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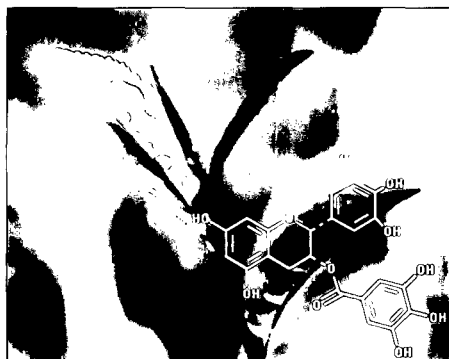
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Study to Promote Industrial Exploitation of Green Tea Polyphenols

Final Report

(UNIDO Project No. TF/GLO/00/105)

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University Institute of
Pharmaceutical Sciences
Panjab University
Chandigarh-India

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KARAN VASISHT
(Project Leader)

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1.0 INTRODUCTION

The man during the history of human civilization has selected three important non-alcoholic beverages namely tea, coffee and cocoa from nature's resources. Among these, tea is the most widely consumed beverage in the world. It is consumed by half of the world population for its attractive aroma, taste and health benefits. It is a safe and easily affordable drink to all sections of the society throughout the world and there is enough evidence that consumption of tea is one of the most important ways to prevent a number of human ailments. It has evoked a great interest in the medical community in the past few decades. Scientific research has validated the useful health effects of tea (especially green tea) and it has shifted its reputation from being the 'cup that cheers' to the 'cup that heals'.

Technically, tea is made from top three leaves, leaf bud and two tender leaves of the tea plant *Camellia sinensis* (L.) O. Kuntze (syn. *Thea sinensis*) family Theaceae (Figure 1-3).

Tea plant is native to Southeastern Asia and was believed to have been discovered accidentally by Chinese emperor Shen Nung around 2737 B.C. Since then, its popularity grew steadily to become China's most favoured drink and its fame spread to other parts of the world. In 1610, tea was taken by the Dutch traders to Europe. In 1835, the East India Company established experimental tea plantations in Assam state of India. The first consignment of eight chests of Indian tea was auctioned in London market in 1839, beginning the advent of Indian tea in the world market. Its cultivation then spread to Sri Lanka in 1857. Encouraged by the worldwide wave, Russia started tea cultivation with plantations in the Caucasus region in 1905. Tea cultivation in Indonesia started in 1910 and soon thereafter tea was grown in Kenya (1925) and other parts of Africa.¹

Types of Commercial Tea

Tea is commercially available in three types: green, black and lesser-known oolong tea. These types vary in their method of manufacture and thereby in their chemical content. Black tea is widely used in India and green tea is popular in China, Japan and Taiwan. Oolong tea is mainly produced in China and Taiwan. Small quantities of green and oolong tea are also produced in India and Sri Lanka. On world basis, 80 per cent of total production is of black tea, 20 per cent of green tea and only 2 per cent of oolong tea.

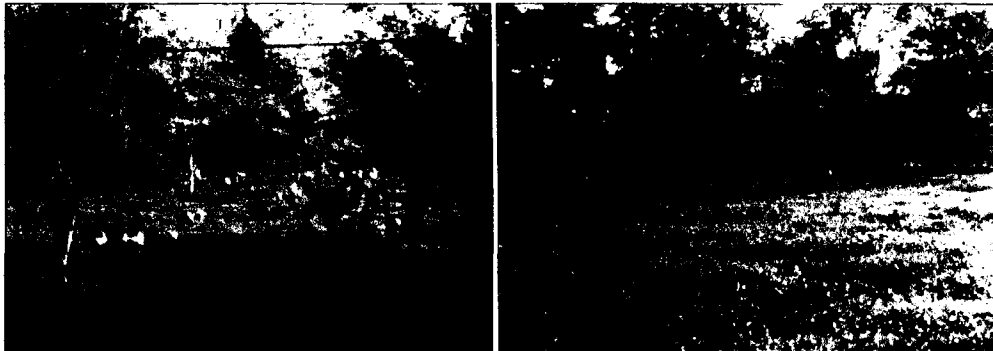


Fig. 1: Field view of tea plantation



Fig. 2: Growing tip of tea plant

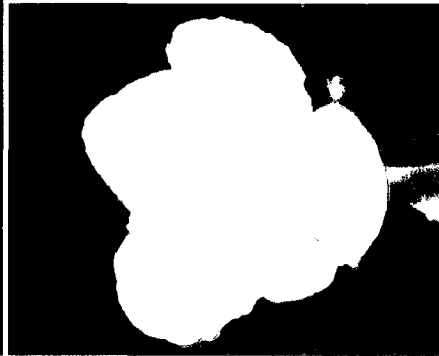


Fig. 3: Flower of a tea plant

Green Tea

It is unfermented type and is the least processed among the three types of tea. In the manufacture of green tea, the freshly harvested leaves are immediately

steamed to inactivate the enzymes, especially polyphenol oxidase (PPO), to prevent oxidation and polymerization of primary polyphenols. In the cells, primary polyphenols are located in the vacuoles and are kept separated from the enzymes that are present in the chloroplast. If the leaf is allowed to ferment, the enzymes and polyphenols come in contact with each other resulting in oxidation and polymerization of polyphenols to form complex compounds.² The enzyme deactivation during manufacturing process prevents polymerization of primary polyphenols, which pass as such into green tea. The green tea infusion has a leafy taste, a smell of fresh vegetables, low caffeine content and no calories.³

Black Tea

It is the most popular among the three varieties of tea. It is a completely fermented type of tea prepared by keeping the tea leaves for fermentation after plucking. This process results in oxidation and polymerization of polyphenols, changing the nature of chemical constituents of tea leaves and forming two groups of colouring polyphenols; the yellow group (theaflavins) and the red group (thearubigins).³ These polyphenols are responsible for briskness, strength, colour, taste, aroma and pungency associated with black tea. The infusion of black tea has a bright red or copper colour, astringent taste and characteristic aroma.²

Oolong Tea

It is a semi-fermented tea where primary polyphenols are partly allowed to oxidize. Oolong tea is not common and is intermediate in characteristics between green and black tea. Immediately after plucking, the tea leaves are partially fermented for about half the time of black tea. It has a colour of black tea and flavour of green tea.

Cultivated Varieties of Tea

There are three cultivated varieties of tea namely, (i) Cambodian, (ii) Chinese (iii) Indian.³ The different tea varieties hybridize freely. Varieties and their important characters are given in Table 1.

Table 1: Common cultivated varieties of tea

Variety name	Taxonomic name	Important Characteristic
Cambodian	<i>Camellia sinensis</i> var. <i>assamica</i> subsp. <i>lasiocalyx</i>	Grows to about 5 m in height. It is used for hybridization with other varieties.
Chinese	<i>C. sinensis</i> var. <i>sinensis</i>	Grows up to 3 m. It is a small leaved, hardy plant and is able to withstand cold winters.
Indian	<i>C. sinensis</i> var. <i>assamica</i>	Grows as high as 3 to 18 m. Cultivated in India and mainly grown in Assam, Darjeeling and Nilgiris.

Grading of Black Tea

The most distinguished tea comes from the year's first bud while twigs and other leaves down the stem tend to yield the products of poor quality. The best tea is hand-harvested.⁴ The freshly gathered shoots are collected and subjected to withering, rolling, fermentation and drying to produce the tea. In the final sorting or grading, tea acquires the colourful names that are used in the tea trade. This nomenclature does not refer to the quality but to the size and appearance of the tea.

There are two major grades of tea; *leaf* (unbroken-leaf) and *broken-leaf*. The leaf grades have larger and intact leaves and fetch a higher price. Broken-leaf tea is next in quality to leaf tea and gives a stronger tea than leaf grades.⁵ Within the broken types there are further grades, including fanning (small leaf tea), and dust (smallest leaf particles). The grades used in tea trade in descending order of quality are given in Table 2.⁶

Table 2: Tea grades in descending order of quality

i) Orange Pekoe (OP)	v) Finest Tippy Golden Flowery Orange Pekoe (FTGFOP)
ii) Flowery Orange Pekoe (FOP)	vi) Supreme Finest Tippy Golden Flowery Orange Pekoe (SFTGFOP)
iii) Golden Flowery Orange Pekoe (GFOP)	vii) Broken Orange Pekoe (BOP)
iv) Tippy Golden Flowery Orange Pekoe (TGFOP)	viii) Broken Orange Pekoe Fanning (BOPF)

2.0 CHEMICAL CONSTITUENTS OF TEA

The constituents of green tea leaves include carbohydrates, proteins, polyphenols, caffeine, theanine, vitamins and minerals. Commercially and biologically, polyphenols and caffeine are more important.

Polyphenolic Constituents

Green tea contains 30-42 per cent polyphenols on dry weight basis.⁴ It contains predominantly flavanols, flavandiols and phenolic acids, such as gallic, coumaric or caffeic acid. The polyphenols are the derivatives of gallic acid and catechin.⁷ Catechin is synthesized in tea leaves through mixed pathways of malonic and shikimic acid. Gallic acid is derived through shikimic acid pathway. The primary polyphenols of green tea are devoid of tanning properties and lack colour in tea infusions. They are bitter in taste and are unique, as most of them are not found elsewhere in plants.⁸

Primary polyphenols are catechin derivatives and were first reported by Roberts and Woods in 1951 using paper chromatography.⁹ The natural polyphenols in tea include (-)-epigallocatechin-3-gallate (EGCG), (-)-epigallocatechin (EGC), (-)-epicatechin-3-gallate (ECG), and epicatechin (EC). The highest concentration is of EGCG followed by EGC, ECG and EC in the decreasing order.¹⁰⁻¹³ Other catechins including (+)-gallocatechin (GC), (-)-gallocatechingallate (GCG), (-)-catechin gallate (CG) and (+)-catechin (C) are present in minor quantities.² A cup of green tea contains about 300 to 400 mg of polyphenols.³

The primary polyphenols oxidize during fermentation process of black tea and transform to compounds having tanning properties. The oxidized polyphenols are present up to 6 percent. The black tea polyphenols are divided into two groups, thearubigin and theaflavin (20 and 2 to 6 per cent respectively on dry weight basis). Thearubigins have higher molecular weight and are chemically poorly characterized. They are partly polymeric proanthocyanidins, and impart colour to the tea infusions.^{3, 14} Theaflavins in black tea are theaflavin 3-*O*-gallate, theaflavin

3'-*O*-gallate and theaflavin 3,3'-di-*O*-gallate.³ They contribute to the briskness of the tea infusion. A good tea must contain a balanced quantity of these compounds.^{4, 15}

A large number of dissimilar chemical reactions initiated by enzymes during fermentation are of practical use in commercial manufacturing of black tea. The chain of reactions in the process of tea fermentation are shown in Figure 4.¹⁵

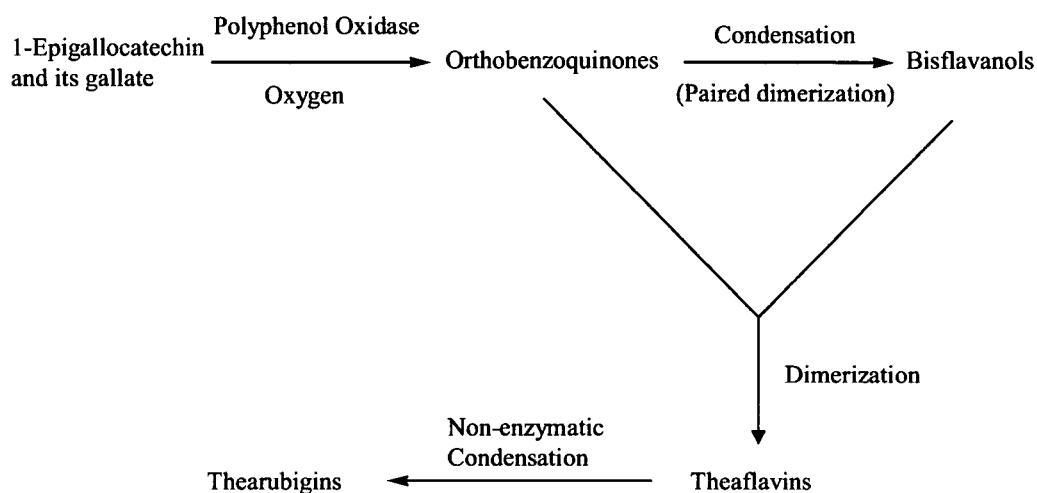


Fig. 4: Reactions involved in the process of tea fermentation

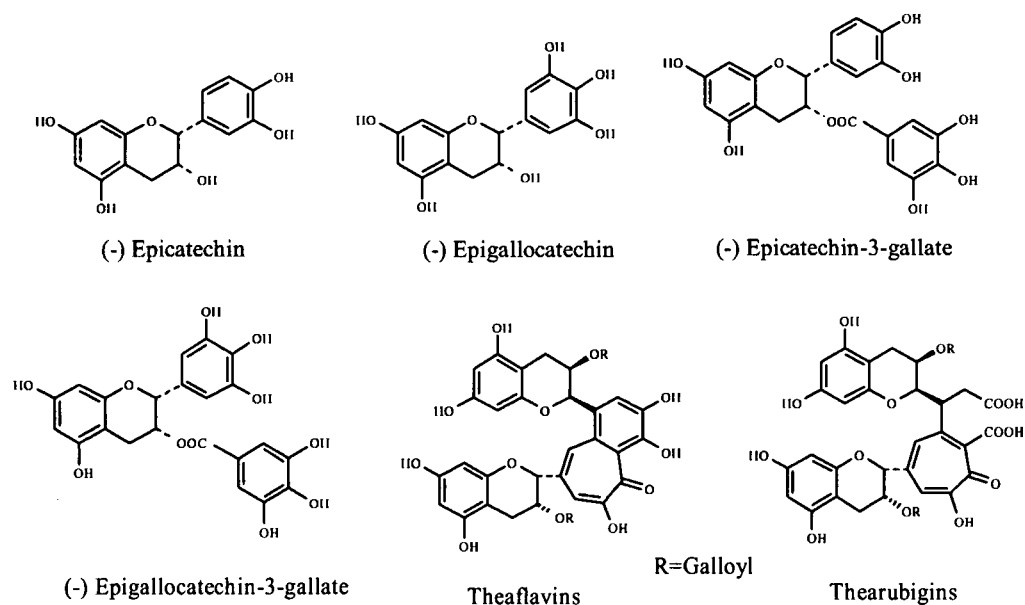
The polyphenol oxidase of tea leaf works best at 28.3 °C and the reaction slows down with increase or decrease in temperature. Oxidation takes place more quickly in fresh leaf than in withered leaf.¹⁶ The action of the oxidase converts 1-epigallocatechin and its gallate to corresponding orthoquinones, after which the enzyme action ceases. The changes that follow are non-enzymatic and the rate of reaction increases steadily with the rise in temperature. The four monomers of fresh tea leaves are epigallocatechin (EGC), epicatechin (EC) and their gallate esters namely epigallocatechingallate (EGCG) and epicatechingallate (ECG).^{4, 16}

Non-Polyphenolic Constituents

Caffeine

Runge first observed occurrence of caffeine in coffee in 1820.⁸ Nakabayashi isolated a similar compound from tea and named it theine which was later identified

to same compound as caffeine.¹⁰ Caffeine content of tea leaves varies from 2 to 5 per cent. Commercially, caffeine is prepared from tea leaves and tea wastes.^{8, 17}



Amino Acid and Other Nitrogenous Compounds

The content of total nitrogen in green tea extract ranges from 4.5 to 6.0 per cent, and about half of it comes from free amino acids.¹⁸ Theanine and glutamic acid are the major amino acids whereas aspartic acid and arginine are the minor amino acids present in the tea.^{19, 20} Theanine is a unique amino acid, produced by tea plant and certain other species of *Camellia*. The rate of metabolism of theanine in tea leaves is slow but its transport from root to leaf is rapid leading to its accumulation in leaves. It is also one of the major components that gives specific taste to green tea and is antagonistic in action to caffeine.²¹

Vitamins

Green tea contains about 0.3 per cent of vitamin C, which is decomposed during manufacture of black and oolong tea. As a result, the vitamin C content of black and oolong tea are very low.²²

Inorganic Elements

Some specific inorganic elements present in the tea are aluminium, fluorine and manganese.^{10, 22} The level of aluminium and fluorine in the tea leaves is relatively higher than in other plants. It is presumed that the tea plant has biochemical mechanism to neutralize the toxicity of aluminium. It has been observed that aluminium in tea leaf exists mainly in a chelated form, indicating that catechins prevent the aluminium toxicity. These findings are important in physiological significance of tea polyphenols.²³

Carbohydrates and Lipids

The carbohydrate content of green tea is about 40 per cent and one third of it is cellulose. Starch is also present which affects the quality of green tea. Tea harvested in the morning has less starch and is better in quality in comparison to afternoon collections. Tea leaves contain about 4.0 per cent of oil.²⁴

3.0 GREEN TEA POLYPHENOLS – EXTRACTION AND ESTIMATION

Extraction Methods

Polyphenols are extracted with water at boiling or near boiling temperature. Mixture of acetonitrile and water (1 : 1) has been used to extract tea polyphenols at room temperature. Different workers have used different ratio of tea leaves to solvent (1 : 40 to 1 : 500) and varying periods of extraction (3 to 60 min). The extract is filtered preferably through 0.45 μ M membrane filter before subjecting to quantitative analysis in HPLC.²⁵⁻³²

Estimation of Tea Polyphenols

Lot of work has been done and reviewed on quantitative estimation of green tea polyphenols.^{33, 34} Tea polyphenols have been estimated by various methods, such as nuclear magnetic resonance,³⁵⁻³⁷ near-infrared reflectance spectroscopy,³⁸ spectrophotometry,³⁹ column liquid chromatography,³⁰ thin layer chromatography

& high performance thin layer chromatography (HPTLC),⁴⁰ liquid chromatography coupled with mass spectroscopy (LC-MS),²⁶ high performance capillary electrophoresis (HPCE)³¹ and high performance liquid chromatography (HPLC).^{41, 42} The HPLC is a common and most frequently used technique for estimating tea polyphenols. Many detection methods have been employed including a post-column reaction with 4-methylaminocinnamaldehyde and detection at 640 nm,³⁹ electrochemical detector,^{43, 44} chemiluminiscent reaction⁴⁵ besides use of conventional UV-detectors.²⁹ In most cases reversed phase HPLC with UV-absorbance detection has been the method of choice.³³ The separation is achieved using a C18 column. End capped, deactivated, monomeric C18 columns are preferred over non-deactivated or polymeric columns. Dalluge *et al*²⁹ compared a variety of stationary phases and elution conditions. They observed that the stationary phases, with ultra pure silica and maximized coverage of the silica support, improved the separation. The presence of an acid in the mobile phase is essential for complete and efficient resolution of catechins, specifically for elimination of peak tailing and its detection at shorter wavelengths.^{26, 46}

Liquid chromatography coupled with mass spectrophotometry provides a reliable approach to the analysis of catechins in pico-molar quantities in the complex matrices. The first report of LC-MS for the identification of catechins from tea appeared in 1993.⁴⁷ This report demonstrated the separation of a mixture of (-)-epigallocatechin-3-gallate (EGCG), (-)-epigallocatechin (EGC), (-)-epicatechin-3-gallate (ECG), and epicatechin (EC), employing thermo-spray ionization mass spectroscopy. Direct MS characterization of catechins in tea extracts without the use of LC has also been demonstrated. Electro-spray ionization, electron impact ionization, and fast atom bombardment mass spectroscopy have been employed to provide both molecular mass and structural information for catechins.^{26, 48}

High Performance Capillary Electrophoresis (HPCE) is another technique that offers the advantage of short analysis time as compared to HPLC. Capillary zone electrophoresis (CZE) and micellar electro kinetic capillary chromatography (MEKC) with UV absorbance detection are the preferred capillary electrophoresis

methods for the determination of catechins. In most cases, uncoated fused-silica capillaries have been used to effect the separations. In general, the MEKC methods provide better separation, resolution and quantification than CZE methods.^{28, 31}

Gas chromatographic (GC) methods utilizing glass columns or fused-silica capillary columns employing flame ionization detector (FID) has been developed for the estimation of catechins. As a prerequisite, a derivatization step is needed to convert catechins to trimethylsilyl (TMS) derivatives. Techniques of GC-MS and capillary column GC-FID have also been used for the separation of catechins.^{49, 50}

Thin layer chromatography of tea has been used for qualitative and quantitative determination of individual catechins. Zhu and Xiao⁴⁰ used silica gel plates and chloroform : ethyl formate : n-butanol : formic acid as a mobile phase to separate catechins and caffeine in green tea samples. The separated components were visualized under UV and quantitative measurements were made based on the areas under the curve of the catechins spots. Four catechins EC, EGC, ECG and EGCG were separated on silica plate using chloroform: methanol: water. Detection of the catechins was done by colour formation with vanillin-hydrochloride acid reagent.

Singh *et al*³⁹ have developed a simple method for separating, identifying and quantifying individual catechins based on two-dimensional paper chromatography. The catechins are identified as bright yellow spots on the chromatographic paper by spraying the paper with diazotized sulfanilamide, a reagent specific and selective for catechins. The sensitivity of visual detection is less than 1 µg of catechins.

The spectrophotometric determinations of catechins are based on the formation of a green coloured complex with 4-dimethylaminocinnamaldehyde (DMACA), which is specific and selective for catechins.³⁹

Total catechins have also been measured using biosensor.⁵¹ Burdock tissue (*Arctium lappa* L., a biennial plant) contains polyphenol oxidase, which catalyses the oxidation of polyphenols. A catechin biosensor has been constructed that uses a slice of burdock tissue. As the catechins in a sample are oxidized, the oxygen electrode measures the amount of oxygen consumption. This biosensor was found

to respond to various catechins namely, EC, EGC, ECG, EGCG, gallic acid, catechol and ascorbic acid. The biosensor has been applied for determination of total catechins in green tea infusions, but it is inadequate for accurate quantification because of the variability of biosensor response to the different catechins.

The quantitative determination of three catechins based on their chemiluminescent emission has also been reported. Catechins are reacted with hydrogen peroxide-acetaldehyde, horseradish peroxidase, which results in a distinct chemiluminescent emission at 630 nm.⁴⁵

4.0 BIOLOGICAL SIGNIFICANCE OF GREEN TEA

Phenolic compounds are widely distributed in food of plant origin and are regarded as effective antioxidants. Several studies suggest that these components may be of importance in reducing the incidence of degenerative diseases like cancer and arteriosclerosis. The most relevant compounds in dietary regimens are cinnamic acid derivatives and flavonoids.

The health benefits of tea have long been recognized and continue to be legendary in China and Japan. Scientific reports during the last two decades have validated many of the beneficial claims of tea. The result of studies to understand the mechanisms of the biological effects have interested scientists all over the world. The majority of beneficial effects have been attributed to polyphenolic constituents. As natural polyphenols remain unchanged in green tea, it can be said that green tea is more beneficial in comparison to black tea, where fermentation during manufacture leads to the oxidation of primary polyphenols. Strong antioxidant potential of tea polyphenols is thought to mediate most of the beneficial effects of tea.⁵²⁻⁵⁵ The health benefits in cancer, arthritis, cardiovascular diseases, diabetes, obesity and dental caries have been focused for scientific investigations in the recent past.

Antioxidant Activity

The most potentially beneficial effects of tea catechins are attributed to their antioxidant properties that sequester metal ions and scavenge oxygen species and

free radicals.⁵⁴ The free radical scavenging property of catechins has been well studied particularly during the last decade. The early evidence of antioxidative property showed EGCG-induced inhibition of soybean lipoxygenase ($IC_{50} = 10$ to $20 \mu M$).⁵⁵ Later it was reported that EGCG inhibited TPA-induced oxidative DNA base modification in HeLa cells,⁵⁶ inhibited Cu^{2+} -mediated oxidation of low-density lipoprotein (LDL),⁵⁷ reduced *tert*-butyl hydroperoxide-induced lipid peroxidation,⁵⁸ and blocked the production of reactive oxygen species derived from NADPH,⁵⁹ cytochrome P-450 mediated oxidation of the cooked meat carcinogen and 1,2-amino-3-methylimidazo [4,5-f] quinoline.^{60, 61} Low concentration of EGCG inhibited Jurkat T-cell DNA damage caused by hydrogen peroxide or 3-morpholinopyridone (a peroxynitrite generator) and at high concentration EGCG itself induced cellular DNA damage.⁶² EGCG is the most abundant component present in tea extract and also the most potent chemical of the epicatechin derivatives tested for biological activity. Along with other tea catechins and polyphenols, it is thought to prevent tumorigenesis by protecting cellular components from oxidative damage by free radical scavenging. Indeed a number of studies have demonstrated the free radical scavenging activity,⁶³⁻⁶⁵ of EGCG as well as its antimutagenic,^{66, 67} antiangiogenic,^{68, 69} antiproliferating and/or pro-apoptotic effects on mammalian cells both *in vitro* and *in vivo*.⁷⁰

Tea catechins have been found to be better antioxidants than vitamin C, E, tocopherols and β -carotene.⁷¹ The antioxidant property of the purified extract was less pronounced than the crude extract suggesting that many constituents contribute to the activity.⁷² The polyphenols block free radical damage to lipids (found in cell membranes and serum lipids), nucleic acids and proteins (like those found as cellular enzymes and structural proteins). Damage to these cell components can lead to tumor formation. The oxidative damage by free oxygen radicals of low-density lipoproteins (LDL) in serum leads to arteriosclerosis and coronary heart diseases. The oxidation of cell membranes and other cell components leads to ageing.⁷³

Highest level of total catechins (72.9 mg / g of tea leaves) has been reported in samples of Chinese green tea and consequently showed maximal antioxidant

potential.⁷⁴ Oolong and black tea show weak protective action. The antioxidant activity of tea polyphenols is not only due to their ability to scavenge superoxides but also due to their ability to block xanthine oxidase and related transducers.⁷⁵ The polyphenols increased the activity of antioxidant enzymes e.g. glutathione peroxidase, glutathione reductase, glutathione-S-transferase, catalase and quinone reductase in small intestine, liver and lungs which are the detoxifying enzymes of the body.⁷⁶ The antioxidant activity of tea gets diminished by the addition of milk to the infusion due to binding of tea polyphenols to milk proteins.

Cardiovascular Activity

Several flavonoids and related phenolics have been reported to inhibit either enzymatic or non-enzymatic lipid peroxidation, an oxidative process implicated in several pathological conditions including atherosclerosis.⁷⁶ In particular, tea polyphenols have been suggested to lower the oxidation of low-density lipoproteins (LDL) cholesterol, with a consequent decreased risk of heart disease.⁷⁷ It is seen that green tea polyphenols significantly reduce the levels of serum LDL, very low-density lipoproteins (VLDL) and triglycerides.⁷⁸ At the same time they increase the levels of high-density lipoproteins (HDL).⁷⁹ A low ratio of triglycerides to HDL is an excellent marker for cardiovascular health.⁸⁰⁻⁸² In a cross-cultural correlation study of sixteen cohorts, known as the seven countries study, the average flavanol intake was inversely correlated with mortality rates of coronary heart disease after years of follow-up.^{83, 84} Adding support to the observation are the finding that in hypercholesterolemic rats green tea polyphenols lowered blood cholesterol levels and reduced blood pressure in spontaneously hypertensive animals.⁸⁵

The volumes of antioxidants in diet are inversely related to the risk of death from heart disease and of non-fatal heart attacks.⁸⁶ Green tea inhibits vascular smooth muscle proliferation, which is another essential factor contributing to the formation of arteriosclerotic plaque.⁸⁷ They also interfere with the absorption of dietary fat and cholesterol.⁸⁸

Green tea polyphenols have been found to play an important role in controlling essential hypertension by inhibiting angiotensin-I converting enzyme (ACE), which converts angiotensin-I to vasoconstrictive angiotensin- II.²

Anticancer Activity

Experimental evidence points to potential protection at several stages of carcinogenesis including cancer prevention,⁸⁹ endogenous carcinogen activation,⁹⁰ DNA damage and destabilization,⁹¹ cell proliferation,⁹² neoplastic growth and metastasis.^{77, 89, 90, 93-97}

Tea, especially green tea reduced the incidence of cancers of stomach,^{98, 99} small intestine,⁹⁸ pancreas,^{100, 101} lung,¹⁰² breast,¹⁰³ skin,¹⁰⁴ urinary bladder,¹⁰³ prostate,¹⁰⁵ oesophagus⁹⁸ and mouth.^{106, 107} Also, it has been shown to reduce tumor size and growth in cancer bearing animals.^{108, 109}

Green tea polyphenols directly inhibited the cytochrome P-450 enzyme systems (phase I enzyme) that play a pivotal role in carcinogen activation.⁶⁰ Concurrently they boost the activity of phase II enzymes (e.g. glutathione transferase) that make xenobiotics hydrophilic for clearance from the body. The process is crucial for carcinogen detoxification.^{93, 110}

Recently, it was proposed that the anticancer activity of EGCG is associated with the inhibition of urokinase, which is one of the most frequently expressed enzymes in human cancers.¹¹¹ However, the practicability of this study was later challenged due to required dose levels.¹¹⁰

Green tea polyphenols also inhibited the reactions that give rise to nitrosamines, both *in vitro* and *in vivo*.¹¹²⁻¹¹⁴ Pretreatment with tea polyphenols resulted in substantial reduction of carcinogen binding to DNA.¹¹⁵ The increased activity of glutathione peroxides and catalase in the intestine, liver and lungs of mice on pretreatment with tea polyphenols suggested that it may suppress mutagenesis mediated by peroxides in the microenvironment of DNA.⁹⁵

Tea polyphenols have also been shown to promote apoptosis of cancer cell lines such as prostate, lymphoma, colon and lung.⁹⁴ Modulating apoptosis is useful

in the management, therapy and prevention of cancer. Reduction and inexpression of tumour necrosis factor (TNF- α) may be the way polyphenols induce apoptosis. TNF- α is an endogenous tumour promoter and a central mediator in chronic inflammatory diseases like rheumatoid arthritis and multiple sclerosis.^{89, 108-110, 116, 117}

Green tea showed protective effect against damage produced by UV radiation and reactive oxygen species to the dermis through apoptosis and inhibiting lipid peroxidation.⁹⁴ Topical application of EGCG decreased UV-induced erythema, edema and hypersensitivity.¹¹⁰ Tea polyphenols also inhibited tumour promoters like okadaic acid and teleocidin in the skin.¹¹⁷

DNA-reactive genotoxic carcinogéns, which affect the DNA of normal cell, are responsible for majority of human cancers. Polyphenols have been shown to stimulate the repair process of nucleotide excision by removing DNA- adducts produced by carcinogens.⁹⁵ The structure of tea polyphenols possess strong nucleophilic centres, which react with electrophilic carcinogens to form an adduct, which results in the prevention of cancer.⁹⁶

Effects of polyphenols on nitric oxide (NO) induction have also been studied. EGCG inhibits the induction of NO synthase (NOS) via down regulation in the transcription nuclear factor, thus inhibiting induction of NOS. The NO is a bioactive molecule that plays an important role in inflammation and carcinogenesis.¹¹⁰

Green tea has been reported to be of great use in the modulation of cancer chemotherapy.^{89, 97, 118} It enhances the effect of antitumour drugs, which in many cases, have resulted in serious complications as a result of effects on normal cells. Green tea has been reported to be beneficial as it increases the drug concentration (e.g. doxorubicin, sulindac) in the tumour cells with no increase in normal cells. As it is a beverage, it reduces the patient burden of taking too many medicines.^{97, 118, 119}

Bioavailability studies of polyphenols (especially EGCG) reported wide distribution in multiple organs (digestive, liver, pancreas, mammary gland, and skin).¹²⁰ After intravenous administration of catechins in rats, it was seen that the half-life of EGCG, ECG and EC were 191, 362 and 45 min respectively.¹²¹ When

pure EGCG was given, a shorter half-life was observed, suggesting the effect of other components in the extract on the plasma concentration and elimination of EGCG.¹²²⁻¹²⁴

Epidemiological studies from Japan and China suggested that relatively high consumption of green tea (over 10 cups a day) is associated with substantial reduction in the risk of cancers of the skin, oesophagus, stomach, breast, lung and bladder.^{117, 122, 124-127} Theaflavins are also reported to inhibit cancer of the lung and oesophagus.^{89, 122} Thus therapeutic levels of polyphenols can exert important antimutagenic and anticarcinogenic effects in humans.

Most reports on cancer prevention were from Asians, who drink predominantly green tea, whereas studies involving black tea in Europeans observed infrequent protective effects.^{122, 128}

Antidiabetic Effect

The ability of polyphenols to lower blood glucose has been confirmed in diabetic rats. Both green tea and black tea were shown to possess antidiabetic activity and were effective in prevention and treatment of diabetes.¹²⁹ Tea polyphenols lower the serum glucose by inhibiting the activity of starch digesting enzyme, amylase. Tea inhibits both salivary and intestinal amylase. As a result starch is broken down more slowly and the sudden rise in serum glucose is minimized. In addition, tea also reduces the intestinal absorption of glucose.¹²⁹⁻¹³¹

Antiarthritic Activity

Polyphenols from green tea have shown exceptional protection against arthritis. The major polyphenols showing antiarthritic effect include epicatechin, epigallocatechin, epicatechin-3-gallate and epigallocatechin-3-gallate.

In a study conducted in mice, green tea polyphenols significantly reduced the incidence of arthritis (33 to 50 per cent). Further, the arthritic mice in polyphenols fed group showed less severe form of the disease. Histopathological examination of arthritic joints of mice from control group revealed extensive cartilage and bone erosions with massive infiltration of mono-nuclear cells and

fibroblasts, whereas green tea polyphenols fed group showed marked reduction in the number of infiltrating cells with no significant cartilage and bone erosions.¹³²

Expression of cyclooxygenase-2 enzyme (COX-2) is upregulated in arthritis. It has been found that green tea polyphenols inhibit the production of COX-2 in the arthritic joints.^{132, 133} There was also a marked reduction in other inflammatory mediators such as IFN- γ and TNF- α . The neutral endopeptidase activity is decreased on the administration of green tea polyphenols.¹³² It was seen that total IgG and type II collagen specific IgG levels were lower in the serum and arthritic joints of green tea polyphenols fed mice.¹³² Catechins inhibited the release of lysosomal enzymes, the chemiluminescence response and the production of free radicals.^{134, 135}

The studies suggested that a polyphenolic fraction of green tea that is rich in antioxidants might be useful in the prevention and severity of this disease.¹³² Since there is no cure for the rheumatoid and therapy aims at controlling the symptoms, a slight modification in the lifestyle and adding green tea to the diet can reduce the risk of this disease. This was demonstrated by a study conducted in mice over a period of 85 days which were administered an extract equivalent to human drinking of 4 cups of green tea per day.²

Antiplaque Activity

In the discovery of antibacterial activities of green tea polyphenols, it was found that these compounds inhibit the growth and adherence of oral bacteria.^{136, 137} Green tea extract acts in three ways. First it was shown to inhibit the growth of periodontal disease producing bacterium, *Porphyromonas gingivilis* and decay causing bacteria such as *Streptococcus mutans*.^{138, 139} Therefore, green tea as a mouth rinse, resulted in less plaque and periodontal disease. Secondly, it inhibited the enzyme amylase present in saliva and starch did not get converted in the mouth to glucose and maltose.^{130, 139} Less nutrition was thus available to decay causing bacteria. Lastly, it increased the resistance of tooth enamel to acid induced erosion.¹⁴⁰

Antiviral Activity

Tea extract has been shown to have virucidal activity against polio, influenza, vaccinia and herpes simplex virus.¹⁴¹

Anti-AIDS Activity

The long-term efficacy of new combination drug therapies for HIV infection is limited by the tendency of transfected HIV to mutate to become drug resistant. Green tea polyphenols are antimutagenic and act as effective adjuvants to drug therapy.⁹⁴ It was also discovered that polyphenols from green tea and their oxidation products could inhibit the reverse transcriptase or polymerase of several types of viruses, including HIV-1 and herpes simplex-1.^{130, 142, 143} However, research in this area is still in its primary stages.

Anorectic Effect

The catechin polyphenols inhibit catechol-*O*-methyl transferase¹⁴⁴ and caffeine inhibits transcellular phosphodiesterase thus stimulating thermogenesis and assisting the management of obesity.^{145, 146} The release of glucose is slowed down by tea and thus harmful spiking of insulin is prevented. Since insulin is the most fattening hormone, fat burning overtakes fat storage.¹³⁰

Antimicrobial Activity

Crude catechins and theaflavins have been found to possess antibacterial activity. They are believed to damage bacterial cell membranes. Tea has been used in the treatment of diarrhoeal infections, cholera and typhus.¹⁴⁷ Polyphenols kill spores of *Clostridium botulinum* and thus display antibacterial activity against food borne diseases and are also effective against heat-resistant bacilli like *Bacillus subtilis*, *B. cereus*, *Vibrio parahaemolyticus*, *Clostridium perfringens*.¹⁴⁸ Tea extracts also inhibited growth of *Staphylococcus aureus*, *S. epidemidis*, *Salmonella typhi*, *Shigella* spp. and *Streptococcus mutans*.¹⁴⁹ However, they show no activity against some common bacilli like *Salmonella enteridis*, *Escherichia coli* and *Yersina enterocolitica*.^{2, 149} Green tea also has protozoacidal properties.^{2, 130}

Other Biological Effects

Recently some reports have been published showing that green tea polyphenols also exhibit neuromuscular, antiangiogenic, antihepatotoxic, antiproliferative/apoptotic and immunomodulatory effects.¹⁵⁰

In summary, it can be concluded that a number of diseases can be prevented by the incorporation of green tea into the diet. It is seen that at least 10 cups a day of 100 mL each (3-4 g of polyphenols) are needed to have a significant beneficial effect. However, reports on this aspect vary.¹⁰⁸

5.0 EXPERIMENTAL

5.1 Materials and Methods

Tea Samples

Samples of different cultivated varieties of green tea were collected from four major tea growing areas of India including Palampur (North India), Assam and Darjeeling (Northeast India), Coonoor and Ooty (South India). The samples of three main cultivated varieties i.e. *C. sinensis* var. *cambodiensis*, *C. sinensis* var. *assamica* and *C. sinensis* var. *sinensis* and their hybrids presently under cultivation in India were collected from these areas. The collection from Palampur was done at three different times of plucking season first in the month of April (beginning of plucking season), then in September (mid and growing burst after rainy season) and finally in October and November (end of plucking season). Darjeeling samples were collected in the month of June and samples from Coonoor and Ooty were collected in October. The details of collections are provided in Table 3. The collected samples were treated as in the manufacture of green tea by subjecting to steam and drying under controlled conditions. Additionally some samples were sun dried without steaming. The leaves were powdered prior to extraction.

Solvents, Reagents and Chromatographic Plates

(-)-Epigallocatechin-3-gallate (EGCG) and (-)-epicatechin-3-gallate (ECG) were obtained from Sigma-Aldrich Chemicals. The solvents, chemicals and reagents were either obtained from E. Merck, or S. D. Fine Chemicals, India. Distilled water was used wherever water is mentioned. Laboratory made TLC plates (silica gel G, 0.2 mm thickness) activated at 110 °C for 30 min were used

for qualitative work. For quantitative work, precoated silica gel G plates (20 x 20 cm, 0.2 mm thickness, plastic base, E. Merck) cut into desired size were used.

Table 3: Collection details of tea samples

Zone	Place of collection	Clone/ Variety	Month & year of collection
North	Palampur	TV-23	Sep. & Nov. 2001, Apr. 2002
		Kangra Asha	Nov. 2001, Apr. & Sep. 2002
		TV-1	Nov. 2001 & Apr. 2002
		Kangra Jawala	Apr. & Sep. 2002
Northeast	Darjeeling	P-126	Jun. 2002
		Takda-7, 8	Jun. 2002
		Tenali-17	Jun. 2002
		TV-1	Jun. 2002
		TV-9	Jun. 2002
		TV-18	Jun. 2002
		TV-20	Jun. 2002
		TV-23	Jun. 2002
		TV-26	Jun. 2002
		China Bush + Clones	Jun. 2002
South	Ooty	China Variety	Oct. 2002
		VP-Clones	Oct. 2002
		BSS-1	Oct. 2002
		C-1	Oct. 2002
		CR-6017	Oct. 2002
		UPASI-3	Oct. 2002
	Coonoor	TRI-2024	Oct. 2002
		TRI-2025	Oct. 2002
		TRI-2026	Oct. 2002
		UPASI-2	Oct. 2002
		UPASI-3	Oct. 2002
		UPASI-8	Oct. 2002
		UPASI-9	Oct. 2002
		UPASI-10	Oct. 2002

Preparation of Tea Extract

The tea extracts were prepared by refluxing tea leaves with the appropriate solvent. At the end of the extraction, the material was filtered under reduced pressure and the marc was washed with specified volume of fresh solvent while

continuing filtration. The final volume of the filtrate was made to a specified volume before subjecting it to analysis. The quantity of tea leaves, its ratio to solvent, extraction period, temperature, volume of solvent used for washing the marc and final volume of the extract were predetermined for each set of extraction. Initial experiments were conducted to optimize extraction procedure with respect to choice of solvent, temperature, period of extraction and particle size of tea leaves. The optimized extraction conditions were subsequently used to determine the amount of polyphenols in different varieties of tea.

Quantitative Analysis

TLC method

All quantitative estimations were made in triplicate, using CAMAG TLC Scanner 3 and CATS software version 4.06. Known amount of tea extract was applied on a precoated TLC plate and the plate was developed in an appropriate solvent system. The plate was dried in a current of hot air and scanned in TLC scanner at 280 nm. The area under the curve (AUC) for peak corresponding to the spot of each polyphenol was noted and the concentration of different polyphenols in the extract was determined from their respective standard plots.

HPLC method

The analytical determinations of EGCG and ECG were carried out using reverse phase-high performance liquid chromatography at isocratic mode. The Waters HPLC system equipped with automated gradient controller, 510 pumps, U 6 K injector, 481 detector and 746 data module was used for the analysis. The Waters μ -bondapak C18 (3.9 x 300 mm) column at ambient temperature 24 to 28 °C, mobile phase water : methanol : acetic acid (70 : 30 : 0.5), flow rate 1.0 mL / min and UV detection at 280 nm were used in the HPLC analysis. All extracts were prepared and analysed in triplicate. The extract was filtered through 0.45 μ m filter before injecting 5 μ L of the appropriately diluted sample.

Stability Studies

The degradation pattern of EGCG was studied in buffers of pH 1.2, 2.0, 4.0, 6.0, 7.0 and 8.0 (Appendix 1) over a period of 48 h. The change in concentration of EGCG in buffered solution was monitored through quantitative TLC using CAMAG TLC Scanner.

5.2 Development of TLC Fingerprint Profile of Green Tea

A methanol extract of green tea was prepared by refluxing 2 g tea leaves in 50 mL methanol for 1 h in water bath at 80 °C. A variety of solvent systems were tried in order to obtain the best resolution. The spots were visualized by spraying the plate with 1 per cent vanillin in sulphuric acid followed by heating for 10 min at 110 °C. A solvent system comprising of chloroform : acetone : formic acid (5 : 4 : 1) produced most resolved profile. TLC fingerprint profile of tea polyphenols and non-polyphenols is shown in Figure 5.



Fig. 5: TLC fingerprint profile of tea polyphenols (A) and non-polyphenols (B)

5.3 Preparation of Standard Plot of EGCG and ECG

Preparation of Standard Plots of EGCG

A standard solution of EGCG was prepared by dissolving 2.41 mg of EGCG in 25 mL methanol. The varying volumes of standard solution 0.5 to 16.0 μL in geometric progression were applied in triplicate on a precoated TLC plate. The plate was developed in chloroform : acetone : formic acid (5 : 4 : 1), dried and then scanned at 280 nm in the TLC scanner. The AUC for each quantity was noted (Table 4, Figure 6) to construct standard plot.

Table 4: Mean peak areas of EGCG in TLC analysis

EGCG (μg)	^a Mean AUC \pm SD
0.0492	313 \pm 5.90
0.0984	631 \pm 26.71
0.1968	1219 \pm 49.08
0.3936	2471 \pm 23.61
0.7872	4808 \pm 190.59
1.5744	8853 ^b

^a $n=3$, ^b $n=1$

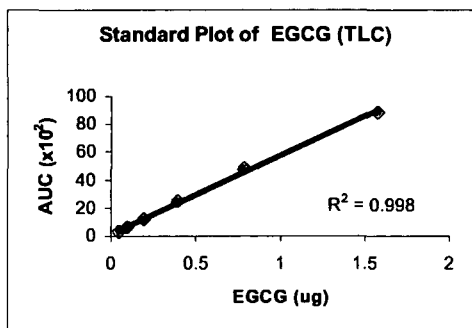


Fig. 6: Standard plot of EGCG in TLC analysis

Standard plot for HPLC analysis was constructed by injecting in triplicate a constant volume of 5 μL of serially diluted concentrations and noting AUC corresponding to each concentration (Table 5 & Figure 7). Reverse phase C18 column and water : methanol : acetic acid (70 : 30 : 0.5) at 1 mL / min flow rate were used for the analysis.

Table 5: Mean peak areas of EGCG in HPLC analysis

EGCG (ng)	^a Mean AUC±SD
9.85	23471 ± 19.0
19.70	46026 ± 855
39.35	88815 ± 36
78.50	175341 ± 2502
157.50	355954 ± 6708
283.20	635194 ± 3077

^an= 3

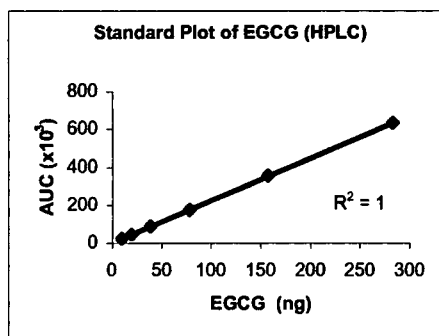


Fig. 7: Standard plot of EGCG in HPLC analysis

Preparation of Standard Plots for ECG

The standard plots of ECG for TLC and HPLC analysis were constructed similarly as described for EGCG using the same conditions of the analysis in TLC and HPLC. Standard solution of ECG was prepared by dissolving 3.02 mg of ECG in 10 mL methanol. In TLC 0.5 to 2.5 μ L of standard solution in increments of 0.5 μ L was applied and AUC was noted (Table 6) to construct standard plot (Figure 8).

Table 6: Mean peak areas of ECG in TLC analysis

ECG(μ g)	^a Mean AUC±SD
0.0302	211±12.28
0.0604	411±9.74
0.0906	571±26.0
0.1208	761±30.26
0.1510	953±31.47

^an= 3

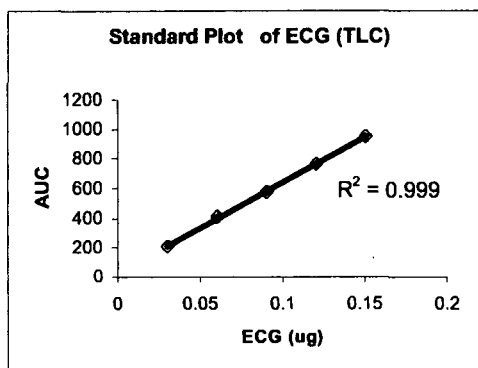


Fig. 8: Standard plot of ECG in TLC analysis

In HPLC analysis standard solution was diluted to inject a constant volume of 5 μ L. The data and standard plot for HPLC are given in Table 7 and Figure 9 respectively.

Table 7: Mean paek areas of ECG in HPLC analysis

ECG (ng)	^a Mean AUC \pm SD
09.40	26898 \pm 736
18.85	48516 \pm 1079
37.50	100503 \pm 2205
75.50	188155 \pm 2252
151.00	378395 \pm 3563
302.00	761380 \pm 5851

^an= 3

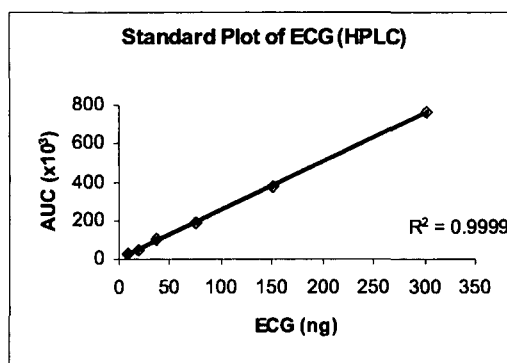


Fig. 9: Standard plot of ECG in HPLC analysis

5.4 Optimization of Extraction Procedure for Tea Polyphenols

Effect of Solvent on Extraction of EGCG

Tea sample (2 g) was refluxed separately in 50 mL each of methanol, water and ethyl acetate for 30 min at 60 °C. The extract was filtered under vacuum while hot. The marc was washed with 20 mL of additional extraction solvent while continuing filtration. The volume of the filtrate was made up to 100 mL. 1 μ L of this extract was applied on a precoated TLC plate, developed in a solvent system consisting of chloroform : acetone : formic acid (5 : 4 : 1) and scanned using TLC scanner. The AUC for peak corresponding to EGCG was noted and the concentration in the extract was calculated from the standard plot. The results are shown in Table 8 and Figure 10.

Table 8: Effect of solvent on extraction of EGCG

Solvent	EGCG (mg / g) \pm SD
Methanol	57.12 \pm 0.68
Water	48.39 \pm 0.20
Ethyl Acetate	4.32 \pm 0.30

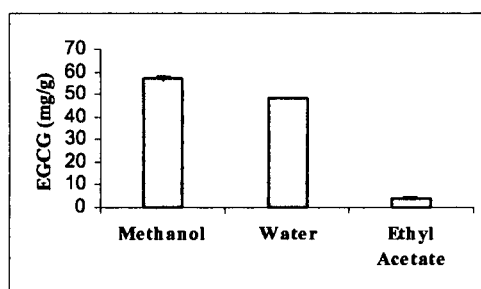


Fig. 10: Effect of solvent on extraction of EGCG

Effect of Time and Temperature on Extraction of EGCG

Tea leaf powder (2 g) was refluxed separately with 50 mL each of methanol and water at different temperatures (40, 60, 80 and 98 °C) on a water bath for different time intervals (15, 30, 45, 60, 75 and 90 min). After completion of each extraction, the final volume of the extract was made to 100 mL. 1 μ L of this extract was applied on TLC plate and the EGCG content was determined using the standard curve. Extraction was carried out in triplicate and further each extract was applied in triplicate. Table 9 shows the extracted amount of EGCG. In one study the marc was re-extracted with fresh solvent to determine if the extraction was complete.

Table 9: Effect of time and temperature on extraction of EGCG

Extraction Time (min)	Solvent	EGCG (mg / g) \pm SD			
		40 °C	60 °C	80 °C	98 °C
15	Methanol	15.25 \pm 0.37	54.12 \pm 0.06	42.73 \pm 1.08	
	Water	16.44 \pm 0.63	40.65 \pm 0.14	42.72 \pm 1.09	53.60 \pm 1.20
30	Methanol	20.55 \pm 0.20	57.12 \pm 0.68	47.55 \pm 0.36	
	Water	21.42 \pm 0.92	48.39 \pm 0.20	53.51 \pm 0.27	48.48 \pm 0.62
45	Methanol	23.00 \pm 1.16	58.24 \pm 0.03	48.22 \pm 0.59	
	Water	24.92 \pm 0.62	46.09 \pm 0.08	53.64 \pm 0.36	49.55 \pm 0.84
60	Methanol	29.98 \pm 0.22	59.06 \pm 0.26	48.68 \pm 0.35	
	Water	23.82 \pm 0.74	50.30 \pm 1.19	54.02 \pm 0.30	48.00 \pm 0.96
75	Methanol	24.14 \pm 0.31	58.74 \pm 0.93	40.66 \pm 0.65	
	Water	27.52 \pm 0.30	49.18 \pm 0.10	53.45 \pm 0.13	49.45 \pm 0.13
90	Methanol	26.52 \pm 0.51	58.70 \pm 0.52	39.96 \pm 0.31	
	Water	23.90 \pm 0.50	48.85 \pm 0.47	53.40 \pm 0.07	49.15 \pm 0.13

Effect of Time on Extraction of EGCG in Water

Tea leaf powder (2 g) was extracted with 50 mL of water at 98 °C for varying period of extraction from 5 to 90 min. The amount of extracted EGCG was determined using TLC method of analysis. The data is shown in Table 10 and Figure 11.

Table 10: Effect of time on extraction of EGCG in water

Extraction Time (min)	EGCG (mg/g) ± SD
5	54.30 ± 0.16
10	53.62 ± 0.07
15	53.60 ± 1.20
30	48.48 ± 0.62
45	49.55 ± 0.84
60	48.00 ± 0.96
75	49.45 ± 0.13
90	49.15 ± 0.13

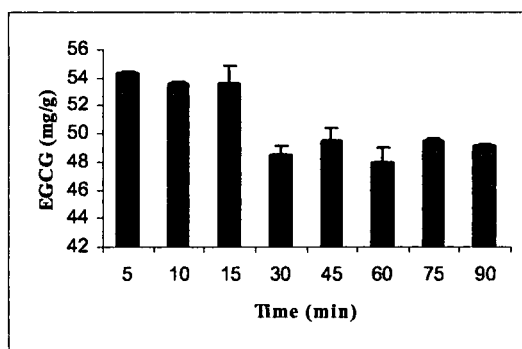


Fig. 11: Effect of time on extraction of EGCG in water

Effect of Sample to Solvent Ratio on Extraction of EGCG

Different ratios of sample to water were used to extract EGCG and other parameters were kept constant as optimized for extraction with water. After completion of each extraction, the quantity of EGCG in the extract was determined by TLC. The effect of volume of water on extraction is given in Table 11 and Figure 12.

Table 11: Effect of sample: water ratio on extraction of EGCG

Sample:solvent (g : mL)	EGCG (mg / g) \pm SD
1:10	40.21 \pm 2.40
1:25	53.27 \pm 0.58
1:50	54.58 \pm 0.18
1:100	55.62 \pm 0.67
1:150	61.01 \pm 2.66
1:200	57.64 \pm 0.60
1:400	24.98 \pm 0.45

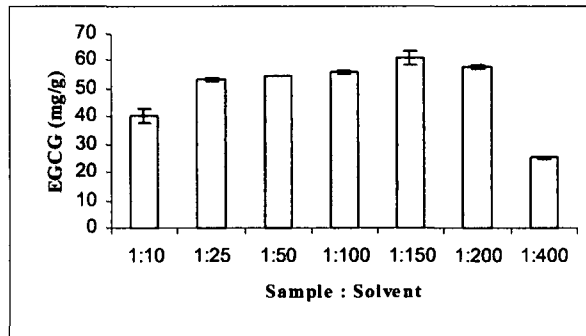


Fig. 12: Effect of sample: water ratio on extraction of EGCG

Effect of Shaking on Extraction of EGCG

Tea leaf (4 g) was extracted with water at 98 °C for 5 min. The flasks were shaken continuously in a shaker. The extraction yield was compared with control. The results are given in Table 12 and Figure 13.

Table 12 : Effect of shaking on extraction of EGCG

Treatment	EGCG (mg / g) \pm SD
Control	14.80 \pm 0.06
Shaking	21.16 \pm 0.03

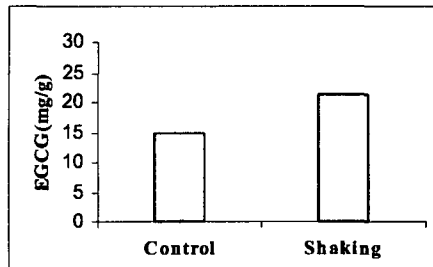


Fig. 13 : Effect of shaking on extraction of EGCG

Effect of Particle Size on Extraction of EGCG

Tea leaf (2 g each) powdered into coarse, moderately coarse, moderately fine and fine powder (Appendix 2) were extracted using in methanol optimized conditions of time and temperature. The results are shown in Table 13 and Figure 14.

Table 13: Effect of particle size on extraction of EGCG

Powder Grade	EGCG (mg / g) ± S.D.
Coarse	55.88 ± 0.73
Moderately Coarse	56.54 ± 0.80
Moderately Fine	58.85 ± 0.49
Fine	56.34 ± 0.70

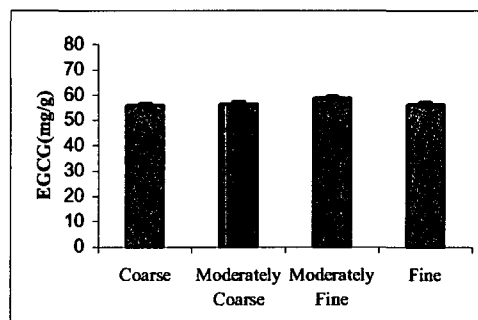


Fig. 14: Effect of particle size on extraction of EGCG

5.5 Studies on Stability of Tea Polyphenols (EGCG)

Effect of pH on Degradation of EGCG

The tea extract was prepared using the optimized method. Different aliquots of 10 mL each of this extract were concentrated to dryness and dissolved separately in 10 mL buffer of pH 1.2, 2, 4, 6, 7 and 8. The buffered extracts were stored at room temperature (26 to 28 °C) and EGCG content was monitored through quantitative TLC at different time intervals (Table 14). The control extract was concentrated to dryness and dissolved in 10 mL of distilled water. The per cent degradation of EGCG at different time intervals is given in Table 15 and Figure 15.

Table 14: Effect of pH on EGCG degradation

pH of Solution	EGCG (mg/g) \pm SD ^a at different time intervals (h)						
	0	1	2	4	8	24	48
Control	40.86 \pm 0.85	36.07 \pm 0.43	32.63 \pm 0.44	30.80 \pm 0.10	29.98 \pm 0.78	25.35 \pm 0.18	23.18 \pm 0.56
1.2	50.88 \pm 0.88	49.55 \pm 0.78	49.20 \pm 0.53	49.18 \pm 1.05	49.12 \pm 0.44	49.14 \pm 1.11	47.22 \pm 0.84
2.0	44.84 \pm 0.81	43.60 \pm 0.52	43.44 \pm 0.85	43.42 \pm 0.96	43.44 \pm 0.80	43.40 \pm 1.10	41.96 \pm 0.53
4.0	61.75 \pm 0.53	60.01 \pm 1.02	59.93 \pm 0.95	59.96 \pm 0.66	59.88 \pm 0.89	59.86 \pm 0.59	46.12 \pm 0.82
6.0	50.65 \pm 0.75	48.98 \pm 0.96	48.98 \pm 0.75	48.78 \pm 1.05	48.75 \pm 1.06	40.70 \pm 1.02	39.54 \pm 0.64
7.0	39.95 \pm 0.37	35.62 \pm 0.26	35.64 \pm 0.77	35.58 \pm 0.34	32.40 \pm 0.54	25.19 \pm 0.95	25.00 \pm 0.46
8.0	28.15 \pm 0.53	22.14 \pm 0.84	19.95 \pm 0.75	17.60 \pm 0.46	16.65 \pm 0.85	12.25 \pm 0.52	09.64 \pm 0.46

^an=3

Stability of EGCG Solution in Ethyl Acetate

The stability of EGCG in tea extracts stored in ethyl acetate was monitored till 48 h (Table 16 & Figure 16).

Table 15: Per cent degradation of EGCG at different pH

pH of Solution	EGCG degradation (%) at time intervals (h)						
	0	1	2	4	8	24	48
Control	0	11.6	19.9	24.3	26.3	37.5	42.8
1.2	0	2.6	3.2	3.3	3.4	3.4	7.1
2.0	0	2.7	3.0	3.1	3.1	3.2	6.3
4.0	0	2.7	2.9	2.9	3.0	3.0	25.1
6.0	0	3.1	3.1	3.5	3.6	19.3	21.6
7.0	0	10.7	10.7	10.8	18.6	36.5	37.0
8.0	0	21.0	28.6	36.8	40.2	55.6	64.7

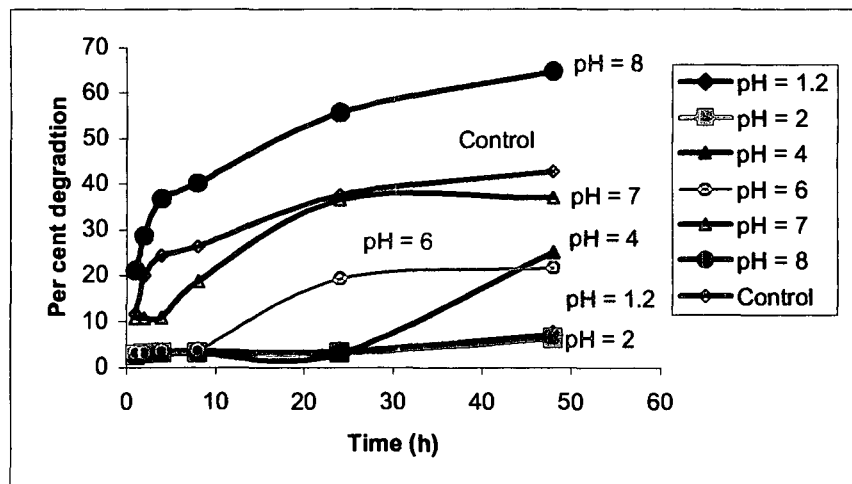


Fig. 15: Effect of pH on stability of EGCG

Table 16: Stability of EGCG in ethyl acetate

Time interval (h)	EGCG (mg / g) \pm SD	Percent degradation
0	14.66 \pm 0.50	0
1	14.50 \pm 0.56	1.1
2	14.48 \pm 0.28	1.2
4	14.48 \pm 0.17	1.2
8	14.48 \pm 0.53	1.2
24	14.48 \pm 0.38	1.2
48	14.43 \pm 0.54	1.5

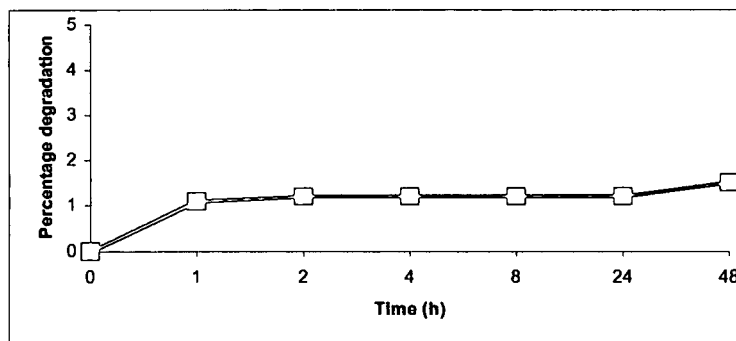


Fig. 16: Stability of EGCG in ethyl acetate extract

5.6 Polyphenol Content in Cultivated Varieties of Indian Tea

The EGCG and ECG content were estimated by HPLC in the different samples of cultivated Indian varieties of tea using optimized procedure of extraction and analysis developed in the laboratory. Accurately weighed (2 g) moderately fine powder (2 g) of tea sample was taken in a vacuum flask and 100 mL of boiling water was added to it. The flask was stoppered and shaken for 5 min. The extract was filtered while hot and the marc was washed with 10 mL of boiling water. The volume of the extract was adjusted to 100 mL with refrigerated water. The extract was diluted (1 to 25) with a mobile phase of water : methanol : acetic acid :: 70 : 30 : 0.5 of HPLC analysis. The diluted extract was filtered through a 0.45 μm filter and used for HPLC analysis (Figure 17). The amount of EGCG and ECG in extract was calculated from the area under the curve corresponding to the peaks of EGCG and ECG and using standard plots (Table 17).

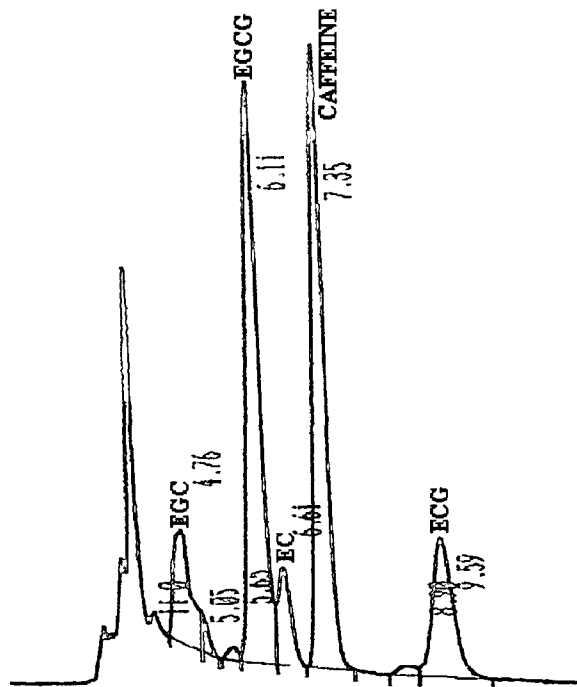


Fig. 17: A typical HPLC chromatogram of green tea extract

Table 17: Polyphenol content of tea samples of different cultivated varieties of India

Zone	Clone/ Variety	Period of collection	Processing method	EGCG (mg/g)	ECG (mg/g)
North	TV-23	Sep. 2001	Sun Drying	2.70 ± 0.50	N.D.
		Sep. 2001	Steam Drying	24.45 ± 2.48	6.36 ± 0.50
		Nov. 2001	Sun Drying	19.82 ± 0.20	9.58 ± 0.11
		Nov. 2001	Steam Drying	43.60 ± 0.25	16.40 ± 0.30
		Apr. 2002	Sun Drying	31.87 ± 1.63	12.18 ± 0.22
		Apr. 2002	Steam Drying	58.52 ± 1.34	18.95 ± 0.33
	Kangra Asha	Jun. 2002	Steam Drying	23.52 ± 0.61	13.71 ± 1.49
		Nov. 2001	Sun Drying	34.94 ± 0.23	8.13 ± 0.16
		Nov. 2001	Steam Drying	32.00 ± 2.93	7.04 ± 0.51
		Apr. 2002	Sun Drying	52.35 ± 1.41	10.62 ± 0.56
		Apr. 2002	Steam Drying	45.61 ± 1.67	9.56 ± 0.32
		Sep. 2002	Sun Drying	23.50 ± 0.85	11.39 ± 0.85
	Kangra Jawala	Sep. 2002	Steam Drying	64.38 ± 1.52	14.04 ± 0.90
		Apr. 2002	Sun Drying	26.64 ± 2.52	6.37 ± 0.58
		Apr. 2002	Steam Drying	68.89 ± 2.77	19.04 ± 0.35

Table contd....

Table contd.....

	Kangra Jwala	Sep. 2002	Sun Drying	27.14 ± 2.07	10.88 ± 0.38
		Sep. 2002	Steam Drying	62.01 ± 4.52	14.30 ± 1.15
	TV-1	Nov. 2001	Sun Drying	29.61 ± 2.74	20.63 ± 2.63
		Nov. 2001	Steam Drying	42.54 ± 0.43	25.36 ± 0.07
		Apr. 2002	Sun Drying	22.16 ± 0.52	15.56 ± 0.67
		Apr. 2002	Steam Drying	43.22 ± 2.80	27.39 ± 2.20
Northeast	TV-1	Jun. 2002	Steam Drying	27.48 ± 2.32	19.64 ± 1.71
	TV-9	Jun. 2002	Steam Drying	33.91 ± 3.67	10.72 ± 1.18
	TV-18	Jun. 2002	Steam Drying	50.18 ± 1.61	16.22 ± 1.52
	TV-20	Jun. 2002	Steam Drying	19.18 ± 0.82	6.60 ± 0.86
	TV-26	Jun. 2002	Steam Drying	18.46 ± 1.40	7.33 ± 0.53
	P-126	Jun. 2002	Steam Drying	12.89 ± 1.24	7.16 ± 0.71
	Takda-7,8	Jun. 2002	Steam Drying	19.48 ± 0.59	6.45 ± 0.30
	Tenali-17	Jun. 2002	Steam Drying	17.62 ± 1.48	5.63 ± 0.69
	China Bush + Clones	Jun. 2002	Steam Drying	30.36 ± 3.49	8.50 ± 0.85
South	China Variety	Oct. 2002	Sun Drying	1.65 ± 0.07	1.37 ± 0.04
	UPASI-3	Oct. 2002	Sun Drying	1.36 ± 0.22	0.32 ± 0.00
	VP-Clones	Oct. 2002	Sun Drying	2.14 ± 0.50	0.71 ± 0.16
	BSS-1	Oct. 2002	Steam Drying	24.32 ± 0.86	8.04 ± 0.56
	C-1	Oct. 2002	Steam Drying	30.87 ± 0.89	10.54 ± 0.55
	China Variety	Oct. 2002	Steam Drying	38.12 ± 2.35	14.24 ± 1.08
	CR-6017	Oct. 2002	Steam Drying	44.18 ± 3.58	11.60 ± 1.20
	TRI-2024	Oct. 2002	Steam Drying	33.43 ± 3.60	12.76 ± 0.89
	TRI-2025	Oct. 2002	Steam Drying	42.31 ± 5.09	10.29 ± 0.82
	TRI-2026	Oct. 2002	Steam Drying	68.35 ± 5.21	22.20 ± 1.91
	UPASI-2	Oct. 2002	Steam Drying	51.47 ± 0.74	12.15 ± 0.33
	UPASI-3	Oct. 2002	Steam Drying	19.75 ± 6.99	5.53 ± 2.02
	UPASI-8	Oct. 2002	Steam Drying	45.44 ± 3.09	14.68 ± 1.26
	UPASI-9	Oct. 2002	Steam Drying	57.82 ± 1.62	14.24 ± 0.98
	UPASI-10	Oct. 2002	Steam Drying	21.64 ± 2.24	8.33 ± 0.76

6.0 SURVEY DATA

The tea producing areas of North, Northeast and South India were surveyed to collect information on cultivated varieties of tea, area under tea cultivation, production, processing and marketing of green tea, and problems of the green tea manufacturers. The information was procured from the Tea Board of India, tea processing companies and other governmental and non governmental organizations working on tea in each zone. The data on the production including total tea produced, area under tea cultivation, tea yield, monthly and annual tea sales in different auctions and average prices of tea (Appendix 3) was provided by Tea Board of India and J. Thomas and Private Company Limited, Kolkata. For obtaining processed green tea samples of different tea varieties for analysis and information regarding tea varieties, marketing channels, problems in green tea processing and marketing various tea companies (Makaibari Tea Estates; Sannyasithan Tea Co. Pvt. Ltd.; Maud Tea & Seed Co. Ltd.; Tea Promoters (India) Pvt. Ltd.; Ambari Tea Co. Ltd.; Sublime Agro. Ltd.; Sepoydhoorah Tea Co. Pvt. Ltd.; Goodricke Group Ltd.; Duncans Industries Ltd.; United Planter's Association of Southern India,; Indcoserve; The Peria Karamalai Tea & Produce Co. Ltd. and Mahalinga Ind. Co. op. Tea Factory Ltd.) and tea gardens were visited and surveyed through a questionnaire (Appendix 4) and the information procured in Appendix 5.

India is the world's largest producer and consumer of tea and is known to produce finest qualities of tea.¹⁵⁰ It also produces largest number of tea varieties. The Indian tea is best known for its flavour, taste and colour. The three world famous types of Indian tea are Assam, Darjeeling and Nilgiri tea. According to an estimate, 673 thousand tonnes of tea was consumed in India in 2001 ranking it at the top among the tea consuming nations of the world.

Tea producing areas in India are located in the north, northeast and south parts of the country (Figures 18-20). The major production comes from the

northeastern parts of India. The tea producing states and regions under each zone are presented in Table 18.

6.1 Cultivated Varieties

In north zone, Palampur in Kangra valley of state of Himachal Pradesh and Nainital in the state of Uttaranchal are engaged in tea production. The region cultivates largely the China variety of tea (*Camellia sinensis* (L.) O. Kuntze var. *sinensis*), which is suited to prevailing agro-climatic conditions of the region.

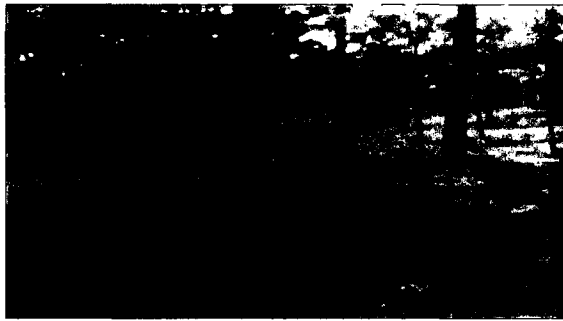


Fig. 18: A tea garden at Palampur (North India)



Fig. 19: A tea garden at Darjeeling (Northeast India)



Fig. 20: A tea garden at Ooty (South India)

Table 18: Tea producing areas of India

Zone	State	District (Region)	
North	Himachal Pradesh	Kangra (Palampur)	
	Uttaranchal	Nainital	
Northeast	Arunachal Pradesh	-	
		Cachar	
	Assam	Darrang*	
		Dibrugarh*	
		Goalpara*	
		Kamrup*	
		Karbi Anlong	
		Lakhimpur*	
		North Cachar	
		Nowgong*	
		Sibsagar **a	
		Kisanganj	
		Bihar	-
		Manipur	-
		Meghalaya	-
Mizoram	-		
Nagaland	-		
Orissa	-		
Sikkim	-		
Tripura	-		
	West Bengal	Darjeeling	
		Dooars ^b	
		Terai ^c	
South	Tamilnadu	Coimbatore	
		Kanyakumari	
		Madurai	
		Nilgiris	
		Tirunelveli	
	Kerala	Cannanore	
		Ernakulam	
		Idukki	
		Kottayam	
		Kozhikode	
		Malapuram	
		Palghat	
		Quilon	
		Trichur	
		Trivandrum	
		Wynaad	
		Karnataka	Chikmagalur
			Coorg
Hassan			

* Collectively known as Assam Valley.

^a Including Mikhir Hills and North Cachar, ^b Including Cooch Behar

^c Including West Dinajpur,

Assam and West Bengal are the main tea producing states in the northeast zone (Table 18). In Assam, Assam variety (*Camellia sinensis* (L.) O. Kuntze var. *assamica*) and its hybrids are cultivated, whereas in Darjeeling and surrounding areas in West Bengal, China variety is cultivated. A number of clones and cultivars have been developed for cultivation in India (Table 19). Department of Tea Husbandry and Technology of Himachal Pradesh Krishi Vishwa Vidhalaya at Palampur, Tocklai Experimental Station at Jorhat and United Planters Association of South India (UPASI), Tea Research Institute at Coonoor have developed these clones.

Table 19: Important cultivars of tea in different tea producing areas of India

Zone	Clone/cultivar	Zone	Clone/cultivar
North	Kangra Asha	South	BSS-1
	Kangra Jwala		C-1
	Takda-7-8		CR-6017
	Tenali-17		TRI 2024
	TV-1		TRI 2025
Northeast	TV-23	TRI 2026	
	TV-1	UPASI-1	
	TV-9	UPASI-2	
	TV-18	UPASI-3	
	TV-19	UPASI-4	
	TV-20	UPASI-5	
	TV-23	UPASI-6	
	TV-26	UPASI-7	
	TV-27	UPASI-8	
	TV-29	UPASI-9	
	TV-30	UPASI-10	
		UPASI-15	
		UPASI-20	

6.2 Tea Production and Area Under Tea Cultivation in India

North and Northeast India

The area under tea production in north and northeast zones from 1998 to 2000 is presented in Table 20.¹⁵¹ No change in area under tea cultivation was

observed in north zone during this period. However, the tea production decreased by 529 tonnes (25.68 per cent) in 1999 while in 2000 a decrease of 20 tonnes (1.30 per cent) was observed.

In northeast zone, the area under tea cultivation during 1998 was 368238 hectares, which increased by 11103 hectares (3.02 per cent) in 1999 and 11263 hectares (2.96 per cent) in 2000. In Assam, the area under tea cultivation increased by 6110 hectares (2.42 per cent) in 1999 and 9657 hectares in 2000 whereas in West Bengal an increase of 936 hectares (0.86 per cent) was observed compared to 3130 hectares (2.96 per cent) in 1999. Except for the state of Orissa (for which no change was observed), all the other states in northeast zone observed an increase in area under cultivation during 1998 to 2000.

All tea producing districts in West Bengal except Terai, states of Manipur and Arunachal Pradesh recorded higher crop production. Maximum increase was observed in Assam (4.22 per cent) and minimum in West Bengal (0.284 per cent) in 2000 (Table 20).¹⁵¹ Production in northeast zone declined from 668598 tonnes in 1998 to 621566 tonnes (7.03 per cent) in 1999 whereas; an increase of 18884 tonnes (3.04 per cent) in tea production was witnessed during 2000 over the level of previous year.

A decrease of 9.8 per cent in yield was observed for North India (north and northeast zones) during 1998 while negligible change (increase of 0.06 per cent) was observed during 2000 (Table 21).¹⁵¹

South India

The state of Tamilnadu witnessed maximum increase in the area under tea cultivation, 5612 hectares in 1999 to 5176 hectares in 2000 (Table 22).¹⁵² In Kerala, marginal increase of 0.04 per cent was observed from 1998 to 2000 and no change was observed in Karnataka. The tea production in South India increased by 1.59 per cent during 1999-2000 and stood at 204522 tonnes in the year 2000. Major increase was reported from Kerala with no or marginal contribution from other tea producing states of South India. The average tea production and yield of South India has been given in Table 22 and 23 respectively.¹⁵¹

Table 20: Production and area under tea cultivation in North and Northeast India

Zone/State/ District (Region)	1998		1999		2000	
	Area (ha)	Quantity (tonnes)	Area (ha)	Quantity (tonnes)	Area (ha)	Quantity (tonnes)
North Zone						
Himachal Pradesh						
Kangra (Palampur)	2325	1711	2325	1222	2325	1247
Uttaranchal						
Nainital	1068	349	1068	309	1068	264
Total	3393	2060	3393	1531	3393	1511
Northeast Zone						
Assamachal Pradesh						
Assam	1953	965	2179	1063	2176	993
Cachar	30565	51850	30938	49119	31116	53722
Darrang	40950	86942	41393	78501	41968	80227
Dibrugarh	83380	164463	88291	150469	93138	155932
Goalpara	3338	6099	3357	5436	3522	5855
Kamrup	3307	5146	3289	4941	3300	5003
Karbi Anlong	1652	1375	1781	1301	2005	1878
Lakhimpur	4809	9995	5046	9413	5230	9701
North Cachar	4516	6743	4523	6367	4524	6521
Nowgong	7746	14684	7850	13559	8014	14073
Sibsagar(a)	71362	119749	71267	113819	74575	118324
Total	251625	467046	257735	432925	267392	451236

Table contd.....

Table 20 contd

Zone/State/ District (Region)	1998		1999		2000	
	Area (ha)	Quantity (tonnes)	Area (ha)	Quantity (tonnes)	Area (ha)	Quantity (tonnes)
Bihar	762	138	1348	473	1350	538
Manipur	536	76	746	97	907	96
Meghalaya	145	127	215	135	351	140
Mizoram	350	23	360	35	391	39
Nagaland	472	29	1012	39	1214	43
Orissa	214	94	214	100	214	105
Sikkim	202	112	296	102	296	105
Tripura	6355	6099	6482	6385	6623	6431
West Bengal						
Darjeeling	17830	10253	17968	9294	18109	9814
Doars	70479	147133	70996	133803	71225	135963
Terai	17315	36403	19790	37115	20356	34947
Total	105624	193789	108754	180212	109690	180724
Total	368238	668598	379341	631566	390604	640450
Grand Total (North & Northeast India)	371631	670658	382734	623097	393997	641961

Table 21: Average tea yield in North and Northeast India

Zone/ State/ District (Region)	Average yield (kg/ha)		
	1998	1999	2000
North Zone			
Himachal Pradesh			
Kangra (Palampur)	736	526	536
Uttaranchal			
Nainital	327	289	247
Northeast Zone			
Arunachal Pradesh	494	488	456
Assam			
Cachar	1696	1588	1727
Darrang	2123	1896	1912
Dibrugarh	1972	1704	1674
Goalpara	1827	1619	1662
Kamrup	1556	1502	1516
Karbi Anlong	832	730	937
Lakhimpur	2078	1865	1855
North Cachar	1493	1408	1441
Nowgong	1896	1727	1756
Sibsagar(a)	1678	1597	1587
Average	1856	1680	1688
Bihar	181	351	399
Manipur	142	130	106
Meghalaya	876	628	399
Mizoram	66	97	100
Nagaland	61	39	35
Orissa	439	467	491
Sikkim	554	345	355
Tripura	975	985	971
West Bengal			
Darjeeling	575	517	542
Dooars	2088	1885	1909
Terai	2102	1875	1717
Average	1835	1657	1648
North & Northeast India	1805	1628	1629

Table 22: Tea production in South India

State/ Districts	1998		1999		2000	
	Area (ha)	Quantity (tonnes)	Area (ha)	Quantity (tonnes)	Area (ha)	Quantity (tonnes)
Tamilnadu						
Coimbatore	11609	32997	11650	32000	11650	32100
Kanyakumari	434	119	434	101	434	120
Madurai	941	3075	941	3600	941	3650
Nilgiris	49759	93972	55330	90375	60506	90729
Tiruneveli	800	1883	800	2012	800	2100
Total	63543	132046	69155	128088	74331	129699
Kerala						
Cannanore	-	-	-	-	-	-
Ernakulam	2	-	2	-	2	-
Idukki	26608	48254	26610	51200	26615	52000
Kottayam	840	244	840	300	840	320
Kozhikode		-				1420
Malapuram			174		174	5468
Malapuram	174	-	174	-	174	-
Palghat	841	2398	841	1800	841	1950
Quilon	1348	366	1348	360	1348	375
Trichur	523	1951	523	1500	523	1650
Trivendrum	965	582	965	440	965	475
Wynaad	5447	12148	5449	12196	5454	12585
Total	36748	65943	36752	67796	36762	69355
Karnataka						
Chikmagalur	1420	3484	1421	3269	1421	3288
Coorg	290	768	290	750	290	760
Hassan	395	1209	395	1408	395	1420
Total	2105	5461	2106	5427	2106	5468
South India	102396	203450	108013	201311	113199	204522

Table 23: Average tea yield in South India

State/ Districts	Average yield (kg/ha)		
	1998	1999	2000
Tamilnadu			
Coimbatore	2842	2747	2755
Kanyakumari	274	233	276
Madurai	3268	3826	3879
Nilgiris	1889	1633	1516
Tirunelveli	2354	2515	2625
Average	2078	1852	1745
Kerala			
Cannanore	-	-	-
Ernakulam	-	-	-
Idukki	1814	1924	1954
Kottayam	290	357	381
Kozhikode	-	-	-
Malapuram	-	-	-
Palghat	2851	2140	2319
Quilon	272	267	278
Trichur	3730	2868	3155
Trivendrum	603	456	492
Wynaad	2230	2238	2307
Average	1794	1845	1887
Karnataka			
Chikmagalur	2454	2300	2314
Coorg	2648	2586	2621
Hassan	3061	3565	3595
Average	2594	2577	2596
South India (average)	1987	1864	1807

Total Tea Production in India

The total area under tea cultivation increased from 490747 hectares in the year 1999 to 507196 hectares in the year 2000. Area wise production of tea in India is given in Figure 21. The tea production increased by 2.67 per cent during the year 2000 over the previous year and was 846483 tonnes in the year 2000, although the yield of tea recorded negligible growth during the same period. Only state of Kerala recorded an increase (5.18 per cent) in yield during the year 2000.

6.3 Production Trends of Different Types of Indian Tea

Production of cut tear curl (CTC) black tea in North India decreased by 35514 tonnes (5.99 per cent) in the year 1999 and increased by 34024 tonnes (5.75 per cent) in the year 2000 (Table 24).^{151,152} In South India however, reverse trend

was observed as CTC tea production increased by 3815 (2.30 per cent) in 1999 and decreased by 1871 tonnes (1.10 per cent) in 2000. On all India basis, production of CTC tea decreased by 31699 tonnes (4.18 per cent) in 1999 and increased in 2000 by 32153 tonnes (4.42 per cent). Orthodox tea manufacture of black tea in North India decreased by 28788 tonnes (41.21 per cent) in 1999 and increased by 39370 tonnes (9.66 per cent) in 2000. Similar trends were observed in south zone and on all India basis.

A continuous decrease in green tea production since 1998 was observed in North India, (6.31 per cent) during 1999 and (5.27 per cent) during 2000. In South India the production of green tea stayed nearly static during 1998 to 2000 and stood at 1819 tonnes in 2000.

Table 24: Production of different types of tea in India

Zone/Tea Type	1998		1999		2000	
	Tonnes	% Share	Tonnes	% Share	Tonnes	% Share
North						
CTC Black	592421	88.5	556907	92.2	590931	92.1
Orthodox Black	69845	10.5	41057	6.8	45027	7.0
Green Tea	6764	1.0	6337	1.0	6003	0.9
Total	669030	100.0	604301	100.0	641961	100.0
South						
CTC Black	165737	82.3	169552	84.2	167681	82.0
Orthodox Black	33786	16.8	29905	14.9	35022	17.1
Green Tea	1852	0.9	1854	0.9	1819	0.9
Total	201375	100.0	201311	100.0	204522	100.0
All India						
CTC Black	758158	87.1	726459	90.2	758612	89.6
Orthodox Black	103631	11.9	70962	8.8	80049	9.5
Green Tea	8616	1.0	8191	1.0	7822	0.9
Total	870405	100.0	805612	100.0	846483	100.0

Amritsar in Panjab, Kolkata and Siliguri in West Bengal, Guwahati in Assam, Cochin in Kerala and Coonoor and Coimbatore in Tamil Nadu are the trading centres of tea in India where tea is sold through open auction. The sale of tea has decreased in Amritsar auctions since 1998. The tea sale price has also followed a similar trend. The different kinds of tea at other markets followed a variable trend both for sale and average price. Orthodox and all leaf tea generally fetched higher price compared to other grades (Table 25 & Figure 22).^{151,152} Sale

and average price trends were variable in south Indian auctions of tea (Figure 23).^{151,152} The average sale price was highest for orthodox tea in all the auctions.

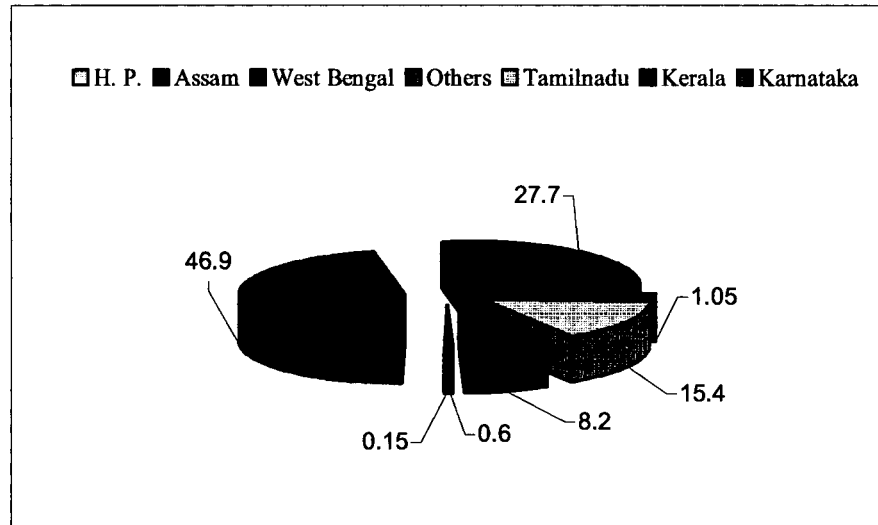


Fig. 21: State wise production share of tea in India (2000)

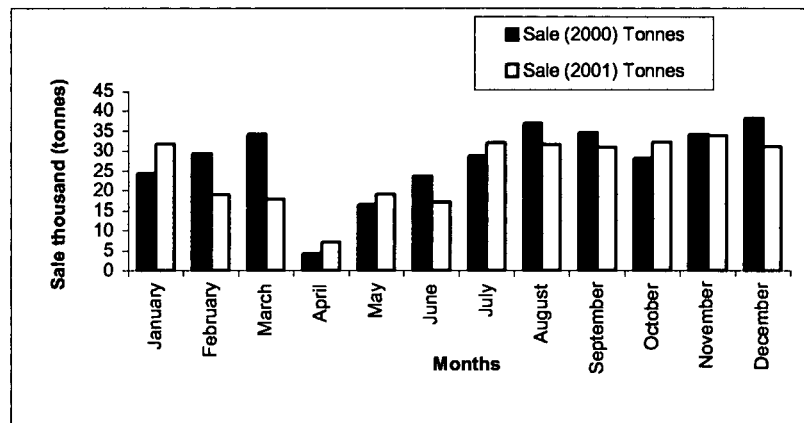


Fig. 22: Monthly sales in north Indian auction for All tea

Table 25: Sales through auctions of different kinds of black tea in India

Market	Type of Tea	1998			1999			2000			2001		
		Quantity (tonnes)	Price (IRs/kg)	Quantity (tonnes)	Price (IRs/kg)	Quantity (tonnes)	Price (IRs/kg)	Quantity (tonnes)	Price (IRs/kg)	Quantity (tonnes)	Price (IRs/kg)	Quantity (tonnes)	Price (IRs/kg)
Amritsar	All Tea	718	50.83	472	49.31	405	48.56	319	43.05				
Kolkata	Orthodox	24469	85.48	11669	94.57	18330	95.77	19277	79.80				
	CTC	34792	84.81	49881	83.98	52147	71.74	47635	73.69				
	All Leaf	67560	91.56	67146	93.51	75674	84.93	71563	81.56				
Guwahati	All Dust	18058	79.61	22026	79.86	22439	70.08	23213	66.48				
	CTC	92377	79.98	98618	82.35	101196	69.99	94368	71.47				
	All Leaf	93222	79.94	99044	82.24	111797	70.08	94995	71.47				
Silliguri	All Leaf	58192	77.29	71630	74.82	64977	63.37	58449	66.71				
	All Dust	14164	69.56	15163	65.09	13859	53.93	13342	57.55				
	Orthodox	14100	80.42	14855	68.03	17664	58.10	13935	56.67				
Cochin	CTC	9607	64.94	7791	52.81	5966	38.09	4845	40.98				
	All Leaf	23707	74.22	22646	62.80	23633	52.87	18780	52.59				
	Orthodox	2110	69.08	1720	59.75	1882	49.89	1663	47.62				
Coonoor	CTC	56384	64.78	68366	53.98	57187	39.12	57351	42.25				
	Orthodox	4038	75.87	4555	64.54	6678	56.32	4023	54.75				
	CTC	7449	65.72	9566	53.71	15834	38.16	11978	40.79				
Coimbatore	All Dust	5759	67.76	6141	56.84	10763	43.64	9117	47.39				

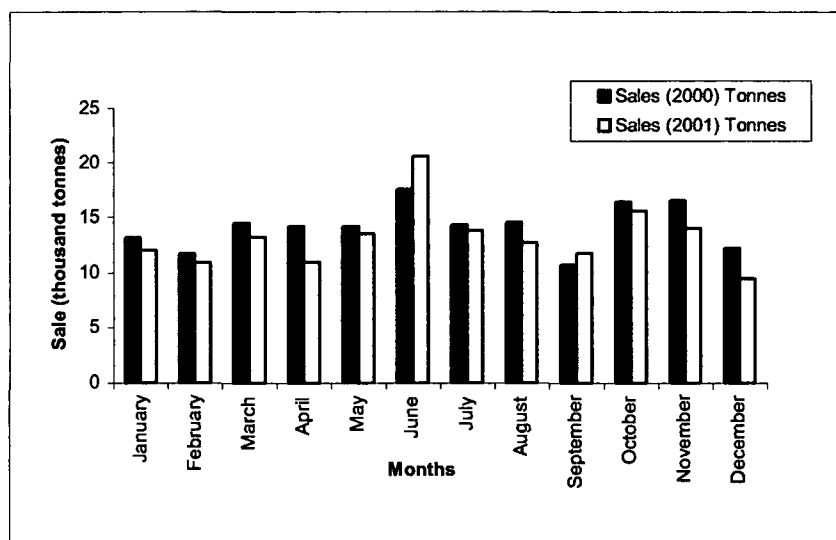


Fig. 23: Month-wise sale of All tea in south Indian auctions

6.4 Green Tea Production in India

In South India, green tea is produced only in the state of Tamilnadu. On all India basis 19.39 per cent decrease in green tea production was observed during a period of 1997 to 99 (Table 26).¹⁵¹

The state of Himachal Pradesh and Uttaranchal in north and state of Assam, West Bengal and Tripura in northeast zone are engaged in production of green tea. West Bengal is the major producer of green tea. The production of green tea has declined in North India since 1997.

6.5 Problems and Prospects of Green Tea Production

Indian green tea had good market in Afghanistan and Morocco and 50 per cent of green tea production in India was exported to these two countries. The export to Afghanistan suffered a major setback following political disturbances in that country. Some other potential markets for Indian green tea are Germany, France, USA, Tunisia, Algeria and middle-east countries. The initiative need to be taken to promote export of green tea. In India, green tea is mainly produced by small tea companies, which are financially weak. As a result, it is not possible for

them to undertake R & D to improve the quality or technique of manufacture of green tea. Old methods of manufacture are used. Green tea is manufactured by steaming followed by rolling. A substantial amount of components are lost from the leaf during the process leading to lower quality, which is the major drawback of this process. The other methods such as frying or panning and devising sophisticated sorting machines, which can remove both smaller and bigger stalks, can produce better grades of tea.

Table 26: Green tea production in India

Zone/ State/District (Region)	Production (Tonnes)		
	1997	1998	1999
North zone			
Himachal Pradesh			
Kangra (Palampur)	755	648	600
Uttaranchal	260	210	175
Total	1015	858	577
Northeast zone			
Assam			
Cachar	212	178	160
Darrang	289	243	233
Dibrugarh	407	342	325
Goalpara	218	185	175
Kamrup	12	10	7
Sibsagar(a)	506	437	415
Total	1644	1395	1315
West Bengal			
Dooars	1926	2755	2221
Terai	3282	1658	1600
Total	5208	4413	3821
Total	6822	5808	5136
South zone			
Tamilnadu			
Madurai	45	43	47
Nilgiris	455	482	453
Total	500	525	500
Total	500	525	500

The lack of proper auction centres for green tea, low consumption of green tea in the domestic market and lack of knowledge regarding the technical aspects of green tea manufacture to the producers, lack of R & D facilities for upgradation of manufacturing processes are some of the main problems faced by the green tea

industry in India. In order to overcome these problems and to increase green tea production, there is a need to identify some potential areas for green tea production in each zone and cultivation of best variety of tea. At present Kashmir valley has annual domestic market of about 500 tonnes. Other markets in the country should be developed through promotional campaigns to increase the domestic consumption of green tea and make people aware of its health benefits. It is urgently required to undertake R & D activities to develop cultivars, suited for green tea production and improve upon the existing manufacturing process of green tea.

7.0 CONCLUSION

Samples of different cultivated varieties of tea were collected from all tea growing areas of the country. In one case, collection was repeated at start, mid and end of the plucking season. The hand-picked tea leaves were processed for sample preparation following usual manufacturing procedure of green tea. The leaves were steamed for 5 min and then dried in an oven at a temperature not exceeding 65 °C. Samples were also prepared by directly drying the leaves in sun immediately after plucking to compare their polyphenol profile.

Quantitative TLC was used for standardizing the extraction procedure and HPLC method was used for estimating the polyphenol content of tea samples. The TLC solvent system effecting best resolution of polyphenols in tea was developed in the laboratory. The best results were obtained using a mixture of chloroform : acetone : formic acid :: 5 : 4 : 1, which gave R_f values of 0.22 for EGCG, 0.32 for ECG, 0.27 for EGC and 0.46 for caffeine. In HPLC analysis, the mixture of water : methanol : acetic acid :: 70 : 30 : 0.5 as mobile phase gave good separation of tea polyphenols in a 15 min run. It was observed that presence of acid in solvent system of TLC and mobile phase of HPLC was essential for obtaining good chromatograms.

All analyses were performed in triplicate. The standard plots were constructed using reference samples of EGCG and ECG obtained from Sigma-Aldrich Chemicals. The estimations of EGCG and ECG in standard solutions showed excellent coefficient of correlation both in TLC ($R^2 = 0.9998$ and 0.999 for EGCG and ECG respectively) and HPLC ($R^2 = 1.0$ and 0.9999 for EGCG and ECG respectively) over a wide range of concentration. It was therefore, satisfactory to use the standard curve to determine the amount of EGCG and ECG in tea extracts. The concentration of extract was adjusted to fall in the middle range of standard plot for carrying out the analysis.

In initial experiments to optimize extraction procedure, EGCG extraction from tea samples was tried using methanol, water and ethyl acetate and it was observed (Table 8 & Figure 10) that methanol was most suitable for extraction of tea polyphenols followed by water, whereas ethyl acetate proved to be a very poor solvent. Therefore, water and methanol were further compared and investigated to optimize period and temperature of extraction.

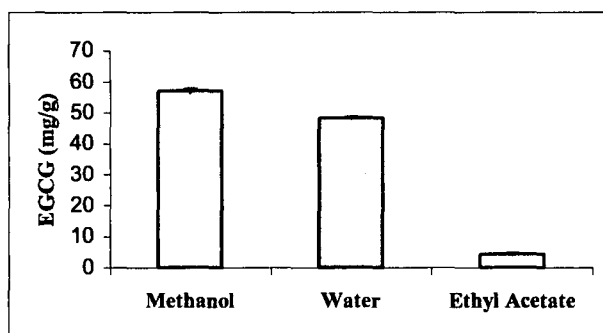


Fig. 10: Effect of solvent on extraction of EGCG

The extractions in these two solvents were done at different temperatures (40, 60, 80 and 98 °C) and time periods (15, 30, 45, 60, 75 and 90 min). In case of methanol, there was a sharp rise in EGCG extraction as the temperature was increased from 40 to 60 °C and maximum extraction was achieved in 60 min before it started declining. In case of water, similar trend was noticed as the temperature was increased from 40 to 98 °C. The maximum extraction was achieved at 98 °C in 15 min and thereafter, it remained more or less static. However, in case of methanol, it started declining after attaining a maxima. These observations indicate different rates of EGCG degradation in water and methanol. The maximum EGCG extracted in methanol was slightly higher than obtained in water but required 60 min extraction as compared to water, which produced maximum extraction in a short period of 5 min. Safety, acceptability and economical parameters of water greatly out weighed small advantage of little higher extraction in methanol. Therefore water became the logical choice as an extraction solvent.

The extraction of EGCG in water for time periods of 5, 10 and 15 min showed that the amount of EGCG obtained in 5 min did not improve further as the

extraction time was increased to 15 min after which it declined significantly. Therefore, extraction in boiling water for 5 min was selected to extract and compare polyphenols content of different tea samples.

The sample to water ratio of 1 : 50 was found most appropriate to extract EGCG. No significant increase was observed on increasing the water proportion, although at lower proportion it resulted in incomplete extraction (Table 11 & Figure 12).

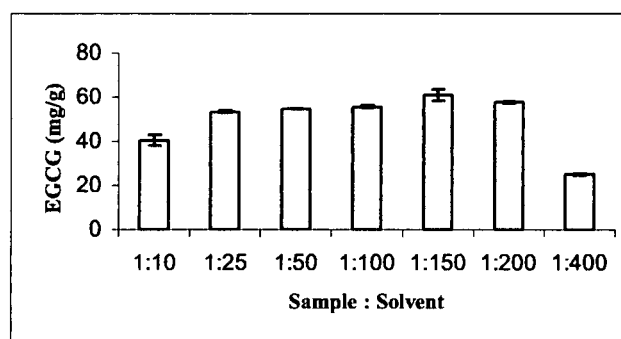


Fig. 12: Effect of sample: solvent ratio on extraction of EGCG

Shaking during extraction resulted in a highly significant increase of 43 per cent in EGCG extraction in comparison to still extraction (Table 12 & Figure 13).

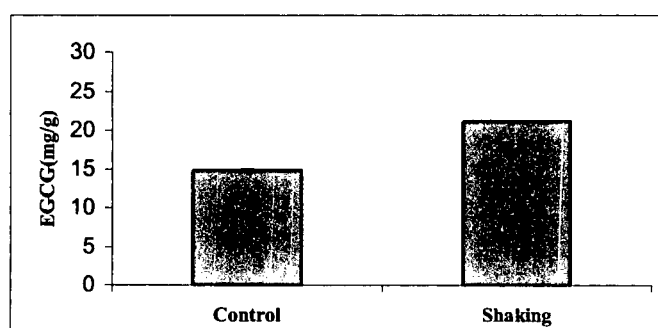


Fig. 13: Effect of shaking on extraction of EGCG

The extraction of EGCG was also affected by the particle size of the powder (Table 13 & Figure 14). Fine powder gave better extraction and moderately fine powder gave the maximum yield. Further reduction in the particle size of the powder adversely affected the extraction. These observations indicate that solvent,

temperature, duration of extraction, particle size and shaking during extraction are critical for extraction of EGCG from tea leaves. The preliminary experiments established that boiling water, 1 : 50 sample to water ratio, 5 min extraction time, moderately fine powder was most appropriate for extracting EGCG from tea leaves.

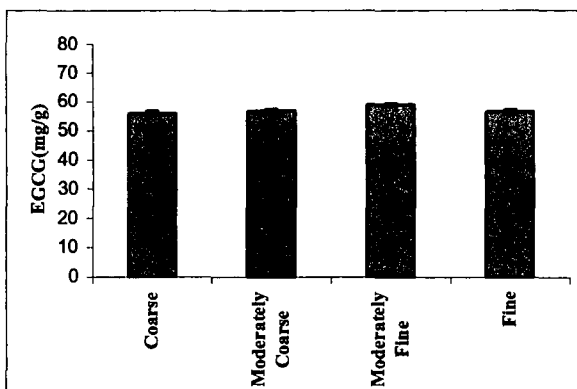


Fig. 14: Effect of particle size on extraction of EGCG

Stability of EGCG as mixture of polyphenols in tea extract was monitored using buffers of different pH. The results indicated that there was marginal (3 per cent) degradation of EGCG at pH 4 or below during first 24 h, which increased to around 7 per cent in next 24 h at pH 1.2 and 2.0 but increased to 25 per cent at pH 4 (Table 15 & Figure 15). The degradation was faster in alkaline pH and 65 per cent of EGCG degraded during first 48 h at pH 8. In water extract, pH 5.5, 43 per cent degradation was observed after 48 h.

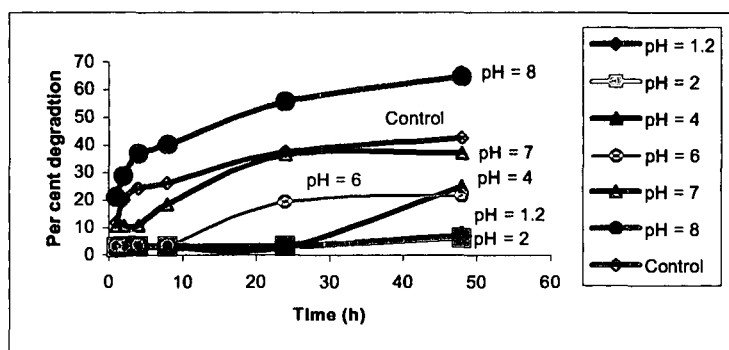


Fig. 15: Effect of pH on stability of EGCG

The EGCG in ethyl acetate extract of tea was stable and it was found that only 1.5 per cent of it degraded during first 48 h (Table 16 & Figure 16). Therefore although ethyl acetate is not a good solvent for extraction but can be used as a solvent for short storage of EGCG solution.

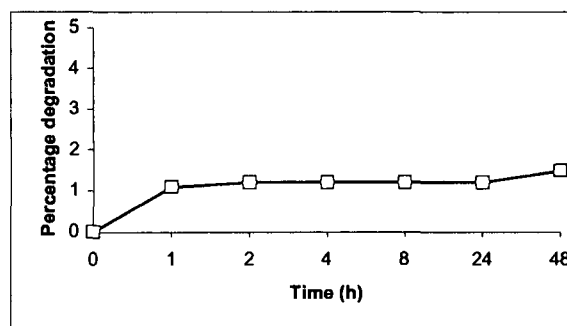


Fig. 16: Stability of EGCG in ethyl acetate extract

The EGCG and ECG content of different varieties were determined using optimized conditions of extraction and analysis developed in the laboratory. The content of EGCG and ECG in analyzed varieties varied over a wide range (0.14 to 6.88 per cent for EGCG and 0.03 to 2.74 per cent for ECG). In general, samples prepared by the method of steaming and drying under controlled temperature showed higher content of these two polyphenols. In samples collected from North India, the content of EGCG and ECG varied from 0.27 to 6.88 per cent and 0.64 to 2.73 per cent respectively. The comparison of EGCG and ECG content in tea samples collected at start and end of the plucking season showed that the polyphenol content were higher in samples collected at start of the plucking season. EGCG and ECG content of samples from northeast ranged from 1.28 to 5.01 per cent and 0.56 to 1.96 per cent respectively, whereas it ranged from 0.14 to 6.83 per cent and 0.03 to 2.22 per cent in samples from South India.

Among different cultivars, steamed samples of Kangra Jawala, a variety developed at Department of Tea Husbandry and Technology, Himachal Pradesh Krishi Vishvavidalya, Palampur (North India) and TRI-2026 developed at United Planters Association of South India showed the highest content of EGCG (6.88 and 6.83 per cent respectively). The TV-1 variety from Palampur showed highest

content of ECG (2.74 per cent) among all the samples analysed. EGCG and ECG content of TV-1 & TV-23 in samples from Palampur was marginally higher than samples from northeast.

8.0 SUMMARY

There are three zones of cultivation and production of tea in India. The black tea constituted 99.07 per cent of total production of tea in the year 2000. The bulk of green tea is produced in northeast zone (West Bengal and Assam) followed by north zone (Himachal Pradesh) and very small quantities are produced in south zone (Tamilnadu). The production figures from 1998 to 2000 are compiled in this report along with the auction prices. The production of green tea has decreased from 8616 to 7822 tonnes during 1998-2000. Various factors responsible for low production of green tea in India have been highlighted in this report and views of some companies visited are produced in the Annexure.

The extraction procedure of EGCG from green tea leaves was optimized with respect to solvent, time, temperature, particle size and sample to solvent ratio. EGCG can be extracted with boiling methanol or water and the extraction is quicker in water. Maximum amount of EGCG is extracted in 50 times by weight of water in 5 min using moderately fine powder of tea leaves. The ethyl acetate has been proved to be a poor solvent for extraction of polyphenols. These constituents were found to degrade rapidly in weakly acidic to alkaline solution.

A large number of cultivars of tea have been developed which are cultivated in India. In the present study, 26 prominent cultivars were analysed. Region wise, Kangra Jawala, grown in Palampur (North India) showed the highest content of EGCG (6.88 per cent) followed by TRI-2026 (6.83 per cent) grown in Coonor (South India) and Kangra Asha (6.44 per cent) grown in Palampur. The lowest content of EGCG was recorded in P-126 (1.29 per cent), Tenali-17 (1.76 per cent) and TV-20 (1.92 per cent) grown in Darjeeling. The samples of two varieties TV-1 and TV-23 were collected from two regions- north (Palampur) and northeast (Darjeeling) and the content of EGCG were lower in samples of northeast, suggesting possible role of agro-climatic conditions. Deactivation of enzymes immediately after plucking by steaming is essential as observed from very low content of EGCG in sun dried samples.

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APPENDIX-1

BUFFERS USED IN STABILITY STUDIES OF EGCG

Hydrochloric acid buffer pH 1.2	Add 85 ml of 0.2 M hydrochloric acid to 50 ml of 0.2 M potassium chloride solution. Make the volume upto 200 ml with water.
Hydrochloric acid buffer pH 2.0	Add 13 ml of 0.2 M hydrochloric acid to 50 ml of 0.2 M potassium chloride solution. Make the volume upto 200 ml with water.
Phosphate buffer pH 4.0	Dissolve 6.8 g of potassium dihydrogen orthophosphate in 700 ml of water. Adjust the pH with 10% v/v orthophosphoric acid. Add water to make 1000 ml.
Phosphate buffer pH 6.0	Dissolve 5.6 ml of 0.2 M NaOH solution to 50 ml monobasic potassium phosphate. Add water to make 200 ml.
Phosphate buffer pH 7.0	Add 29.1 ml of 0.2 M NaOH solution to 50 ml monobasic potassium phosphate. Add water to make 200 ml.
Phosphate buffer pH 8.0	Add 46.1 ml of 0.2 M NaOH solution to 50 ml monobasic potassium phosphate. Add water to make 200 ml.

APPENDIX -2

PARTICLE SIZE DESCRIPTION OF POWDER USED IN EXTRACTION

Coarse powder	All passes through 2000 μ sieve and not more than 40 % passes through 350 μ sieve.
Moderately coarse powder	All passes through 710 μ sieve and not more than 40 % passes through 250 μ sieve.
Moderately fine powder	All passes through 355 μ sieve and not more than 40 % passes through 180 μ sieve.
Fine powder	All passes through 180 μ sieve.

APPENDIX-3

MONTHLY SALES OF ORTHODOX, CTC, ALL LEAF AND ALL DUST TEA IN AUCTION MARKETS OF NORTHEAST ZONE (KOLKATA, GUWAHATI AND SILIGURI).¹⁵²

Monthly sales of Orthodox tea in Kolkata auction market

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	3232	91.48	1672	67.63	1314	84.50	2328	66.05
February	2296	89.02	1571	65.02	765	78.30	2379	58.23
March	822	76.94	789	64.74	358	83.10	851	57.33
April	592	93.39	372	62.28	178	101.32	244	75.64
May	1526	80.23	486	96.84	1246	99.65	719	83.46
June	2468	85.16	373	106.74	1192	99.41	874	108.59
July	2779	95.12	602	133.74	2227	121.77	1845	104.18
August	3110	96.37	1086	116.79	2110	111.98	1905	95.79
September	1692	84.81	764	115.89	2423	104.55	2224	91.02
October	1315	80.08	871	112.48	2065	100.37	2053	77.14
November	2484	80.24	1644	102.90	2148	87.11	1719	70.88
December	2153	72.98	1439	89.88	2304	77.24	2136	68.12

Monthly sales of CTC tea in Kolkata auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	2970	101.41	5116	74.46	5588	77.74	5684	88.80
February	1668	95.29	5134	72.01	5666	64.27	4588	82.13
March	1511	83.95	3401	63.63	3480	57.79	3998	63.51
April	1502	90.06	832	80.58	703	60.61	1070	72.69
May	2585	83.47	1857	99.74	2544	78.26	2846	79.54
June	3185	87.47	3834	91.20	2966	77.23	2350	84.75
July	3025	80.92	4413	88.73	4978	82.13	4228	80.33
August	4140	80.41	6311	83.90	4503	75.33	3887	70.90
September	2722	81.51	3798	88.08	4544	74.83	4154	66.51
October	2007	81.20	4134	93.34	5064	74.58	4861	59.85
November	4915	79.13	6095	91.01	5874	67.40	4529	64.56
December	4562	73.97	5356	81.09	6237	70.82	5440	70.73

Monthly sales of all leaf tea in Kolkata auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	6551	98.54	7261	73.73	7313	80.93	8400	84.26
February	4136	92.86	7011	70.75	6648	66.54	7143	74.60
March	2400	83.53	4358	65.06	3990	60.72	4957	64.11
April	2284	111.62	1310	98.39	957	95.21	1497	96.85
May	4619	95.57	2567	117.39	4169	99.16	3904	91.28
June	6201	93.67	4613	106.41	4590	92.85	3532	98.29
July	6415	99.34	5589	110.30	7933	106.77	6818	99.27
August	7939	92.10	8095	95.49	7187	92.66	6338	85.14
September	4798	87.46	5048	98.39	7614	88.63	6833	77.57
October	3714	85.88	5535	101.69	7678	84.28	7331	67.09
November	8012	82.45	8358	96.96	8555	75.85	6635	68.12
December	7254	75.76	7401	87.63	9040	75.61	8175	72.24

Monthly sales of all dust tea in Kolkata auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	1561	97.19	2314	65.35	2522	74.09	3081	86.47
February	1457	90.67	2091	58.20	2347	59.63	2368	74.67
March	775	75.00	1753	51.26	2056	50.28	2346	50.21
April	322	86.06	231	63.47	324	50.27	486	47.26
May	938	78.62	536	100.61	658	83.93	855	77.89
June	1270	82.97	1319	93.51	1039	76.80	1049	86.16
July	1524	76.86	1817	91.93	1985	82.56	2183	77.45
August	2441	73.52	2974	83.99	2024	72.22	1764	66.77
September	1728	76.40	1969	89.99	2550	75.63	1839	62.40
October	767	77.67	2006	92.58	1751	73.98	2116	56.84
November	3157	72.94	2767	88.45	2300	70.86	2147	55.50
December	2118	67.73	2249	79.06	2883	70.80	2979	56.22

Monthly sales of CTC leaf tea in Guwahati auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	8509	98.33	8292	70.90	8279	75.73	9322	87.27
February	6477	93.87	8014	68.67	9882	66.05	4323	76.60
March	4114	75.64	7076	63.41	13036	55.12	3960	56.79
April	4035	82.97	1070	85.32	1246	68.07	2081	79.02
May	7758	77.50	2964	99.12	6767	76.95	6704	78.16
June	9240	81.71	8960	89.59	7598	76.13	6262	83.18
July	8457	76.11	9711	82.31	9118	80.39	11403	75.73
August	8885	76.06	14802	80.42	12787	71.40	9343	68.40
September	9131	74.44	9496	89.17	10960	69.31	9947	64.27
October	8529	76.69	7499	92.42	9738	68.07	11132	59.36
November	8513	75.63	10770	88.20	10674	63.71	10493	62.34
December	8729	70.82	9964	78.68	10981	69.00	9398	66.53

Monthly sales of all leaf tea in Guwahati auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	8597	98.30	8333	70.94	8313	75.69	9405	86.99
February	6495	93.67	8057	68.50	9905	66.04	4391	75.97
March	4114	75.74	7093	63.43	13063	55.14	3982	56.70
April	4092	82.99	1095	84.14	1257	67.98	2091	78.90
May	7809	77.44	2973	99.04	6817	76.96	6718	78.17
June	9377	81.55	8999	89.50	7632	76.13	6292	83.13
July	8545	76.13	9727	82.32	9187	80.39	11478	75.66
August	8937	76.12	14859	80.43	12859	71.48	9416	68.37
September	9172	74.46	9555	89.16	11035	69.34	10015	64.27
October	8679	76.63	7526	92.48	9857	69.17	11179	59.37
November	8568	75.61	10825	88.22	10803	63.72	10560	62.36
December	8837	70.64	10002	78.72	11069	68.96	9468	66.47

Monthly sales of all leaf tea in Siliguri auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	5760	91.86	5740	70.72	4926	69.76	5092	78.01
February	3142	83.87	5213	65.73	5724	59.52	2246	75.04
March	1532	72.01	4893	57.88	6752	50.57	3016	59.74
April	2009	86.79	2286	79.76	891	71.08	1866	71.78
May	4952	77.94	2670	86.96	2433	70.36	4705	72.96
June	3593	82.07	5182	82.19	6061	70.64	3218	77.73
July	6300	74.54	7968	75.95	4650	68.58	5549	69.43
August	5741	72.78	7094	71.29	7567	62.83	7354	63.73
September	6007	72.23	10228	75.77	6937	60.22	6234	56.94
October	6179	72.89	5928	82.21	3554	61.41	5909	54.63
November	6601	70.77	6634	78.15	6647	55.90	7983	59.93
December	6376	69.75	7794	71.26	8835	59.57	5277	60.86

Monthly sales of all dust tea in Siliguri auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	1500	88.02	1110	56.99	846	66.91	1260	66.11
February	827	78.31	1273	48.22	955	55.24	625	64.69
March	421	67.78	1413	43.3	1612	39.82	718	49.53
April	238	82.50	376	60.87	150	49.42	350	57.94
May	1000	70.80	401	80.94	375	64.82	812	66.38
June	888	70.22	894	76.07	1215	64.08	723	71.91
July	2025	61.23	1815	70.47	1168	60.85	1232	62.33
August	1746	60.95	1281	64.59	1873	52.23	2017	53.70
September	1570	63.95	2446	66.61	1626	47.62	1514	49.94
October	1365	65.75	1475	72.98	833	49.30	1198	45.65
November	1393	63.38	1304	72.52	1576	45.49	1835	49.19
December	1191	61.83	1375	67.53	1630	51.39	1058	53.30

MONTHLY SALES OF ALL TEA (BLACK AND GREEN) IN AUCTION MARKET OF NORTH ZONE (AMRITSAR).¹⁵²

Monthly sales of all tea in Amritsar auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	45	32.36	21	36.18	38	40.26	14	24.53
February	54	29.54	10	32.44	11	34.68	14	26.49
March	46	17.52	30	23.41	8	23.64	7	26.56
April	30	89.25	4	111.29	9	111.05	3	82.34
May	89	65.09	22	69.74	79	71.39	38	64.62
June	101	60.41	34	57.43	87	50.38	31	59.39
July	66	60.10	51	66.64	47	58.94	33	53.28
August	52	56.81	31	64.85	27	52.69	45	44.89
September	77	58.19	90	47.84	28	45.41	32	50.25
October	64	54.11	65	49.34	27	44.69	36	39.18
November	36	46.33	78	44.84	21	25.66	38	25.38
December	58	40.26	36	37.05	23	24.02	28	19.76

Total sales of different types of tea in Amritsar auction

Year	Quantity (tonnes)			Average Price (IRs. / kg)		
	Black tea	Green tea	Total	Black tea	Green tea	Total
1997	23	721	744	18.24	42.65	60.89
1998	55	662	717	18.42	54.74	73.16
1999	10	546	556	47.31	12.48	59.79

Monthly sales of different types of tea in Amritsar auction

Months	BLACK TEA					
	Quantity in tonnes.			Average Price (IRs. / kg)		
	1997	1998	1999	1997	1998	1999
January	9	18	2.70	23.91	24.88	36.64
February	8	-	-	12.60	-	31.06
March	5	32	5.70	12.01	13.76	25.64
April	-	-	-	-	-	111.29
May	-	-	-	-	-	69.74
June	-	-	-	-	-	57.42
July	1	-	-	-	-	66.64
August	-	3	-	28.02	27.95	64.89
September	0	-	-	37.04	-	38.80
October	-	1	-	-	23.36	49.20
November	-	-	2.00	-	-	44.78
December	-	1	-	-	17.62	42.98
				GREEN TEA		
January	44	27	36.20	22.10	37.36	12.50
February	17	54	15.10	29.65	29.58	-
March	24	14	24.90	25.25	26.32	12.35
April	11	30	3.40	73.87	89.24	-
May	62	89	22.10	59.01	65.09	-
June	79	102	34.00	45.98	60.42	-
July	80	65	51.50	39.21	60.10	-
August	49	49	31.10	41.31	58.64	-
September	60	77	143.20	41.32	58.18	-
October	137	63	65.40	42.43	54.35	-
November	83	36	100.50	40.88	46.41	12.48
December	75	56	18.50	49.62	40.64	-

**MONTHLY SALES OF ORTHODOX, CTC, ALL LEAF AND ALL DUST TEA
IN AUCTION MARKETS OF SOUTH ZONE (COCHIN, COONOR AND
COIMBTORE).¹⁵²**

Monthly sales of CTC leaf tea in Cochin auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	1376	82.42	651	58.64	542	43.99	595	45.25
February	927	83.26	837	59.17	642	41.48	473	49.11
March	837	82.46	716	55.03	402	40.9	398	49.21
April	864	69.1	664	51.58	329	38.11	365	44.16
May	602	56.89	539	51.46	653	36.36	676	38.92
June	660	55.76	550	47.59	462	37.11	537	37.37
July	946	55.03	588	49.14	417	37.48	350	38.98
August	667	64.2	607	53.8	640	38.3	301	40.16
September	884	52.62	562	56.88	341	38.62	315	37.66
October	647	53.55	549	50.28	582	36.98	362	35.2
November	571	63.1	857	54.46	500	33.56	264	36.65
December	626	60.94	671	45.79	458	34.3	209	38.2

Monthly sales of Orthodox leaf tea in Cochin auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	1535	87.89	1062	69.57	1285	61.54	1922	53.49
February	1135	92.2	1094	71.38	1110	69.37	1229	56.87
March	1062	93.62	1043	67.79	1411	63.55	1101	58.79
April	1082	87.49	1002	67.81	1909	54.6	1055	57.45
May	746	80.54	911	63.63	1997	54.44	1303	55.35
June	1197	80.17	1364	57.08	1490	54.23	1835	49.79
July	1640	81.23	1058	65.99	1407	56.74	856	50.52
August	1093	84.35	1083	76.3	1533	58.22	530	59.65
September	1271	71.15	1278	78.33	813	61.07	975	62.98
October	1342	62.05	1546	72.47	2040	60.37	1068	59.66
November	825	71.51	1798	67.81	1411	51.81	1194	55.49
December	1172	72.51	1616	58.3	1258	51.30	867	60.03

Monthly sales of all leaf tea in Cochin auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	2912	85.3	1712	65.42	1827	56.34	2517	51.54
February	2061	88.2	1932	66.09	1752	59.15	1702	54.71
March	1899	88.71	1760	62.6	1814	58.52	1499	56.25
April	1946	79.32	1665	61.34	2238	52.18	1420	54.04
May	1348	69.93	1450	59.06	2649	49.99	1979	49.74
June	2455	71.72	1914	54.39	1952	50.17	2372	46.98
July	1989	71.46	1646	59.97	1824	52.34	1206	47.17
August	1759	76.7	1690	68.23	2173	52.35	831	52.58
September	2155	63.54	1840	71.74	1154	54.44	1290	56.79
October	1989	59.23	2095	66.66	2621	55.18	1430	53.47
November	1396	68.12	2655	63.53	1912	47.03	1458	52.08
December	1798	68.46	2287	54.63	1717	46.76	1076	55.8

Monthly sales of CTC leaf tea in Coonoor auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	6445	82.37	5381	61.89	4705	45.47	2945	46.21
February	4276	84.35	4015	62.45	2785	44.07	3284	51.73
March	3919	82.82	5321	56.24	4644	42.18	4740	52.08
April	4597	70.14	4969	52.75	5021	36.68	3996	47.63
May	3140	57.32	4909	51.96	3835	38.44	4611	42.23
June	4063	59.49	5759	47.97	6749	37.55	8571	39.41
July	5514	58.84	8108	51.44	5132	37.66	5772	40.21
August	4664	61.69	5957	55.1	4377	40.48	5244	40.64
September	4983	53.64	6763	57.77	3969	38.91	4321	38.91
October	5784	53.92	6613	53.64	5443	38.83	5731	34.25
November	4575	50.46	3667	52.29	6165	33.26	4974	36.49
December	4424	62.39	6904	44.35	4362	35.95	3162	37.3

Monthly sales of orthodox leaf tea in Coonoor auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	200	81.07	130	60.78	134	56.35	114	46.39
February	109	84.77	62	64.12	79	63.44	82	45.69
March	188	79.18	140	60.15	169	55.11	146	49.84
April	276	69.97	164	56.97	225	45.89	102	46.00
May	128	65.83	158	55.58	102	50.69	122	47.31
June	167	67.28	137	49.63	180	48.65	263	44.22
July	192	69.22	182	57.65	135	48.08	128	44.01
August	118	76.77	118	66.16	148	46.81	104	46.83
September	173	61.81	147	70.95	134	52.47	98	52.13
October	291	55.40	156	63.85	162	47.28	125	50.49
November	163	58.55	109	59.23	240	43.79	220	47.95
December	105	59.19	217	51.97	174	40.12	159	50.58

Monthly sales of CTC leaf tea in Coimbatore auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	1193	83.06	899	61.37	1026	45.49	785	44.86
February	318	85.67	703	61.71	1146	42.87	692	50.35
March	634	82.66	628	56.72	1498	41.17	1155	50.23
April	530	70.81	722	53.14	1055	35.84	845	46.43
May	423	57.05	416	50.27	1143	37.02	1069	39.60
June	261	56.62	373	47.12	1822	36.06	1449	37.30
July	634	58.15	638	51.19	1514	36.89	1173	39.33
August	493	61.67	549	54.57	1479	38.66	1262	38.40
September	645	53.90	816	57.36	1078	38.10	930	37.67
October	895	52.79	1336	52.72	1262	38.68	1110	33.53
November	588	64.65	1558	52.33	1494	32.94	897	35.80
December	835	61.64	928	46.13	1317	34.29	611	36.00

Monthly sales of orthodox leaf tea in Coimbatore auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	683	85.68	384	64.20	389	61.73	212	50.42
February	228	89.64	173	64.91	494	68.69	352	52.47
March	441	87.56	353	60.42	736	61.52	480	57.13
April	250	81.58	325	59.87	526	51.89	304	54.47
May	222	76.51	305	59.25	587	54.14	283	55.29
June	325	75.16	215	55.23	704	52.82	771	49.85
July	391	77.95	197	64.52	612	55.88	407	50.74
August	214	81.08	165	74.98	430	58.04	238	57.75
September	340	64.48	506	76.99	519	58.34	199	63.62
October	400	56.76	472	72.72	620	57.28	235	58.70
November	238	66.98	854	65.91	614	49.34	304	52.86
December	306	67.08	606	58.49	447	46.18	238	53.72

Monthly sales of all dust tea in Coimbatore auctions

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	595	81.38	597	61.76	799	50.93	622	52.09
February	276	85.32	377	58.22	697	50.18	605	54.96
March	416	78.58	350	58.88	1039	44.72	902	48.16
April	412	74.52	465	57.88	615	40.75	529	46.00
May	439	68.71	459	55.13	662	43.74	502	47.00
June	497	57.54	514	51.22	1131	41.23	971	42.25
July	676	55.06	590	53.21	1075	39.47	706	46.11
August	349	65.36	417	53.96	1030	42.58	763	49.47
September	487	61.01	386	62.35	918	42.81	764	49.27
October	676	59.39	659	56.20	877	41.71	932	44.21
November	432	63.17	666	59.17	1086	39.87	1231	44.60
December	504	63.19	661	54.21	833	45.71	590	44.59

Monthly sales of North Indian tea in auctions (All Tea)

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	28084	96.58	28494	69.34	27429	75.39	31820	83.33
February	19432	90.09	27891	64.68	29382	63.49	19034	73.85
March	11496	76.14	23543	58.91	34201	52.25	17908	56.57
April	10023	90.29	5961	80.78	4219	70.45	7176	75.61
May	22915	80.05	10149	99.53	16559	81.47	19221	78.49
June	25908	83.29	24468	91.11	23636	77.53	17325	85.19
July	29776	78.95	31140	86.38	28948	84.87	32121	78.77
August	32203	77.49	41462	81.65	37032	72.54	31617	68.97
September	28771	75.54	33923	84.91	34621	71.10	30982	64.45
October	25406	76.53	26273	91.06	28125	72.61	32225	58.73
November	32189	75.52	34812	87.90	34299	65.48	34037	60.54
December	29478	70.85	33393	78.08	38331	67.66	31214	64.36

Monthly sales of South Indian tea in auctions (All Tea)

Months	1998		1999		2000		2001	
	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)	Tonnes	Price (IRs)
January	16397	83.71	12321	63.26	13147	51.26	12040	51.31
February	10883	85.55	10559	63.13	11714	52.07	10879	53.69
March	10952	83.06	12235	58.61	14527	47.74	13193	52.54
April	11671	75.07	11753	57.00	14238	42.70	11003	49.62
May	9109	63.81	11508	54.92	14199	44.58	13528	46.13
June	12952	61.66	13841	51.39	17602	41.53	20536	41.92
July	13681	60.54	15581	53.77	14299	42.25	13775	43.61
August	10701	66.18	12472	57.99	14602	45.43	12753	45.39
September	12375	59.29	13829	61.77	10733	45.40	11789	46.35
October	14518	57.41	15363	58.09	16443	42.00	15653	41.27
November	11344	64.99	13243	58.78	16599	40.90	14007	41.81
December	12097	64.45	16474	50.15	12270	43.35	9512	43.87

APPENDIX- 4

QUESTIONNAIRE FOR SURVEY

- 1 **Name of the Company/firm:**
- 2 **Tea brands marketed:**
 - Green Tea
 - Black Tea
- 3 **Total tea production:**

Type of Tea	Production (tonnes)	Year
Green Tea		
Black Tea		
- 4 **Tea varieties cultivated:**

Variety/clone	Area under cultivation (ha.)
- 5 **Total area (ha) under tea cultivation:**
- 6 **Tea consumption in the market:**
 - National:
 - International:
 - Countries to which tea is exported:
- 7 **Production processes for green tea:**
- 8 **Problems related to green tea:**
- 9 **Additional information, if any:**
- 10 **Contact address:**

APPENDIX-5

- 1. Name of the firm:**
Makaibari Tea Estates
Tea brands marketed:
Makaibari Tea (both for green and black tea).
Total tea production during (2001):
110 tonnes of which 40 % is green tea.
Tea varieties cultivated:
Chinese type, Assam type and their clones.
Area under cultivation (ha)
570.12 ha.
Tea consumption in the market (national & international):
80% of the total green tea production is exported to Japan, USA, UK and Germany. Only 20% is consumed in the domestic market.
Production processes for green tea:
Steaming is the basic step in green tea manufacturing.
Problems related to green tea production, plucking, packing and marketing:
Green tea is produced against the order. The main importer is Japan.
Additional information:
Major green tea grades produced by the company are: Tips Golden Flowery Orange Pekoe (TGfOP); Golden Flowery Orange Pekoe (GFOP); Broken Orange Pekoe (BOP); Orange Fanning (OF) and dust. TGfOP is the costliest grade while dust is the cheapest.
Contact address:
Mr. P. N. Banerjee
Director
Makaibari Tea Estates
Flat No. 7, 184, Lenin Sarani, Kolkata-700 013
- 2. Name of the firm:**
Sannyasithan Tea Co. Pvt.Ltd.
Tea brands marketed:
Hind Tea
Total tea production during (2001):
300 tonnes/annum of green tea.
Tea varieties cultivated:
Chinese seed, clones developed by Tocklai Research Association
Area under cultivation (ha)
125 ha.
Tea consumption in the market (national & international):
Almost all the green tea produced by the company is auctioned at Amritsar market. Kashmir is the main domestic market for the product. Small quantities are also exported to Europe.

Production processes for green tea:

Problems related to green tea production, plucking, packing and marketing:

Additional information:

Contact address:

Steaming is the basic step in green tea manufacturing. Lack of awareness of the health benefits among the consumers, organized marketing, auction centers and market statistics for green tea. Manufacturing is costly. Selling price is lower than manufacturing price.

- Mr. P. Bhartia

C.F.A.

Sannyasithan Tea Co. Pvt.Ltd.

B- ½, Gillanders House, 1st Floor

8, Netaji Subhas Road, Kolkata- 700 001

3. Name of the firm:

Tea brands marketed:

Total tea production during (2001):

Tea varieties cultivated:

Area under cultivation (ha)

Tea consumption in the market (national & international):

Production processes for green tea:

Problems related to green tea production, plucking, packing and marketing:

Additional information:

Contact address:

Maud Tea & Seed Co. Ltd. (also known as Biotea Estates Ltd.)

Directly sell tea in bulk from the gardens.

2500 tonnes, including 30 tonnes of green tea.

China variety in Darjeeling gardens and Assam variety in Assam gardens. 1700 ha.

80% of the production is exported to Germany (50%), USA (20-30%), UK, Japan and Australia.

Steaming is the basic step in green tea manufacturing.

Green tea is produced against the order. Production is costly. Crop losses are high. Green tea is not advertised properly. People are not used to the taste of green tea.

Green tea is made from the leaves taken just before the 'Banji period.'

Mr. S. Lohia and Y. Lohia

Maud Tea & Seed Co. Ltd.

1 & 2, Old Court House Corner

Kolkata- 700 001

4. Name of the firm:

Tea Promoters (India) Pvt. Ltd.

Tea brands marketed:

Total tea production during (2001):

Tea varieties cultivated:

Area under cultivation (ha)

Tea consumption in the market (national & international):

Production processes for green tea:

Problems related to green tea production, plucking, packing and marketing:

Additional information:

Bulk green tea producer.

100 tonnes, which includes 55 tonnes of green tea.

China type in Darjeeling gardens and Assam type and its hybrids in Assam gardens.

900 ha.

All the tea production is exported to Europe mainly Germany, Italy, Switzerland and UK.

Steaming or roasting are the basic steps used in green tea manufacturing.

No market for green tea.

Started green tea production in 1988. Since then production has increased significantly. Almost all (99.9 %) green tea production is exported to Europe and rest is auctioned in Amritsar market. Selling price is IRs 300 per kg for leaf grade and 150 per kg for Fannings. Green tea is mainly produced in July. Company also produces organic tea made from plants that are cultivated without fertilizers.

Mr. Binod. K. Mohan

Director

Tea Promoters (India) Pvt. Ltd.

17, Chowringhee Mansions

30, Jawaharlal Nehru Road

Kolkata- 700 016

Contact address:

5. Name of the firm:

Tea brands marketed:

Total tea production during (2001):

Tea varieties cultivated:

Area under cultivation (ha)

Ambari Tea Co. Ltd.

Cooch Behar green tea, Longview/Snow view green tea.

380 tonnes (300 tonnes green tea + 80 tonnes black tea) from Cooch Behar Tea Estates, 710 tones (300 tonnes green tea + 410 tonnes black tea) from Longview Tea Estates.

Assam type and its hybrids developed by Tocklai Research Association. 700 ha. (both the gardens).

Tea consumption in the market (national & international):

Production processes for green tea:

Problems related to green tea production, plucking, packing and marketing:

Additional information:

Contact address:

90% of the total production (green + black tea) is consumed locally and rest is exported to Germany and Japan.

Steaming is the basic step used in green tea manufacturing.

Costly production processes, sorting of green tea is slow and expensive. High stocks of green tea are held due to lack of market. From a given quantity of tea leaves 45% of black tea can be made. However, only 21% of green tea is produced resulting in considerable crop losses, which is due to loss of water. Steaming and firing are also the costly steps.

Green tea is sold at IRs. 80 per kg.

Mr. S. Agarwal

Director

Ambari Tea Co. Ltd.

5/2, Garstian Place

Kolkata- 700 001

6. Name of the firm:

Tea brands marketed:

Total tea production during (2001):

Tea varieties cultivated:

Area under cultivation (ha)

Tea consumption in the market (national & international):

Production processes for green tea:

Problems related to green tea production, plucking, packing and marketing:

Additional information:

Contact address:

Sublime Agro. Ltd.

Bulk producer of tea

250 tonnes green tea + 100 tonnes black tea

Assam type, China type, and clones (TV1-30, Tenali-17)

300 ha.

Exported directly to Germany (about 3 tonnes of green tea), the USA, Japan

Steaming

Mr. Raja Ram Upadhyay

Supervisor

Dagapur Tea Estate

Siliguri

- 7.**
- Name of the firm:** Sepoydhoorah Tea Co. Pvt. Ltd.
Tea brands marketed: Chamling both for green and black tea
Total tea production during (2001): 100 tonnes and small quantity of green tea
Tea varieties cultivated: Assam type (14%), China type (32.3%), hybrids (53%) and clones (TV-9, TV-29)
- Area under cultivation (ha)** -
Tea consumption in the market (national & international): Auctioned in Amritsar market
- Production processes for green tea:** Steaming
Problems related to green tea production, plucking, packing and marketing: Low demand in the market.
Additional information: Started green tea production in 1998. The green tea production has been abandoned since last three years.
Contact address: Mr. R. G. Vyas
 Director
 9C, Lord Sinha Road
 1st Floor, Kolkata- 700 001
- 8.**
- Name of the firm:** Goodricke Group Ltd.
Tea brands marketed: Goodricke Tea
Total tea production during (2001): 27830 tonnes
Tea varieties cultivated: -
Area under cultivation (ha) -
Tea consumption in the market (national & international): Green tea is produced against the order.
Production processes for green tea: Steaming, panning, roasting
Problems related to green tea production, plucking, packing and marketing: No big market for green tea.
Additional information: -

Contact address:

Mr. A. N. Singh
14, Gurusadaya Road
Kolkata- 700 001

9. Name of the firm:

Duncans Industries Ltd.
Duncans Tea, Sargam, Shakii etc.

Tea brands marketed:

17 000 tonnes

Total tea production during (2001):

Tea varieties cultivated:

TV-1, TV-20, TV-27, TV-19

Area under cultivation (ha)

7000 ha.

Tea consumption in the market (national & international):

Domestic market.

Production processes for green tea:

- Do not produce green tea.

Problems related to green tea production, plucking, packing and marketing:

Additional information:

Contact address:

Mr. M. Gurbaxani
31, Netaji Subash Road
Kolkata- 700 001

10. Name of the firm:

The Peria Karamalai Tea & Produce Co. Ltd.

Tea brands marketed:

CTC- producers

Total tea production during (2001):

4500 tonnes per year

Tea varieties cultivated:

Assam light leaf, Vetijan- khorijan, UPASI-clones, Akamalai clones

Area under cultivation (ha)

Appx. 1600 ha. in Annalai hills

Tea consumption in the market (national & international):

Local consumption and also exported to Russia

Production processes for green tea:

Do not produce green tea

Problems related to green tea production, plucking, packing and marketing:

Crop loss and marketing problems due to fine plucking.

Additional information:

-

Contact address:

Mr. Prakash Prashar
Vice President
Cowcoody Chambers,
234-A, Race Course Road, Coimbatore- 641018

11. Name of the firm:

United Planter's Association of Southern India (UPASI)
32 varieties has been recommended for cultivation in South India

Tea brands marketed:

Total tea production during (2001):

Tea varieties cultivated:

- UPASI-02, 03, 08, 09, 10
TRI- 2024, 2025, 2026
CR-6017 and Sri Lankan clones

Area under cultivation (ha)

Tea consumption in the market (national & international):

Production processes for green tea:

Problems related to green tea production, plucking, packing and marketing:

Additional information:

Contact address:

-
-
- Lack of market and technical know how of green tea production.
-
Mr. J. D. Hudson
Assistant Director
UPASI Tea Research Foundation
Regional Centre
Glennview, Coonoor- 643 101

12. Name of the firm:

The IndcoServe

Tea brands marketed:

CTC and Orthodox

Total tea production during (2001):

Data provided

Tea varieties cultivated:

UPASI varieties

Area under cultivation (ha)

Tea consumption in the market (national &

- Most of the green tea production is exported to Russia

- international):**
Production processes for green tea: Roasting and steaming. Roasting is preferred over steaming.
Problems related to green tea production, plucking, packing and marketing: Lack of basic knowledge on green tea production technology.
Additional information:
Contact address: Different grades of tea has been provided for analysis.
 Mr. K. Swaminaathan
 General Manager
 The Indcoserve
 Church Road
 Coonoor- 643 101
- 13. Name of the firm:** Mahalinga Ind. Co. Tea
Tea brands marketed: CTC and Orthodox
Total tea production during (2001): 700 tonnes
Tea varieties cultivated: China bush and VP-clones
Area under cultivation (ha) 1.22 ha.
Tea consumption in the market (national & international): Produces green tea by order.
Production processes for green tea: Steaming
Problems related to green tea production, plucking, packing and marketing: Lack of market.
Additional information: -
Contact address: Mr. Pitchai Pillai
 Special Officer
 Mahalinga Ind. Co. Tea, Coonoor- 643 101