



CIMMYT

International Maize and Wheat Improvement Center

REPORT ON THE
**Asian Maize Biotechnology Network
(AMBIONET) Training Course**

Molecular Marker Applications to Plant Breeding

9 November to 4 December 1998

CIMMYT, El Batán, Mexico



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SUMMARY

1. Funding: Asian Development Bank
2. Duration: 9 November to 4 December, 1997
3. Venue: CIMMYT, El Batán, Mexico
4. Number of trainees: 19
5. Participants/countries: 5 research teams consisting of 2-4 members from China, India, Indonesia, Philippines and Thailand
2 in-house participants from Kenya and Zimbabwe
6. Course emphasis: Practical applications of molecular marker technology
7. Course content: 23 lectures
24 laboratory sessions
13 computer sessions
Trainees' presentations, group discussions
8. Faculty 15 CIMMYT scientists and six support staff
9. Training materials AMBIONET Training Course Manual
Laboratory Protocols Manual
Reprint Collection
10. Course evaluation Post-course Questionnaire

Background

The application of biotechnology is key in a strong breeding program as breeders seek to develop new maize varieties that are high-yielding, stress-tolerant and input-efficient. Biotechnology holds a lot of promise for maize breeders, enabling them to achieve results more quickly and efficiently, and to attain breeding goals that may not be possible using only traditional methods.

Molecular marker technology is an important tool for identifying parts of the genetic material associated with important agronomic traits, allowing the location and manipulation of genes of interest for maize improvement. Molecular markers are now being used to identify chromosome segments associated with resistance to diseases and insect pests, and tolerance for abiotic stresses such as drought and low nitrogen conditions. Because these traits can be mapped and marked on maize chromosomes, the progress of conventional breeding is greatly accelerated. In CIMMYT, biotechnology tools are now being applied to develop high-yielding maize varieties with desirable traits through marker-assisted selection.

The Asian Maize Biotechnology Network (AMBIONET) is a partnership between CIMMYT and national agricultural research systems (NARS) in Asia, where there has been relatively little opportunity to use biotechnology tools for maize improvement. The AMBIONET was established to enhance the capacity of Asian NARS to integrate biotechnology in their search for solutions to problems affecting maize production, and to conduct collaborative research that will eventually result in improved maize varieties. The AMBIONET training program is aimed at the transfer of biotechnological tools to teams of scientists working in the national maize improvement programs in China, India, Indonesia, Philippines and Thailand. The 1998 training course was specifically designed to introduce members of the AMBIONET teams to concepts in plant genome analysis and their applications to plant breeding.

Course objectives

1. Training: train core members of AMBIONET teams on molecular marker technology, with emphasis on their applications to maize breeding, and
2. Team building: facilitate team work among the members of the AMBIONET research teams and encourage the spirit of collaboration and cooperation within the Network.

Trainees

Seventeen members of AMBIONET teams from five countries and two CIMMYT in-house Visiting Scientists participated in the 1998 AMBIONET training course (Annexes 1 and 2). Among the AMBIONET participants, four were from China, three from India, two from Indonesia, four from the Philippines, and four from Thailand. The average age of the trainees

was 36, with the age ranging from 23 to 56 years. There were 16 males and 3 females. The trainees' educational background included nine with Ph.D., seven with M Sc., and three with B. Sc. degrees. Their areas of specialization included plant breeding, genetics, plant pathology, agriculture, crop science, horticulture and botany. All the trainees had experience working with maize, but the majority of the trainees had medium to low levels of awareness about the subjects covered in the course.

Training course staff

- **Organization and conduct of the course**

Training Coordinator:	M. Khairallah
Lecturers:	15 Scientists from CIMMYT's Applied Biotechnology Center, Maize Program and Wheat Program (Annex 3)
Laboratory staff:	J. J. Olivarez M. Mendoza E. Huerta G. Palacios M. Crosby B. Luna
Computer support:	F. Hernandez

- **Preparation of training materials**

Manual, handbook, reprint collection:	M. Khairallah
Statistical analysis programs:	Software Development Unit

- **Logistics and travel arrangements**

ABC administrative support staff
Training Office
Visitors and Conference Services
CIMMYT-China, CIMMYT-Thailand, IRRI-Indonesia liaison offices
AMBIONET Coordinator

Course content

The course consisted of lectures, hands-on laboratory and computer exercises, demonstrations, participants' presentations and group discussions (Annex 4).

The **lectures and hands-on exercises** covered the following topics:

- generation of different marker systems in maize (non-radioactive RFLP technology, PCR-based markers and chemiluminescent AFLPs)
- marker data entry (both segregating and diversity data) with special software
- genetic analysis of segregating marker data and linkage map construction
- tagging of major genes and mapping of QTL
- schemes and strategies of marker-assisted selection for major genes and for QTL
- analysis of genetic diversity and fingerprinting data
- methods of fingerprinting the downy mildew pathogen
- downy mildew and potyviruses diseases and resistance
- breeding for drought and acid soil tolerance
- heterotic patterns in maize and predicting heterosis using MM
- overview of other activities in the Applied Biotechnology Center
- introduction to the public database available for maize

The **participants' presentations** covered each participant's current research involvement and their contribution to the activities of the AMBIONET in their respective countries. As well, the participants presented the progress of their team. This presentation allowed all members of the Network to become familiar with the workplans of each team and opened up opportunities to discuss issues related to the planned work.

The **group discussions** provided opportunities to exchange ideas on activities that are common to the teams, and to further develop and coordinate the field and lab activities within the Network. The topics of group discussions were:

- setting up a molecular marker laboratory
- different marker types: comparisons and applications
- AMBIONET activities on downy mildew
- experimental designs for mapping and line improvement for disease resistance and drought tolerance.

Training materials

The trainees were given an AMBIONET training course handbook, a manual of laboratory protocols developed by CIMMYT's Applied Molecular Genetics Laboratory, and a compilation of over 50 relevant reprints.

Additional materials distributed to the trainees

- Five CIMMYT publications selected by each trainee
- A set of 95 maize SSR primers and 312 RFLP probes as requested by the individual teams
- PCR primers and RFLP clones for downy mildew fingerprinting, as well as DNA from downy mildew infected plants from Mexico, Thailand and the Philippines (India, Philippines, Thailand)
- Software (HyperMapData, HyperMAS, and Composite Interval Mapping programs developed by ABC)
- AMBIONET t-shirt and bag/CIMMYT coffee mug /cooler
- Diploma and group photo

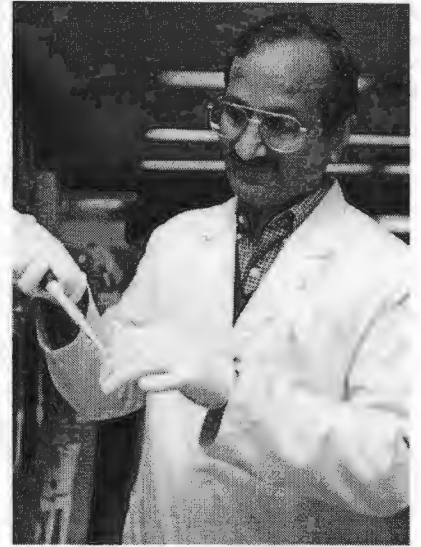
Course evaluation

At the end of the training course, the trainees met with the staff in a question and answer session to clarify remaining issues. They also answered a post-course evaluation questionnaire. About 70% of the trainees felt that the length of the course was about right while the rest felt that the course was too short. The training methods used were found to be useful to very useful. Although the training course schedule was found to be hectic, the majority of the trainees felt that the balance of time spent among the different activities was about right. The coverage and presentation of the topics in the course were considered satisfactory to very satisfactory although some topics were found to be more difficult than others. Topics, which the trainees suggested would need more emphasis, were AFLP, MAS and QTL analyses. The course manual was found to be satisfactory to very satisfactory in terms of usefulness, contents, clarity, length and balance. The trainees found the instructors and laboratory staff well prepared, approachable and helpful. In general, the trainees were pleased with the coverage, organization and conduct of the course. The CIMMYT facilities and services were rated satisfactory to very satisfactory. All the participants felt that the training course will help them to contribute to the activities of the AMBIONET teams in their own countries, but they anticipate facing problems in their work due to lack of adequate laboratory facilities and resources.

Financial Support

The AMBIONET training course was supported by the Asian Development Bank.

AMBIONET TRAINEES IN ACTION





AMBIONET trainees, trainers and other CIMMYT staff who participated in the course **“Molecular Marker Applications to Plant Breeding”** held in El Batan, Mexico, 9 November to 4 December, 1998. L to R, front row: E. Huerta, G. Palacios, C. Pascual, S. Tangsakul, A. Chanmuang, M. Crosby, E. Espinosa; second row: M. Khairallah, M.L. George, B. Luna, J.J. Olivarez, R. Kumar, R. Gadag, A. Salazar, M. Mendoza; third row: F. Kasim, V. Ruanjaichon; G. Chigeza, Z. Li, X. Li, S. Singh, M. Dalan, P. Guzman, S. Velasquez, M.C. Lopez; fourth row: S. Zhang, G. Hernandez, S. Wanchana, M.S. Li, Z. Muthamia, B. Calilung, L. Rodriguez, M. Cruz; fifth row: O. Leblanc, E. Peroti, D. Hoisington, N. Bohorova, M. Warburton

ANNEXES

Annex 1. AMBIONET Training course participants

Annex 2. Directory of 1998 AMBIONET Training course participants

Annex 3. Directory of Lecturers and Resource Scientists

Annex 4. Course schedule

Annex 5. Results of training course evaluation

Annex 1

**AMBIONET 1998 Training Course
Participants**



Shihuang Zhang
CHINA



Ming Shun Li
CHINA



Cecilia Pascual
PHILIPPINES



Peter Guzman
PHILIPPINES



Zhiwu Li
CHINA



Xielai Li
CHINA



Artemio Salazar
PHILIPPINES



Bienvenido Callong
PHILIPPINES



Ravendra Godag
INDIA



Rajesh Kumar
INDIA



Samar B. Singh
INDIA



Auyin Chuanmuang
THAILAND



Saowaree Tangsakul
THAILAND



Marsunt Dahlan
INDONESIA



Firdaus Kasim
INDONESIA



Yiocharn Ruanjaichon
THAILAND



Samart Waichans
THAILAND



Godfree Chigera
ZIMBABWE



Zachary Muthama
KENYA

**AMBIONET 1998 Training Course
Participant Directory**

AMBIONET PARTICIPANTS

Dr. Xinhai Li
Research Associate
Institute of Crop Breeding & Cultivation
Chinese Academy of Agricultural Science
30 Baishiqiao Road
Beijing, 100081, P.R. CHINA
Ph: 086-10-6 891 8621
Fax: 086-10-6 2174865

Mr. Ming Shun Li
Assistant Professor
Institute of Crop Breeding & Cultivation
Chinese Academy of Agricultural Science
30 Baishiqiao Road
Beijing, 100081, P.R. CHINA
Ph: 086-10-6 891 8596
Fax: 086-10-6 217 4865

Mr. Zhiwu Li
Research Assistant
Institute of Crop Breeding & Cultivation
Chinese Academy of Agricultural Science
30 Baishiqiao Road
Beijing, 100081, P.R. CHINA
Ph: 086-10-6 891 8608
Fax: 086-10-6 217 4865
Email: zwli@z63.net

Dr. Shihuang Zhang
Director of Maize Program
Institute of Crop Breeding & Cultivation
Chinese Academy of Agricultural Science
30 Baishiqiao Road
Beijing, 100081, P.R. CHINA
Ph: 086-10-6 891 8596
Fax: 086-10-6 217 4865

Dr. Raveendra Gadag
Scientist
Division of Genetics
Indian Agricultural Research Institute, Pusa
New Delhi, INDIA
Ph: 91-11-578 3077
Fax: 91-11-576 8195 / 578 1481
Email: rng_gene@iari.emet.in

Mr. Bienvenido J. Callung Jr.
Research Associate
Institute of Plant Breeding
University of the Philippines at Los Baños
College, Laguna 4031
PHILIPPINES
Ph: 63-049-536 2339 ext 232
Fax: 63-049-536 2512
Email: bjcj@ipb.uplb.edu.ph

Dr. Peter S. Guzman
Assistant Professor/Maize Breeder
Department of Agronomy
Institute of Plant Breeding
University of the Philippines at Los Baños
College, Laguna 4031
PHILIPPINES
Ph: 63-049-536 2466
Fax: 63-049-536 2468
Email: psg@ipb.uplb.edu.ph

Ms. Cecilia Pascual
Researcher
Institute of Plant Breeding
University of the Philippines at Los Baños
College, Laguna 4031
PHILIPPINES
Ph: 63-049-536 2512
Fax: 63-049-536 3438
Email: cbp@ipb.uplb.edu.ph

Dr. Artemio M. Salazar
Maize Breeder
Institute of Plant Breeding
University of the Philippines at Los Baños
College, Laguna 4031
PHILIPPINES
Ph: 63-049-536 3304
Fax: 63-049-536 3438
Email: ams@ipb.uplb.edu.ph

Ms. Auytin Chanmuang
Agronomist
Petchaboon Field Crop Experiment Station
Petchaboon 67000
THAILAND
Ph: 066-056-721 507
Fax: 066-056-720 687

Annex 2

Dr. Rajesh Kumar
Technical Officer
Division of Genetics
Indian Agricultural Research Institute, Pusa
New Delhi, INDIA
Ph: 91-11-578 3077 / 578 1481
Fax: 91-11-576 8195 / 576 6420

Dr. Samar Bahadur Singh
Research and Coordination Scientist
Directorate of Maize Research
Indian Agricultural Research Institute, Pusa
New Delhi, INDIA
Ph: 91-11-578 2372
Fax: 91-11-576 8195 / 576 6420
Email: nns_dmr@iari.emet.in

Dr. Marsum Dahlan
Maize Breeder
Research Institute for Maize & Other Cereals
(RIMOC)-AARD
Baliťjas, JL Ratulangi 274
Maros 90514, S. Sulawesi
INDONESIA
Ph: 62 411 371 529
Fax: 62 411 371 961
Email: baliťjas@upandang.wasantara.net.id

Dr. Firdaus Kasim
Maize Breeder
Research Institute for Maize & Other Cereals
(RIMOC)-AARD
Baliťjas, JL Ratulangi 274
Maros 90514, S. Sulawesi
INDONESIA
Ph: 62 411 371 529
Fax: 62 411 371 961
Email: baliťjas@upandang.wasantara.net.id

IN-HOUSE PARTICIPANTS

Zachary K. Muthamia
Maize Breeder
Kenya Agricultural Research Institute
N.A.R.C. Muguga
Box 30148
Nairobi, KENYA
Ph: 254-154 32880
Fax: 254-154-32348
Email: dmuthoka@africaonline.co.ke

Mr. Vinitchan Ruanjaichon
DNA Fingerprinting Unit
National Center for Genetic Engineering &
Biotechnology
Khamphangsaen Campus, Kasetsart University
Nakhorn Pathom, THAILAND
Ph: 066-34-281093
Fax: 066-34-281093
Email: R.vinitchan@mailcity.com
Ball@dna.kps.kv.ac.th

Ms. Saowaree Tangsakul
Banmai Samrong Field Crops Experiment Station
Sikhui Nakhon Ratchasima 30340
THAILAND
Ph: 066-44-325 048
Fax: 066-44-279 063

Mr. Samart Wanchana
Assistant Researcher
DNA Fingerprinting Unit
National Center for Genetic Engineering &
Biotechnology
Khamphangsaen Campus, Kasetsart University
Nakhorn Pathom, THAILAND
Ph: 066 34 281093
Fax: 066 34 281093
Email: samart@dna.kps.ku.ac.th

Godfree Chigeza
Maize Breeder
Scientific and Industrial Research and Development
Centre of Zimbabwe (SIRDC)
P. Bag 8640
Harare, ZIMBABWE
Ph: 263-4-860 321/9
Fax: 263-4-860 333/5
Email: Gopo@icori.brimaize.co

Annex 3

AMBIONET 1998 Training Course Directory of CIMMYT Lecturers and Resource Scientists

Dr. Mireille Khairallah

AMBIONET Resource Scientist (biotic stresses)
Senior Scientist & Molecular Geneticist
Applied Biotechnology Center, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: m.khairallah@cgiar.org

Dr. Manilal William

Associate Scientist & Molecular Geneticist
Applied Biotechnology Center, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: m.william@cgiar.org

Dr. Daniel Jeffers

AMBIONET Resource Scientist (maize pathology)
Senior Scientist & Pathologist
Maize Program, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: d.jeffers@cgiar.org

Dr. Yves Savidan

Senior Scientist & Cytogeneticist
Applied Biotechnology Center, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: y.savidan@cgiar.org

Dr. Dave Beck

Senior Scientist & Leader for Highland maize
Maize Program, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: d.beck@cgiar.org

Dr. Gregory Edmeades

Principal Scientist & Crop Physiologist
Maize Program, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: g.edmeades@cgiar.org

Dr. Olivier Leblanc

Scientist & Geneticist
Applied Biotechnology Center, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: o.leblanc@cgiar.org

Dr. Paul Fox

Senior Scientist
Wheat Program, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: p.fox@cgiar.org

Dr. Marilyn Warburton

AMBIONET Resource Scientist (germplasm characterization)
Scientist & Molecular Geneticist
Applied Biotechnology Center, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: m.warburton@cgiar.org

Dr. Natasha Bohorova

Senior Scientist & Cell Biologist
Applied Biotechnology Center, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: n.bohorova@cgiar.org

Dr. Jean Marcel Ribaut

AMBIONET Resource Scientist (abiotic stresses)
Scientist & Molecular Geneticist
Applied Biotechnology Center, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: j.ribaut@cgiar.org

Mr. Gael Pressoir

Graduate Student
Applied Biotechnology Center, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: g.pressoir@cgiar.org

Dr. Shivaji Pandey

Director
Maize Program, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: s.pandey@cgiar.org

Dr. Daniel Grimanelli

Scientist & Molecular Geneticist
Applied Biotechnology Center, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: d.grimanelli@cgiar.org

Dr. Enrico Perotti

Scientist & Molecular Biologist
Applied Biotechnology Center, CIMMYT
Lisboa 27, Apdo. Postal 6-641
06600, Mexico, D.F. Mexico
Email: e.perotti@cgiar.org

Annex 4

AMBIONET First Training Course
Molecular Marker Applications to Plant Breeding
9 November - 4 December, 1998
CIMMYT Headquarters, El Batán, Mexico

Week 1 (November 9-15)

	Lectures (Sasakawa)	Practice (Wheat Pathology Training Lab)
Monday Nov. 9 am	Welcome (20 min) TGR/SP/DH Intro'd'n to course and logistics (30 min) MK/GH Genome, chromosomes and DNA (35 min) MK RFLPs: DNA digestions and electrophoresis (35 min) MK	Lab orientation Start large DNA extraction
pm		DNA extractions, sap extraction Demo tissue grinding
Tuesday Nov. 10 am	Introduction to the Applied Biotech Center at CIMMYT (30 min) DH??	DNA quality and digestions
pm	RFLPs: Southern transfer and hybridization, cloning and transformation, probe labeling (1 hr) MK/MWi <i>Individual pictures in the photography area</i>	Practice double gel loading Start bacterial cultures Stop digestions
Wednesday Nov. 11 am	NO lectures Only discussions when time permits	Plasmid mini-preps Prepare gels for electrophoresis
pm		DNA electrophoresis
Thursday Nov. 12 am	NO lectures Only discussions when time permits	Stain gels, Southern transfer
pm	Trainees presentations, CHINA (1 hr) Downy mildew pathogen identification (30 min) MK	PCR labeling of probes Run probe gel Blot probe gel
Friday Nov. 13 am	NO lectures Only discussions when time permits	Fix DNA, prehybridize Detect probes blot Prepare probes for hybridization
pm	Trainees presentations, INDIA (1 hr) Downy mildew and viral diseases of maize (1.5 hrs) DJ	Hybridize
Saturday Nov. 14		Stringency washes and detection Miniprep results Expose blots to X-ray films
Sunday Nov. 15		Develop X-ray films To the pyramids! (Optional visit)

Annex 4

Week 2 (November 16 - 22)

	Lectures (Sasakawa)	Practice (Wheat Pathology Training Lab)
Monday Nov. 16 am	PCR/based markers: RAPDs, STSs and ASAs (2 hrs) MWi	Stripwash blots and prehybridize them
pm	SSRs, status of maize SSRs (1 hr) MK	Discuss RFLP results Hybridize blots with second set of probes
Tuesday Nov. 17 am	AFLPs (1 hr) MK Comparison of markers	AFLP restriction digests (enzyme 1) Set up SSRs and DM STSs AFLP restriction digests (enzyme 2)
pm	Trainees presentations, INDONESIA (30 min)	Gel of digests Prepare SSR gels
Wednesday Nov. 18 am	NO lectures Only discussions when time permits	AFLP ligations Run SSR and STS gels AFLP pre-amplifications
pm	Trainees presentations, PHILIPPINES (1 hr)	Stain SSR and STS gels Interpretation of PCR results
Thursday Nov. 19 am	Progress in apomixis research (1 hr) YS	Visit Apomixis Lab AFLP selective amplifications
pm	Trainees presentations, THAILAND (1 hr)	Prepare AFLP gel
Friday Nov. 20 am	<i>Group picture in front of main bldg at 10:30 am</i>	Run AFLP gel
pm		Blot AFLP gel
Saturday Nov. 21 am		Detect AFLP blot Expose to X-ray film
pm		Develop AFLP film
Sunday Nov. 22	Free Time/ Optional visit to Mexico City	

Annex 4

Week 3 (November 23 - 29)

	Lectures (Sasakawa)	Practice (room B-116)
Monday Nov. 23 am	Genetic diversity and fingerprinting (2 hrs) MWa	Demonstration of HyperBlot (1 hr) EH
pm		Sequencer demonstration Genetic diversity data entry
Tuesday Nov. 24 am	Multivariate analyses (2 hrs) MWa	Analyses of diversity data
pm	Maize heterotic patterns (1 hr) SP?? Use of MM to predict heterosis (1 hr) MWa	Analyses of diversity data
Wednesday Nov. 25 am	Maize drought tolerance (2 hrs) GE	Analyses of diversity data
pm	AMBIONET presentations, CHINA (1 hr)	Introduction to the Maize DataBase (2hrs) OL Introduction to ICIS (1hr) PF
Thursday Nov. 26 am	Maize genetic engineering (1 hr) NB Explanation of RFLP segregation patterns (2 hrs) JMR	Visit to AGE Lab
pm	AMBIONET presentations, INDIA (1 hr)	Introduction to HyperMapdata MK Entering and verifying segregating data
Friday Nov. 27 am	Genetic segregation, X2 tests Populations used in mapping Creating maps: linkage groups, assigning link. groups to chromosomes (2 hrs) OL	Map construction JMR
pm	AMBIONET presentations, INDONESIA? (1 hr)	Map construction
Saturday Nov. 28	Free time	
Sunday Nov. 29	Free time / Optional visit to Xochimilco	

Annex 4

Week 4 (November 30 - December 6)

	Lectures (Sasakawa)	Practice (room B-116)
Monday Nov. 30 am	QTL mapping: simple ANOVA, interval mapping, composite interval mapping (2 hrs) GP	ANOVA for QTL analysis
pm	AMBIONET presentations PHILIPPINES (1 hr)	Composite Interval Mapping
Tuesday Dec. 1 am	Gene tagging, bulk segregant analysis	Composite Interval Mapping
pm	Marker-assisted selection: schemes, strategies and efficiency (3 hrs) JMR	
Wednesday Dec. 2 am	MAS for SWCB resistance (1 hr) MK MAS for ASI (1.5hr) JMR	
pm	AMBIONET presentations, THAILAND (1 hr)	HyperMAS demo: graphical genotypes, do selections MK/JMR
Thursday Dec. 3 am	Comparative mapping and functional genomics (2 hrs) DG/EP	
pm	Graduation (1 hr)	
Friday Dec. 4 am	Questions/Answers session (2 hrs) All staff Evaluation of the course (0.5-1 hr)	
pm	Delivery of documents EEL	
evening	Farewell dinner: Noche Mexicana	
Saturday Dec. 5	Departures	
Sunday Dec. 6	Departures	

TGR Tim Reeves, DH Dave Hoisington, MK Mireille Khairallah, GH Gilberto Hernández, Mwi Manilal William, MWa Marilyn Warburton, DJ Dan Jeffers, GE Greg Edmeades, SP Shivaji Pandey, JC José Crossa, PF Paul Fox, JMR Jean-Marcel Ribaut, GP Gael pressoir, DG Daniel Grimanelli, EP Enrico Perotti, EEL Elsa Espinoza López

Molecular Marker Applications to Plant Breeding Training Course
Summary of Trainees' Responses to Post-Course Questionnaire-1998

CONGRATULATIONS!! You have completed your training at CIMMYT. For the past four weeks we have been giving you a great deal of information and opportunities to practice in the labs. Now it is your turn to give us some information. We are especially interested in your feelings about a number of things, including: the topics covered, time spent in various activities, general course organization, training aids and materials, allowances and facilities, and how the course impacts your work in AMBIONET.

The purpose of this questionnaire is to collect information, which we can use to improve future training courses in AMBIONET. Please think carefully about each of the questions indicating your true opinion. We suggest that you first read through the entire questionnaire and then go back to the beginning and start answering the questions.

Thank you for your valuable cooperation!!!

Some information about you:

Highest degree you hold / specialty:

Highest degree	Disciplines	No. of participants	Percentage
Ph.D.	Plant Breeding, Genetics, Plant Pathology, Agricultural Botany	9	47%
M. Sc.	Genetics, Plant Pathology, Agriculture	7	37%
B. Sc.	Plant Pathology, Crop Science, Horticulture	3	16%

Age:

Range	23-56
Average	36
Median	34

Gender:

Gender	No. of participants	Percentage
Male	16	84%
Female	3	16%

What was your level of awareness about the subjects covered in the course?

Level of awareness (subjects)	No. of participants	Percentage
Low	3	16 %
Medium	14	74 %
High	2	10 %

What was your level of practical experience about the molecular subjects covered in the course?

Level of practical experience (molecular subjects)	No. of participants	Percentage
Zero	2	10%
Low	6	32%
Medium	11	58%
High	0	0

Annex 5

What was your level of practical experience about the data analysis subjects covered in the course?

Level of practical experience (data analysis subjects)	No. of participants	Percentage
Zero	3	16%
Low	9	47%
Medium	6	32%
High	1	5%

I. General organization of the course

1. In general, what are your feelings about the length of the course?

Course duration	Percentage
Too long	0
About right	68 %
Too short	32 %

2. If you did not feel the course length "About Right", what do you think a better length would be?

- 6 weeks
- 6-8 wks
- 2 month
- 3 months

3. In this course, a number of methods, techniques and analyses were presented and used. The general categories of activities are listed below. Please indicate how useful each of these categories were in helping you to learn, and also if you think the course should make more, less, or the same use of these categories in the future (Very Useful (VU); Useful (U); Somewhat Useful; and Not Useful (NU)).

Methods and materials	Usefulness				Emphasis		
	VU	U	SU	NU	More	Same	Less
Demonstrations	42%	53%	5%		29%	71%	
Lab work	79%	16%	5%		50%	50%	
Analysis	58%	21%	11%		59%	41%	
Discussions- Marker types & usefulness	47%	37%	11%		41%	59%	
Discussions- Est. of a molecular marker lab	47%	37%	11%		25%	63%	12%
Discussions- Downy mildew study	61%	22%	17%		24%	65%	12%
Exercises	35%	53%	12%		60%	40%	
Computer use	67%	28%	5%		59%	35%	6%
Lectures	50%	50%			31%	63%	6%
Trainee Presentations	17%	50%	33%		20%	60%	20%
AMBIONET Presentations	33%	50%	17%		20%	67%	13%
Self-study	31%	56%	13%		54%	46%	

Annex 5

3. What do you think of the **balance** of time that you spent in lectures, laboratories and data analysis?

Too much time in classroom	Too much time in the laboratory	Balance of time in and out of classroom about right
12%		88%

4. If you were in charge of **organizing** this course, what changes would you make in the future?

- Give more lab work and more practical exercises to students
- Make course longer and give more details especially in data analysis
- I would have organized the course during the cropping season so that the participants would have an opportunity to see the standing crop in the field
- Give a detailed preview of all techniques, especially the analysis part in the beginning (first day) so that when practical hands-on exercises are undertaken, one would be aware of how data will be analyzed subsequently
- Increase the time for computer lab about MAS, QTL mapping, CIM. I think this may be another course which is separate from molecular genetic course
- Give more emphasis on lab work
- More laboratory exercises and computer data analysis
- Emphasize more lab work but leave time for trainees to read books and discuss with instructors
- Lab work, data analysis, and lectures of research progress should be emphasized
- Further subdivision of topics which are too broad, topics should be split for better comprehension
- Incorporation of visit to the field for better appreciation of relationship between field and lab works
- I would just change the length of time, because there is too much knowledge to study
- Assign competent people on CIM lecture, somebody who would explain clearly and concisely the subject matter, one that could go down one level and to explain it in a simple way
- Break time in between lectures and lab works
- I would add more time in the laboratory, would encourage more participation of trainees with plenary sessions in the evenings, include 1 lecture or two on quantitative genetics

5. What did you like **least about the organization** of the course?

- None
- Practical training in the laboratory
- The content of the classroom lecture is more than the entire AMBIONET
- Everything was okay but more emphasis should be placed on lab work, data entering and analysis. The time for practicing these was not enough
- Some demonstration is too fast and not everybody can follow (people who stands in the back)
- Not enough time to do everything
- Overlapping of lab work, sometimes it is just overwhelming but overall, the course was really handled well
- Food should also be considered (oriental food for Asians)
- Specific topics without handouts

6. What did you like **most about the organization** of the course?

- Highly impressed with methodology of teaching in general, particularly M. Khairallah
- Lab work practice for each person
- Good blend of lecturers and practical hands-on experience. Persistence, patience and hard work of most of the instructors, in general, especially Mireille Khairallah
- Practical training and teaching methodology by Mireille Khairallah
- A good personnel resource
- Lectures and lab work
- Practice in the lab
- The training materials, references, and the able people in the lab and knowledgeable instructors
- Very good lectures.
- The treatment and sequence of the topics are very logical
- Content of the course
- Hands-on lab and computer work, lectures
- Lab work and analysis of data
- Well-organized and systematic lectures and lab work, and a very good teacher in the lab
- Quality of lectures, patience of the lecturers

II. **Topics**

A large number of topics were presented during the course. Some of these may have been more useful and better presented than others. Now is your chance to let us know how you felt.

(Very satisfactory (VS), Satisfactory (S); Somewhat Satisfactory (SS); Not Satisfactory (NS))

Topics	Rating			
	VS	S	SS	NS
Genome, chromosomes and DNA	56%	34%	5%	5%
RFLP	74%	21%	5%	
Maize downy mildew and potyviruses	26%	58%	16%	
PCR-based markers: RAPDs, STSs, ASAs	10%	80%	10%	
SSRs	67%	33%		
AFLPs	67%	28%	5%	
Progress in Apomixis research	32%	47%	21%	
Genetic diversity	29%	57%	7%	7%
Multivariate analyses	28%	50%	17%	5%
Heterotic patterns in maize	39%	51%	5%	5%
Use of molecular markers in heterotic pattern determination	53%	42%	5%	
Breeding for drought tolerance	63%	26%	5%	5%
Maize genetic engineering	5%	90%	5%	
Explanation of RFLP segregating patterns	47%	47%	6%	
Genetic mapping	47%	29%	24%	
QTL mapping	69%	26%	5%	
Breeding for acid soil tolerance	21%	63%	11%	5%
Marker assisted selection	42%	58%		
Comparative mapping and functional genomics	5%	48%	42%	5%

III. Manual

What ratings would you give to the manual following the items on the table below?
(Very satisfactory (VS), Satisfactory (S); Somewhat Satisfactory (SS); Not Satisfactory (NS))

	VS	S	SS	NS
Usefulness	84%	16%		
Contents	32%	63%	5%	
Clarity	28%	61%	11%	
Length	21%	42%	37%	
Balance	16%	63%	21%	

IV. Instructors

1. Did you find the instructors approachable and willing to answer your questions?

- Yes. (13)
- Yes. All of the instructors were kind to me.
- Yes, I did. But I have spare time in the evening and sometimes it was not easy to contact them.
- Yes, I did. Every instructor I met was kind and willing to answer my questions.
- Yes. Most had a lot of patience.

2. Any specific comments you wish to address to specific instructors?

- None. (4)
- Lecture of M. Williams could have been more comprehensive.
- Lecture of O. Leblanc could have been clearer in respect to theoretical aspects of quantitative analysis.
- Yes, Martha and Juan Jose have done an excellent job in the lab in assisting all the trainees in various practical exercises.
- Some instructors should speak slowly because some students are from non-English speaking countries.
- Concern and attention to our country needs were given by Mireille, thanks a lot.
- I want to know more about fingerprinting, how I can get assistance on fingerprinting Zimbabwe maize germplasm, also computer programs, hypermap data, SAS, HyperMAS
- Some lectures were very fast, very difficult to take notes on the tables presented such as MAS-population size
- Gael's lecture needs to have more organization.
- O. Leblanc's lecture was too fast.
- O. Leblanc needs to undergo a teaching seminar. He is knowledgeable but his presentation needs improvement.
- Khairallah and Ribaut are very good, they could be excellent university professors
- Some instructors assumed that our background is like theirs.
- Some instructors lectured really fast.

V. Resources, services and facilities

We would also like about the facilities and services provided during your stay in Mexico. In the following table are listed a number of factors over which CIMMYT has some control. Please tell us your degree of satisfaction with each of these where it applies. (Very satisfactory (VS), Satisfactory (S); Somewhat Satisfactory (SS); Not Satisfactory (NS); Not Applicable (NA)).

Service, resource, facilities	VS	S	SS	NS	NA
Dormitory accommodations	70%	18%	12%		
Laundry	70%	24%	6%		
Meals	12%	53%	29%	6%	
Health service	25%	56%	12%		7%
Telecommunications	33%	33%	17%	11%	6%
Recreation	25%	25%	44%		6%
Administrative services	50%	50%			
Travel arrangements	50%	50%			
Sightseeing tours	39%	56%			5%

VI. AMBIONET-RELATED comments

1. In general, do you feel that attending this course will improve your ability to do your work when you return?

YES	Somewhat	NO
100%		

2. What new technologies of those learned will you use and for what purposes?

- MAS for biotic and abiotic stresses (insects and drought)
- RFLPs for identification of DM pathogen, QTL for drought tolerance
- PCR-based techniques (molecular markers) for diversity analysis, fingerprinting, mapping
- Analysis of molecular marker data for clustering and mapping
- PCR markers to be used in identifying heterotic patterns
- RFLPs and PCR-based molecular markers can be used to identify transgenic plants
- Fingerprinting to avoid redundancy of my materials
- MAS and interpretation of molecular data
- MAS for biotic and abiotic resistance/tolerance breeding; MAS for downy mildew work
- SSRs and RFLPs for heterotic grouping of commercial lines, RFLP for virus resistance mapping
- Nonradioactive RFLP method, SSR method, CIM (data analysis), SSR and RFLP for heterotic grouping. Data analysis software will be utilized for QTL mapping.
- RFLP and PCR-based protocols, genetic diversity studies, mapping and data analysis, MAS
- RFLP and PCR-based markers for line conversion and determination of heterotic patterns
- I will use SSRs and RFLPs for heterotic grouping and virus resistance study
- Diversity (and fingerprinting) for understanding of heterotic patterns
- MAS-could complement my conventional breeding work. All the technologies would help me in my teaching work, i.e. revision of courses, propose new courses, etc.
- RFLP and AFLP for fingerprinting

Annex 5

3. What constraints will you face in using and applying molecular markers in your research in AMBIONET?

- Maybe computer packages
- Non availability of modern equipment required for molecular marker technique. Problems regarding good chemicals in our country
- Downy mildew pathogen, it is difficult to isolate and identify
- Inadequate lab equipment, chemicals, reagents of high quality present a problem
- General infrastructure
- A fully functional laboratory has not been established. For the time being we will have to depend on other laboratory and take help
- Budget/funds (4)
- Facilities, primers and probes always can't be gotten easily.
- I might have problems of immediate resource personnel to help me and also long-term financial support of such project, computer programs and packages for data analysis
- Biotechnology lab is not available in our institute.
- Data analysis and utilization of software
- Data analysis method and software, especially method that can be used to analyze interaction between loci
- The capacity of our staff
- Pathogen characterization, resistance evaluation
- Setting up the lab with appropriate provisions for waste disposal, water and power equipment, sustainability of funding support
- Primers, probes
- Equipment, trained technician/personnel
- Markers to differentiate isolates of *P. philippensis*

4. What solutions do you suggest to overcome these constraints?

- Good link with CIMMYT
- Early development of infrastructure of lab required for molecular techniques, request to CIMMYT
- Sharing of some equipment with other labs working in the same area
- Strengthening of infrastructure with additional resources
- To contact other molecular biotechnology lab functional in the institute
- More communication with other researchers
- Presently I only see communication with CIMMYT as the only solution
- Cooperation with other institute/university
- Hold another workshop or training course for Asia area for data analysis
- More time should be spent and new software should be used if these software have been made
- To establish good working relationship within country members and more consultation with CIMMYT resource scientists
- Current and new protocols for pathogen characterization
- Come up with concrete useful results to convince the higher ups for continuous or more support
- Explore other funding agencies, public, private and international and other agencies, propose studies that would provide equipment, conduct training course
- Request funds from the government
- Your support

Annex 5

5. Did the course provide you with the necessary information to decide when to apply/not apply molecular markers in your research?

- Yes. (15)
- Yes, course has provided all necessary information at present required for molecular marker technology.
- Yes, the course did provide me with the necessary information to decide when to apply molecular markers.
- Yes, the course helped me to modify our workplan.
- I think so.

6. Have you been introduced to enough alternative strategies for MAS to help you better define your workplan?

YES	Somewhat	NO
65%	35%	

7. Did the course reach the following objectives related to AMBIONET?

Objectives	VS	S	SS	NS	NA
Introduce concepts of molecular markers and their applications	84%	16%			
Break the breeding/biotechnology language barrier	32%	58%	10%		
Provide awareness of AMBIONET research teams and themes	47%	53%			
Build the spirit of teamwork	39%	50%	11%		

8. From what you know about your workplan and team members, what training courses are still needed by your team?

- Data analysis and cloning
- Intense course on data analysis and interpretation (the course was really great, congratulations to the group especially Mireille. Keep it up, Mireille!!)
- I think the computer software about analyzing genetic maps is still needed
- More specific practical experience/hands-on in lab and data analysis/interpretation, maize breeding course
- Biometrics for QTL analysis, screening methods for disease resistance to various pathogens
- Biometrics but more of visiting scientist type
- Data analysis and software, SSR
- Training for our technicians in lab work
- Lab organization, computer data analysis
- More detailed data analysis, SSR method, Computer software training
- A joint effort of breeders, pathologists and physiologist is required to execute the project. A more detailed training on molecular assisted selection
- Analysis of molecular marker data
- Molecular genetics techniques, genetic engineering
- More practical training required for AFLP and in-situ hybridization
- Data interpretation and analysis

