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Project GE/GLO/89/001 - Five-Year Work Programme of ICGEB.
UNIDO Contract No. 93/107
ICGEB Practical Course "Preparation, Analysis and Applications of Synthetic Oligonucleotides". CINVESTAV-IPN, Mexico City, Mexico, July 5-17, 1993.

FINAL REPORT

1. Introduction.

The ICGEB Practical Course "Preparation, Analysis and Applications of Synthetic Oligonucleotides" was held in Mexico City, July 5-17, 1993, sponsored by UNIDO contract No. 93/107. The course was organized by Profs. Gabriel Guarneros and Francisco M. De La Vega of the Department of Genetics and Molecular Biology of the Centro de Investigación y de Estudios Avanzados del Instituto Politécnico Nacional (CINVESTAV-IPN), an ICGEB Affiliated Centre via CONACYT (National Council of Science and Technology), Mexico.

The course venue was the Nucleic Acids Unit of the Department of Genetics and Molecular Biology at CINVESTAV. To hold all the participants to the practical sessions, a separate laboratory within the Department was also conditioned for that purpose. Course lectures were carried out in the departmental conference room. The equipment and infrastructure used were mostly these of the Nucleic Acids Unit, but several departmental facilities and Dr. Guarneros' own laboratory were used also.

2. Additional Resources Obtained.

Besides the main sponsorship awarded by ICGEB/UNIDO, the organizers obtained additional support sources that were indispensable for the success of the course. Very important was the support received from Applied Biosystems Division of Perkin Elmer, USA, that consisted in demonstration equipment and supplies, as well as the paid participation of two scientists as course staff. Furthermore, the Department of Genetics and Molecular Biology allocated a small budget, and CONACYT, Mexico, also contributed to fund the participants' expenses. Numerous reagent gifts were received as donations from a number of comercial companies (listed in the course manuals cover).

An overall balance of the course costs follows; a more detailed financial balance of UNIDO grant expenditures is appended to this report.

INCOME

ICGEB/UNIDO grant (First Payment)	US$16,000
Dept. of Genetics and Molecular Biology	US$6,165.05
CONACYT, Mexico	US$3,500
Total	US$25,665.05

EXPENSES

Course Staff Travel and Subsistence costs	US$9,900.78
Participants local expenses	US$8,158.76
Reagents	US$6,094.44
Small equipment	US$3,156.75
Social events	US$1,448.20
Secretarial Assistance	US$500
Management costs and various	US$477.09
Total	US$29,736.02

Balance due:	*(US$4,070.97)*[1]

Considering an additional estimated contribution of around US$8,000 from Applied Biosystems support (travel and local expenses of ABD scientists, transportation costs for demonstration equipment and reagents donated), the total estimated cost for the Course is around US$38,000.

3. Applications & Selection Procedure.

Advertisement for the course was done via a small poster sent to all the focal points of ICGEB member countries, ICGEB Trieste Component, selected Mexican and Latin American Institutions and Research Centers, and through BIOSCI E-Mail newsgroups and ICGEBnet BBS. As a result of this effort, 51 applications were received from throughout the world.

[1] This deficiency was defrayed temporarily through a loan from the Dept. of Genetics until UNIDO second payment is received.

Applicants from Latin American countries:

Mexico	15
Argentina	7
Brazil	1
Bolivia	1
Colombia	5
Costa Rica	1
Chile	1
Ecuador	1
Peru	2
Venezuela	1

From the rest of the world we received:

Algeria	2
Egypt	1
Germany	1
Hungary	1
India	4
Italy	1
Pakistan	4
Sudan	1
Thailand	1

We selected 15 participants for the practical course based in the following criteria:

a) Examination of the C.V. and list of publications
b) Citizenship to ICGEB Member Countries
c) Previous experience in the basics required for the course and in scientific research (this was an advanced course).
d) Direct involvement in research where the knowledge acquired in the course could be applied.
e) Possibility of local benefit due to the transfer of new technologies (at the Institutional and country levels).
f) A balance of countries in order to meet the proposed goal of 5 mexican, 5 latin american and 5 from the rest of the world participants.

From the first selected group two candidates resigned which were substituted by two runner-ups. Another participant did not arrived, and was substituted within the second day by a Mexican participant. One participant was selected from a non-ICGEB

member country (Germany) due to his high academic merits. Finally, the course participants were the following:

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16132 Genova, ITALIA. TEL: (010) 3534.1 - centralino; FAX: (010) 35 29 99

Alvaro Pedro Miquel Poblete, Departamento de Bioquímica, Facultad de Medicina, Universidad de Chile, Casilla 70086, Santiago 7, Chile. FAX: (562) 737 63 20

Beatriz Eugenia Jiménez Moraila, CINVESTAV-Irapuato, Apado. Postal 629, Irapuato, Gto. TEL: (462) 5 16 00; FAX: (462) 512 82

Eva Adilia Avila Muro, INtituto de Investigación en Biología Experimental, Facultad de Química Universidad, Universidad Autónoma de Guanajuato, Noria Alta S/N, 06350 Guanajuato, Gto. TEL: (473) 243 02 - 249 96; FAX: (473) 242 50

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Jorge Florin-Christensen, Universidad del Salvador, Tucumán 1845, Piso 10, 1050 Buenos Aires, Argentina. FAX: (541) 664 75 56

Kailash Chand Gupta, Nucleic Acids Research Laboratory, Centre for Biochemical Tecnology, Mall Road, Delhi Univ. Campus, Delhi 110007, INDIA. TEL: (91 11) 725 72 98; FAX: (91 11) 436-28 84

Mahmoud Rifaat, Agricultural Genetic Engineering Research Institute, Agricultural Research Center, AGERI, ARC, 9 Gamao Street, Giza, Egypt
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Pablo Daniel Ghiringhelli, Instituto de Bioquímica y Biología Molecular, Facultad de Ciencias Exactas, Universidad Nacional de La Plata, 47 y 115, (1900) La Plata, Argentina. TEL: (54 21) 25 04 97; FAX: (54 21) 25 92 23

Rocío de María Inga, Instituto de Medicina Tropical "Alexander Von Humboldt", Universidad Peruana Cayetano Hereria, Apdo. Postal 4314

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Rubén Paul Gaytan Colín, Instituto de Biotecnología, Universidad Nacional Autónoma de México, Apdo. Postal 510, 62271 Cuernavaca, Mor. TEL: (73) 13 88 11, 17 27 99; FAX: (73) 17 23 88

Thomas Rupp, European Molecular Biology Laboratory, Meyerhofstr, 1 6900 Heidelberg-Germany. TEL: (49) 6221-38 73 94; FAX: (49) 6221-38 73 06

Victor Jimenez Cabriales, Centro de Ingeniería Genética y Biotecnología, P. O. Box 6162, Cd. de la Havana, Cuba. FAX: (537) 21 80 70

4. Course Staff.

The organizers contacted outstanding scientists in the field to compose the staff of the course. Previous experience in oligonucleotide research and in international teaching of the staff guaranteed the success of the course. They designed a series of lectures and practical sessions in accordance with the course goals previously set by the organizers.

The course staff was composed as following:

Prof. Dr. Hartmut Seliger, Sektion Polymere, Universität Ulm, Albert-Einstein-Allee 11, D 7900, Ulm, Germany. Tel. (731) 502-3161; Fax (731) 502-2038.

Dr. J. Flavio Raimaho Ortigao, Sektion Polymere, Universität Ulm, Albert-Einstein-Allee 11, D-7900, Ulm, Germany. Tel. (731) 502-3161; Fax (731) 502-2038; E-Mail ortigao@rz.uni-ulm.dbp.de

Dr. Michael Meklenburg, Department of Pure and Applied Biochemistry, Chemical Center, Univerity of Lund, P.O. Box 124, S-221 00, Lund, Sweden

Dr. Alexander Andrus, DNA and Peptide Synthesis R & D Manager, Applied Biosystems Division of Perkin Elmer, 850 Lincoln Centre Drive, Foster City, CA 94404, USA. Tel. (415) 570-6667; Fax (415) 572-2743.

Dr. Frank Stephenson, Training Department Manager, Applied Biosystems Division of Perkin Elmer, 850 Lincoln Centre Drive, Foster City, CA 94404, USA. Tel. (415) 570-6667; Fax (415) 572-2743.

Technical Assistance:
Jose Manuel Galindo. Dept. of Genetics, CINVESTAV-IPN, A.P. 14-740, Mexico D.F. 07000, Mexico. Tel. (525) 754-0200; Fax (525) 752-6330;

5. Logistics.

A secretary was hired to help with the course organization, orders, accounting and eventual chores. A file is kept with all the correspondence, applications, copies of orders, receipt and invoices, and several other relevant information regarding the course organization. The original invoices and receipts covering the expenses of the course are kept at CINVESTAV's Accounting Dept., and are available for examination upon request. The services of a travel agency were also required to obtain airfares and accommodations for both professors and students. Professors and participants were housed in a hotel located near downtown Mexico City, and at about 20 mins from CINVESTAV. Accommodations for the participants were held from Sunday, July 5, to Sunday, July 18. Transportation was arranged daily to and from the hotel. Arrangements were made to have lunch at CINVESTAV's dinning room at the participants expense. Pocket money was made available to participants in three payments (US$20 per day). A welcome cocktail and a closing dinner were carried out as social activities. In the closing dinner diplomas were delivered to participants (a sample of it is included in Apendix 2).

6. Programme.

The course consisted of about 100 hours of lectures and practical sessions that covered the topics proposed previously. The work schedule covered Monday through Friday, 9-18:30 hrs and Saturdays 9-13 hrs, including one hour for lunch and coffee break.

The final course lecture and practical programme follows.

Monday, 5 July

09:00	**Welcome Address**	**G. Guarneros**
09:20	**Introduction to oligonucleotide applications**	**F. Raimaho**
10:00	**Solid-phase oligonucleotide chemistry**	**H. Seliger**
10:45	**Coffe Break**	

11:00	Practical Session: Preparation of synthetic oligonucleotides and troubleshooting	**F.Stephenson/ F. Raimaho**
13:30	Lunch	
15:00	Practical Session: Contd.	**F. Raimaho/ F. Stephenson**

Tuesday, 6 July

09:00	Antisense technology and chemistry	**F.Raimaho/ H. Seliger**
10:30	Coffe Break	
11:00	Practical Session: Labeling & Purification of oligonucleotides	**F. Raimaho/ M. Meldenburg**
13:30	Lunch	
15:00	Practical Session: Contd.	**F. Raimaho/ M. Meldenburg**
17:30	Disscusion Session	

Wednesday, 7 July

09:00	Oligonucleotide hybridization	**F. Raimaho**
10:00	Coffe Break	
10:30	ABCs of PCR	**M. Meldenburg**
11:30	Practical: Antisense inhibition	**F. Raimaho**
14:00	Lunch	
15:00	Practical: Contd.	**F. Raimaho**

Thursday, 8 July

09:00	Gene synthesis & Genetic Polymers	**F. Raimaho**
10:45	Coffe break	
11:00	Automated fluorescence DNA sequencing	**F. Stephenson**
13:30	Lunch	

15:00 Practical: PCR for sequencing — **F. Stephenson**

17:30 Discussion Session

Friday, 9 July

09:00 Solid phase DNA sequencing — **F. Ralmaho**

10:15 Coffe Break

10:30 Practical Session: Automated sequencing — **F. Stephenson**

13:30 Lunch

15:00 Practical Session — **F. Stephenson**

17:30 Discussion Session

Saturday, 10 July

09:00 Tools to design primers for sequencing & PCR — **F. De La Vega**

10:00 Coffe Break

10:15 Practical Session: RNA synthesis — **F. Ralmaho**

14:00 Lunch

Monday, 12 July

09:00 Automated RNA synthesis — **A. Andrus**

10:30 Coffe Break

10:45 Ribozyme studies — **F. Ramalho**

12:00 Practical Session: Capillary Electrophoresis — **F. Stephenson**

13:30 Lunch

14:30 Practical: Ribozyme experiment — **F. Ralmaho**

18:00 Discusion Session

Tuesday, 13 July

09:00 Oligonucleotide Labeling Methods	**A. Andrus**
10:00 Coffe Break	
10:15 Cloning systems	**M. Meklenburg**
11:30 Practical: PCR cloning and expression	**M. Meklenburg**
13:30 Lunch	
14:30 Practical: Oligonucleotide directed mutagenesis	**M. Meklenburg**
17:30 Discussion Session	

Wednesday, 14 July

09:00 Analysis and Purification of oligonucleotides	**A. Andrus**
10:15 Coffe Break	
10:30 Electrochemical luminiscence detection	**J. Oprandy (IGEN)**
11:00 Practical Session	**M. Meklenburg**
13:30 Lunch	
14:30 Practical Session	**M. Meklenburg**

Thursday, 15 July

09:00 PCRrevisited	**M. Meklemburg**
10:15 Coffe Break	
10:30 Oligonucleotide Ligation Assay	**F. Stephenson**
11:30 Practical Session: OLA	**F. Stephenson**
13:30 Lunch	
15:00 Large scale oligonucleotide synthesis	**A. Andrus**
16:00 Practical Session	**M. Meklenburg**
18:00 Discussion Session	

Friday, 16 July

09:00	Expression/ general amplification systems	**M. Meklenburg**
10:15	Coffe Break	
10:30	Practical: Immuno-PCR	**M. Meklenburg**
13:30	Lunch	
14:30	Combinatorial libraries of oligonucleotides	**H. Seliger**
15:30	Practical: Insert detection/expression	**M. Meklenburg**

Saturday, 17 July

09:00	Workshop/round table	
11:00	Closing Words	**F. De La Vega**

7. Course Outcome.

The time schedule for the course resulted very tight. For this reason some rescheduling of experiments was done on the first week resulting in one hour increase in course sessions for most of the second week. However, most experiments were carried out successfully. All equipment and reagents worked properly and all theoretical sessions were carried out as planned. About 10 more local researchers were admitted after selection from the Mexican runner-ups, only to the course lectures. Even this partial information transfer was judged by these people as valuable to their research subjects, thus increasing the local course benefit. Course manuals were distributed to participants the first day. Additional bibliography was distributed throughout the course. All these material concentrated a wealth of valuable information not easily available together nowadays. A proposal for later publishing of the course protocols by a prestigious publisher was received and is currently analyzed by the course staff.

The expert help of Jose Manuel Galindo as technical assistant solved most critical situations, thus contributing to the success of the course. Some of the participants had the opportunity to discuss their own research data during informal sessions carried out during practicals void time. Practical work was accomplished dividing the participants in work teams. Participants with experience in some areas were encouraged to work in other techniques and to share technical tips with other participants. Personal contact between participants and course staff was encouraged

in order to help participants to pose particular questions relevant to their own research subjects.

Furthermore, a few original research questions were proposed by course staff and advanced participants, that were partly attacked during the course, and to be continued in Drs. Seliger and Ramalho laboratory. Even if the participants' background was diverse, most of them came to the course with specific research questions that could be solved through application of course material. Therefore, the impression of the course staff was that the proposed goals were fulfilled successfully. Besides, direct benefit to the hosting Affiliated Centre was ensued, and various research problems of the host laboratory are currently being attacked through techniques developed within the course.

8. Conclusions.

The ICGEB practical course "Preparation, Analysis and Applications of Synthetic Oligonucleotides" was carried out successfully, and the proposed goals were fulfilled as judged by the course staff. Participants returned to their countries and institutions loaded with a series of new technologies that will help to solve their particular research questions and, together with the careful selection of participants, will promote the dissemination of this set of new methodologies into ICGEB member countries and Affiliated Centres. The benefit to the host Institution was also substantial and currently these new techniques are being assimilated.

Mexico City, August 31, 1993

Prof. Gabriel Guarneros Peña **Asst. Prof. Francisco M. De La Vega**

The Organizing Committee