrived at similar results by processing the bran at temperatures between 77° C and 110° C; again, he did not specify the conditions under which the bran was stored.

Working in the Philippines, using the same kind of machine, Harper and others [60] processed three commercial types of bran without any difficulty. These were the common or coarse type, which is a mixture of bran, husk and milling residues, the medium type, containing a small proportion of husk and the fine type, without husk, the size of the particles varying from 60 to 80 mesh (250-177  $\mu$ m). The bran was processed at various temperatures (from 138° C to 171° C) at a rate of 295 kg/h. The higher temperatures used made it necessary to work with a very narrow gap in the outlet, thus causing the feed to clog. The addition of 1 per cent water (the moisture content of the bran is not indicated) facilitated the operation, but made it impossible to exceed a temperature of 141° C. Details are not given of the effects of the treatment on the stability of the bran. The authors point out the abrasive effect to the bran on the extruder screw, which became highly polished in the course of the operation. The extruded bran was in the form of chips measuring about 6 mm in diameter.

Lin and Cater [42], working with an extrusion cooker driven by a 100-hp tractor, treated bran with a 9.3 per cent moisture and 18.8 per cent oil content at various temperatures from 104° to 152° C and evaluated the effects by determining the residual peroxidase activity and the increase in FFA during storage under well-defined conditions. The samples treated at 122° C or above, packed in waterproof polythene bags, showed no increase in the FFA at all after 23 days of hermetic storage at ambient temperature (23°-28° C), during which time the moisture content was maintained at below 6 per cent. Subsequent storage of the bran, for a period of four extra days, at 35° C and 100 per cent relative humidity, in open containers, produced an increase in the FFA of samples treated at temperatures below 132° C, but not in those treated at a higher temperature. Details are not given of any more prolonged storage. During the four extra days the moisture of the samples rose to 12.0-12.5 per cent. The residual activity of the peroxidase in bran extruded at 132° C was about 30 per cent (see figure 14).

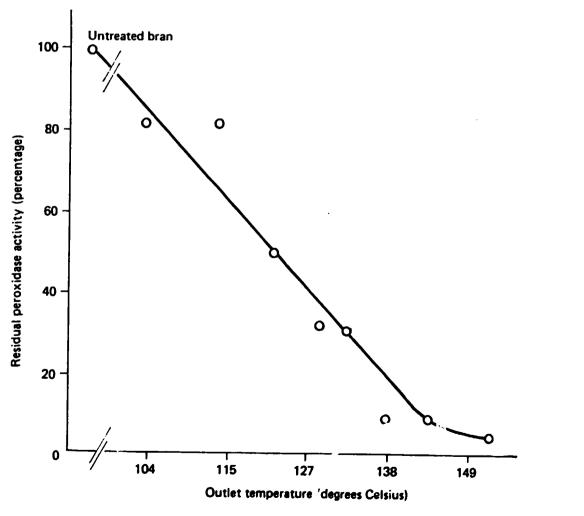
In a subsequent investigation, Pablo and Sangalang [57], confirmed that when bran was extruded at 138° C, the moisture fell from 11.9 per cent to 5.54 per cent and the treated product, packed in polythene bags at 29° C for 80 days, showed no increase in FFA.

The MFM-KIST extrusion cooker [41] consists of an extruder screw 63.5 mm in diameter, with constant thread pitch. The body of the machine is divided into three areas and has no cooling or heating devices. The inner surface of the body has four flanges, which run parallel to the longitudinal axis. The machine is driven by a 30-hp motor and is fitted with a three-speed gearbox. The technical characteristics of the machine [41] are summarized below:

# Technology for the stabilization of rice bran

Main motor	1,750 rpm, 22 kW (30 hp)
Bran feeder motor	1,710 rpm, 1.5 kW (2 hp)
Electrical supply	220 V, three-phase, 60 Hz
Transmission	(1) 1:11.5
	(2) 1:6.5
	(3) 1:4.0
Speed of screw	(1) 150 rpm
	(2) 273 rpm
	(3) 430 rpm
Diameter of the screw	63.5 mm
Maximum production capacity	80-100 kg/h
Length $ imes$ height $ imes$ width	200 × 200 × 190 cm





Source: Lin and Cater [42].

187

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Treatment of the rice bran at 130° C, with a total dwell-time of 18 seconds without the addition of water, reduced the peroxidase activity to 23 per cent of its original value. The treated bran, however, with 8.1 per cent of residual moisture (11.1 per cent in the untreated bran), packed in cotton sacks and stored at 30° C and 87 per cent of relative humidity, doubled the FFA content in less than three weeks from the original 8 per cent.

A different model (KIST-EO 3030-100), with a capacity of 150 kg/h, has a screw 100 mm in diameter, which revolves at 600-1,000 rpm driven by a 30-hp motor in a body with four parallel grooves. The addition  $\neg$ f water is not required. The consumption of energy, in terms of production capacity and the extrusion temperature, as the bran leaves the machine is given in table 6. The temperature falls as production increases, and the consumption of energy increases as the temperature rises. Extrusion reduces the moisture content of the bran from 13.8 per cent to 6-9 per cent depending on the temperature, and has a marked inactivating effect on the peroxidase. At 130° C, residual peroxidase is only 16 per cent of the original amount, the bran so treated showed a markedly lower tendency to form FFA. After two weeks' storage at 32° C and 75 per cent relative humidity, however, the FFA content increased from approximately 9 per cent to 14 per cent. Other characteristics of the extruded bran used for the extraction of oil (dealt with elsewhere) were also reported to be favourable.

## Heat stabilization with the addition of water

Some stabilization processes call for the addition of water to the bran, generally in the form of steam. The steam has a dual role: (a) as a means of heating the bran, with a high coefficient of heat transfer; and (b) as a means of increasing the water activity in the bran and reducing the thermo-resistance of the enzymes and the micro-organisms that they contain. These two aspects have not always been taken into account when steam has been used. As a result, while in many cases a rapid transfer of heat has been achieved with an acceptably uniform temperature, hydration has been slow and the distribution of moisture uneven, and thus very different results have been obtained in treatments involving a similar ratio of time to temperature.

It seems logical to classify the processes into those that are designed to obtain a thorough mix of bran and steam and those that are not.

Any process that treats the bran while still on the grain, with or without husk, constitutes a special case.

## Classification of processes

In the following sections, therefore, processes based on treatment with direct steam are classified into three types: (a) stabilization of the bran while still on the rice, before milling; (b) stabilization of the bran in a fixed bed; and (c) stabilization of the bran in a moving bed. In the latter case it is useful to distinguish processes operating at normal pressure from those that use extrusion cookers.

#### Stabilization of the bran on the grain, before milling, by direct steam treatment

Roberts and others [68] and Houston, Hunter and Kester [69] report that the treatment of recently harvested paddy with steam at 100° C, for one minute, is sufficient to prevent the formation of FFA in the husked rice with a 13 per cent moisture content when stored for 15 days at 25° C. Lipase activity, although not precisely determined, was considerably reduced. Viraktamath and Desikachar [34] obtained similar results not only for recently harvested paddy, with a 20 per cent moisture content, but also for paddy with a 12-14 per cent moisture content. None of these authors succeeded in completely checking the formation of FFA when storage extended beyond 10-15 days. Nevertheless after treatment for 5 and 10 minutes, the increase in FFA when the product was stored for 80 days at 37° C was significant.

Many alternative ways of parboiling [70, 71] have been shown to destroy rice enzymes. The degree of inactivation depends on the conditions under which the processing is carried out and particularly on the steam treatment. Nawab Ali and Ojha [72] have pointed out that, in general, the different methods of parboiling rice use steam saturated at 1-5 kg/cm<sup>2</sup> for the treatment of soaked paddy, and treatment times vary from 2-3 minutes for loads of small size to 20-30 minutes for larger loads. In view of the high moisture content of the rice when undergoing steam treatment, 2-3 minutes would probably be enough at an actual temperature of 100° C to inactivate most, if not all, of the enzymatic activity of the grains. Viraktamath and Desikachar [36] determined the effects of different methods of parboiling, using the conditions specified in table 7. The bran obtained from parboiled rices proved to be reasonably stable. Its FFA content (the only characteristic evaluated) was maintained at between 3 and 5 per cent during 50 days' storage at 37° C (humidity not indicated). Similar results were arrived at by lengar and others [73] for bran with an 8 per cent moisture content, packed in glass bottles, derived from parboiled rices prepared by different processes (table 7). It should be noted that the bran from rice parboiled under pressure (table 7, process No. 7) was, by comparison, more stable.

In industrial practice, and indeed generally, the results obtained are not as a rule so satisfactory, although, with modern methods of parboiling, bran with

TABLE 7.	PROCESSING CONDITIONS FOR PARBOILING RICE SPECIFIED IN BRAN
	SUITABILITY STUDIES

Process No	Soaking	Length of steam treatment
1	At cold temperatures, 72 hours	10 minutes, at atmospheric pressure
2	At cold temperatures, 72 hours	Not specified <sup>a</sup>
3	At cold temperatures, 72 hours, 0.1 per cent potassium dichromate	Not specified <sup>a</sup>
4	At 45°-50° C, 24 hours	10 initutes, at atmospheric pressure
5	At 70° C, 3-3.5 hours	10 minutes, at atmospheric pressure
6	At 70° C, 3-3.5 hours	Not specified <sup>a</sup>
7	10 minutes with steam at atmospheric pressure	20 minutes at 0.356 kg/cm <sup>2</sup>
	• • • • •	followed by 5 minutes
		at 1.758 kg/cm <sup>2</sup>

Source: For processes Nos. 1, 4 and 5, Viraktamath and Desikachar [36]; for processes Nos. 2, 3, 6 and 7, Iengar and others [73].

<sup>a</sup>Probably until the glumes have opened.

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a 14 per cent moisture content may sometimes be maintair.ed for a fortnight in the winter with a 2-4 per cent FFA [74].

There may be several reasons for poor storage properties in parboiled rice bran, the following being the most important: (a) insufficient heat treatment, in which the required ratio of time to temperature is not reached (sometimes the steam injection time may be very short), or uneven steam treatment, which does not affect all the grains of rice equally; (b) storage of parboiled paddy in conditions favouring contamination and the formation of micro-organisms that produce lipase: this can occur during drying or storage; (c) contamination of the bran by micro-organisms producing lipase and the growth of these under favourable conditions. Viraktamath and Desikachar [36] have reported a specific case in which insect infestation of bran stabilized by heat treatment in a sealed container, without the addition of water, was the principal cause of the increase in FFA.

It should be noted that the stabilization of bran is not even considered to be a secondary aim of parboiling and is only a fortuitous result of the process, which is therefore frequently carried out under inadequate or unsuitable conditions. It would not seem to be too complicated a matter to adapt these conditions so as to inactivate the bran and avoid recontamination, and this would greatly facilitate storage.

## Stabilization of bran by direct steam treatment in a fixed bed

Different authors have encountered widely varying conditions for stabilizing bran with steam in a fixed bed (table 8). While in some cases satisfactory results are obtained by subjecting the bran to 100°-105° C temperature for 10-15 minutes, others require 118° C for 30 minutes or 134° C for 1 hour. The apparently divergent results are probably due to the causes mentioned in the previous section. The moisture content of the bran during heat treatment can also make a great difference. Because of the lack of data on what are very important parameters, no valid interpretation can be made of the results.

## Stabilization of bran by direct steam treatment in a moving bed

The processes that use direct steam treatment to stabilize bran do so by adding water to the bran either at normal or at high pressure by means of water or steam injection. Both methods result in a thorough mix of steam and bran, which is of the greatest importance, since it increases the transfer of heat and the speed of hydration of the bran and facilitates a uniform treatment of the whole mass. Not unsurprisingly, most authors have recommended very similar processing conditions (table 9). In general, about three minutes' exposure to direct steam at normal pressure is sufficient to inactivate the bran enzymes. An increase in the moisture content of the bran of 3-5 percentage units is assumed, but this may be counteracted by subsequent drying to restore the initial moisture. Some authors recommend that the moisture content of the stabilized bran should be reduced to 3 per cent [44] or 2-5 per cent [33]. If, however, the enzymes have really been inactivated by the treatment (which must be demonstrated by subjecting the bran to the appropriate chemical analyses), the precaution only results in higher processing costs and, in all probability, additional deterioration of the useful components of the bran. Admittedly,

	Autoclave tre	aiment			
T <mark>emperatur</mark> (degrees Celsius)	e Time	Bran bed	Remarks	Storage	Reference
121	2 h	On trays	Additional drying for 1 h at 60° C	At 21° C, no increase in FFA: After 41 days with 13.7 per cent moisture content After 13 days with 14.6 per cent moisture content After 7 days with 26.4 per cent moisture content	Loeb and Mayne [43]
134	1 h	On trays	Initial moisture content 11.30 per cent	Moisture content 11.11 per cent In polythene bags at ambient temperature— no increase in FFA after 50 days	Sidhom, El-Tabey Shehata and Mohasseb [32]
118	30 min			In hermetic tins—no increase in FFA after 15 days	lengar and others [73]
105	10 min	3 cm layer	In laboratory conditions	Moisture content 8-10 per cent In cloth sacks at ambient temperature 20°-30° C 40-90 per cent relative humidity 6.5 per cent FFA after 50 days	Ratanapunvorakul and Hermans [75]
128	7.5 min	3 cm layer	In pilot plant Additional drying	In cloth sacks At ambient temperature 23°-32° C 50-90 per cent relative humidity	Hermans, Pichitakul and Bhuntumkomol [76]
100 <sup>a</sup>	15 min	0.5-1 cm layer	Additional drying in stove until <10 per cent moisture content	FFA <10 per cent after 80 days at 37° C	Viraktamath and Desikachar [36]
100	20 min	In cylindrical tank with conical bottom; central tube steam injection 250 kg bran	Temperature of bran on leaving 95°-97° C Increase in moisture 2-3 per cent Additional drying	Moisture content 6.8 per cent FFA ≈10 per cent after 25 days	Viraktamath (77)

## TABLE 8. RECOMMENDED CONDITIONS FOR THE STABILIZATION OF RICE BRAN BY DIRECT STEAM TREATMENT IN A FIXED BED

Technology for the stabilization of rice bran

101

<sup>a</sup>Steam at atmospheric pressure.

# TABLE 9. STABILIZATION OF RICE BRAN BY DIRECT STEAM TREATMENT IN A MOVING BED

	Conditio	ns			
Temperature <sup>a</sup>	Time	Remarks	Results	Reference	
100° C	15 min (for 1 t) 8 min (for 250 kg)	Bran temperature on exit 95°-97° C Increase in M. 2-3 percentage units Additional drying	$M \simeq 6$ per cent, FFA $\simeq 10$ per cent in 15 days	Viraktamath (77)	
100° C	3 min	Increase in M. 1.5-2 percentage units Additional drying	Total inactivation of lipase	Srimani and others [33, 34]	
100° C <sup>a</sup>	4.5 min	Increase in M. 3-5 percentage units Additional drying	Maintains FFA at acceptable levels for at least one month	Burns and Cassidy [78]	
100° C <sup>a</sup>	2.5 min <sup>b</sup>	Increase in M. 7-8 percentage units Additional drying to $M \approx 12$ per cent	Total inactivation peroxidase	Barber and others [92]	
100° C	3 min	Additional drying to $M \simeq per cent$	At $M \approx 3$ per cent, bran keeps for several months	Yokochi [44]	

*Note:* M = moisture content.

<sup>a</sup>Steam at atmospheric pressure.

<sup>b</sup>For a final moisture content of 15-16 per cent, 3 minutes are recommended.

under industrial conditions, it is difficult to prevent microbiological contamination of the stabilized bran. On the other hand, the lower the water activity in the bran, the less risk there is that lipase-producing micro-organisms in the bran will multiply. In any case, the bran will have to acquire a moisture content that will be in balance with the environmental conditions and that must be maintained below the minimum level necessary for the rapid growth of lipase-producing micro-organisms. The use of extrusion cookers with water or steam injection [11, 56] calls for: (a) a very short dwell-time; (b) high pressures; and (c) immediate loss of part of the water content in the decompression stage, leading to expansion of the product. According to Williams and Baer [56] the conditions required to inactivate the bran are a moisture content of the bran to be extruded of 27 per cent, an internal temperature of  $115^{\circ}$  C, a final temperature of  $82.5^{\circ}$  C and a final moisture content of 23 per cent.

## Machines in use

The stabilization of bran by treatment of the whole rice grain is merely incidental to the manufacture of parboiled rice, so no special machinery has been developed for the purpose. Neither has any specific machinery been developed to treat the bran with direct steam in a fixed bed. Generally, the bran has been heated in autoclaves or, on an experimental basis, in devices designed for other purposes [73, 77].

There are three basic types of machine for treating bran with direct steam in a moveable bed: (a) screw conveyors, conveyors with buckets or other mixing devices; (b) a fluidized bed; and (c) extrusion coolers. In each case the steam treatment is followed by a drying stage.

## Screw conveyors for direct steam treatment in a moving bed

For direct steam treatment, screw conveyors [33, 77, 78, 79] or blade conveyors [78] have been used.

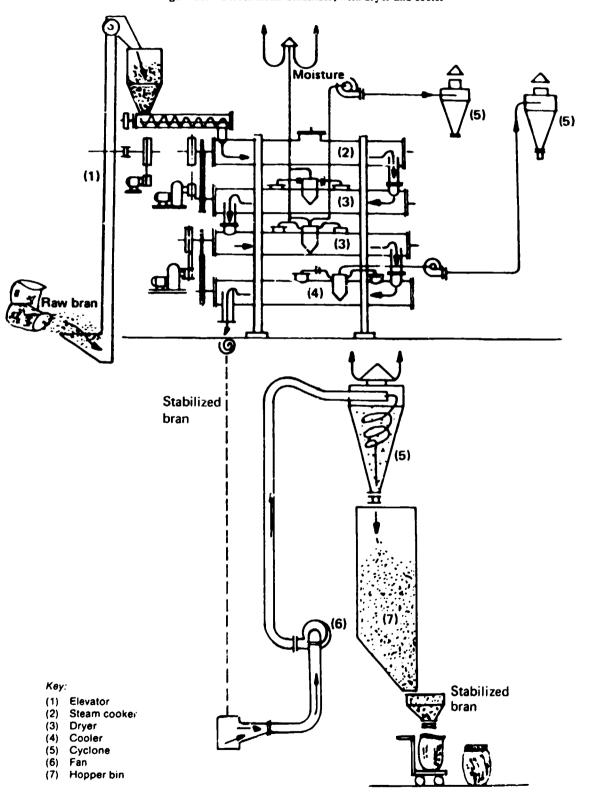
Viraktamath [77] has carried out tests on an industrial scale, using conventional oil-mill equipment. In one case he used a conditioner with a capacity of 1 t/h, consisting of a cylinder drum fitted with a steam jacket, direct or of steam injector and bucket conveyors to circulate and mix the bran simultaneously. In another case he used semi-cylindrical conditioners (1 t/h and  $\frac{1}{4}$  t/h capacity), also equipped with a steam jacket, direct steam injector and bucket conveyors. Working continuously, the dwell-times were 15 minutes in the 1-tonne units and 8 minutes in the  $\frac{1}{4}$ -tonne unit. The final temperature was 95°-97° C in all cases. During the treatment the moisture increased by 2-3 percentage points but the increase was subsequently eliminated by drying and cooling in the shade. The bran, which was packed in jute sacks and stored in unspecified ambient conditions, showed an increase in acidity, but it took more than 15 days to reach 10 per cent FFA.

Burns and Cassidy [78] have patented a procedure for stabilizing rice bran, based on the use of a screw conveyor 30 m long, fitted with a hopper and with a small screw for feeding the material. In the first section of the screw, which is 6.6 m long and thermally lagged, steam is injected at 100 lb pressure througn a perforated tube. The moisture content increases by 3-5 percentage points, the bran having been subjected to 100° C for  $1\frac{1}{2}$  minutes. In the second section, which is 20 m long and also thermally lagged, steam is injected and radiant heat provided in order to maintain the temperature of the product at  $102^{\circ}-104^{\circ}$  C, the dwell-time being about 3 minutes. Finally, in the last section, which is about 3.3 metres long and equipped with a duct of ample draught to eliminate the steam, the product is allowed to cool down partially. The installation requires other ancillary devices (for example a dryer), which are not described.

Yokochi [79, 80] has designed an installation for stabilizing bran (see figures 15 and 16) with the following characteristics: (a) feed system, equipped with bucket elevator, hopper and loading screw; (b) horizontal cooker, with steam jacket and direct injection of steam, fitted with agitator system; (c) horizontal dryer, with steam jacket, fitted with agitator; (d) horizontal dryer-cum-cooler, with steam jacket, fitted with agitator; (e) pneumatic conveyor with cyclone separator and hopper; (f) transmission systems; and (g) control systems. The bran is treated for 3 minutes in the cooker and then dried to reduce the moisture content from 14 per cent to 3 per cent, at which level the bran should then be stored.

The design in figure 17 offers an alternative method. The system is composed of: (a) a cooker using direct injection of steam and a conveyor, with a retention-time of 5 minutes and an outlet temperature of 110° C; (b) a pelletizer; (c) an elevator; and (d) a column dryer with cooling chamber [81].

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Figure 15. Direct steam stabilizer, with dryer and cooler

Source: Yokochi [79].

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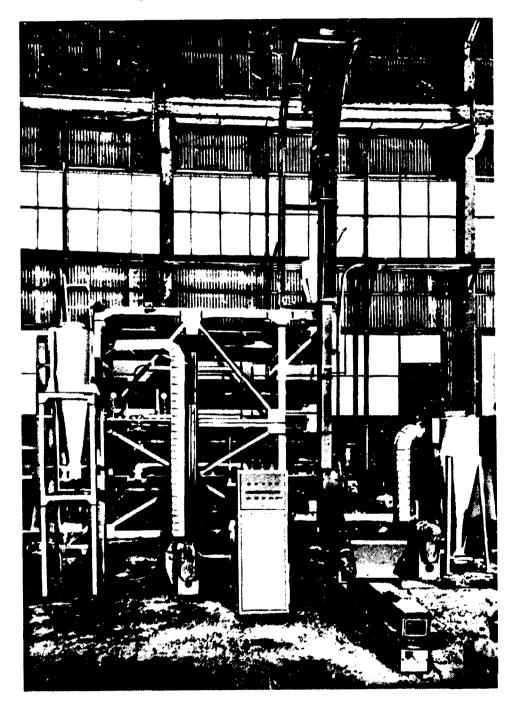
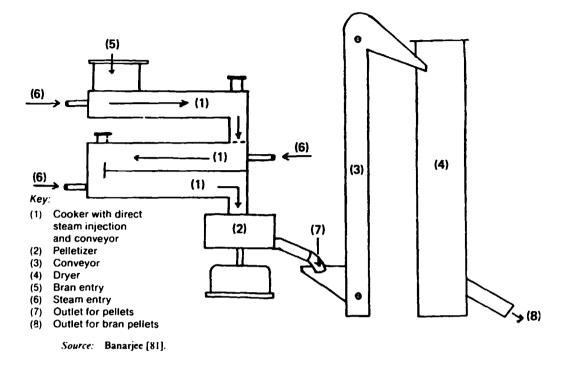


Figure 16. Bran stabilization unit being assembled

Source: K. Yokochi [80].

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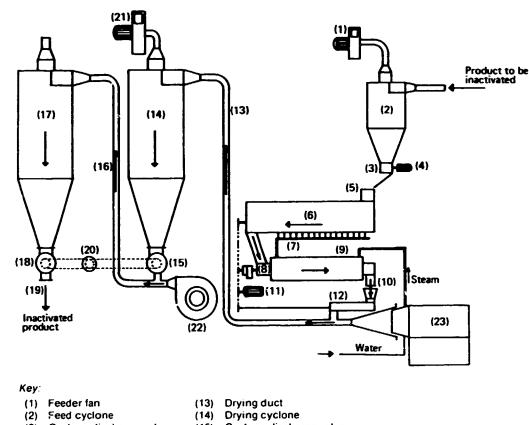


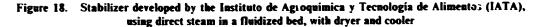
#### Figure 17. Alternative type of stabilizer

## Devices for direct steam treatment in a fluidized bed

There is only one reference to a device that meets the specifications for treatment of bran with direct steam in a fluidized bed. This is the stabilizing unit developed by the Instituto de Agroquímica y Tecnología de Alimentos of Valencia.<sup>9</sup> The device is distinctive in that it uniformly and almost instantly humidifies and heats each of the discrete particles which make up the bran, keeping them constantly on a fluidized bed. Thus, under time, temperature and moisture conditions that can be easily regulated, every one of the bran particles is rapidly and totally inactivated. The installation consists basically of an inactivating unit, a drying unit and a cooling unit, together with control and other auxiliary and optional devices (see figures 18 and 19). The inactivating unit is composed of a metering hopper and the inactivator itself, with a fluidized bed. The hopper may be loaded by a pneumatic or other system. It can also act as a feed-regulating tank, when the stream of bran comes straight from the whitening cones. The inactivator includes a steam injection system with mechanical agitation for fluidization. It can also include a second, retaining, unit, consisting of a screw conveyor with steam jacket, to combine the treatment time with the feed capacity. The drying and cooking unit consists of a ducting and cyclone dryer, ducting and cyclone cooker, valves at the cyclone outlets, fans for the drying and cooling circuits and the necessary electrical drive motors. Figure 18 shows how the installation works. The feed cyclone (2) receives the bran and meters it out continuously to the inactivation

<sup>&</sup>lt;sup>9</sup>"Patrinato Juan de la Cierva", for scientific and technical research, Spanish patent No. 401.685 of 16 May 1974.





- Cyclone discharge valve (3)
- (4) Valve motor
- (5) Hopper
- (6) Inactivation unit
- Steam distributor (7)
- Screw conveyor
- (8) (9) Steam jacket
- (10) Hopper
- Inactivation unit: motor (11)
- and screw conveyors
- (12)Screw conveyor

- Cyclone discharge valve (15)
- (16)Cooling duct
- (17) Cooling cyclone
- Cyclone discharge valve (18)
- Outlet for inactivated product (19)
- Motor for valves of cooling (20)
  - and drying cyclones
- (21) Drying fan
- (22) Cooling fan
  - Equipment for the production (23)
  - of steam and hot air

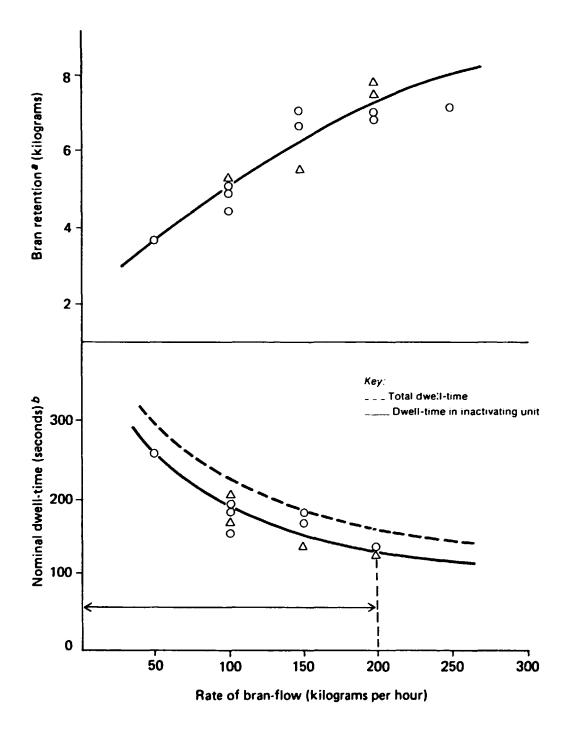
unit (6); in the latter, the bran receives a direct injection of steam, and this, in conjunction with the mechanical agitation system, maintains the product in perfect fluidization for  $2\frac{1}{2}$ -3 minutes at about 100° C, with a moisture content 4-5 percentage points higher than at the start. The inactivated bran is extracted continuously by the worm gear for drying. If the plant includes the optional retention unit (steam jacket) (9) the speed of the feed and the retention time in each phase can be adjusted. On leaving the inactivator, the bran is discharged into the dryer, where it is carried along by the stream of warmed air produced by the fan (21) in conjunction with the steam and hot-air equipment (23). In the cyclone (15), the dry powder is separated and extracted through the lower part of the cyclone. The fan (22) then drives the dry bran through the ducting (16), cools it, and afterwards collects it in the cyclone (17), from which it is extracted



Figure 19. IATA stabilizer

for storage, in sacks or in bulk, in moisture and temperature conditions similar to those at the point of entry. Figure 20 illustrates the variation in the nominal dwell-time and retention of bran in the inactivator in terms of product flow. It must be pointed out that the dwell-time does not vary linearly with the reciprocal of the flow-rate; the weight of the material that is retained increases appreciably with the growing flow-rate. Consequently, the dwell-time is greater for higher flow-rates, thus widening the operating range of the machine: 155 seconds was sufficient to destroy the peroxidase activity of the bran completely.

Table 10 gives the operating details for an industrial prototype with a 500 kg/h capacity.





Source: Fito and others [83].

<sup>a</sup>Weight of bran retained in the inactivating unit.

<sup>b</sup>Calculated according to the equation DT = M/F, *M* being the weight of bran retained in the inactivating unit (kg) and *F* the bran-flow (kg/s). The dwell-time in the optional retention body was 30 s, whence total DT = 30 + (M/F).

			Characteristics of stabilized bran			
Processing conditions				Moisture	Peroxidase activity <sup>b</sup>	
Run No.	Flow-rate of bran (kg/h)	Steam injected into bran (kg/h)	Ratio of steam to bran	content <sup>a</sup> (percentage, wet basis)	Raw bran	Stabilized bran
I	290	34	0.117	11.9	12.2	0.00
2	360	36	0.100	12.0	10.6	0.00
3	410	44	0.107	12.7	8.2	0.01
4	470	47	0.100	12.5	11.4	0.00
5	510	55	0.108	12.5	12.5	0.01

# TABLE 10. STABILIZATION OF BRAN USING AN INDUSTRIAL PROTOTYPE OF THE IAT & STABILIZER

Source: Fito and others [83].

"Initial moisture content of the bran: 12.2-13.2 per cent wet basis; drying air at  $115^{\circ}-120^{\circ}$  C; air-flow 1,700 m<sup>3</sup>/h.

<sup>b</sup>Absorption units per gram of bran, wet basis, determined according to the method used by Vetter. Steinberg and Neison [82].

#### Extrusion cookers using water or steam injection

In addition to the extrusion cookers described earlier, there are other types equipped with devices for injecting steam or water directly into the mass of material that is transported under pressure. Williams and Baer [56] have used one of these machines, a pilot model of the "Expander" cooker, to stabilize bran. The machine had an inner diameter of about 11.5 cm and was about 1.30 m long. The die had a single gap of 15.88 mm and the shaft revolved at 216 rpm, driven by a 25-hp motor. The water injector was situated 1 m from the outlet and the steam injector was about 30 cm away from it. The freshly milled bran was fed into the machine through a system composed of a hopper and screw, conveyed about 15 cm and then injected with water. After moving along about another 75 cm, it was injected with steam. Under the conditions specified by the authors, the bran acquired a moisture content of close to 25 per cent and reached a temperature of about 115° C on combining frictional heat and the heat introduced by the injected steam. The fall of pressure when the bran was discharged led to rapid vaporization of the water (which had remained liquid under the high pressure inside the casing) and expansion of the product bran. The bran lost up to 25 per cent of its moisture content with instant cooling, and was finally dried to the required moisture level in a suitable dryer. Typical data on the stabilization of bran with this type of extrusion cooker are summarized below.

Feed specifications

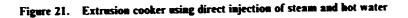
Moisture content of the bran, 9 per cent Load capacity, 5.8 kg/h Injection of water, 10 kg/h Injection of steam, 4.5 kg/h

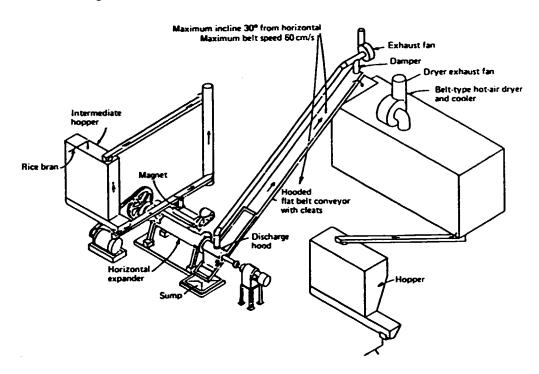
Expansion specifications

Moisture content of the bran, 27 per cent Inside temperature, 115° C Temperature on discharge, 82.5° C Production, 72.5 kg/h Product specifications

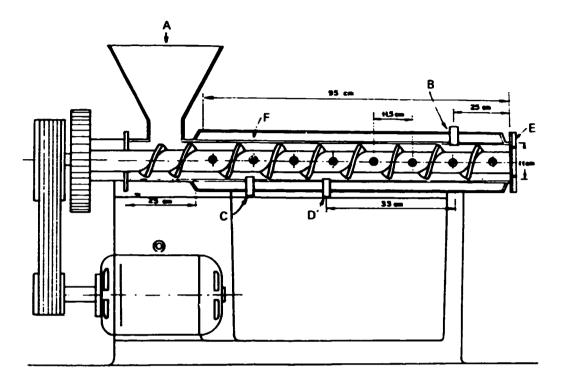
Moisture content of the bran, 23 per cent Density in bulk, 400 kg/m<sup>3</sup> Coefficient of expansion, 1.24

Figure 21 illustrates a typical installation of this type. Plants with a capacity of 9-270 t per 24-hour day, or even larger, are available.





Other models have been designed, constructed and tested on an experimental basis [11, 83] but there are no reports of their having been used in industry. Filho, Germany and Melo [11] have described an extrusion cooker for the stabilization of bran that also uses direct damp steam (see figure 22). It is composed of a cylinder with a double wall for indirect heating, 120 cm long, with an inside diameter of 11 cm, with water and steam injection valves. At the points along the shaft where the thread is interrupted, bars are fixed on the cylinder to break up the mass. The extrusion cooker has a die with six vents 5 mm in diameter. The shaft revolves at 100 rpm and is driven by a 20-hp motor. In the experiment, the initial moisture content of the bran was 11.7 per cent, which was raised to 14 per cent in order to increase the machine to efficiency. The temperature before the bran left the machine was about  $100^{\circ}$ - $110^{\circ}$  C. The moisture content of the bran when it left the extrusion cooker was reduced to 12 per cent, so for satisfactory storage the bran then had to be dried until the moisture content reached 9 per cent.





#### Key:

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- Bran feed
- D Steam injection
- E Plate with outlet holes for the bran F Steam jacket
- C Water injection F

Steam injection

Source: Filho, Germany and Melo [11].

## Criteria for the evaluation of stabilization methods

The ultimate object of stabilization is to convert bran into a durable product, under normal ambient and storage conditions, preserving as far as possible the good qualities of the original product. The most important way of evaluating stabilization methods, therefore, is to measure the quality of the stabilized product and determine to what extent the objectives pursued in the process have been attained. The adaptation of the method to industrial requirements and availabilities is another criterion, which may have either a technical or an economic basis. In some cases, the two aspects are closely related, so that the one cannot be separated from the other.

The concept of quality embraces such aspects as: (a) the stability of the treated bran; (b) the degree of retention of the desirable properties of the untreated bran; and (c) improvements in other functional properties of the bran.

## Criteria for evaluating the stability of treated bran

The most important criteria for evaluating the stability of treated bran are: (a) moisture content and temperature; (b) residual enzymatic activity; (c) total count of micro-organisms and, in particular, of moulds producing lipase; (d) other causes of biological change; and (e) storage life.

#### Moisture content and temperature

In order to ensure satisfactory storage, moisture content and temperature must be kept below critical values. A product is said to be in a "dry" or "safe" condition [84] when it has a moisture content at which, taking into account the likely range of temperature to be experienced during storage and transport, there is no risk of appreciable metabolic activity of the grain itself, nor of attack by moulds or other micro-organisms and other pests. The moisture range for which there is a minimal risk of change varies according to the condition of the consignment of bran, and depends on the temperature and duration of storage. In general it may be assumed that a "safe" moisture content is below the level at which it is in equilibrium with a relative humidity of 65 per cent. If a stabilization method requires that the treated bran should have a lower moisture level to be effective, the stabilization will not be adequate.

In a consignment of bran, the periodical changes in the relative humidity of the atmosphere particularly affect the surface layers. If the consignment is a large one, the relative humidity of the interstitial air inside is chiefly controlled by the initial moisture content and temperature of the stabilized bran. The relative humidity and moisture content gradients are determined by continuing differences in the temperature between the surface and the inside, caused by changes in the external temperature or internal heating. The moisture migrates to the coldest area, and so raises the moisture content of the bran in that area.

High temperatures in the treated bran can lead to oxidative reactions in the fats during the storing period, and if the bran is stored without cooling there is a risk of spontaneous combustion. Furthermore, if cooling takes place quickly, without ventilation, the relative humidity of the interstitial air increases dangerously.

To sum up, therefore, the temperature of the bran at the end of the stabilization process should be as close as possible to the ambient temperature and the moisture content should be somewhat lower than the one that would be in equilibrium with the 65 per cent humidity relative to the ambient temperature. Otherwise, the bran should be subjected to additional stages of drying or cooling, and these must be incorporated into the overall stabilization process if the latter is to be acceptable.

## Residual enzymatic activity

In bran that is adequately stabilized, the lipase activity must be nil. While this condition is necessary, it may not be sufficient, as other harmful enzymes

may remain at functioning levels of activity. As has been indicated elsewhere, peroxidase is the most resistant of all known vegetable enzymes. Therefore, determination of the peroxidase activity gives a good indication of the degree of inactivation of the bran. If the peroxidase activity is nil, the others, including the lipase, will also be nil.

The methods for determining lipase in bran consist in evaluating the acidity developed when the bran, or better, an aqueous extract of it, is allowed to act on oil or a fatty acid ester under controlled conditions. In one method [85] the bran is defatted with hexane at 25° C, is then extracted with distilled water, and an aliquot of the extract, after centrifuging, is made to react with Tween 2010 at pH 7.2; the pH is readjusted periodically with alkali for 30 minutes, and the lipase activity is calculated from the volume consumed. The activity is expressed in units of lipase per kilogram of bran, a unit being the quantity of lipase which catalyses the formation of a milli-equivalent of acid per minute. In another method [46], based on the procedure established by San Clemente and Vadhera [86], the extract containing the enzyme is prepared directly from the undefatted bran by vigorously agitating it with water at 4° C; an aliquot of the extract, after it has been through the centrifuge, is allowed to act for 24 hours on an aqueous emulsion of refined olive oil at pH 8 and 35° C. The fatty acids thus liberated are evaluated with alkali, after adjustment of the pH to 8 6, on the sample and on a blank. The lipase activity is expressed as micro-equivalents of acid produced per minute per cubic centimetre of enzyme solution.

The method used by Vetter, Steinberg and Nelson [82] to determine peroxidase activity gives good results. It is based on colorimetric determination of the colour developed by the o-phenylendiamine oxidized by the enzyme in the presence of hydrogen peroxide. An aqueous extract of the enzyme is prepared using a phosphate-citrate buffer, and to an aliquot of the filtrate is added o-phenylendiamine and hydrogen peroxide, allowing it to react for 5 minutes at 25° C. The reaction is halted with bisulphite and the absorption of the sample is measured against a blank. With bran, there is no interference from starch, so the solutions are perfectly clear, which simplifies the test.

The absorption curve of the oxidized colour shows a peak at 430 nm. The colour developed by the oxidized o-phenylendiamine follows Beer-Lambert's law. The peroxidase activity is expressed in units of absorption per gram of bran. A simplification of this method, in which the spectrophotometric measurement is replaced by visual comparison of the sample and the blank in Nessler tubes, makes it possible to detect very quickly the pressure of peroxidase in the bran. Since it is easily able to distinguish between activated and inactivated bran, this method is very useful for in-plant control of stabilization in industrial processing.

It must be noted that the determination of FFA during storage is not an adequate measurement of the effectiveness of the stabilization procedure. In the first place, it takes a very long time. In the second place, the development of FFA depends on many factors, some of which are exogenous and occur after stabilization. On the other hand, the FFA content is a good index of the fitness or suitability of the storage conditions.

<sup>&</sup>lt;sup>10</sup>Soluble ester of lauric acid and polyoxyethylenated sorbitan. The use of this watersoluble substrate avoids the difficulties usually presented by other methods that use water-oil systems, since the degree of dispersion of the substrate is an important factor.

## Total count of micro-organisms and lipase-producing moulds

Very little information is available on the parallel destruction of enzymatic activities and micro-organisms, and, in particular, on the typical bran flora to be found in different rice zones and on the identity and incidence of lipaseproducing moulds, the heat-resistance of their spores etc. Nevertheless, under the conditions in which peroxidase is destroyed in the bran, the microflora are reduced to such low levels that there is no danger of deterioration, provided the moisture and temperature conditions are kept within safe limits.

Determination of the microbial count, particularly moulds, in the stabilized bran is very useful for evaluating a particular stabilization method under certain local conditions, at least until some familiarity with the microflora peculiar to the area has been gained.

## Other causes of biological deterioration

Deterioration in the bran can also be caused by other living organisms, of which insects and mites are the most important. However, the conditions necessary for the inactivation of peroxidase, in particular by heat treatment, exceed the levels of resistance of any of the vegetative forms of these organisms. These biological causes can acquire special significance when specific agents are employed to destroy the enzymes, and their possible presence ought to be taken into account.

## Storage life

The criteria for evaluating the stabilization process are not independent of each other but closely interrelated. An industrial batch of bran may have considerable residual lipase activity, but if its moisture content is maintained at a low level and there is no biological infestation, it will have a long storage life. On the other hand, under industrial conditions, the factors that determine the success of the stabilization process may only approximate the safest and most favourable extreme values, for example, if the residual microflora are at a low level but have a definite potential for growth. A combination of circumstances determines whether or not the residual levels of micro-organisms, moisture and enzymes are operative and how long the storage life of the bran will be. Lastly, the usefulness of the process also depends on whether there is any possibility of the bran being contaminated during subsequent operations, after it has been treated to destroy the causes of change. The cooling of the treated bran is a very risky stage. The air is a favourable vector for contamination; bran acts as a filter, retaining a large proportion of the micro-organisms and insects contained in it. Changes in the temperature and relative humidity of the environment also play an important role.

The storage life of the treated bran may be predicted from the values of the characteristics specified below (criteria (a) (i)-(iv)), taking into account the risks of recontamination and local environmental conditions. While some knowledge has been gained of the influence of these factors, storage tests are advisable. To be of any use, they must satisfy two conditions: first, the characteristics determining the

potential stability of the stabilized bran must be known; and secondly, all the environmental conditions that influence the storage life of bran must be controlled. A third condition, which is optional but strongly recommended, is that the test conditions simulate as far as possible the conditions prevailing in industry or, should that already be the case, that they make it possible to predict with reasonable certainty the behaviour of the bran under local conditions. Specialized bibliographies list numerous tests that have been done on the storage of treated bran, but they are not of any great use since they do not meet the above conditions. A very common practice in evaluating the value of a stabilization process is to carry out a simple storage test, in which the development of FFA-the only index of quality considered—has been followed and compared in both the treated sample and the original sample. The difference between the two is of little practical significance, and the values acquired by the treated sample depend on factors that are not controlled: initial lipase activity, recontamination, increase in moisture content during storage, temperature fluctuations etc. The value of this information also depends on the size of the sample, the type of container and the circumstances in which the bran is stored. Results obtained with samples of 250 g packed in small cloth sacks and kept under laboratory conditions, which are generally not specified but are supposedly hygicnic and ventilated, cannot be extrapolated to those obtained under industrial conditions. They do not make it possible to predict the behaviour of the same bran in batches of several tonnes, packed in jute sacks, probably re-used, and stowed in a warehouse with limited ventilation.

In short, storage tests, far from being the simplest way of evaluating the value of a stabilization process, are a complex procedure and many variables must be taken into account and controlled if the tests are to be of any use. On the other hand, it is strongly recommended that they be carried out in order to determine the fitness of any new stabilization process under local conditions.

It is suggested that the following factors should be taken into account:

- (a) Characteristics of the stabilized bran;<sup>11</sup>
  - (i) Residual lipase activity;<sup>12</sup>
  - (ii) Mould count;
  - (iii) Presence of insects;
  - (iv) Moisture content;
  - (v) Temperature;
- (b) Environmental conditions and means of storage:
  - (i) Relative humidity;
  - (ii) Temperature;
  - (iii) Lighting;
  - (iv) Ventilation;
  - (v) Type of container and stowage;
  - (vi) Size of sample;
  - (vii) Characteristics of the premises in which the bran is stored.

<sup>&</sup>quot;The consignment must be analysed to ensure that these characteristics are homogeneous,

<sup>&</sup>lt;sup>12</sup>Residual lipase activity does not need to be determined if the peroxidase activity is nil and if there is no evidence of lipases of bacterial origin that are more thermo-resistant than peroxidase.

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## Criteria for evaluating the loss of valuable properties in the bran during stabilization

The most important existing application of commercial stabilized bran are as a source of edible oil and as an ingredient in feeding-stuffs. Among the most promising possibilities are its use in the manufacture of foodstuffs, and probably also as a raw material for certain pharmaceutical products. For each of these purposes, bran possesses some natural qualities which it is important to preserve, and which must therefore be controlled. The absence of toxicity is a prerequisite for all applications in foods or feeding-stuff.

## Bran used as a source of *zdible* oil

In evaluating the loss in quality during stabilization when bran is used as a source of edible oil, the chief criteria to be taken into account are: (a) total yield of oil; (b) speed of extraction; (c) behaviour of the crude oil during refining, particularly the ease with which it can be decolorized; (d) particle size.

It is very important to know the total yield of crude oil. Before and immediately after the stabilization treatment, similar values are generally obtained.

Speed of extraction and particle size are important attributes, and the known stabilization procedures involve the use of conditions that may well have either a positive or a negative effect on those qualities. Granulation of the bran and expansion of the granules during stabilization by extrusion can eliminate the subsequent need to pelletize the bran in order to avoid the problem of fines and improve the speed of extraction.

It is also important to know how the oil behaves during the decolorization process. Some methods of stabilization, such as heat treatment without the addition of water, darken the oil. They are said to "fix" the colour. The pigments are relatively easy to eliminate with the use of neutralization and, in particular, decolorization treatments; the highly coloured products of degradation, which are largely responsible for the darkening and poor colour, are, however, very hard to eliminate. Therefore, it is more important to measure the colour of the decolorized oil than that of the crude oil, under simulated industrial conditions. The colour differences of the crude oils do not always correspond to those of the decolorized oils. The oil, once decolorized, must satisfy the same quality specifications as oil extracted from crude and untreated bran if the bran has just been manufactured and is suitably refined.

The colour of the oil is generally measured with a Lovibond Tintometer [87], comparing the colour of the bran oil (placed in a glass column 154 mm long and 19 mm in diameter and filled to the 133.35 mm mark) with the colour of several standard glasses, in three graduated series: yellow, red and blue. Generally, a combination of the red and yellow glasses is enough; the blue glasses are necessary only if the oil has a green tinge. The red glasses are standardized against the Priest-Gibson N colour scale. The yellow glasses are not standardized, since the human eye can only detect relatively large variations in yellow, and when comparing the colour of the oil sample the colour need only be approximated to obtain a satisfactory comparison with the red glasses.

For bran oil, as for most oils, the depth of colour may be satisfactorily expressed in terms of "red" units. The Lovibond system for measuring colour is not suitable, however, for highly coloured oils or for colour other than red and yellow.

The colour of the oil may also be measured spectrophotometrically, and this method is widely used in investigations and in routine control of production. The spectral transmission of the oil at 525-550 nm is not only more reproducible than the Lovibond red values, but is usually related to them. The American Oil Chemists Society (AOCS) method determines the optical absorption of the oil at wavelengths of 460, 550, 620 and 670 nm, in cells with a 21.8 mm inner diameter. It uses the following equation to calculate the photometric colour, which approximates the colour expressed in Lovibond red units:

Photometric colour =  $1.29A_{460} + 69.7A_{550} + 4.12A_{620} - 56.4A_{670}$ where A is the absorbency [88].

From the industrial point of view, some additional characteristics of the stabilized bran may be worth considering, inasmuch as they can reflect negative aspects of the stabilization process. These include the product's predisposition to oxidative reactions during storage and handling. Stabilization under certain conditions can cause a significant loss of tocopherols, with their anti-oxidant properties. This, in association with low moisture levels and the presence of oxygen in the large volume of interstitial air, encourages oxidative changes in the fats of the treated bran. Furthermore, in some machines stabilization can lead to the formation of particles of greatly varying sizes, even, occasionally, large lumps, which make it difficult to handle the product. In processes involving the use of chemicals the possibly harmful or unpleasant effects on workers who handle the treated product should be borne in mind.

### Bran used as an ingredient of feeding-stuffs

The value of bran as an ingredient of feeding-stuffs lies in its calorific value, its protein value and its vitamin value. The relative importance of these three components depends on whether the bran is used in the formulation of compound, nutritionally well-balanced feeding-stuffs or whether it is used as a normal component of an animal diet that is not scientifically formulated, as is the case with rural feeding. In the first case, the vitamin content is not of overriding importance, as the bran is not used in place of vitamin additives; in the second case, however, it may be important if the daily vitamin requirements of the animal can only be met by retaining the vitamins in the bran. In this sense, thiamine and the tocopherols are important. Both are sensitive to heat, and severe heat treatment can result in significant losses. Thiamine is destroyed by the action of  $SO_2$ .

It is not likely that the energy value of the bran will suffer significant losses in any of the known stabilization processes.<sup>13</sup>

<sup>&</sup>lt;sup>13</sup>Some authors have suggested that the extraction of fats could be a means of increasing the stability of bran. Defatting cannot, however, be regarded as a stabilization procedure, quite apart from the fact that acidification of the residual fat continues to be a problem in defatted bran and it has been pointed out that heat treatment is necessary to arrest the process [89].

The protein value of the bran depends on two main factors: the total concentration of proteins and the distribution of the amino-acids that make up those proteins. The proportion of the protein swallowed that is actually digested and absorbed is largely determined not only by the protein ration, in terms of its composition in amino-acids, but also by the availability of the amino-acids. Stabilization processes, and particularly heat processes, are not expected to affect either the concentration of proteins of the bran or its aminoacid composition. Depending on the severity of the treatment, however, they can reduce the availability of some of the amino-acids. The available lysine is a good index of amino-acid retention during stabilization. Lysine is sensitive to the necessary conditions of enzymtic inactivation and, above all, is a limiting amino-acid in bran. Moreover, acceptable chemical methods for determining its presence are available.

The available lysine can be determined by the direct fluorodinitrobenzene method known as Carpenter's method [90] as modified by Booth [91]. The procedure involves converting the lysine residues with free  $\varepsilon$ -amino groups in the proteins into yellow 2,4-dinitrophenyl derivatives by treating them with an alkaline solution of 1-fluoro-2,4-dinitrobcnzene (FDNB). The dinitrophenyl derivatives of other *a*-amino-acids are soluble in ether (except when derived from *a*-arginie), and are eliminated by extraction with this solvent, the absorption rate of the residual aqueous phase being measured afterwards. The interference of the dinitrophenyl derived from *a*-arginine is corrected by making the  $\varepsilon$ -dinitrophenyl derived from the lysine react with methyl chloroformate (methoxy-carbonyl chloride), which gives rise to a compound that can be extracted in ether. The dinitrophenyl derived from the *a*-arginine remains in the aqueous phase, and the absorption is again measured. The initial reading minus the final reading gives the quantity of available  $\varepsilon$ -lysine present in the bran.

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# VI. The storage of rice bran

### Storage in practice

Rice bran is a product which, under normal conditions, deteriorates rather rapidly and must therefore be stored for as short a time as possible before being used or sent for subsequent industrial processing (mainly for the production of feedstuffs and the extraction of oil). In practice, however, circumstances vary greatly from one area to another and the time that elapses between production of the bran in the mill and its consumption or use in a later process may vary from a few hours to several months. In many rural areas, the bran is produced in a small local mill, sold or distributed daily, and consumed the same day or within 48 hours. The same is true of integrated extraction plants combined with large mills. In other areas, however, located far from the production centres, such as those of Bombay, the bran being processed has been produced several months earlier. It is collected in small batches from small village mills by intermediaries, transported some 200-400 kilometres, and distributed to the extraction plant as soon as possible, and then quite possibly several more days will elapse before it is processed. Thus, even taking into account the fact that the miller, the intermediaries and the users try to keep delays to a minimum, in practice the bran will still have to be stored. Because of the instability of the product itself, major changes will occur even in a short storage period.

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Because bran is a by-product, it has to be dispatched or consumed quickly, and the technology is still at an early stage, storage is generally somewhat haphazard. After passing through the whitening machines, the bran is left to accumulate, sometimes just in a heap on the ground, or is transported to a store or silo in the larger mills.

Bran stored in silos usually creates problems because of its tendency to cake and so clog the outlet. It is often possible to see dents in the wall of the silo or store near the outlet, caused when workers have banged the surface with sticks to force out the bran.

From the store or silo, the bran is discharged directly into lorries for transport in bulk. In many areas of the world, the bran is usually handled and transported in sacks. The sack is an important factor because of its economic and social implications in terms of labour and because of its role in preserving the bran.

## Factors that influence changes in the bran

A consignment of bran, in industrial and commercial practice, is a very complex ecosystem. It contains the germ of the seed, which retains its respiratory and germinative capacity. It also contains fragments of tissue with large numbers of highly active enzymes. Its cellular structure is largely destroyed, allowing its constituents to establish contact with one another and with the oxygen in the large area of interstitial air that it occludes. The microbial content is generally high. Bran is also a carrier of insect eggs or various adult species of insects. The internal and external biological activities and chemical reactions that may occur will depend upon a number of factors. The most important of these are: (a) moisture content; (b) temperature; (c) time; (d) composition of the interstitial atmosphere; and (e) physical and chemical characteristics of the bran particles and their interrelationships. Moisture (in terms of both relative humidity and water activity) plays a fundamental role, and without an adequate knowledge of this role, the behaviour of bran during storage cannot be properly understood. This study of the changes that take place in the bran will therefore start with a study of the role of water.

## The role of water: sorption isotherms

Molecules of water can form hydrogen bridges:<sup>1</sup> (a) between other water molecules, to form the aggregates characteristic of the liquid state or the crystalline structures characteristic of ice; and (b) with polar organic groups (-OH, -COOH, -NH) (see figure 1). The latter are frequently found in bran; starch and sugars, which are the main constituents of the by-product, are characterized by the large number of hydroxyl groups present, and proteins,

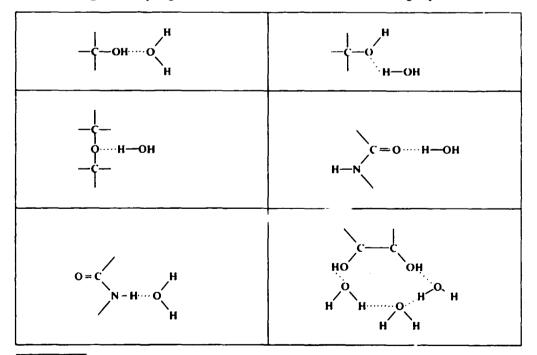


Figure 1. Hydrogen bonds between water and various functional groups

<sup>&</sup>lt;sup>1</sup>The linkage through electrostatic hydrogen bonds may be formed between an electronegative atom (O, N, F, S, Br, I, Cl), which has a surplus electron pair, kn we as the donor, and a hydrogen atom, known as the acceptor. This is a weak bond of low energy (17-46 kJ/mole) compared with the covalent bond (about 419 kJ/mole).

another major constituent of bran, also contain a large number of polar and ionic groups in their amino-acid side chains (-OH, =NH,  $-NH_2$ ,  $-NHCNHNH_2$ , -COOH,  $-CONH_2$ ), all of which are capable of being linked through the formation of hydrogen bonds. Water can also form structures around nonpolar groups ( $\equiv CH$ ,  $=CH_2$ ,  $-CH_3$ ) for which it shows no affinity: the polar groups, connected to water through hydrogen bonds, force the non-polar groups into water-free zones within the macromolecule. The affinity of a biological material for water depends upon the ratio of the number of polar groups to non-polar groups, on its accessibility and on its orientation.

The affinity of water for bran is shown graphically by the so-called sorption curve (see figure 2). The curve represents the quantity of water adsorbed by a substance (bran) at a constant predetermined temperature (hence the term "isotherm"), as a function of the relative humidity of the atmosphere in equilibrium with the product. It is also possible to express it as a function of the activity of the water or of the equilibrium vapour pressure. The water activity  $a_w = p/p_o$  is a function of the relative humidity at equilibrium with the product (RHE); where p is the vapour pressure exerted by the water in the substance at a predetermined temperature, and  $p_o$  is the saturation vapour pressure of water at the same temperature. There is a different meaning for  $a_w$ and RHE: RHE refers to the gaseous and  $a_w$  to the condensed phase.

The typical sorption isotherm is in the form of a sigmoid. It has three well-defined sections: one in which  $a_w$  is between 0 and approximately 0.10, and in which it is assumed that the water molecules are strongly bonded at the free and accessible polar centres; a second section, in which  $a_w$  lies approximately between 0.10 and 0.65, corresponding to the straight part of the plot, and in which it is assumed that the molecules of water attach themselves to the preceding ones or to the polar centres that are made accessible by the structure

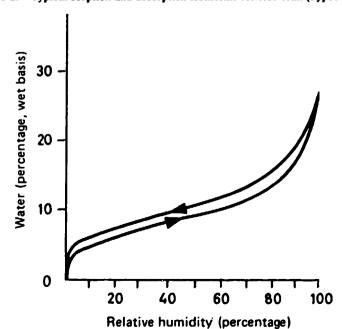
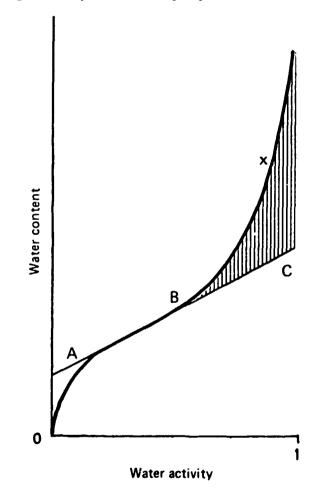


Figure 2. Typical sorption and desorption isotherms for rice bran (hypothetical)

of the substrate; and a final section with  $a_w$  values of over 0.65 and with a marked slope, which suggests the accumulation of additional molecules of water in secondary and tertiary structures of the macromolecules, also as a result of capillary and osmotic effects at high  $a_w$  values; in this area, the water molecules possess high mobility. As reported by Multon and Bizot [1], Guilbot and Lindenberg [2] demonstrated that the difference in order between the sigmoidal section and the prolongation of the straight section (the shaded area in figure 3) represents the fraction of the adsorbed water retained by low energy bonds with solvent properties. The ordinate of the straight part extrapolated to  $a_w = 1$  represents strongly bonded water, without solvent power.



#### Figure 3. Sorption curve showing the presence of solvent water

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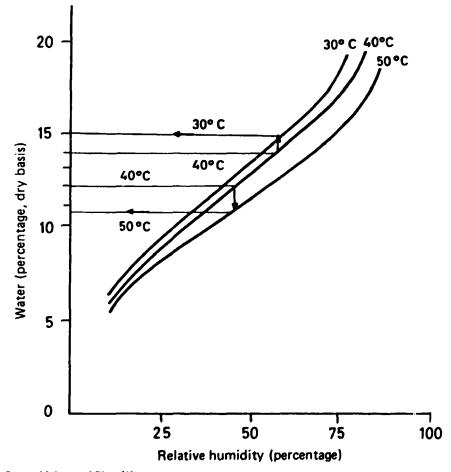
- A Point corresponding to the quantity of strongly bonded water
- B Point at which solvent water appears
- C Point corresponding to the total quantity of non-solvent water
- x Available water

Source: Guilbot and Lindenberg [2].

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The sorption and desorption curves do not coincide (see figure 2). The latter moves towards the origin of the co-ordinates. The moisture content corresponding to a given relative humidity may have one of two values, depending upon whether the material is gaining or losing moisture. At the same RHE, bran, like other cereal products, has a higher moisture content when it is being dried than when it is acquiring moisture. This phenomenon, known as hysteresis, may represent differences of 1-2 percentage points in the moisture content for any given RHE value. In some cases it will thus be important to know whether the data refer to sorption or to desorption.

If, instead of considering a single temperature, a range of temperatures is considered, a series of isotherm curves is obtained (see figure 4). The vapour pressure exerted by the moisture content of the product changes in such a way that the ambient atmospheric humidity represents an almost constant fraction of that of a saturated atmosphere at this temperature. As the temperature increases, the affinity of the bran for water decreases. Consequently, if the bran is heated it loses water and, conversely if it is cooled it absorbs water.





Source: Multon and Bizot [1].

Under the conditions represented by the final section of the isotherm, beginning with  $a_w$  values of around 0.75 and 0.80, storage will be risky. From this point onward, water will be available, and this can be used by microorganisms, thus facilitating their proliferation. Above an  $a_w$  value of 0.75, moulds develop rapidly and produce heat, which will increase the possibility of deterioration. To avoid any risk of degradation, it is therefore necessary to keep  $a_w$  below 0.75. With some exceptions, no enzymatic reaction (except for lipase) and no major bacterial growth will take place without a minimum of solvent water.

If the wrapping is not moisture-proof, the product can lose or absorb water; in the first case, there will be a loss in weight and, in the second case, if  $a_w$  exceeds 0.75, there will be a risk of degradation. On the other hand, if the wrapping is moisture-proof, the weight-loss will be avoided but, if the temperature falls, the RHE will increase and, depending upon the amount, may give rise to condensation. Similar problems may also arise in products kept in stores and silos. Condensation on the walls or on the product itself will increase the water activity in that region and, for this reason, increase the risk of degradation. When the interstitial air is moved from a heated area to another and colder zone, as part of the natural convection circuit, it gives up its moisture to the product. These transfers of moisture are frequently the cause of mould and caking at the top of the pile of bran when it is stored in bulk.

## Temperature

Temperature is just as important as moisture content for the storage of bran. It has an effect on all types of chemical reactions. The rate of 2 chemical reaction is proportional not to the total number of molecules present but to the number that has the activation energy needed for the reaction: this activation energy depends on the temperature. The Arrhenius equation  $k = A \exp(-\Delta E/RT)$  relates three important factors: the rate of reaction (through the rate constant k), the activation energy E and the absolute temperature T (R is the universal gas constant). The equation indicates that the relation between the k and the T is of the exponential type, which emphasizes the effect of the T. The logarithmic form of the equation,

$$\log k = \frac{-E}{2.303 R} \times \frac{I}{T} + \log A$$

indicates that the logarithm of k is inversely proportional to T. The Arrhenius equation thus requires that the graph of log k against I/T should be a straight line. Reactions in food products generally meet this requirement over an intermediate range of temperatures, although they show deviations at more extreme values. When log k is plotted against I/T, the slope of the line equals -E/2.303 R, and from this the activation energy E can be readily calculated.

To express the effect of temperature on the rate of enzymatic reactions it is usual to employ the term  $Q_{10}$ , which is defined as the ratio of the rate of reaction at temperature T + 10 to the rate of reaction at temperature T, and this has a value close to 2, which indicates that the rate of reaction approximately doubles for every rise in temperature of 10 K, at least over intermediate temperature ranges, where the reaction time does not produce any significant degradation.

In many rice-growing regions, the ambient temperature reaches relatively high values, with maximum temperatures of over 40° C in the shade, within the intermediate temperature range. At these levels the changes in the rate of reaction brought about by the changes in temperature that can occur in practice are already considerable. Thus, temperature plays a decisive role in the chemical, enzymatic and biological changes in bran, which will be described later.

#### Other factors

Although water and temperature activity are the most important factors in the changes that take place in the bran, together with the time available for the reactions to occur, others, such as the composition of the interstitial atmosphere and particle size, are also worth noting.

The composition of the interstitia! atmosphere is important in respect of the availability of oxygen. Oxygen takes part in some enzymatic reactions: in purely chemical oxidation it determines the aerobic or anaerobic nature of the metabolism of the micro-organisms, and has a marked influence on the survival of insects. The concentration of  $O_2$ , combined with the proportion of  $CO_2$  or  $N_2$  in the atmosphere, provides a means of combating insects.

Particle size has not been studied in relation to the reactions that characterize changes in the rice bran. It nevertheless plays an important role. Processing breaks down the natural barriers that protect the lipids from lipase in the caryopsis. The organized cellular structure of the external layers of the grain is destroyed, and friction of the particles causes the constituents thus freed to be brought into intimate contact. The surface area of the bran that is exposed to oxygen in the interstitial atmosphere favours oxidative reactions, which are only inhibited by natural anti-oxidants, the tocopherols, which are lost. In this context, the variations in particle size to be expected in industrial practice do not seem to alter the nature of the changes but only their rate: this may help to explain the considerable differences in the values published for certain types of change in bran.

Thermal conductivity is also a factor to be considered. In bran it is very low and encourages the accumulation of heat (so facilitating the formation of thermal foci where changes take place). This lack of conductivity means that fluctuation, in outside temperature would not be very likely to reach the interior of bran stored in bulk, although bran is never stored in bulk on the same scale as rice.

## Causes of changes in the bran

Changes in the bran can be of chemical, enzymatic or biological origin, the latter being mainly in the form of micro-organisms, insects and rodents.

## **Chemical reactions**

Under normal storage conditions purely chemical reactions appear to be of limited importance in the case of raw bran. Their importance may be more marked in bran from parboiled rice; this is particularly true of oxidative changes in the lipids in very dry media, since the presence of water provides some degree of protection. Maillard reactions generally require high temperatures or prolonged storage time. In the latter case, they may occur at temperatures above 20° C with  $a_w$  between 0.5 and 0.75, but at higher  $a_w$  values they are less likely [1].

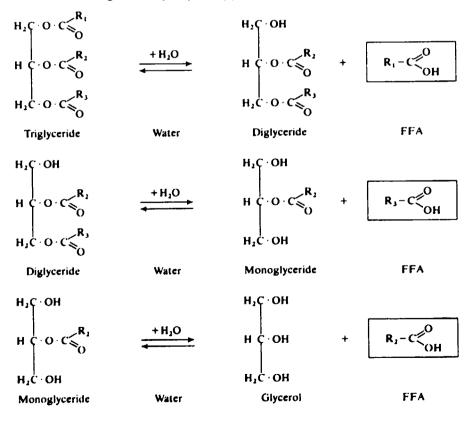
## Enzymatic reactions

As has been shown earlier, bran contains a large number of active enzymes, of which the lipases are the most important from the point of view of storage. The second most important enzymes, although they only have a secondary role, are the oxyreductases—lipoxygenase and peroxidase. The reactions catalysed by the lipases are of a hydrolytic nature (see figure 5).

Free fatty acids are liberated as a result of the enzyme action. Lipolysis takes place only at the interface between water and oil.

Water, in addition to acting as a substrate, also acts as a solvent medium tor diffusion of the enzyme, the reactants and the reaction products. It is not surprising, then, that most enzyme reactions only occur above the threshold at which solvent water appears, and as a function of the available

## Figure 5. Hydrolysis of glycerides catalysed by lipase



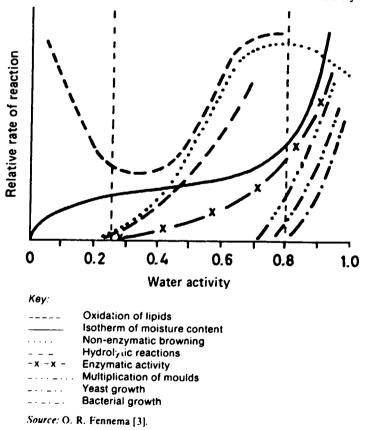
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quantity of solvent water. Thus, in general, most enzymatic activities start to become apparent at  $a_w = 0.75$ -0.88. Lipase, however, acts at much lower levels  $(a_w \approx 0.20)$  (see figure 6). The explanation is that the substrate (oil), being a liquid, facilitates renewal of the contacts between enzymes and substrate. It should be noted that any increase in the surface area of the fat will bring about an increase in the rate of lipolysis.

The action of heat on lipolysis, as on other enzymatic reactions, is twofold: at the kinetic level it accelerates the reaction, whilst at the protein structure level it inactivates the enzyme. Below the optimum temperature for rice lipases,<sup>2</sup> any increase in temperature accelerates the rate of reaction. Above the optimum temperature, the denaturing effect is predominant, and the subsequent rise in temperature causes a decrease in the reaction rate.

Lipolysis in rice bran is catalysed by lipases of two distinct origins: vegetable and microbial. The rice grain has its own lipolytic activity. It may, however, also contain lipases produced by micro-organisms. There are a number of such micro-organisms, and many are frequently found contaminating rice. Although some of them produce intra-cellular lipases, most produce extra-cellular lipases. Moulds, which are the most important sources, include *Penicillium, Aspergillus, Rhizopus* and *Mucor. Xanthomonas* are among





<sup>2</sup>Aizono and others [4] found that the optimum temperature for rice lipase, at pH 7.5, was  $37^{\circ}$  C. In subsequent investigations [5] they isolated a lipase II with an optimum temperature of  $27^{\circ}$  C at the same pH.

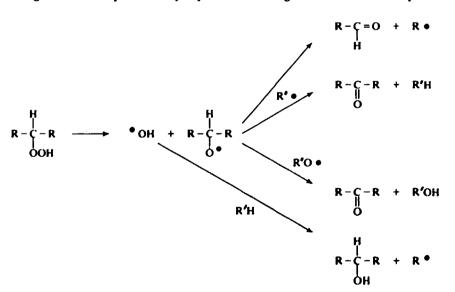
the most prominent lipase-producing bacteria. The microbial lipases are true lipases, since they are not inhibited by di-isopropyl fluorophosphate. Like the natural lipases, those of microbial origin hydrolyse various natural oils and fats, and this characteristic is used in analysing them. Not all substrates are hydrolysed at the same rate and to the same extent, however, and not all lipases have the same effect. The lipase from *Rhizopus oligosporus* and from *Mucor sufu* catalyse hydrolysis of the ester link in positions 1 and 3, while that from *Aspergillus flavus* also catalyses the link in position 2.

Lipoxygenase (lipoxidase) catalyses the oxidation, by molecular oxygen, of polyunsaturated fatty acids which contain a *cis*, *cis*-1,4-pentadiene group. The acids of this type which are found in bran are linoleic (9,12-octadecadienoic acid), linolenic (9,12,15-octadecatrienoic acid), and arachidic (5,8,11,14-eicotetraenoic acid). Substrates for lipoxidase catalysis are in the form of either FFA or glycerides, although the reaction rate is faster in the former. The reaction results in the formation of an organic peroxide; one of the double bonds jumps to a conjugate position and the compound takes the form of a *cis-trans*-isomer. Hydroperoxides are the main products of the initial reaction with oxygen. They decompose rapidly to form radicals, which contribute to the chain oxidation reaction or form other products such as aldehydes, alcohols, ketones and acids (see figure 7).

The peroxidase catalyses the reaction ROOH +  $AH_2 - H_2O + ROH + A$ . It catalyses the peroxidative degradation of unsaturated fatty acids, giving rise to the formation of volatile carbonyl compounds, with their own odour, which contribute to the odour of the oxidized product. With some H donors, such as dihydroxyfumaric acid and molecular oxygen, peroxidase may hydroxylate the amino-acids tyrosine and phenylalanine.

Both lipoxygenase and peroxidase have a destructive effect on a variety of food components, probably through the radicals that are generated by the decomposition of the hydroperoxides.

#### Figure 7. Decomposition of hydroperoxides resulting from the oxidation of lipids



Source: O. R. Fennema [3].

It is not easy to measure the state of oxidation of the oil. The peroxide index provides some indication, provided there is not hydroperoxide decomposition. The content of carbonyl compounds can also be employed, always provided that there have been no secondary reactions or vaporization. The tiobarbituric acid (TBA) index is not a good indicator in low moisture systems such as rice [6, 7].

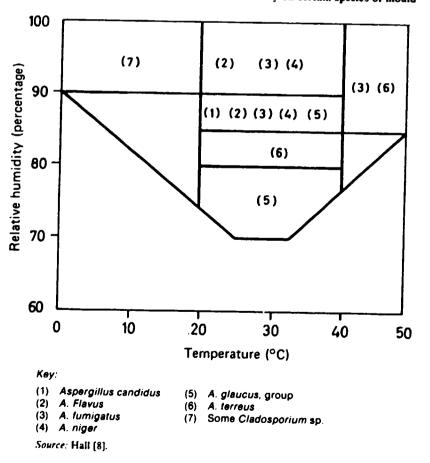
#### **Biological** changes

Micro-organisms, insects and rodents are among the more important sources of biological change.

#### Micro-organisms

Bran, because of its origins and the method by which it is produced, usually has a rich and varied microbial population. Nevertheless, only the moulds constitute a real risk of deterioration, when the bran is stored and handled in the usual way. Above  $a_w = 0.70-0.75$ , moulds multiply rapidly, particularly as the temperature rises (within the range 20°-40° C). Some mould spores may proliferate at  $a_w \approx 0.62$  [1]. Figure 8 shows the usual limits for

Figure 8. Effect of temperature and relative humidity on certain species of mould

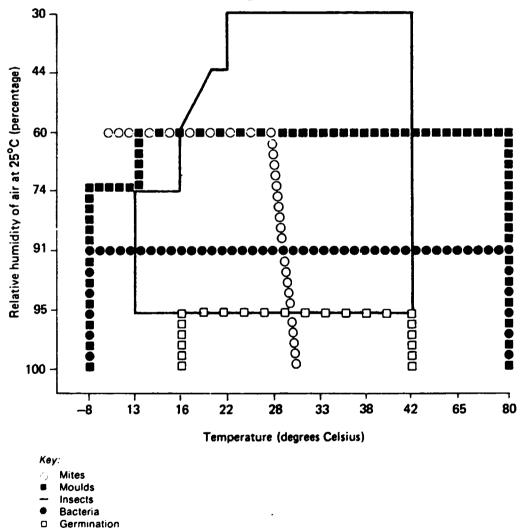


relative humidity and temperature within which some of the principal species of moulds found in bran multiply [8]. Figure 9 shows the physical limits within which some biological species grow. A knowledge of the conditions governing the growth of moulds makes it possible to predict the moisture levels at which the risk of deterioration disappears under prevailing temperature conditions.

Mesophilic bacteria require a minimum  $a_w$  value of 0.90-0.95 for multiplication; yeasts require 0.88 and bacteria and osmophilic yeasts 0.70-0.75 and 0.62-0.65 respectively [1].

The composition of the microflora undergoes changes during storage, the pattern depending on the ambient conditions. Average levels of water activity encourage the multiplication of storage moulds (*Penicillium* and *Aspergillus*), which persist at low  $a_w$  values at intermediate temperatures. In modified atmospheres (vacuum, CO<sub>2</sub>) the patterns are different and the moulds decrease. Bacteria also decrease, but some may grow at high humidity levels [1].

Figure 9. General limits of temperature and relative humidity for the multiplication of biological agencies



Source: Hall [8].

One important consequence of the proliferation of micro-organisms in bran (or in the rice at an earlier stage) is the production of enzymes, which have been discussed earlier.

Some typical mould flora are capable of producing substances that are toxic to both man and domestic animals (see table 1). Under optimal conditions, some micro-organisms, such as *Aspergillus flavus*, may produce toxins within 24 hours. It is important to note that the mere presence of a species that will produce toxic metabolites in bran does not mean that the bran itself will be toxic. First, not all varieties have the same capacity to produce toxic substanc<sup>5</sup>s and, secondly, the toxicity will depend upon the degree to which the micro-organism has developed in the product. A large number of mycotoxins are known which produce different pathological effects (see table 2). The sensitivity of animals to mycotoxins varies widely from one to the other: trout and duck, for example, are very sensitive. In tests carrried out with doses from 0.5 to 4 g of aflatoxin B-1, the degree of sensitivity of the subjects,

TABLE I.	MOULD FLORA CAPABLE OF PRODUCING TOXIC
	METABOLITES IN BRAN

Species	Rice husk <sup>a</sup>	Rice produced by Spanish mills <sup>b</sup>
Aspergillus chevalieri	+	
A. candidus <sup>c</sup>	+++	+
A. nidulans <sup>c</sup>	+	
A. flavus-orizae <sup>c</sup>	+++	+++
A. fumigatus <sup>c</sup>	+	
A. niger	+++	+++
A. ochraceous	+	+
A. glaucus	+	+
A. wentii	+	+
A. terreus <sup>c</sup>	+	+
Penicillium citreo-viride <sup>c</sup>	+	
P. notatum <sup>c</sup>	+	
P. islandicum <sup>c</sup>	+	
P. urticae <sup>c</sup>	+	
P. expansum		+
P. italicum	+++	+++
P. digitatum		+
Rhizopus nigricans	• +	+++
Rhizopus niger		+
Fusarium sp. <sup>c</sup>	+	+
Alternaria sp. <sup>c</sup>	+	+++
Mucor sp.	+	+
Absidia sp.	+	+
Streptomyces sp.	+	+
Sacharomyces sp.		+
Cladosporium sp.	+	+
Tricholecium roseum		+

Key: +++ Frequently found

+ Less frequently found

<sup>a</sup>Boller and Schroeder [10], Iizuka [11] and Kurata and others [12].

<sup>b</sup>Hernandez and others [13, 14].

<sup>c</sup>Main species producing toxic metabolites.

Toxin	Micro-organism	Selected pathological effects
Aflatoxins	Aspergillus flavus	Can cause carcinoma of the liver and kidney proliferation of the bile duct, infiltration of liver fat in animals
Aspergillic acid	A. flavus	Antimicrobial and toxic to mice
Cojic acid	A. flavus	Antimicrobial and toxic to mammals
Betanitropropanoic acid	A. flavus	Toxic to man and animals
Cubratocin	A. ochraceus	Can cause liver and kidney diseases in rats
Luteoskyrin	Penicillium islandicum	Can cause liver toxicity, haematoma
Chloridopherous peptide	P. islandicum	Hepatotoxin, can cause hepatoma in animals
Islandotoxin	P. islandicum	
Citreo-viridin	P. citreo-viride	Paralysis in mammals
Patulin	P. expansum	Antimicrobial, phytotoxic; rat carcinogen
Fusarenone	Fusarium nivale	Inhibition of protein synthesis in mice
Nivalenol and		
Desoxinivalenol	F. nivale	Inhibition of DNA synthesis

#### TABLE 2. SOME MYCOTOXINS PRODUCED BY MOULDS AND THEIR PATHOLOGICAL EFFECTS

Source: Food and Agriculture Organization of the United Nations [15].

in descending order, was as follows: ducks, turkeys, geese, pheasants and chickens [9]. The climate of the rice-growing regions lends itself to the development of moulds, and the presence of mycotoxins in the bran must be foreseen. Very precise analytical techniques are now available, which allow the presence of mycotoxins to be detected at levels of one part per billion. In view of the impossibility of completely avoiding contamination by mycotoxins, tolerance limits in foods and feedstuffs have been established for some of these substances.

Protection of the bran from contamination by moulds and mycotoxins must start with the rice itself. Contamination may be prevented most efficiently by: (a) harvesting the rice when it is fully ripened; (b) avoiding damage from machines when the rice is harvested; (c) drying the rice immediately after harvesting; (d) ensuring that the grain does not get wet again; and (e) avoiding infestation by insects, which deposit and transmit moulds and their spores and form pockets of high humidity where moulds are bound to grow [15]. The drying of parboiled rice in the sun, if done under unsatisfactory conditions, may give rise to serious infestation. Essentially the same control methods are used to prevent the bran from becoming mouldy; in other words, the product to be stored must be in a stable state, it must be stored under suitable temperature and humidity conditions that reduce the risk of contamination by insects or animal pests to a minimum, and it must be examined frequently to ensure that it remains in good condition and, if necessary, chemical means must be used to control pests.

In practice it is not always possible to protect all consignments, so that some may become contaminated by aflatoxins or other mycotoxins. Although much effort has been devoted to developing methods for decontaminating grain and grain products, there is no efficient and reliable process that can be applied to bran. The aflatoxins, being the commonest form of mould, have received the most attention, and it has been found that they can be only partially inactivated by heat (boiling at atmospheric pressure or roasting). Another alternative is to mix the mouldy and contaminated product with other ingredients, thus reducing the toxin concentration to tolerable levels; this method entails some risk.

#### Insects

Most of the insects found in both rice and bran have a four-stage growth cycle: egg, larva, pupa and adult. Each species has its own characteristic periods of development. But the ambient conditions (basically humidity and temperature) determine to a great extent how long it takes to pass from one stage to the next, provided that food is available. The weevil (Sitophilus oryzae L.) can complete the whole of its growth cycle in four weeks if conditions are favourable. The confused flour beetle (Tribolium confusum J. du Val) at 32° C takes the same amount of time, but at 22° C the growth cycle slows down and takes some three months [16]. At temperatures below 15° C, egg-laying, incubation and development of the larvae are a slow and difficult process. The rates of development and reproduction increase as the temperature rises, but temperatures above 35° C are unfavourable. Insects reproduce more readily as the humidity increases, but only up to the point at which micro-organisms begin to intervene. The humidity requirements naturally differ from one species to the next. The weevil will not reproduce at moisture contents below 9 per cent. Because of the powdery consistency of bran, which makes its nutrients readily useable, insect life is tolerated at lower levels than it is in whole grain.

Mention must be made of the cockroach (*Blattella germanica* L., *Blatta orientalis* L. and *Periplaneta americana* L.) since experience has shown that *Blattella germanica*, at least, is attracted to rice bran. Confirmed invasions by this insect have been detected in bran stabilization plants. Cockroaches carry dangerous micro-organisms and transmit others through their faeces, including species of *Salmonella* [8].

#### Changes in the composition and properties of bran during storage

Changes that take place in the bran during storage are one of the most widely studied yet at the present time least known facets of the science and technology of this by-product. The principal use of bran is as a source of oil for food, but it has one essential disadvantage, namely the rapid development of FFA, which starts from the moment the bran is produced in the mill. For this reason, much of the very large amount of work carried out on the effects of storage has been limited to a study of the development of the FFA. Unfortunately, this is not the only disadvantage. In practically none of the cases studied have the factors and causes controlling the formation of FFA been taken into account. Similarly, many data are hard to interpret, let alone compare. The information available therefore provides a general picture of the actual situation, but is actually of limited value.

#### Colour, taste and odour

No systematic studies seem to have been carried out on the effects of storage on the colour, taste and odour of bran. Data on a storage experiment on raw bran packed in 5-kg cloth sacks stored at very moderate moisture and temperature conditions (see table 3) indicate no major changes in colour over a period of four months. A musty odour and taste become noticeable after two months. The most marked changes, however, consist of the appearance of bitter substances and of a somewhat disagreeable taste, which sticks in the throat; these changes take place very rapidly, and may appear in a few days if the temperature is high. The formation of bitter substances appears to be either directly or indirectly associated with enzymes. Scalding with steam inhibits the development of a bitter taste in the rice germ [17], and stabilization with moist heat has a similar effect on the bran [19]. The formation of bitter components in oats is accelerated by the presence of FFA, which are more susceptible to oxidation than the triglycerides; peroxides appear to act as precursors of the bitter substances [20]. In this sense, oxidases may play an important role but, as suggested by various authors, other enzymatic agents, such as proteases, must be taken into account in discussing the reasons for the presence of bitter substances in bran and degraded germ. Saponins, which are also present in rice

	Chai	nge during storag	e
Characteristic	0 months	2 months	4 months
Colour			
Trichromic components			
(Hunter values):			
L	63.8	63.7	64.5
а	0.2	0.3	1.0
ь	14.3	13.8	13.9
Visual estimation:			
Whiteness	3.6	3.2	3.8
Odour			
Stale	8.6	7.3	7.3
Mouldy	9.0	9.0	9.0
Taste			
Stale	9.0	7.0	6.7
Bitter	8.4	4.0	2.7
Catches the throat	8.5	5.3	4.7
Humidity (percentage)	13.8	10.8	11.1

TABLE	EFFECTS					
	CHARACTE	RIST	ICS OF RAW	RICE	BRAN	

Source: Tortosa and Benedito de Barber [18].

Note: The bran was commercial bran degerminated in an industrial mill; degree of milling about 9 per cent. It was stored in 5-kg cloth sacks at room temperature with temperature variations from 15° to 25° C and relative humidity 40 per cent to 90 per cent. Scale for visual colour estimation: 9 = white; 7 = creamy white; 5 = cream; 3 = light brown; 1 = dark brown. Scale for estimation of odour and taste; 9 = none; 7 = just perceptible; 5 = clearly perceptible; 3 = moderately strong; 1 = strong.

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bran, do not appear to play an important role; although they are water-soluble, their extraction does not eliminate the bitter taste [21].

The effects of storage on the volatile constituents of the aroma of rice bran have been studied in stored samples<sup>3</sup> at two different temperatures,  $-20^{\circ}$  C and  $+25^{\circ}$  C [22].

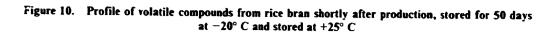
The profile of the volatile constituents of the bran was determined by direct gas chromatography,<sup>4</sup> without prior enrichment of the volatile constituents, employing techniques that require a sample weighing at least 1 gram. Eight constituents were identified in each of the samples: methanol, acetaldehyde, ethanol, acetone, *n*-pentanal, *n*-hexanal, 1-hexanol and *n*-dodecane; of these, acetone, *n*-hexanal and *n*-dodecane were found in large quantities (see figure 10). It is interesting to note that storage of the bran at  $-20^{\circ}$  C for 50 days results in changes in some of the constituents identified, such as ethanol, acetone and 1-hexanol, as well as in others with a higher molecular weight. Storage at  $+25^{\circ}$  C markedly increases the volatile content, particularly hexanal and hexanol and many other contituents with a higher molecular weight that were not identified in the study.

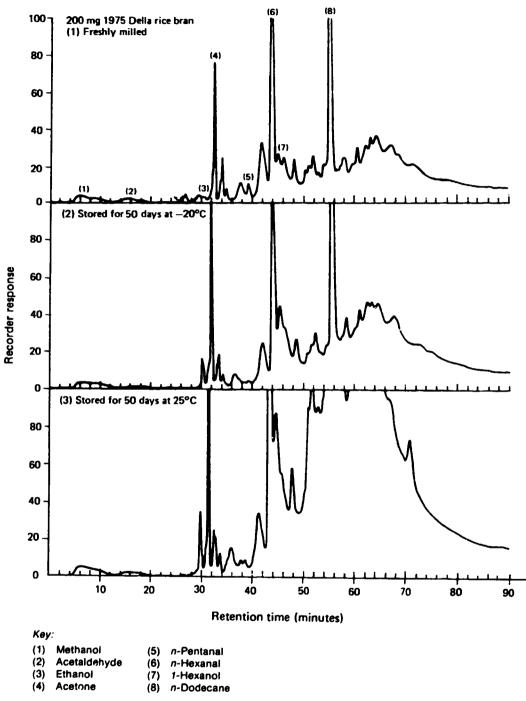
#### Average chemical composition

The average chemical composition of the bran does not undergo any significant changes when the bran is stored for short periods under normal conditions, provided that it has not been infested by micro-organisms and insects. In bran samples stored at 27°-28° C for one month, in brown glass jars with tightly fitting screw caps, no change was found in the total nitrogen content, fat, ash or raw fibre [23]. Identical results were obtained when samples of raw and parboiled rice bran, packed in hermetically sealed glass bottles, were stored at ambient temperature (not specified) and over a period of months; nevertheless, after 10 months there was found to be a slight reduction in the percentage of oil that could be extracted with hexane [24]. It should be noted, however, that significant changes have been recorded in the quantity of oil that can be extracted with hexane (see figure 11), with petroleum ether [25] or under pressure [26] (see table 4). The latter results relate to storage in sacks under really severe temperature conditions, which nevertheless correspond to conditions encountered in practice. Various hypotheses have been advanced to explain why there is a reduction in the quantity of extractable oil [24]. One such hypothesis attributes this reduction to the formation of polar compounds containing oxygen and polymers, which decrease the solubility of the glycerides in the non-polar solvent (hexane) used for extraction. Others associate it with the appearance of forces that combine glycerides and bran, probably related to the fact that the fatty acids have the ability to form complexes with amylose. Although data are not available for bran, data for prepared rice indicate that the content of fat-by-hydrolysis remains constant through the storage period [27].

<sup>&</sup>lt;sup>3</sup>The study does not include any other data on the storage conditions or initial characteristics of the samples.

<sup>&</sup>lt;sup>4</sup>Some of the constituents were identified by combining gas chromatography and infra-red spectroscopy of the materials.





Source: Legendre and others [22].

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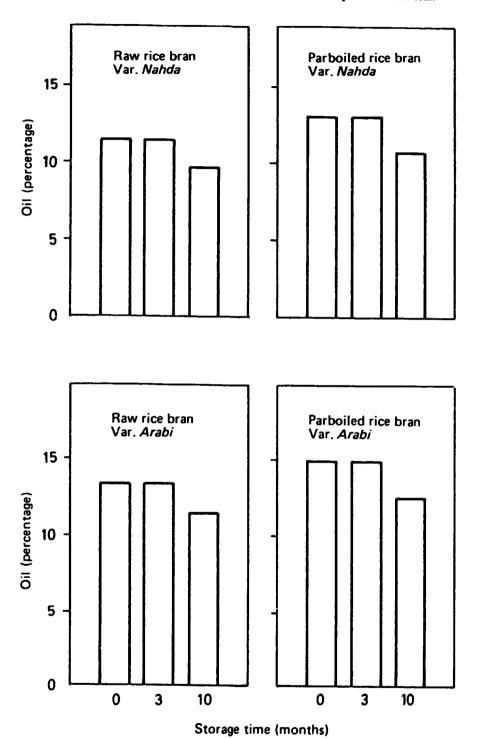


Figure 11. Effects of storage on the oil content of raw and parboiled rice bran

Source: Shaheen, El-Dash and El-Shirbeeny [24].

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Note: The oil content of the bran was determined as follows: 20 g of bran was mixed with 500 ml of hexane, at a temperature of  $40^{\circ}$ -50° C, and agitated for three hours. The hexane was separated, further hexane added and the process repeated. The oil recovered from the two extracts was determined after removing the last traces of hexane by drying the oil at a temperature of 70° C under vacuum.

#### TABLE 4. EFFECTS OF STORAGE<sup>a</sup> ON YIELD AND COLOUR OF THE OIL EXTRACTED FROM THE BRAN BY PRESSING

	Storage co	onditions				
Storage time (days)	Average temperature <sup>b</sup> (degrees Celsius)	Relative humidity <sup>b</sup> (percentage)	Moisture content (percentage)	Oil yield <sup>c</sup> (percentage)	Colour of oil	
0	28.7	74	11.86	11.43	Yellow	
7	28.2	85	9.62	10.40	Yellow	
21	36.3	63	10.32	9.63	Dark yellow	
42	36.3	63	9.23	8.97	Dark yellow	

Source: K. Yokochi [26].

<sup>a</sup>The bran was packed in sacks with a 33.75-kg capacity.

<sup>b</sup>Measured at 11 a.m.

<sup>C</sup>Extracted in presses in two stages, at a pressure of 1,000 psi (70 bar) for 15 min and a pressure of 3,800 psi (260 bar) for 5 min.

# Composition and characteristics of the chemical constituents of the bran

#### Carbohydrates

The limited amount of information available on the sugar content of bran indicates that in some varieties the percentage of reducing sugars may increase and that of non-reducing sugars decrease (see table 5). Similar changes occur in rice during storage [28].

## TABLE 5. SUGAR AND MOISTURE CONTENT OF RICE BRAN BEFORE AND AFTER STORAGE

#### (Percentage)

	Non-reduc	ing sugars	Reducin	g sugars	Moisture		
Variety of rice	Before	After	Before	After	Before	After	
Diaja	50.54	47.99	1.07	1.30	10.85	10.90	
Inapon	55.79	56.89	0.82	0.94	13.41	13.89	
Intan	51.28	52.01	0.84	0.79	13.68	14.17	
Makapiña	50.02	51.90	0.54	0.63	11.13	13.13	
Malagkit	46.35	47.61	1.07	1.30	13.16	14.15	
Margate	60.10	60.40	0.96	1.18	!0.26	11.21	
Milagrosa	53.24	54.70	0.94	1.16	12.72	13.43	

Source: Cea and Sutaria [23].

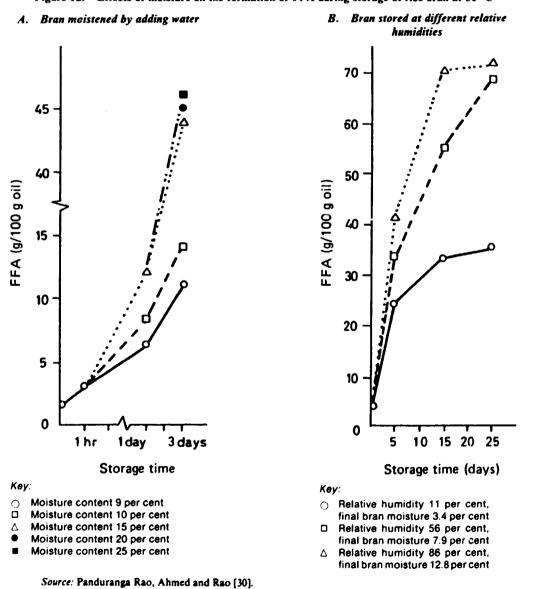
Note: The samples were packed in brown glass bottles with tightly sealed screw-caps and stored at 27°-28° C for one month.

#### Lipids

During storage, the bran lipids undergo hydrolytic and oxidative changes [29]. Hydrolytic change normally occurs in bran produced and stored under normal conditions. The FFA content of the bran oil increases as a function of time, and this increase can be detected in as little as one or two hours. The rate

at which FFA are formed depends, amongst other things, on moisture, temperature and the presence of microflora and insects. The FFA increases more rapidly with a higher moisture content (see figure 12A). Moisture content is important, and differences of 1 or 2 percentage points may account for very varied increases in FFA after only a few hours of storage (see table 6). The relative humidity of the environment in which the bran is stored also plays a role in determining the level of water activity. The increase in the FFA grows with the relative humidity (see figure 12B). Here the effects depend upon the speed with which the bran takes water from the surrounding atmosphere or loses it to the atmosphere. And it should also be pointed out that, on an industrial scale, a consignment of bran provides areas of rapid interchange (on the surface, where it is in contact with the air) and others of very slow

Figure 12. Effects of moisture on the formation of FFA during storage of rice bran at 30° C



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Sample	Moisture (percentage)	FFA four hours from receipt of the bran <sup>a</sup> (g per 100 g oil)
1	5.9	4.5
2	6.3	8.4
3	8.2	11.8

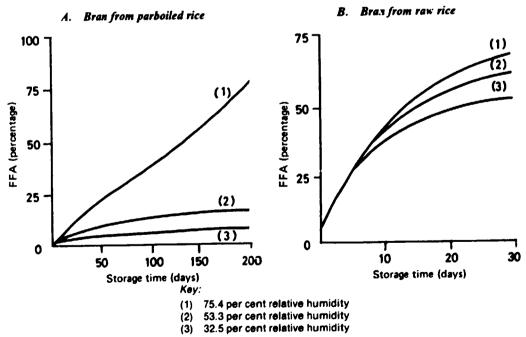
ABLE 6.	EFFECT OF MOISTURE ON THE RATE OF FORMA-
TION OF	FFA DURING STORAGE OF RICE BRAN AT 35° C

Source: Meinke, Holland and Harris [31].

<sup>a</sup>Zero time was some four hours after manufacture in the mill.

interchange (on the inside), resulting in a heterogeneous mass with foci that are more or less favourable to hydrolytic action. The effect of the relative humidity also varies according to the type of bran involved (see figure 13), not only because of the rapidity of moisture exchange but also because of the level of lipolytic activity. The formation of FFA in the stored bran depends to a great extent on the temperature. The higher it is, the greater the increase in FFA (see figure 14). The formation of FFA takes place at temperatures as low as  $3^{\circ}-5^{\circ}$  C [32, 33]. Under these conditions, according to laboratory data on small samples, the increase in FFA in the first few days varies from between 0.2 to 1.5 per cent a day [31, 33]. Increases in the FFA content have also been recorded at temperatures below 0° C [34]. Raw bran with a 15.9 per cent oil content and 11.7 per cent moisture content increased from 2.4 to 8.4 and 10.5 g of FFA per 100 g of oil in 10 and 20 days respectively at  $-3.3^{\circ}$  C.

Figure 13. Effects of storage at 25° C and at different levels of relative humidity on the formation of FFA in rice bran



Source: Loeb, Morris and Dollear [32].

Note: Humidity reached equilibrium in approximately five days.

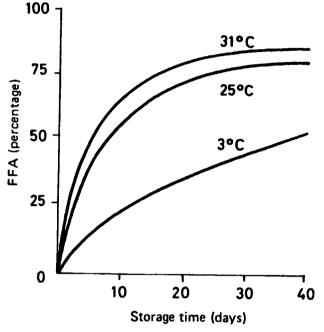


Figure 14. Effects of storage at different temperatures on the formation of FFA in rice bran

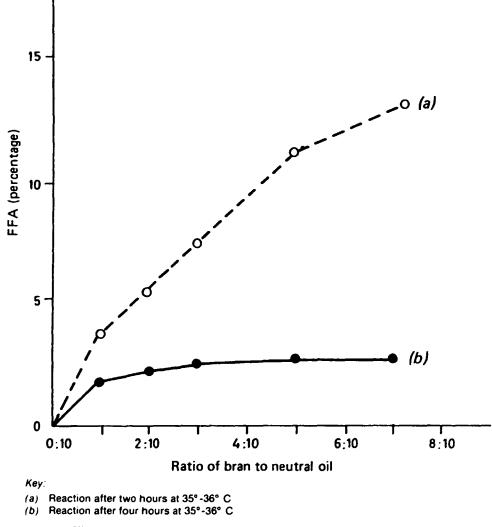
Source: Loeb, Morris and Dollear [32].

There is also some evidence that the size of the bran particle affects the rate of formation of FFA, since the increase in FFA is greatest when the particles are smallest [30, 33]. A smaller particle generally suggests greater cellular disintegration, and hence a greater opportunity for contact between enzyme and substrate. The subject has not, however, been studied systematically to provide data that are unaffected by other factors, such as composition (associated with particle size) or humidity.

El Hinnawy [35] studied the formation of FFA in neutral rice-bran oil, which was induced by adding various quantities of raw bran to the oil; he noted that the greater the proportion of bran, the more rapid the increase in FFA (see figure 15). As the amount of enzyme that acts on the substrate is increased, the result of its lipolytic action becomes more pronounced. In an industrial consignment of bran, the amount depends on the actual opportunity for contact between enzyme and substrate. At the same time, it also depends on the concentration of enzyme in the bran and on its specific activity. Even though both may vary from one consignment to another, depending on the variety of the bran, the degree of processing it has been subjected to, the presence of microflora etc., the formation of FFA as a function of the initial activity of different consignments of bran has still not been evaluated.

Some data, however, suggest that the increase in FFA (expressed as grams of oleic acid per 100 g of oil) is greater in bran from the first cone than in bran from the second cone (see figure 16). The greater reactivity of the layers of bran nearest the outside has been pointed out, not only in relation to the change in lipids but also to the change in other contituents [36].

Loeb and Mayne [37] have studied the influence of moisture content on the proliferation of micro-organisms and the formation of fatty acids during

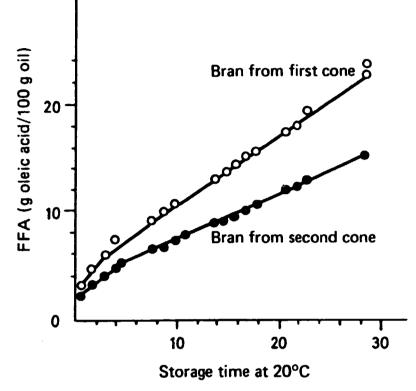


#### Figure 15. Formation of FFA in neutral rice-bran oil after the addition of bran

Source: El Hinnawy [35].

storage, and have shown that the microbial population of the bran contributes to the increase of the fatty acids. In one experiment, they treated bran at  $121^{\circ}$  C in an autoclave for two hours, dried it for one hour at 60° C, allowed it to cool overnight to ambient temperature and, using sterile water, prepared various samples with different moisture contents, which they stored in different desiccators. The samples with 10.7 per cent and 13.7 per cent moisture content showed no growth in micro-organisms and the FFA content did not change. The sample with a 14.6 per cent moisture content showed neither proliferation of micro-organisms nor increase in FFA during the first 13 days, but after this period, colonies of *Aspergillus glaucus* developed and the FFA began to rise simultaneously; no bacterial growth was detected. In one sample with a 26.4 per cent moisture content, moulds (*Rhizopus* sp.) appeared on the sixth day, coinciding with an increase in FFA (see table 7). The suggestion of a relationship between micro-organisms and the production of FFA was





Source: Instituto de Investigaciones Tecnológicas (38). <sup>a</sup>Using 6-kg samples in sealed metal containers.

TABLE 7. CHANGES IN MOULD AND MICROBIAL COUNT AND FFA CONTENT
DURING THE STORAGE OF BRAN TREATED FOR TWO HOURS IN AN AUTOCLAVE
AT 121° C AND DRIED FOR ONE HOUR AT 60° C

	Moisture 13.7 pei		Moisture 14.6 per		Moisture 26.4 pei		Bacteria (10*/g)
Storage time (days)	FFA (perceniage)	Moulds <sup>d</sup> and yeasis (10 <sup>3</sup> /g)	FFA (perceniage)	Moulds <sup>d</sup> and yeasis (10 <sup>y</sup> /g)	FFA (perceniage)	Moulds <sup>d</sup> and yeasts (10 <sup>3</sup> /g)	
0	2.3		2.3		2.3		
6	2.3	0e	2.2	0e	12.6	0.15	0e
13	2.6	0e	2.8	0e	18.5	I	0.02
20	2.6	0¢	3.1	0.1	25.3	10	0.5
27	2.6	0e	18.1	155	79.4	100	
34	2.7	0¢	38.2	1 100			

Source: Loeb and Mayne [37].

<sup>a</sup>Relative humidity 75.5 per cent.

<sup>b</sup>Relative humidity 80.3 per cent.

CRelative humidity 93 per cent.

 $d_{\rm Not}$  infected with bacteria.

Lowest dilution 1:20.

confirmed in a second experiment described below. The bran was treated at 121° C for three hours, after which it was dried for three hours at 85° C and finally allowed to cool. Various samples with different moisture contents were then prepared, using sterile water, and a series of these was inoculated with  $53 \times 10^3$  spores per gram, with a Thom and Churuch strain of Aspergillus chevalieri (Mangin), previously isolated from bran of the Bluebonnet variety.<sup>5</sup>

The samples were stored in desiccators and the changes in FFA and mould count were studied over a period of 20 days. The samples that were not inoculated showed no growth in micro-organisms and no increase in FFA. The inoculated samples showed marked changes affecting the micro-organisms and the FFA simultaneously (see table 8).

It should be noted that in regular untreated bran, in which the normal lipolytic activity of the grain coexists with that of the microflora, the FFA content may increase even without the development of micro-organisms. On the other hand, the greatest proliferation of micro-organisms may not correspond to the greatest increase in FFA. Thus, it is known that the formation of FFA is greater in species of *Rhizopus* and *Aspergillus* than in *Penicillium* or *Bacillus* [39]. Even different strains of *Aspergillus* have been found to have different capacities for producing FFA (see table 9).

Pillaiyar [40] has published data suggesting a parallel between the proliferation of insects and the increase of FFA in bran during storage (see figure 17). No systematic study appears, however, to have been made of the contribution of insects to the formation of FFA. It is not yet clear whether the reasons for the parallel between FFA and the presence of insects are: (a) that conditions favouring higher lipase activity also favour the proliferation of insects; (b) that the growth of insects, with the resultant increase in moisture and heat, directly favours the activity of lipases or promotes the development of microflora; or (c) that insects have the capacity to produce acidic metabolites or lipolytic enzymes.

As a result of the group and stereochemical specificity of lipase and of the composition of the glycerides in terms of fatty acids, the different fatty acids are not liberated by lipolysis in the same proportions. Canale, Sarra and Caramello [42] noted marked differences in myristic, palmitic, stearic, oleic, linoleic and linolenic acids (14:0, 16:0, 18:0, 18:1, 18:2 and 18:3 respectively) during the storage of rice from the first and second whitener cones in the dark at  $22^{\circ} \pm 2^{\circ}$  C (see figure 18). The rate of formation decreased in the following order: 18:2, 18:1, 16:0, 18:3, 14:0 and 18:0. The pattern of the changes was the same in the two brans, although there were quantitative differences. The relative proportion of the different acids varied, with a marked increase in the proportions of 18:2, 18:1 and 16:0 (see table 10). During the storage of milled rice, the most important changes in the FFA fraction also occur in the 18:2, 18:1 and 16:0 [27] in exact proportion to the corresponding losses in the neutral fats fraction.

The rice germ lipids are hydrolysed during storage, as are those of bran. The rate of hydrolysis is much less, however, in the former than in the latter<sup>6</sup> (see figure 19). The greater stability of the germ is known and appreciated at ſ

<sup>&</sup>lt;sup>5</sup>Although the optimum temperature for growth was 30° C, the spores grew well at 21.1° C.

<sup>&</sup>lt;sup>6</sup>Other researchers have furnished data on the increase in FFA during storage of the germ, which is much smaller than the increase normally found in bran [43, 44].

	Treated bra	n with 11.5 p	er cent moisti	ure content <sup>a</sup>	Treated bra	Treated bran with 14.5 per cent moisture content <sup>b</sup>				Treated bran with 33.5 per cent moisture content <sup>c</sup>			
	Non-inoculated		Inoculated with A. chevalieri		Non-inoculated		Inoculated with A. chevalieri		Non-inoculated		Inoculated with A. chevalieri		
Storage time (days)	FFA (percen- iage)	Moulds and yeasts (10 <sup>3</sup> /g)	FFA (percen- iage)	Moulds and yeasis (10 <sup>3</sup> /g)	FFA (percen- lage)	Moulds and yeasis (10 <sup>3</sup> /g)	FFA (percen- lage)	Moulds and yeasts (10 <sup>3</sup> /g)	FFA (percen- lage)	Moulds and yeasts (10 <sup>3</sup> /g)	FFA (percen- lage)	Mouids and yeasis (10 <sup>3</sup> /g)	
0	2.3	0	-			—	_	_	_	_	_	_	
1	2.6	0	2.9	1	2.9	0	2.9	12	2.6	0	3.2	4	
2	_	0	2.7	62	_		2.5	8	_	_	2.9	2	
3	2.4	0	2.9	8	2.8	0	3.4	285	2.8	ð	3.3	5 200	
6	2.7	0	3.4	8	2,4	0	15.2	4 250	2.7	0	7.0	5 900	
13	2.7	0	4.9	295	2.8	0	40.3	560	2.8	0	32.6	3 900	
20	2.4	0	25.8	4 600			59.3	3 800	_	_	_		

 TABLE 8. CHANGES IN THE MOULD COUNT AND FFA CONTENT OF NON-INOCULATED STERILIZED BRAN AND BRAN INOCULATED WITH ASPERGILLUS CHEVALIERI DURING STORAGE AT DIFFERENT LEVELS OF MOISTURE CONTENT

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Source: Loeb and Mayne [37].

Note: Lowest dilution 1:20.

<sup>a</sup>Relative humidity 68 per cent.

<sup>b</sup>Relative humidity 80.3 per cent.

<sup>c</sup>Relative humidity 100 per cent.

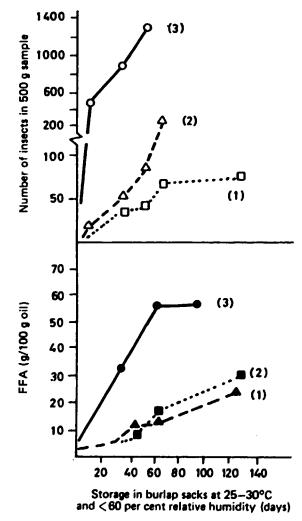
	FFA after incubation at 28° C for				
Aspergillus strain	Four days	Five days	Six days		
Sp <sub>3</sub>	2.17	3.10	23.25		
Sp, Sp,	3.10	4.34	23.71		
Sp <sub>11</sub>	3.87	5.42	29.45		
Sp11 Sp10	14.10	17.51	23.25		

TABLE 9.	FFA-PRODUCING CAPACITY OF VARIOUS STRAINS
OI	F ASPERGILLUS ISOLATED FROM RICE BRAN

(mg oleic acid/100 g culture medium)

Source: Chattopadhyay and Srimani [41].

#### Figure 17. Changes in FFA content and insect infestation of bran during storage



Key:

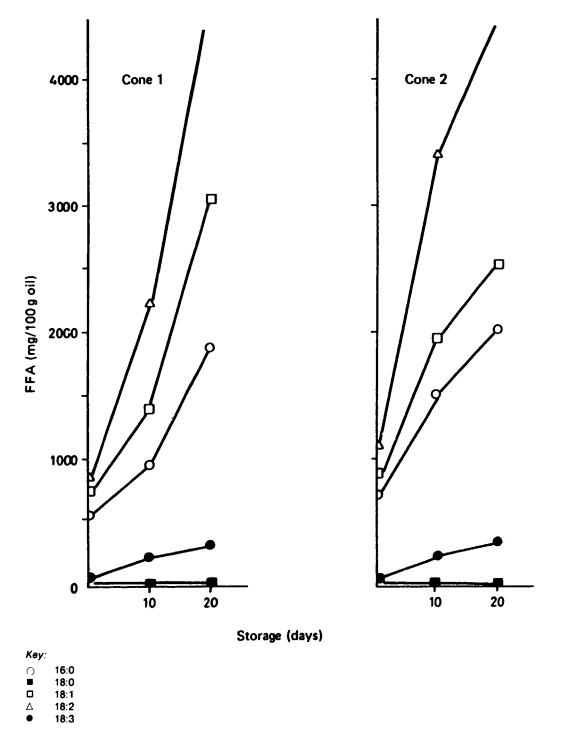
Parboiled rice bran from huller mill (7.0 per cent oil)
 Parboiled rice bran from rubber-roll huller and huller mill as whitener (17.2 per cent oil)

Raw rice bran (19.0 per cent oil). (3)

Source: Pillaiyar [40].

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Figure 18. Development of FFA in bran from the first and second whitener cones during storage

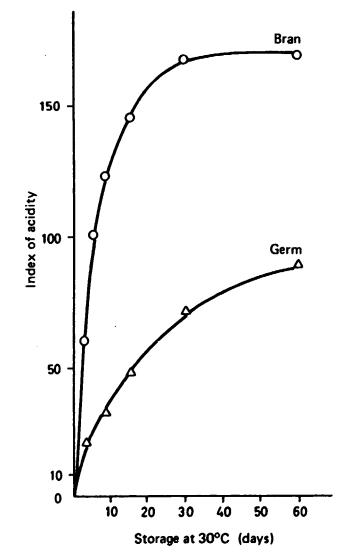


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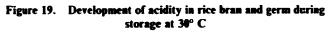
UT OT							
Source of bran	Length of storage (days)	14:0	16:0	18:0	18:1	18:2	18:3
First whitener	0	1	50	1	70	75	5
	20	1	75	1	120	185	12
Second whitener	0	I	45	1	50	65	5
	20	1	80	1	100	175	15

TABLE 10. CHANGE IN RELATIVE PROPORTIONS OF FFA IN BRAN DURING STORAGE

Source: Canale, Sarra and Caramello [42].



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Source: Gómez Fabra and Primo Yufero [45].

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the commercial level, particularly in countries where the separation and sale of bran on an industrial scale is common practice. Although detailed studies have not been carried out, there is evidence that the stability of the germ depends to a large extent on cellular deterioration during processing. Intact germ can be stored much better than damaged germ. The greater degree of stability has been attributed to the fact that the lipase is still encapsulated in the cellular cytoplasm and cannot act freely on the fats present [46]. As a consequence, differences in stability are to be expected in the germ produced from different varieties in different whiteners and under different conditions.

De Rege [46] has indicated that he has in no case detected FFA levels above 90 per cent, even in samples of bran where hydrolysis has been brought about experimentally in the laboratory. In general, he suggests a hydrolysis limit of around 85 per cent for bran and around 65 per cent for uncrushed germ. Probably the pH of the medium is too low to allow the enzyme reaction to proceed. This fact is of interest in relation to the theoretical study of lipolysis [46, 47, 48]. Some authors have detected a decrease in FFA content in bran lipids in an advanced stage of hydrolytic change.

Bran or rice with FFA-free oil does not seem to have been obtained on an industrial scale or in the laboratory. Yokochi [26] has indicated that the FFA content in bran immediately after manufacture is normally 2-4 g per 100 g of oil, when it is produced from the current year's rice harvest, and about 10 g/100 g of oil, or higher, if the bran is obtained from a crop two years old. These values are, of course, no more than indicative, and may be noticeably affected by the usual events that take place during the storage of hulled rice: heating-up, development of microflora etc. With regard to germ (from raw rice) of industrial origin, values from 1 to 2 g per 100 g of oleic acid are normal [49].<sup>7</sup>

On the other hand, Sarda [50] has reported that in India (West Bengal), using modern systems for parboiling rice, it has proved possible to manufacture bran on an industrial scale with 2-4 per cent FFA, which can be stored for a fortnight in winter without any marked changes.

Oxidative deterioration of bran has received little study despite the fact that it causes undesirable foreign (e.g. stale) odours and tastes, and leads to the formation of oxyacids, which must be separated during refining and thus contribute to overall losses in the yield of edible oil. Storage of the bran causes an increase in the peroxide index of the oil, which rises to a maximum and then decreases. The content of carbonyl compounds increases at first, and decreases thereafter if the changes continue. The iodine number decreases from the start. Similar changes seem to occur in rice [36]. The bran from raw rice and parboiled rice show, in principle, two entirely different situations from the point of view of oxidative changes. Bran from raw rice is characterized by the presence of lipoxydases, which catalyse oxidation but are accompanied by a number of natural antioxidants (tocopherols) and largely globular oil substrate. Parboiled bran loses its lipoxidase activity during heat treatment of the rice but, at the same time, loses its antioxidant capacity, in proportion to the severity of the treatment, as a result of the thermal degradation of the tocopherols; the fat

<sup>&</sup>lt;sup>7</sup>The lowest value recorded in the 1970 campaign, in which consignments from various mills were studied from November 1970 to June 1971, was 0.73 g per 100 g of oil.

in the parboiled rice bran, to a greater or lesser extent, loses its globular form and is found to be dispersed, so presenting a greater surface for oxidation.

In storage studies carried out with bran from raw and parboiled rice,<sup>8</sup> packed in polythene sacks, not hermetically sealed, in diffuse light at ambient temperature, it has been found that the peroxide index in the raw bran increases more quickly and reaches higher values than in parboiled bran. The calculations were made at 12-hour intervals over a 120-hour period [51]. These results seem to conflict with the known fact that parboiled rice is more susceptible to oxidative change than raw rice.

In a recent paper, Sowbhagya and Bhattacharya [52] have confirmed that raw rice changes less and more slowly (lower values for the peroxide index and for the carbonyl index) than parboiled rice when both are stored under comparable conditions, whether in the light or in the dark. It must be pointed out, however, that when both types of rice are triturated to a sieve size below 80 mesh and then stored in the dark at 60° C in open containers, the autoxidation rate in raw rice increases very markedly, while in a sample of parboiled rice it barely increases at all. As a consequence, both flours show very similar behaviour, both qualitatively and quantitatively.

Apparently the relationship between the carbonyl index and the detection of the stale odour in bran has not been determined. In parboiled rice, the stale odour is detected at around 100-200 M per gram of fat, while in raw rice it is already noticeable at 30 M per gram of fat.

One fortunately sporadic phenomenon that occurs during the storage of bran is spontaneous ignition and combustion. This also occurs in some other oil-bearing raw materials. It appears to be associated with the formation of unsaturated fatty-acid oxidation products. High temperatures, humid atmospheres, fine bran powder and incomplete removal of solvent from defatted bran are considered to be favourable conditions for ignition. Although the problem does not often arise, the dangers it entails in storage and transport should not be underestimated, since they may have serious consequences. Despite the danger, no detailed knowledge of the phenomenon is available, and its true causes do not appear to have been evaluated. This prevents the establishment of preventive rules other than the general one of avoiding the conditions cited above.

#### **Mycotoxins**

While the presence of mycotoxins in rice has been the subject of considerable research, relatively little work has been done on mycotoxins in bran, either at the time of production in the mill or during prolonged storage, despite the fact that, as demonstrated by Schroeder, Boller and Hein [53], between 60 and 80 per cent (by weight) of the toxins in milled rice pass into the bran and polish during processing. These fractions may contain toxins in concentrations more than 10 times higher than in milled rice. The chance or intentional incorporation of husk in the bran will aggravate the situation even further.

A small-scale sample from Spanish mills in 1971 gave negative results in all cases (Barber and others [54]). The samples varied in moisture content from

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<sup>&</sup>lt;sup>a</sup>Brown rice, after having been soaked in water overnight, is cooked in water for one hour and then dried in the sun.

7 to 16 per cent and in FFA content from practically zero to 23 per cent (see table 11). Repeated evidence of the presence of mycotoxins in rice in many countries, coupled with the fact that, in any consignment, mycotoxins may grow rapidly and in great quantities if the conditions are favourable to the growth of micro-organisms, would seem to warrant making an adequate study in some areas, to allow the existing risks to be evaluated.

Sample	Moisture (percentage)	FFA <sup>a</sup> (percensage oleic acid)	Aflatoxin B1	
Bran				
1	11.3	15.1	Negative	
2	11.8	14.0	Negative	
3	9.2	3.5	Negative	
4	12.7	12.5	Negative	
5	14.6	1.6	Negative	
6	10.7	12.0	Negative	
7	9.5	11.3	Negative	
8	10.4	11.5	Negative	
9	9.1	9.2	Negative	
10	8.7	10.0	Negative	
11	10.0	7.7	Negative	
12	10.2	8.3	Negative	
13	15.9	23.4	Negative	
Husk				
1	10.7	0.1	Negative	
2	6.7	0.7	Negative	

TABLE	11.	RESULTS	OF	SAMPLING	CARRIED	OUT	IN
SPANIS	H RIO	CE MILLS T	O D	ETECT THE	POSSIBLE P	RESEN	ICE
	(	OF AFLATO	XIN:	S IN BRAN AI	ND HUSK		

Source: Barber and others [54].

<sup>a</sup>Additional data supplied by J. Botey.

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