ISTA News Bulletin No. 132 October 2006

IN THIS ISSUE

COVER ARTICLE
The 28th ISTA Congress
3–14
How to become a Member of the ISTA Executive Committee
15
How to become an ISTA Voting Delegate at the Ordinary Meetings
16

ASSOCIATION NEWS
Report of the ISTA Annual Meeting 2006
17
ISTA Membership Categories
18

RULES DEVELOPMENT
Report on Performance of Grinders
23

ACCREDITATION
Evaluation of Non-Conformities
29
6th ISTA Proficiency Test on GMO Testing on Brassica napus L.
31

SEED SCIENCE
Priming: A Technique for Improving Seed Quality
38
Water Concentration Considerations in Recalcitrant/Non-Orthodox Seeds
43
Seed Science and Technology goes online
47

TRAINING & EDUCATION
Workshop Announcements
48
ISTA Workshops Reports
49–60
Editorial

By Michael Muschick,
ISTA Secretary General

Dear Reader,

In this second and last issue of the year, a lot of the contents of Seed Testing International unsurprisingly is focusing on events taking place in the forthcoming year. We are proud to present a calendar 2007 which is filled with a variety of ISTA Workshops – still the biggest event on the agenda for 2007 is the triennial ISTA Congress next May. It will be the 28th ISTA Congress, for the first time to be hosted by Brazil, upon the kind invitation of the Ministry of Agriculture and in conjunction with the XV Congresso Brasileiro de Sementes.

The Congress is the center of attention of this issue of Seed Testing International as it is one of the major events of its kind – not only for ISTA, but also generally seen in relation to seed science and technology on the international level.

The cover article presented in this issue gives valuable information on all pre-congress events (a list of five ISTA Workshops on diverse technical subjects addressed by the Association’s experts in the areas) and the Congress programme itself which includes ISTA Technical Committee Meetings where decisions on the future working programmes of ISTA Technical Committees and Task Forces will be made; the three-days ISTA Seed Symposium – the biggest happening of the Congress presenting papers and posters on scientific investigations and technical expertise in the testing and evaluation of seed quality by the leading scientist in the area of seed science and technology; the ISTA Technical Committee Presentations giving an overview on the work already achieved by these working groups; and finally the ISTA Ordinary Meeting where votes are cast on proposals for enhancement and extensions to the ISTA International Rules for Seed Testing by the ISTA voting delegates, also deciding on the future of the Association in adopting the final Draft ISTA Strategy which will be presented at this occasion as well. The Ordinary Meeting concludes the working part of the Congress, nevertheless some exciting post-congress tours are offered for those people who have the chance to book in on one of those organised excursions.

The last issue of the year is also a time to look back on the past: I would like to thank everyone who attended and collaborated to the ISTA Annual Meeting in Zurich in June. It was an exciting meeting and good progress was made in many areas of the Association. The Draft ISTA Strategy discussions was a very productive one, allowing the Executive Committee to come up with a final draft version for adoption next year in Brazil. The Rules Proposals 2006 which were adopted at this meeting will come into force on January 1st, 2007 – the corresponding Rules Updates will be distributed to all members within short or can be obtained from the ISTA Secretariat. The ISTA Voting Delegates also adopted the new proposed membership categories (see page 18) as presented in the proposed ISTA Constitution Changes 2006. There is quite an interest in the new membership categories noticeable – our records today already show six new subscriptions for associate membership with ISTA.

Now I have been telling you all about meetings, nevertheless, there are more interesting articles to read about in this issue also from the other business areas: in the Rules Development we have a contribution about Grinders, Accreditation stands for an evaluation of non-conformities, conductivity testing for Phaseolus vulgaris and the newly released results of the 6th ISTA Proficiency Test on GMO, including a call for participation in the 8th and 9th test rounds. Seed Science is represented with three scientific articles and an announcement for ‘SST Online’, the ISTA journal ‘Seed Science and Technology’ goes on the internet.

Now I wish you a pleasant reading and a joyful season and happy New Year to follow!

Yours sincerely,

Michael Muschick

PS: We will be looking forward to receiving any interesting articles which you would like to contribute to the next issue of Seed Testing International latest by February 15, 2007.
# CONTENTS

## PRESIDENT'S REPORT

2

## COVER ARTICLE

The 28th ISTA Congress 2007 3
- Programme Overview 4–5
- Detailed Programme 6–14

How to become:
- nominated for the Executive Committee 15
- an ISTA Voting Delegate at the Ordinary Meetings 16

## ASSOCIATION NEWS

Report of the ISTA Annual Meeting 2006 17
ISTA Membership Categories 18
Centennial – the 1st International Conference for Seed Testing 19
Membership Updates 22

## RULES DEVELOPMENT

Report on the Performance of Grinders used in OSTS 23

## ACCREDITATION

Auditors' Meeting Report 28
Evaluation of Non-Conformities observed during ISTA accreditation audits 29
6th Proficiency Test on GMO Testing 31
Call for Registration for 8th and 9th Proficiency Test on GMO Testing 32
Report of the GMO Task Force Meeting 34
Conductivity Test for Phaseolus vulgaris 35
Member Lab Update 37

## SEED SCIENCE

Primming: A Technique for Improving Seed Quality 38
Identification of region of provenance for a rationale management of Common Ash Fraxinus excelsior L. Seeds 41
Water Concentration Considerations in Recalcitrant/ Non-Orthodox Seeds 43
Seed Science and Technology goes online 47

## TRAINING AND EDUCATION

Workshop Announcements 48
ISTA Workshops Reports 49–60
Calendar 61

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The 28th ISTA Congress 2007
Find all relevant information and the final programme for the ISTA Congress 2007 in Iguassu Falls, Brazil
Pages 3 to 16

ISTA Membership Categories
Read about the types of new membership categories available now
Page 18

6th ISTA Proficiency Test on GMO Testing
The results of the 6th ISTA Proficiency Test on GMO Testing on Brassica napus L. are released
Page 31

Seed Science and Technology goes online
Page 47

Workshop Reports
Read the reports of the various ISTA Workshops
Pages 49 to 60
President’s Report

By Pieter Oosterveld, ISTA President

As president I am very proud to announce that up-to-date ISTA has reached the number of 100 laboratories it has accredited. The latest accreditation has been granted to NCVESC Seed Testing Laboratory in Hanoi, Vietnam. ISTA began the accreditations programme, as it stands now, in 1995. Since then, all the ISTA laboratories worked very hard to set up their quality manual, meeting the ISTA standards. ISTA audit teams have travelled around the world, performing the audits. ISTA audits are unique. The auditors are specialists in quality management – above all in seed testing. The analysts of the laboratories, that have been assessed, report that the ISTA audits helped them to improve their work. I congratulate the laboratory all the accredited laboratories, and of course especially the Seed Testing Laboratory in Hanoi, Vietnam, the ISTA staff and auditors for the success of the programme.

The annual meeting in June this year in Zurich was a successful event. Over 150 persons participated in the meetings and discussions. We invited Dr. Gisbert Kley from the European Seed Association (ESA) to present the developments of the seed trade in Europe over the past decennia and his forecast for the future. Dr. Kley has a successful career in plant breeding and has since retired from some of the important positions in international breeding associations. Dr. Kley showed us some data indicating the importance of the seed trade for the economies of the European countries. He also clearly indicated the importance of ISTA and the role our association has played in increasing the quality of seed. A summary of the speech of Dr. Kley can be found on the ISTA Website.

The participants were impressed by the presentations made by the chairpersons of the Technical Committees. Again, this response has us convinced that the Annual Meetings should continue. Some of the Technical Committees make good progress. The Amalgamation of the Rules is part of the work of the programme of the Technical Committees. As expected, it is a difficult task to fulfil. However, the work progresses on schedule.

During the accreditation session we were informed about the success of the ISTA Accreditation Programme. The presented data showed clearly that the performance of the ISTA accredited laboratories is significantly better than the non-accredited laboratories. Also the results of the Proficiency Testing Programme on GMO were presented. After six rounds we conclude that this programme is an equally successful one. The performance of the laboratories that participated in a number of rounds, is consistent, especially with regard to the qualitative tests.

The draft strategic plan of the Association was already published in the April edition of this magazine. The draft was presented to the Ordinary Meeting for preliminary discussion. The Executive Committee is grateful for the comments and suggestions. The amended draft strategic plan is going to be presented for discussion and decision at the 2007 ISTA Congress. A ‘hot’ topic was the discussion on the proposal by the Executive Committee to discontinue with the issuance of the Green Certificate. In principle, ISTA members seem to agree on the proposal. However, they clearly expressed that the ISTA Rules concerning the criteria for the issuance of ISTA certificates were not clear. Therefore, they asked the Executive Committee to come up with a new draft of Chapter 17 of the ISTA Rules with clear guidelines.

In Zurich we also had a meeting with representatives from laboratories in Africa. Together with the working group on tropical seeds the representatives agreed to develop methods for testing of seeds of species for which we have not yet accepted methods in the Rules. Furthermore we talked about a possible training programme for seed testing in Africa. In this field ISTA works together with FAO. ISTA is trying to find sponsors for financing such a training programme.

In August, I attended the meeting of the members of the OECD Seed Schemes in Brazil. Many items about seed certification have been discussed during the meeting. OECD invited ISTA to participate in a working group discussing the items varietal identity and varietal purity. ISTA appreciates the invitation of OECD. It shows that both organisations are very keen to work together and serve the international seed trade in an optimal way.

The ISF Congress in Copenhagen was a great success as well. About 1300 people attended this congress, that included many presentations and meetings of the committees. For ISTA it was an unique opportunity to meet the representatives of the seed trade companies and seed trade associations from all parts of the world.

Already during many years ISTA and AOSA made efforts to come to some harmonisation of their rules for seed testing. In Budapest 2004 the associations agreed to invite their stakeholders to highlight the differences that bothered them most. This action resulted in a more efficient action plan. The chairs of the rules committees of both associations were asked to coordinate the harmonisation process. During the meeting of the ISTA Moisture Committee it became clear that both associations had plans to produce a handbook for moistures testing. Having discovered that AOSA and ISTA shared a common goal, discussions started to elaborate a joint handbook. The idea was fully supported by the ISTA Executive Committee, which worked out a concrete proposal for collaboration for decision by the AOSA Executive Committee. Unfortunately, the ISTA Executive Committee has been informed by the President of AOSA, that AOSA will produce its own AOSA handbook. The clear message from the AOSA President unfortunately leaves no room for further discussions on that subject.

The ISTA Executive Committee is going to meet from 26 to 29 November 2006. It will be a great pleasure for me and my colleagues to host the meeting in Emmeloord, The Netherlands. As usual, we have quite a number of interesting items to discuss. We will go through the draft Generic Method Validation Programme after consultation of the chairs of the Technical Committees followed by chapter 17, including the status of the Green Certificate. We will of course talk about ISTA’s strategy, financial issues and the preparation of the upcoming ISTA Congress 2007 in Iguassu Falls, Brazil.

I like to invite you to come to Brazil for the 2007 Congress, including Workshops, the Symposium, the meetings of the Technical Committees and of course the Ordinary Meeting. In other words, it is worth to attend the Congress, it is worth to visit Brazil. Register and book your flight in time.

See you in Brazil! Your President, Pieter Oosterveld
Announcement & Programme

28th ISTA Congress
XV Congresso Brasileiro de Sementes

May 5–11, 2007
Iguassu Falls, Brazil

www.seedtest.org
www.abrates.org.br
The 28th ISTA Congress is coming closer in big steps. As usual the ISTA Congress will include a set of different events to be held in the hosting country. In the year 2007 this will be Brazil.

These different events will include a number of Training and Education Workshops for seed analysts; the meetings of all ISTA Technical Committees to discuss the progress in their work and do the planning of the strategic working programme for the next three years; a seed symposium where seed scientists from all over the world exchange the results of their recent research; the presentations of the work of their committees by the Technical Committee chairs; the Ordinary meeting of ISTA, as the businesses meeting of ISTA where the voting delegates of ISTA, who are nominated by their respective governments, are discussing and making decisions on the affairs of the Association; and finally not to forget the pre- and post-congress tours organised by the hosting country, which will allow you to see all the beauty of this wonderful and exciting country in guided tours.

Table 1 gives you a detailed overview on the different events and a number of additional important information including the page numbers where to find the full information about single events in this issue of Seed Testing International.

Technical Workshops (between April 25 and May 3)

Five Technical Workshops in different locations are scheduled in conjunction with the 28th ISTA Congress. Some will be held far away from the Congress venue, some closer and some at the congress venue itself:

• a Variety Workshop to be held from April 25-27 in Pelotas, around 500 km away from Iguacu Falls.
• a Tetrazolium and Germination Workshop on tropical & subtropical seeds to be held from April 30-May 3 in Curitiba, around 600 km away from Iguacu Falls.

• a Workshop on Statistical aspects of GMO detection to be held from May 1-3 in the Raffain Hotel, Iguacu Falls, the congress venue itself.
• a Seed Health Workshop to be held from May 1-3 in the Raffain Hotel, Iguacu Falls, the congress venue itself.
• a Seed Vigour Workshop to be held from May 1-3 in Cascavel, around 100 km away from Iguacu Falls

ISTA Technical Committee Meetings (May 5 and 6)

Currently ISTA has 16 Technical Committees and 1 Task Force dealing mainly with the establishment of seed testing methods which are validated and then proposed to the voting delegates of ISTA to be included in the ISTA International Rules for Seed Testing.

All committee members should meet in person at least every three years to evaluate the performance of the committee, select or confirm the chairperson of the committee and to elaborate a strategic working programme for the upcoming triennium.

All meetings of the Technical Committees are open meetings and any interested person can participate in these meetings to listen to the discussion of the committee or to actively give input to the evaluation of the committee or the strategic working programme for the next triennium.

ISTA Seed Symposium (May 7 to May 9)

The ISTA Seed Symposium is a scientific symposium and aiming at bringing leading seed scientist together to discuss the latest progress in research. Six sessions are scheduled for the ISTA Seed Symposium 2007 with the following topics:

• Session 1 – Diversity with and among seed lots and species
• Session 2 – Problems associated with the domestication and use of on crop species
• Session 3 – Diversity in contaminating organisms
• Session 4 – Seed development, dormancy and Germination: Physiology and methods
• Session 5 – Vigour and Invigoration
• Session 6 – Seed storage and genetic conservation

Each session will be chaired by a leading seed scientist in that field, who also will be the lead speaker in that session and will present the latest results in his research. Next to the oral presentations there will be an extensive poster session organised with more than 500 posters about all of the aspects of seed science and technology.

Technical Committee Presentations and Ordinary Meeting (May 10 and 11)

The Technical Committee presentations will inform the voting delegates of ISTA (see page 16 ‘How to become an ISTA voting Delegate at the Ordinary Meetings’) about the work and activities of all the ISTA Technical Committees. These reports of the Technical Committee work will be approved by the ISTA voting delegates in a vote during the Ordinary Meeting of ISTA. Insofar the Technical Committee presentations and the Ordinary Meeting fit in together.

The ISTA Ordinary Meeting is the business meeting of the Association. In this meeting all decisions regarding the affairs of the Association are made. The Executive Committee will be discharged, the financial report and the budgets will be approved and as in all ISTA Ordinary Meetings in conjunction with the ISTA Congress (every third year) a new Executive Committee will be elected (see page 15 ‘How to become a Member of the ISTA Executive Committee’).

See you in Brazil!
<table>
<thead>
<tr>
<th>DATE</th>
<th>EVENT</th>
<th>PLACE</th>
<th>Registration Fees (in US$)</th>
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<td>25-27 April</td>
<td>ISTA Variety Workshop</td>
<td>Laboratório de BioSementes</td>
<td>ISTA members $270 non-ISTA members $405</td>
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<tr>
<td></td>
<td>Paulo Dejalma Zimmer</td>
<td>Faculdade de Agronomia Pelotas, Brasil</td>
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<td></td>
<td>E-mail: <a href="mailto:djzimmer@ufpel.edu.br">djzimmer@ufpel.edu.br</a></td>
<td>Phone: +55-53 8118-4171</td>
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<tr>
<td>30 April – 3 May</td>
<td>ISTA Tetrazolium and Germination Workshop on Tropical &amp; Subtropical Seed</td>
<td>Laboratório Oficial de Análise de Sementes LASO</td>
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<td></td>
<td>Osvaldo de Castro Ohlson</td>
<td>Curitiba, Cabral, Paraná, Brasil</td>
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<td>E-mail: <a href="mailto:lascuritiba@terra.com.br">lascuritiba@terra.com.br</a></td>
<td>Phone: +55-41 3254-6444</td>
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<td>1-3 May</td>
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<td>Leopoldo Baudet</td>
<td>Cascavel, Paraná, Brasil</td>
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<td>E-mail: <a href="mailto:lmbaudet@ufpel.edu.br">lmbaudet@ufpel.edu.br</a></td>
<td>Phone: +55-53 9983-0448</td>
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<td>1-3 May</td>
<td>ISTA Statistical Aspects of GMO Detection</td>
<td>Rafain Hotel</td>
<td>ISTA members $450 non-ISTA members $700</td>
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<td>Maria Laene Moreira de Carvalho</td>
<td>Foz do Iguaçu, Paraná, Brasil</td>
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<td>E-mail: <a href="mailto:marialaene@uol.com.br">marialaene@uol.com.br</a></td>
<td>Phone: +55-35 3829-1318</td>
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<td>Validation Aspect Prof. José da Cruz Machado</td>
<td>Foz do Iguaçu, Paraná, Brasil</td>
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<td>E-mail: <a href="mailto:machado@ufla.br">machado@ufla.br</a></td>
<td>Phone: +53 358291470</td>
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<td>5-6 May</td>
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<td>ISTA Ordinary Meeting</td>
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<td>12 May</td>
<td>POST – CONGRESS TOURS</td>
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<td>Duration: one day Maximum number of participants: 45</td>
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<td>Agricultural tour (visit of the Seed Companies COODETEC and</td>
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<td>Application deadline: April 15, 2007</td>
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<td>COOPAV EL, production fields, seed processing units and seed</td>
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<td>laboratories situated in the state of Paraná)</td>
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<td>To register please contact : <a href="http://www.centraltours.com">www.centraltours.com</a></td>
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<td>Departure: from Hotel Rafain Time: 7 am</td>
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<td>email : <a href="mailto:congress@istactbscongress.com.br">congress@istactbscongress.com.br</a></td>
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<td>Lunch and dinner are included.</td>
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<td>Pantanal North (visit of the Chapada dos Guimarães)</td>
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<td>To enjoy these tours, airplane tickets must be purchased before arriving into Brazil. The Brazil air pass allows up to 3 stops. Details can be found at <a href="http://www.centraltours.com.br">www.centraltours.com.br</a></td>
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<td>Fernando de Noronha (trip to the Island at the Atlantic Ocean Beach)</td>
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<td>Rio de Janeiro (city tour)</td>
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<td>Manaus (visit of the Tropical Jungle)</td>
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### ISTA Variety Workshop, 25-27 April 2007

**Location:**
Laboratório de BioSementes, UF Pel
Faculdade de Agronomia / Fitotecnia
C.P. 354, CEP: 96010-900
Pelotas, Brasil

**Lecturers:**
Prof. Dr. Norbert Leist,
ISTA GMO Task Force Chair
Mr. Rainer Knoblauch,
ISTA Variety Committee Chair

**Number of participants:** 15

**Application deadline:** 31/01/07
**Payment deadline:** 01/03/07

**Registration fee:**
ISTA Members US$ 270
Non-ISTA members US$ 405

**Workshop programme:**
- Variety testing, general principles and methods
- Demonstration of conventional methods: morphology, anatomy, chemical and fluorescence
- Isoelectric focusing, principles and practical use in seed testing
- Evaluation of IEF gels, principles and actual results
- Method development with IEF
- Statistics and tolerances in variety testing
- Presentation of the International Seed Testing Association
- Identification, homogeneity test and hybrid determination – practical part with Zea mays, Oryza sativa, Triticum aestivum, Helianthus annuus; Lycopersicon esculentum

**To register please contact:**
Paulo Dejarma Zimmer
E-mail: dzimmer@ufpel.edu.br
Phone: +55 - 53 8118-4171

### ISTA Tetrazolium and Germination Workshop on Tropical & Subtropical Seed, 30 April – 3 May 2007

**Location:**
Laboratório Oficial de Análise de Sementes/Curitiba
– LASO/Curitiba
João Américo de Oliveira rf 330
Anexo ao TECPAR
Caxias, CEP: 80.035 – 060
Paraná, Brasil

**Lecturers:**
Mrs. Stephanie Krämer,
ISTA Tetrazolium Committee Chair
Prof. Dr. Norbert Leist,
ISTA GMO Task Force Chair
Dr. Ronald Don,
ISTA Germination Committee Chair

**Number of participants:** 24

**Application deadline:** 31/01/07
**Payment deadline:** 01/03/07

**Registration fee:**
ISTA Members US$ 335
Non-ISTA members US$ 503

**Workshop programme:**
- Preparation for Germination testing in the laboratory
- Germination Testing and evaluation
- Germination, evaluation of abnormal seedlings in mono- and di-cotyle dons
- Use of tolerance tables, calculation and reporting of hard and fresh seed
- Tetrazolium testing for the determination of un-germinated seed
- Viability, Germination and Vigour
- practical part - Glycine, Phaseolus, Zea, Brachiaria, Coffea, Triticum, Avena, Triticosecale and Hordeum testing

**To register please contact:**
Osvaldo de Castro Ohlson
E-mail: lascuritiba@terra.com.br
Phone: +55 - 41 3254-6444

### ISTA Vigour Workshop, 1-3 May 2007

**Location:**
COODETEC Laboratory
Coodetec BR 467-km 98
Cx Postal 301 CEP 85813-450
Cascavel Parana Brazil

**Lecturers:**
Dr. Alison Powell,
ISTA Vigour Committee Chair
Dr. Stan Matthews,
University of Aberdeen, Scotland
Dr. Roberval Daiton Vieira
UNESP, Brazil

**Number of participants:** 20

**Application deadline:** 31/01/07
**Payment deadline:** 01/03/07

**Registration fee:**
ISTA Members US$ 400
Non-ISTA members US$ 600

**Workshop programme:**
- Background to seed vigour
- Importance of seed vigour in crop production
- Two ISTA validated vigour tests: Accelerated ageing test for Glycine max and Conductivity test for Pisum sativum
- Controlled deterioration test for small seeded vegetables
- Cold test for maize
- Rate of germination as a vigour assessment, illustrated using maize
- Precision in vigour testing
- Tolerances

**To register please contact:**
Leopoldo Baudet
E-mail: lmbaudet@ufpel.edu.br
Phone: +55 - 53 9983-0448
ISTA Workshop on Statistical Aspects of GMO Detection, 1-3 May 2007

Location:
Rafain Hotel
Av. Olímpio Rafagnin, 2357
CEP 85862-210
Foz do Iguaçu, Paraná

Lecturers:
Dr. Sylvain Grégoire,
ISTA Statistics Committee Chair
Dr. Enrico Noli,
ISTA GMO Task Force Information Working Group Chair
Mr. Kirk Remund,
ISTA Statistics Committee Vice-Chair
Mr. Jean-Louis LaFont,
ISTA Statistics Committee Member

Workshop programme:
• determining the appropriate testing plans to thresholds or quality levels
• getting an estimate of % GM in a sample when either a quantitative or qualitative assay is used
• robust testing plans
• multi-stage testing plans
• outlier testing
• ISO 5725: repeatability and reproducibility
• checking purity in reference material
• ISTA proficiency testing rating system
• a refresher on statistical tests
• discussion of statistical distributions found in living material
• Seedcalc, QualiStat and an Access program software (all available on the ISTA website) are the primary software packages that will be used during this workshop
• PCR generalities (screening and specific detection methods)
• quantification methods (semi-quantitative and real time analysis)
• good laboratory practice.

Number of participants: 20
Application deadline: 31/01/07
Payment deadline: 01/03/07

Registration fee:
ISTA Members US$ 440
Non-ISTA members US$ 660

To register please contact: Maria Laene Moreira de Carvalho
E-mail: marialaene@uol.com.br
Phone: +55-35 3829-1318

ISTA Seed Health Workshop and Method Validation Aspect, 1–3 May 2007

Location:
Rafain Hotel
Av. Olímpio Rafagnin, 2357
CEP 85862-210
Foz do Iguaçu, Paraná

Lecturers:
Dr. Valerie Cockerell,
ISTA Seed Health Committee Chair
Prof. José da Cruz Machado,
ISTA Seed Health Committee Member

Workshop programme:
• introduction to method validation for seed health testing
• Method Validation from the perspective of ISO 17025
• the ISTA Seed Health Method Validation Programme
• Problem solving – Infected seed?
• How many seeds, samples, or laboratories required
• Data handling
• fundamentals of writing test plans and validation reports
• sampling strategies
• measuring uncertainty
• reporting results
• personal method validation issues and plans

Number of participants: 20
Application deadline: 31/01/07
Payment deadline: 01/03/07

Registration fee:
ISTA Members US$ 400
ISTA non-members US$ 600

To register please contact: Prof. José da Cruz Machado
E-mail: machado@ufu.br
Phone: +55 353 29 14 70

Complete details and the registration form can be downloaded at: www.seedtest.org/en/workshop.html
# ISTA TECHNICAL COMMITTEE MEETINGS AND OPENING CEREMONY

## SATURDAY, MAY 5

<table>
<thead>
<tr>
<th>Time</th>
<th>Event</th>
</tr>
</thead>
<tbody>
<tr>
<td>07:30 – 21:00</td>
<td>Registration of Participants at Rafain Palace Hotel</td>
</tr>
<tr>
<td>08:00 – 09:00</td>
<td>Bulking and Sampling Committee Meeting</td>
</tr>
<tr>
<td>09:00 – 10:00</td>
<td>Flower Seed Committee Meeting</td>
</tr>
<tr>
<td>10:00 – 10:30</td>
<td>Coffee break</td>
</tr>
<tr>
<td>10:30 – 11:30</td>
<td>Forest Tree and Shrub Committee Meeting</td>
</tr>
<tr>
<td>10:30 – 11:30</td>
<td>Advanced Technologies Committee Meeting</td>
</tr>
<tr>
<td>11:30 – 12:30</td>
<td>Germination Committee Meeting</td>
</tr>
<tr>
<td>12:30 – 13:30</td>
<td>Lunch</td>
</tr>
<tr>
<td>13:30 – 14:30</td>
<td>Seed Moisture Committee Meeting</td>
</tr>
<tr>
<td>13:30 – 15:30</td>
<td>Nomenclature Committee Meeting</td>
</tr>
<tr>
<td>13:30 – 15:30</td>
<td>Task Force on Seed Analyst Training (Executive Meeting)</td>
</tr>
<tr>
<td>15:30 – 16:00</td>
<td>Coffee break</td>
</tr>
<tr>
<td>16:00 – 17:00</td>
<td>Proficiency Test Committee Meeting</td>
</tr>
<tr>
<td>17:00 – 18:00</td>
<td>Auditors’ Meeting (Executive)</td>
</tr>
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</table>

## SUNDAY, MAY 6

<table>
<thead>
<tr>
<th>Time</th>
<th>Event</th>
</tr>
</thead>
<tbody>
<tr>
<td>07:30 – 21:00</td>
<td>Registration of Participants at Rafain Palace Hotel</td>
</tr>
<tr>
<td>08:00 – 09:00</td>
<td>Purity Committee Meeting</td>
</tr>
<tr>
<td>09:00 – 10:00</td>
<td>Seed Health Committee Meeting</td>
</tr>
<tr>
<td>10:00 – 10:20</td>
<td>Coffee break</td>
</tr>
<tr>
<td>10:20 – 11:10</td>
<td>Statistics Committee Meeting</td>
</tr>
<tr>
<td>11:10 – 12:10</td>
<td>Seed Storage Committee Meeting</td>
</tr>
<tr>
<td>12:10 – 13:10</td>
<td>Lunch</td>
</tr>
<tr>
<td>13:10 – 14:10</td>
<td>Tetrazolium Committee Meeting</td>
</tr>
<tr>
<td>14:10 – 15:10</td>
<td>Seed Vigour Committee Meeting</td>
</tr>
<tr>
<td>15:10 – 15:30</td>
<td>Coffee break</td>
</tr>
<tr>
<td>15:30 – 16:30</td>
<td>Variety Committee Meeting</td>
</tr>
<tr>
<td>16:30 – 17:30</td>
<td>GMO Task Force Meeting</td>
</tr>
</tbody>
</table>

### OPENING CEREMONY

<table>
<thead>
<tr>
<th>Time</th>
<th>Event</th>
</tr>
</thead>
<tbody>
<tr>
<td>18:30 – 20:00</td>
<td>• Officialization of opening of the Congress by the Ministry of Agriculture of Brazil</td>
</tr>
<tr>
<td>18:30 – 19:30</td>
<td>• Opening address by the ISTA President &amp; ISTA Secretary General</td>
</tr>
<tr>
<td></td>
<td>• Opening address by ABRATES</td>
</tr>
<tr>
<td>19:30 – 20:00</td>
<td>Presentation of the global seed industry by Dr. Bernard Le Buanec, ISF Secretary General</td>
</tr>
<tr>
<td>20:00</td>
<td>Welcome Cocktail</td>
</tr>
</tbody>
</table>
## ISTA SEED SYMPOSIUM

Disclaimer: at the time of printing, the following presenters were selected for the oral sessions.

### MONDAY, MAY 7

<table>
<thead>
<tr>
<th>Time</th>
<th>Event</th>
</tr>
</thead>
<tbody>
<tr>
<td>08:00 – 08:20</td>
<td>Welcome and Presentation by ISTA</td>
</tr>
<tr>
<td>08:20 – 08:30</td>
<td>Opening Seed Symposium</td>
</tr>
<tr>
<td>08:30 – 10:00</td>
<td>SESSION 1 - DIVERSITY WITHIN AND AMONG SEED LOTS AND SPECIES</td>
</tr>
<tr>
<td></td>
<td>GM testing; varietal identification; identification of germplasm for breeding; seed lot heterogeneity and sampling; purity; automated and computer-based methods for seed identification and assessment. Chair and lead speaker: Michael Kruse University of Hohenheim, Stuttgart, Germany</td>
</tr>
<tr>
<td></td>
<td>This session will include a selection of offered papers on an approach for semi-quantitative testing for GM presence in conventional seed lots, sampling and testing for seed moisture content in tropical and oily seeds and a hydrotine model of germination.</td>
</tr>
<tr>
<td>10:00 – 10:30</td>
<td>Coffee break</td>
</tr>
<tr>
<td>10:30 – 11:30</td>
<td>SESSION 1 continuation –</td>
</tr>
<tr>
<td>11:30 – 12:30</td>
<td>POSTER SESSION 1</td>
</tr>
<tr>
<td>12:30 – 13:30</td>
<td>Lunch</td>
</tr>
<tr>
<td>13:30 – 15:00</td>
<td>SESSION 2 - PROBLEMS ASSOCIATED WITH THE DOMESTICATION AND USE OF NON-CROP SPECIES Seed production and processing; germination; dormancy; contamination with other organisms; seed-borne pathogens. (This session includes flower, ornamental, tree, shrub and medicinal species) Chair and lead speaker: Mirian Eira Embrapa, Brasilia, Distrito Federal, Brazil</td>
</tr>
<tr>
<td></td>
<td>The offered, selected papers will include germination and dormancy breaking in ornamental species, Brazilian tree species and medicinal plants, and the influence of provenance and season on variations in longevity in Australian native species.</td>
</tr>
<tr>
<td>15:00 – 15:30</td>
<td>Coffee break</td>
</tr>
<tr>
<td>15:30 – 16:30</td>
<td>SESSION 2 continuation -</td>
</tr>
<tr>
<td>16:30 – 18:00</td>
<td>SESSION 3 - DIVERSITY IN CONTAMINATING ORGANISMS Detection and effects of seed-borne pathogens; weeds; other species and parasitic plants; seed treatments; conventional and organic methods; effects on seed performance Chair and lead speaker: Gary Harman Cornell University, Geneva NY, US</td>
</tr>
<tr>
<td></td>
<td>The papers included in this session will discuss seed health and physical sanitation methods for sustainable farming, immuno-chemical and PCR methods for detection of fungal contamination of organic seeds and biocontrol of seed-borne pathogens</td>
</tr>
</tbody>
</table>
# ISTA SEED SYMPOSIUM (CONTINUED)

## TUESDAY, MAY 8

<table>
<thead>
<tr>
<th>Time</th>
<th>Event</th>
</tr>
</thead>
<tbody>
<tr>
<td>08:00 – 08:30</td>
<td>Presentation about the Brazilian Seed Industry</td>
</tr>
<tr>
<td>08:30 – 09:30</td>
<td>SESSION 3 continuation -</td>
</tr>
<tr>
<td>09:30 – 10:00</td>
<td>SESSION 4 - SEED DEVELOPMENT, DORMANCY AND GERMINATION: PHYSIOLOGY AND METHODS</td>
</tr>
<tr>
<td></td>
<td>(ISSS collaborative session)</td>
</tr>
<tr>
<td></td>
<td>Seed development and maturation; influence of seed production factors; viability; germination; dormancy; dormancy breaking; acquisition of desiccation tolerance</td>
</tr>
<tr>
<td></td>
<td>Chair and lead speaker: Roberto Benech-Arnold</td>
</tr>
<tr>
<td></td>
<td>University of Buenos Aires, Buenos Aires, Argentina</td>
</tr>
<tr>
<td></td>
<td>The use of proteomics to detect the onset of germination, biochemical tests to assess physiological quality, genomic analysis of germination, screening for physical dormancy and the use of digital scanning technology to assess development will be topics included in papers in this session</td>
</tr>
<tr>
<td>10:00 – 10:30</td>
<td>Coffee break</td>
</tr>
<tr>
<td>10:30 – 12:30</td>
<td>SESSION 4 continuation -</td>
</tr>
<tr>
<td>12:30 – 13:30</td>
<td>Lunch</td>
</tr>
<tr>
<td>13:30 – 15:00</td>
<td>SESSION 5 - VIGOUR AND INVIGORATION</td>
</tr>
<tr>
<td></td>
<td>Causes of vigour differences (seed production, processing, physiological); vigour testing; impact of vigour on emergence and storage; priming and other invigoration treatments</td>
</tr>
<tr>
<td></td>
<td>Chair and lead speaker: Kent Bradford</td>
</tr>
<tr>
<td></td>
<td>Seed Biotechnology Center, University of California, Davis CA, US</td>
</tr>
<tr>
<td></td>
<td>Offered papers to be included in this session will discuss prediction of seed vigour in peppers, mean germination time, vigour and metabolic repair in maize, a trait led investigation of seed vigour in Brassica oleracea using natural genetic variation, radish seed vigour and field performance and conductivity testing in Brassica</td>
</tr>
<tr>
<td>15:00 – 15:30</td>
<td>Coffee break</td>
</tr>
<tr>
<td>15:30 – 16:30</td>
<td>SESSION 5 continuation -</td>
</tr>
<tr>
<td>16:30 – 17:30</td>
<td>POSTER SESSION 2</td>
</tr>
</tbody>
</table>
## SESSION 6 - SEED STORAGE AND GENETIC CONSERVATION

Desiccation sensitivity and alternative storage methods for recalcitrant seeds; orthodox seed storage: processing, drying and optimum conditions for long term storage; predicting storage potential; seeds for genetic conservation; physiological basis of seed deterioration.

Chair and lead speaker: Hugh W. Pritchard  
Seed Conservation Department, Royal Botanic Gardens Kew, GB

Offered and selected papers to be presented will include the impact of vegetative stress on longevity, seed storage and dormancy release in Brachiaria brizantha, drying of soyabean seeds using dry air of differing relative humidities and desiccation sensitivity.

<table>
<thead>
<tr>
<th>Time</th>
<th>Activity</th>
</tr>
</thead>
<tbody>
<tr>
<td>08:30 – 10:00</td>
<td>SESSION 6 - SEED STORAGE AND GENETIC CONSERVATION</td>
</tr>
<tr>
<td>10:00 – 10:30</td>
<td>Coffee break</td>
</tr>
<tr>
<td>10:30 – 11:30</td>
<td>SESSION 6 continuation –</td>
</tr>
<tr>
<td>11:30 – 12:00</td>
<td>Symposium Conclusion</td>
</tr>
<tr>
<td>12:00 – 13:00</td>
<td>Lunch</td>
</tr>
<tr>
<td>13:00 – 13:30</td>
<td>Editorial Board Meeting (Seed Science and Technology)</td>
</tr>
<tr>
<td>13:30 – 14:00</td>
<td>Accreditation Session</td>
</tr>
<tr>
<td>14:00 – 15:00</td>
<td>Ordinary Meeting Preparation Strategy</td>
</tr>
<tr>
<td>15:00 – 15:30</td>
<td>Coffee break</td>
</tr>
<tr>
<td>15:30 – 18:00</td>
<td>Rules Committee Session (discussion of Proposed Rules Changes 2007)</td>
</tr>
<tr>
<td>19:30</td>
<td>OFFICIAL DINNER</td>
</tr>
</tbody>
</table>
### TECHNICAL COMMITTEE PRESENTATIONS

**Presentations of Working Programmes 2007–2010 and activities**

#### THURSDAY, MAY 10

<table>
<thead>
<tr>
<th>Time</th>
<th>Presentation</th>
</tr>
</thead>
<tbody>
<tr>
<td>07:30 – 18:00</td>
<td>Registration of Participants at Rafain Palace Hotel</td>
</tr>
<tr>
<td>08:30 – 08:45</td>
<td>Opening Technical Committee Presentations</td>
</tr>
<tr>
<td>08:45 – 09:15</td>
<td>Bulking and Sampling Committee Presentation</td>
</tr>
<tr>
<td>09:15 – 09:45</td>
<td>Purity Committee Presentation</td>
</tr>
<tr>
<td>09:45 – 10:15</td>
<td>Germination Committee Presentation</td>
</tr>
<tr>
<td>10:15 – 10:45</td>
<td>Coffee break</td>
</tr>
<tr>
<td>10:45 – 11:15</td>
<td>Tetrazolium Committee Presentation</td>
</tr>
<tr>
<td>11:15 – 11:45</td>
<td>Vigour Committee Presentation</td>
</tr>
<tr>
<td>11:45 – 12:15</td>
<td>Moisture Committee Presentation</td>
</tr>
<tr>
<td>12:15 – 12:30</td>
<td>Editorial Board Presentation (Seed Science and Technology)</td>
</tr>
<tr>
<td>12:30 – 13:30</td>
<td>Lunch</td>
</tr>
<tr>
<td>13:30 – 14:00</td>
<td>Statistics Committee Presentation</td>
</tr>
<tr>
<td>14:00 – 14:30</td>
<td>Seed Health Committee Presentation</td>
</tr>
<tr>
<td>14:30 – 15:00</td>
<td>Proficiency Test Committee Presentation</td>
</tr>
<tr>
<td>15:00 – 15:30</td>
<td>Variety Committee Presentation</td>
</tr>
<tr>
<td>15:30 – 16:00</td>
<td>Coffee break</td>
</tr>
<tr>
<td>06:00 – 17:00</td>
<td>GMO Task Force Presentation</td>
</tr>
<tr>
<td>17:00 – 17:30</td>
<td>Flower Seed Committee Presentation</td>
</tr>
<tr>
<td>17:30 – 18:00</td>
<td>Forest Tree and Shrub Seed Committee Presentation</td>
</tr>
<tr>
<td>18:00 – 18:15</td>
<td>Nomenclature Committee Presentation</td>
</tr>
<tr>
<td>18:15 – 18:45</td>
<td>Seed Storage Committee Presentation</td>
</tr>
<tr>
<td>18:45 – 19:00</td>
<td>Advanced Technologies Committee Presentation</td>
</tr>
</tbody>
</table>

#### ORDINARY MEETING

**FRIDAY, MAY 11**

<table>
<thead>
<tr>
<th>Time</th>
<th>Presentation</th>
</tr>
</thead>
<tbody>
<tr>
<td>08:30 – 10:00</td>
<td>Opening Ordinary Meeting</td>
</tr>
<tr>
<td>1.</td>
<td>Call to order</td>
</tr>
<tr>
<td>2.</td>
<td>President's address</td>
</tr>
<tr>
<td>3.</td>
<td>Roll call of Designated Members entitled to vote</td>
</tr>
<tr>
<td>4.</td>
<td>Reading and acceptance of Minutes</td>
</tr>
<tr>
<td>5.</td>
<td>Report of the Executive Committee</td>
</tr>
<tr>
<td>6.</td>
<td>Report of the Secretary General</td>
</tr>
<tr>
<td>10:00 – 10:30</td>
<td>Coffee break</td>
</tr>
<tr>
<td>10:30 – 12:30</td>
<td>7. Election of Officers and Members-at-large of the Executive Committee</td>
</tr>
<tr>
<td>8.</td>
<td>Fixation of annual subscriptions</td>
</tr>
<tr>
<td>12:30 – 13:30</td>
<td>Lunch</td>
</tr>
<tr>
<td>13:30 – 15:00</td>
<td>9. Constitution changes</td>
</tr>
<tr>
<td>10.</td>
<td>Consideration and Adoption of the proposed Rules Changes 2007</td>
</tr>
<tr>
<td>11.</td>
<td>Consideration and Adoption of Reports</td>
</tr>
<tr>
<td>15:00 – 15:30</td>
<td>Coffee break</td>
</tr>
<tr>
<td>15:30 – 18:00</td>
<td>12. Announcement of the place and date for the next Ordinary Meeting</td>
</tr>
<tr>
<td>13.</td>
<td>Any other business raised by a Member, of which notice in writing has been</td>
</tr>
<tr>
<td></td>
<td>received by the Secretary General two months prior to the date of the meeting</td>
</tr>
<tr>
<td>14.</td>
<td>Any other business raised by consent of the Executive Committee</td>
</tr>
<tr>
<td>15.</td>
<td>Discharge of the Executive Committee</td>
</tr>
<tr>
<td>16.</td>
<td>Installation of new Officers</td>
</tr>
<tr>
<td>17.</td>
<td>President's closing address</td>
</tr>
<tr>
<td>18.</td>
<td>Adjournment</td>
</tr>
</tbody>
</table>

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*Seed Testing International*  No. 132 October 2006
REGISTRATION FORM
28th ISTA Congress
Iguassu Falls, Brazil, May 5 – 11, 2007

Title: ..............................................................................................................................
Name: ..........................................................................................................................
First Name: ....................................................................................................................
Company: ......................................................................................................................
Position: .......................................................................................................................
Address: ......................................................................................................................
Postal Code: ...................................................................................................................
City: ..............................................................................................................................
Country: ......................................................................................................................
Phone: .........................................................................................................................
Fax: ...............................................................................................................................
E-mail: .........................................................................................................................

I am : ISTA Member ☐ ISTA Technical Committee Member ☐ Non-ISTA Member ☐

Name of accompanying person/ s :
..........................................................................................................................
..........................................................................................................................

I register for:
• ISTA Congress ☐
• Symposium only ☐
• Agricultural Tour ☐
• Sightseeing Tour ☐
  ⚫ Manaus  ☐  ⚫ Pantanal North ☐  ⚫ Fernando de Noronha ☐  ⚫ Rio de Janeiro

For Pre-Congress Workshops registration, please contact respective workshops organisers (see details on Pages 6 – 7)

Please send your registration to:
Dr. Silmar Peske
Rua Maestro Bandeira 237  
96 055-650 Pelotas, RS, BRAZIL.  
Tel: +55 53 327 57 289  
Fax: +55 53 327 32 827
E-mail: congress@istacbscongress.com.br

☐ We wish to receive an invoice
☐ We wish to pay by credit card (please give details)
VISA/MasterCard: ..................................................  Expiry date: ..............
Cardholder’s name: ..................................................
Date: ........................................  Signature: .............................................

For on-line Congress registration: www.abrates.org.br or www.seedtest.org

Methods of Payment
VISA, Mastercard, AMEX, Diners and Swift payment (bank transfer) are accepted.

Cancellation Policy
The organisation will hold 20% of the registration payment in case of cancellation
GENERAL INFORMATION

Organising Committee
Silmar Teichert Peske
President of the Organising Committee
Second Vice President of ISTA
Orlando Antonio Luca filho
Antonio Carlos Albuquerque Barros
Leopoldo Baudet
Francisco Amaral Villela
Paulo Dejamaí Zimmer
Maria Angela André Tillmann
Antonio Eduardo Loureiro da Silva
Airton Lange
José Newmar Francelino
Jorge Szczypior

Congress Venue
Rafain Palace
Hotel & Convention Center
Av. Olimpio
Rafagnin, 2357
CEP 85662-210
Foz do Iguaçu - Brazil
Phone: +55 45 3520-9494
Fax: +55 45 3526-3030
www.rafainpalace.com.br

Symposium Convenor
Dr. Alison Powell
E-mail: a.a.powell@abdn.ac.uk

Scientific Correspondence
ISTA Secretariat - Seed Symposium
Zürichstrasse 50, CH-8303 Bassersdorf,
Switzerland
Phone: +41 44 838 6000, Fax: +41 44 838 6001
E-mail: SeedSymposium@ista.ch

Exhibition
Parallel to the Congress an exhibition of scientific and technical equipments, plant and seed materials will be organised. This will offer excellent opportunity for international exposure and direct communications between suppliers and users of equipments, plants and seeds.

Fee: a space of 3m x 3m is US$ 2500, which entitles exhibitor entry into any place of the event and participation in the Cocktail, Dinner and three complimentary lunch.

For reservations of exhibition booths and for further information contact the local organizers at: abrates@abrates.org.br.

Accommodation
Foz do Iguaçu is one of the most pursued destinations in the whole world. This is why it bears a first-class hotels and services infrastructure, with accommodation facilities ranked among the top ones in Brazil.

The main high-category hotels of the city are supporters of the Iguassu Convention & Visitors Bureau, which unequivocally shows they are nothing but businesses with guaranteed quality and in all cases in accordance with the most demanding international standards.

Hotel fees range from approximately 30 to 100 US$ for a single room. Detailed information about hotels are available on the Congress Website at www.abrates.org.br or www.centraltours.com.br/home/

Location
It is hard, if not impossible at all, to resist the countless natural beauties and the ecological appeal of this wonderful region internationally known as Iguassu. More than 70 different ethnic groups, a spate of languages and diverse customs merge in harmony within paradisiacal scenery, framed by the largest cluster of waterfalls in the whole world, the Iguassu Waterfalls.

The large and exuberant reserve of rainforest that makes up the Iguassu National Park, declared by the UNESCO as part of its heritage’s list in 1986, gathers a wide biodiversity, with hundreds of animal and vegetal species. Iguassu is a region of intimate contact between people and nature, which turns the Waterfalls into an unforgettable experience for anyone.

Travel Information
The best and usual way to enter Brazil is through São Paulo. From there it is another hour by plane to Foz do Iguaçu.

Passport, Visa, Invitation Letters
An entry visa is not required for holders of passports from European or South American countries. A visa is required for those from North America. For further information about the necessary documentation and where visas can be obtained, consult a travel agent. Passports must be valid for at least six months after the planned date of arrival in Brazil.

Currency
Brazil has Real as its currency and is rated around two Reals for one US Dollar. It would be advisable to change some money at the airport (taxis may not accept foreign currency). International credit cards are accepted in most hotels, shops and restaurants.

Languages
The official language of the congress will be English. During the Seed Symposium there will be translation to Portuguese (Brazil speaks Portuguese).
How to become a Member of the ISTA Executive Committee

By Michael Muschick, ISTA Secretary General

The ISTA Executive Committee members have a high responsibility towards the ISTA Membership regarding the future development of the Association. The concrete tasks of the ISTA Executive Committee are laid down in the ISTA Constitution Article VII.

The Executive Committee of ISTA holds personal meetings three times a year, two meetings in conjunction with the ISTA Annual Meeting and a three day business meeting at the beginning of the year.

The Executive Committee of ISTA consist of the President, the First Vice-President, the Second Vice-President and eight Members-at-Large.

If you want to become a member of the ISTA Executive Committee you need

- to become a Personal Member of the Association
- to be nominated by your Government to represent this government in the affairs of ISTA - meaning you need to become a ‘Designated Member
- to be nominated as candidate for the Executive Committee during the ISTA Congress
- to be elected by the ISTA Ordinary Meeting as member of the Executive Committee

How to become a Personal Member

If you want to become a personal member of ISTA, you have to apply for personal membership through the ISTA Secretariat.

The Secretariat will send you the appropriate application documents. The Executive Committee decides on your application to become an ISTA Member.

How to become a Designated Member

Designated Members are ISTA Personal Members, who have been nominated by their Government to represent this Government in the affairs of ISTA.

The ISTA Secretariat can provide the Personal Member with the address of the relevant Governmental Authority (= Designated Authority) in their country which have the authority to designate ISTA Personal Members as Designated members. The designation process for becoming a Designated Member is a country specific procedure, where ISTA bodies do not have any involvement.

How to become nominated as candidate for the ISTA Executive Committee

All Designated Members can be nominated as candidates for the ISTA Executive Committee or the position of the First Vice-President through the support of two other Designated Members from the same or from a different country.

At the beginning of each ISTA Congress, application forms for the nomination of candidates for the Executive Committee and the First Vice-President can be found at the booth of the ISTA Secretariat.

To fill out the application form the name of the candidate and his personal ISTA code (e.g. XXDM01) must be stated. Additionally the name and the personal ISTA code of two other Designated Members from the same or from a different country supporting the candidate need to be stated on the application form.

Only if this is given the application form is valid. Designated Member of ISTA can be identified by the corresponding indication on the name tag.

The filled out application forms need to be returned to the booth of the ISTA Secretariat latest by 15:00 hours local time on the day before the Ordinary Meeting starts.

How to become a member of the ISTA Executive Committee

From the application forms received, the ISTA Secretariat will check the validity and establish out of the list of valid applications a list of candidates for the Executive Committee and the position of the First Vice-President.

This list will be presented to the ISTA Voting Delegates and ISTA Members at the beginning of the Ordinary Meeting of the Congress.

After presentation of the list, each candidate has time to present himself during the Ordinary Meeting.

After this presentation of the candidates, the election process takes place. The identified Voting Delegates will receive two voting sheets for the position of the First Vice-President and the eight Members-at-large together with their voting documents.

They have to fill in the name of the person they wish to elect for the position of the First Vice-President and the eight names chosen form the presented list they want to have in the new Executive Committee.

First the election of the First Vice-President takes place. Proposed Designated Members not being elected as First Vice-President automatically continue to be candidates for the Executive Committee members-at-large.

The First Vice-President is elected by majority of votes. The eight candidates receiving the highest number of votes will be elected as new members of the ISTA Executive Committee.

The newly elected ISTA Executive Committee is installed at the end of the Ordinary Meeting and meets for the first time on the day directly after the Ordinary Meeting for their constitution meeting.
All decision making power within ISTA lie in the hands of the Governments (Article IX (a) ISTA Constitution).

It is up to a single Government to decide whether it wants to execute its voting rights.

This decision of the Government has no influence on the decision of a laboratory or a person to become a member of the Association and to use the services provided by the Association or to participate in the work of the Association.

If a Government wants to execute its voting rights within ISTA, this Government needs to:

• assure that at least one seed testing expert in the country is an ISTA Personal Member
• designate at least one ISTA Personal Member to represent the Government in the affairs of the Association (designated ISTA Personal Members are referred to as ‘Designated Member’).
• nominate one Designated Member to execute the voting rights on behalf of that Government for each ISTA Ordinary Meeting.

In other words, ISTA Personal Members wanting to execute voting rights on behalf of their Governments in the ISTA Ordinary Meeting need to:

• have a designation from their Government to represent this Government in the affairs of the Association ( = being a Designated Member)
• be nominated each year by their Government to execute the voting rights on behalf of their Government at the ISTA Ordinary Meeting

How to become a Personal Member

If you want to become a personal member of ISTA, you have to apply for personal membership through the ISTA Secretariat.

The Secretariat will send you the appropriate application documents. The Executive Committee decides on your application to become an ISTA Member.

How to be nominated to execute the voting rights on behalf of the Government in a particular year

Always in the beginning of each calendar year, the ISTA Secretariat sends out letters to each Designated Authority asking them to nominate one Designated Member executing the voting rights in ISTA Ordinary Meeting to be held in this year.

Therefore right before the ISTA Ordinary Meeting, the ISTA Secretariat has a list of names of Designated Members, who have been entitled to vote in this year’s meeting on behalf of the Government of that country.

If a Designated Authority does not give any indication as to who should execute the voting rights in this year, a Designated Member wanting to execute the voting rights for a country has to provide a letter from its Designated Authority which indicates that he has been nominated as voting delegate for this meeting. This letter must be submitted to the Secretary General of ISTA at least 24 hours before the start of the ISTA Ordinary Meeting.

During the roll call in the Ordinary Meeting the names of the nominated Voting Delegates will be called by name to receive their voting cards. The voting rights can then be executed on behalf of that Government.
The location of the ISTA Annual Meeting 2006 was Zurich, Switzerland – the homebase of the ISTA Secretariat.

Switzerland showed its character once again from its most beautiful side with warm temperatures and beautiful sunshine. We are very proud to report that a record number of 154 delegates and 8 accompanying persons from 54 countries travelled to Switzerland to attend this year’s ISTA Annual Meeting.

The meeting began with the Welcome Cocktail on Sunday evening sponsored by the Swiss Federal Office for Agriculture. After the welcome speech of the ISTA President, Ir. Pieter Oosterveld, the participants had their first opportunity to exchange information among each other besides enjoying the delicious bits of various drinks and food offered.

On Monday morning the sessions traditionally started with the Technical Committee presentations, during which each of the 16 ISTA Technical Committees informed about the current activities, developments and future projects of their committees.

The programme on Tuesday was filled by sessions on current important issues such as the sessions of the GMO Task Force, Accreditation Department and Rules Committee, in which the Rules Proposals to be voted on the next day by the ISTA Designated Members entitled to vote on behalf of their Governments were discussed.

The Ordinary Meeting on Wednesday was opened by a welcome of the ISTA President, Ir. Pieter Oosterveld, followed by a speech of Dr. Gisbert Kley from the European Seed Association (ESA) on the development of the Seed Industry in Europe and the role of ISTA, which was very well received by the participants.

After the roll call of ISTA Designated Members entitled to vote, reading and acceptance of minutes, report of the Executive Committee and the report of the Secretary General, the ISTA Draft Strategy was presented and discussed with the delegates in preparation for the vote on the ISTA Strategy in 2007.

The next item were the proposed changes to the ISTA Constitution (published in Seed Testing International No. 131), that included the alignment of the ISTA Membership categories with the present subscription categories and the introduction of new Membership categories (Corporate Member, Associate Member and Honorary Life Member). All motions were adopted by the voting delegates, The Corporate Member Fee is Swiss Francs CHF 10'000 per year and the Associate Member Fee is Swiss Francs CHF 200 per year.

As point 11., the consideration and adoption of the Proposed Rules Changes 2006 was next on the agenda. All motions were adopted into the ISTA Rules by the voting delegates, with the exception of items 2a, 3c, 3e-j, 8a and b which had been withdrawn from voting as a result of the discussions that took place during the Rules Committee Session earlier in the week. The new ISTA Rules including the changes adopted during this Ordinary Meeting will be coming into force on January 1, 2007.

Prof. Silmar Peske as Second Vice-President of ISTA announced the place and date for the next Ordinary Meeting in Iguacu Falls, Brazil in conjunction with the 28th ISTA Congress to be held May 5-11, 2007.

The meeting was concluded on Thursday afternoon after the closing address of the ISTA President.

The detailed minutes of the Ordinary Meeting have been circulated to all participants of the meeting, and to all ISTA Members and stakeholders. In addition the document can be downloaded from the ISTA Website or can be requested from the ISTA Secretariat.

The proposed fixation of annual subscriptions for the new membership categories were also adopted by the voting delegates. The Corporate Member Fee is Swiss Francs CHF 10'000 per year and the Associate Member Fee is Swiss Francs CHF 200 per year.

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ISTA Membership Categories

At the ISTA Annual Meeting 2006 held in Zurich, Switzerland, the ISTA voting delegates decided on the installation of two new ISTA membership categories, ‘Associate Member’ and ‘Corporate Member’.

The ISTA Constitution, adopted at the ISTA Ordinary Meeting in Zurich 2006, defines both previous and newly installed categories in the Article IV, clause ‘(e)-(g)’. This article provides detailed description of each ISTA membership category, describing rights and services to which ISTA members are entitled.

MEMBER LABORATORY
It is engaged in the testing of seed which supports the Association and its objectives and is admitted by the Association.

Member Rights
The Member Laboratory can participate in the ISTA Proficiency Test Programme, and once accredited by ISTA, be authorised to issue ISTA International Seed Analysis Certificates.

All staff members of the Member Laboratory receive priority for any ISTA event and can enjoy reduced membership registration fees for ISTA Congresses, Ordinary Meetings, Symposia and Workshops.

A Member Laboratory membership includes one Personal Member, whose postal address has to be the same as that of the laboratory. The Personal Member of the Laboratory is its representative in the affairs of the Association. The Personal Member may be designated by their Designated Authority as a Designated Member, and be authorised to execute the country’s voting right on behalf of its Government at ISTA Ordinary Meetings.

ISTA Services
The Member Laboratory receives a copy of the ‘International Rules for Seed Testing’ and subsequent updates, any ISTA Handbooks and proceedings published after admission, ‘Seed Testing International’ and a CD subscription to ‘Seed Science and Technology’. Additional copies and ISTA publications which predate the membership can also be purchased at member’s reduced price.

PERSONAL MEMBER
It is a person engaged in the science and practice of seed testing or in the technical control of such activities. He/She supports the Association, its objectives and is admitted into the Association.

Member Rights
A Personal Member receives priority at any ISTA event and can enjoy reduced membership registration fees for ISTA Congresses, Ordinary Meetings, Symposia and Workshops.

A Personal Member may be designated by their Designated Authority as a Designated Member, and be authorised to execute the country’s voting right on behalf of its Government at ISTA Ordinary Meetings.

ISTA Services
A Personal Member receives a copy of the ‘International Rules for Seed Testing’ and subsequent updates, any ISTA Handbooks and proceedings that are published after admission as member, ‘Seed Testing International’ and a CD subscription to ‘Seed Science and Technology’. Additional copies and ISTA publications which predate the membership can also be purchased at member’s reduced price.

ASSOCIATE MEMBER
It is a person who is not a Personal Member, but who supports the Association and its objectives, and is admitted into the Association.

Member Rights
An Associate Member receives ‘Seed Testing International’ and has priority for membership of an ISTA Technical Committee over individuals who are not ISTA Members.

ISTA Services
An Associate Member can purchase the ‘International Rules for Seed Testing’ and subsequent updates and the ISTA Handbooks and proceedings at member’s reduced price and is entitled to enjoy the preferential reduced membership registration fees for ISTA Congresses, Ordinary Meetings, Symposia and Workshops.

CORPORATE MEMBER
It is an organisation which supports the Association and its objectives, and through paying an annual fee, provides sponsorship to the Association.

Member Rights
A Corporate Member will nominate one representative to act on its behalf in ISTA. That representative is subjected to pay ISTA Member registration fees for ISTA Congresses, Ordinary Meetings, Symposia and Workshops, but cannot be designated as a Designated Member, and therefore prohibited to vote or hold office in the Association.

ISTA Services
A Corporate Member receives two copies of all ISTA publications: ‘International Rules for Seed Testing’ and subsequent updates, the ISTA Handbooks and proceedings, ‘Seed Testing International’ and a CD subscription to ‘Seed Science and Technology’.

A Corporate Member will have his or her name published annually in the ISTA Activity Report under the heading “Corporate Members”, and it will also appear prominently under the same heading at ISTA Ordinary Meetings and Congresses.
By A. M. Steiner and M. Kruse
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In 1869 Friedrich Nobbe published his “Statute concerning the Control of Agricultural Seeds” and founded the first seed testing laboratory of the world in Tharandt, Saxony. This was the beginning of a rapid and broad development. Already in 1875 the “First Assembly of the Directors of Seed Testing Stations and of other Persons Interested in this Matter” took place in Graz, Austria, with 31 participants from Central-European countries. At that time there were 12 seed testing stations in Germany, 2 each in Austria-Hungary and Belgium and 1 each in Denmark, Russia and the United States. In 1876 Nobbe published his famous “Handbook on Seed Testing” presenting among others the seed testing methods which he had proposed and discussed and which were recommended for standard use in Graz. In a follow-up meeting in Hamburg in 1876 the motto “Uniformity in Seed Testing” was coined, later becoming the logo of ISTA. In 1877 Nobbe started the first comparative test with Poa pratensis L. germination determination. As a consequence of these activities and of the actual need of seed testing, only in the years 1876/77 more than 20 new seed testing laboratories were founded. In 1896, a good quarter of a century after Nobbe’s initiation, Rostrup in Copenhagen discovered a total of 119 seed testing laboratories in 19 countries. At that time, Möller-Holst in Copenhagen, Stebler in Zürich, Eidam in Breslau, Kirchner and Michalowski in Hohenheim, Rodewald in Kiel, v. Weinzierl in Vienna and Wittmack in Berlin were esteemed as pioneers of seed testing.

At the II. International Botanical Congress 1905 in Vienna, v. Weinzierl invited to a meeting of agricultural botanists. Just 30 years after the meeting at Graz the discussion was resumed to aim at internationally approved methods and standards for seed testing and, as the case may be, to initiate uniform application.

Subjects of discussion were “Testing of Sugar Beet Seed”, “The Weight Method in Germination Determination”, “Organisational Questions of Seed Testing” and “Cereal Seed Testing and Cereal Breeding”. Other Congress activities prevented more detailed reviewing of these topics. Hence, it was resolved to convene for a specific International Conference
from the beginning and are still strongholds of seed testing as e. g. Copenhagen founded in 1871, Munich 1876, Zürich 1876, Hohenheim 1878, Örebro 1880, Vienna 1881 Budapest 1882 and Wageningen 1887. Many once famous other places were relocated or disappeared for one reason or the other as Tharandt the birthplace of seed testing.

In the sessions participated: For Denmark Director K. Dorph-Petersen – Copenhagen, Norway Director O. Qvam – Kristiania (Oslo), Sweden Director M. J. Widén – Örebro, Russia Prof. Dr. B. Issatschensko – St. Petersburg, Austria Aulic Council Dr. Ritter von Weizsäcker – Vienna, Hungary Dr. A. von Degen – Budapest, Switzerland Director F. G. Stebler – Zürich, England Dr. Glässow of the Royal Agricultural Society – London; the Republic of Argentina had conferred procurement on Prof. Dr. A. Voigt – Hamburg. In the last moment, among others, unfortunately the representatives of the United States of America and the Netherlands, Brown and Bruinning, were prevented from participation as also Director E. Schirbaux – Paris having been assigned to lecture on sugar beet seed in trade. By the way, the representative of Italy received the approval of the ministry for participation only at the last day of the Conference and two representatives of Russian-Poland had difficulties getting passports; similar problems we face sometime or other even today.

The German Agricultural Society was represented by: Prof. Dr. P. Hillmann – Berlin and Prof. Dr. H. Rodewald – Kiel, the Association of Agricultural Experiment Stations in the German Empire by Privy Economy Council Prof. Dr. R. Heinrich – Rostock and Prof. Dr. W. Edler – Jena, Bavaria by Administration Council Dr. L. Hiltner – Munich, Württemberg by Prof. Dr. O. Kirchner – Stuttgart, Saxony by Dr. J. Simon – Dresden, the Association of Seed Traders of Germany by Dr. Th. Waage – Berlin, the Seed Traders of Austria by Mr. Fanta – Prague, the Society of Seed Trade Hamburg by its Chairman Mr. Blumenau and the Society of Seed Trade Stettin by Dr. Waage.

In addition, the Conference was attended by: Dr. A. Atterberg – Bernburg, Dr. O. Appel – Dahlem: “On the Importance of Phytopathology in Seed Testing and in Field Experiments.” Director L. Kühle – Gunsleben: “On the Influence of Dehulling of Beta Seed on Germination (Mechanical Removal of the Perigon).” Prof. Dr. J. Vaihla – Brno: “Quality Testing of Malting Barley.”

for Seed Testing in Autumn 1906 in Hamburg in connection with the Meeting of the Association of Applied Botany. For preparing this Conference, a committee was formed consisting of: Mr. E. Brown, Botanist in Charge U. S. Department of Agriculture in Washington, D. C., F. F. Bruining, jr., Director of the Rijksproefstation voor Zaadcontrole in Wageningen, Administrative Council Dr. L. Hiltner, Director of the Agrícolaturalbiotechnischen Anstalt in Munich, Prof. Dr. A. Voigt, Head of the Abteilung für Samenkontrolle an den Botanischen Staatsinstituten in Hamburg and Aulic Council Dr. Th. Ritter von Weizsäcker, Director of the K. K. Landwirtschaftlich-botanischen Versuchs- und Samenkontrollstation in Vienna. Bruining jr. was appointed as Chairman and Voigt as Executive Secretary.

Primarily, the tasks of the projected Conference should be the following ones: “I. The already existing relations between the institutions of individual states should be improved and extended for the benefit of the international seed trade as well as the agricultural producers and consumers, and II. By discussing the scientific basis of the work of the seed testing laboratories becoming year by year more important, to reach step by step uniform standards in seed testing.”

Voigt invited about 150 persons covering practically the total field of seed science, technology and trade of that time. 25 of them promised to participate, 25 announced likely participation, 70 felt sorry not being in the position to participate, the rest did not respond. Under those deeply regretting not being able to attend was Privy Council Prof. Dr. Friedrich Nobbe having retired from work in 1904 at the age of 74. He had held the leading position in the field for 45 years and suffered temporarily from health problems. Yet, he sent a message of compliments to the Conference which was communicated in the introductory session.

Following, the list of the 35 participants with their places of work is shown as it is presented in the Proceedings, because names are history. Many of the participants had gained great merit contributing to seed science and technology in the years before or were gaining great merit in the years to come and even later within ISTA. In addition, it should be noted that the political map of Central-East-Europe was very different in those days from the present situation, which gives interesting insights into relationships and developments. Some of the places had been pioneering
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# ISTA Membership Changes

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Report on the Performance of Grinders used in OSTS for Scotland

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Introduction
The ISTA Rules state that moisture tests should be carried out on two independently drawn samples. In the view of ISTA auditors this means that where species are required to be ground for the test, the grinding should be carried out on two independent samples. Due to changes in Scottish Seed Marketing Regulations we have been carrying out an increased number of moisture content tests on cereals and have been having difficulties without of tolerance replicate results. Oats and barley samples were particularly problematic and anecdotal evidence indicated that the second replicate of a test invariably had a lower moisture content. There was also an indication that the grinder temperature increased progressively during continual operation.

It was decided to investigate the reasons for the out of tolerance replicate results.

Materials and Methods
In the OSTS we have two grinders that are used for the preparation of moisture content tests where grinding is required: a Regent-Maskin and Janke & Kunkel.

Two varieties of barley, oats and wheat were ground in each grinder and the particle sizes of the grindings assessed by sieving the samples using calibrated metal sieves. For each species, each variety was equilibrated to three different moisture contents prior to grinding. The moisture content of the test samples were measured using a calibrated Dickey John GAC 2100 Grain Analysis Computer (Figure 1).

Table 1: Moisture contents of cereal seed used in grinder performance trial.

<table>
<thead>
<tr>
<th>Species</th>
<th>Variety</th>
<th>Moisture Content</th>
<th>Moisture Classification</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hordeum vulgare</td>
<td>Siberia</td>
<td>15.5</td>
<td>High</td>
</tr>
<tr>
<td></td>
<td>Siberia</td>
<td>13.1</td>
<td>Medium</td>
</tr>
<tr>
<td></td>
<td>Siberia</td>
<td>6.4</td>
<td>Low</td>
</tr>
<tr>
<td></td>
<td>Chalice</td>
<td>16.4</td>
<td>High</td>
</tr>
<tr>
<td></td>
<td>Chalice</td>
<td>14.5</td>
<td>Medium</td>
</tr>
<tr>
<td></td>
<td>Chalice</td>
<td>6.7</td>
<td>Low</td>
</tr>
<tr>
<td>Avena sativa</td>
<td>Winston</td>
<td>15.9</td>
<td>High</td>
</tr>
<tr>
<td></td>
<td>Winston</td>
<td>13.7</td>
<td>Medium</td>
</tr>
<tr>
<td></td>
<td>Winston</td>
<td>7.3</td>
<td>Low</td>
</tr>
<tr>
<td></td>
<td>Argyle</td>
<td>15.6</td>
<td>High</td>
</tr>
<tr>
<td></td>
<td>Argyle</td>
<td>13.5</td>
<td>Medium</td>
</tr>
<tr>
<td></td>
<td>Argyle</td>
<td>7.5</td>
<td>Low</td>
</tr>
<tr>
<td>Triticum aestivum</td>
<td>Robigus</td>
<td>16.3</td>
<td>High</td>
</tr>
<tr>
<td></td>
<td>Robigus</td>
<td>14.2</td>
<td>Medium</td>
</tr>
<tr>
<td></td>
<td>Robigus</td>
<td>7.6</td>
<td>Low</td>
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<td></td>
<td>Malacca</td>
<td>16.0</td>
<td>High</td>
</tr>
<tr>
<td></td>
<td>Malacca</td>
<td>13.9</td>
<td>Medium</td>
</tr>
<tr>
<td></td>
<td>Malacca</td>
<td>8.7</td>
<td>Low</td>
</tr>
</tbody>
</table>

Figure 1: The Dicky John GAC 2100 Grain Analysis Computer used to measure the moisture content of the cereal seed samples used in this grinder performance trial.
THE REGENT MASKIN GRINDER

With the Regent-Maskin Grinder (Figure 2, 3 and 4) set at No 1 the performance of the grinder was assessed and the results are shown in Figure 5.

The Regent-Maskin grinder did not grind the samples fine enough to satisfy the criteria of the ISTA Rules, i.e. that 50% pass through a sieve of mesh 0.5 mm and less than 10% be retained on a sieve of mesh 1.0 mm. Oat samples were a particular problem with more than 50% being retained on a sieve of 1.0 mm (Figure 6a). For all the barley samples and all the wheat, apart from the Robigus sample at high moisture content, more than 50% passed through the 1.0 mm sieve but more than 10% was retained on the 0.5 mm sieve (Figure 6b).

When one looks at the grindings it is clear that for oats in particular the material that is retained on the 1.0 mm sieve is husks. Material passing through both the 1.00 mm and 0.5 mm sieves is extremely fine (Figure 6b and c). Indeed, it has to be “brushed” through the 0.5 mm sieve with gravity and shaking not being sufficient to separate the ground samples into different sized particles.
THE JANKE AND KUNKEL GRINDER

Using the Janke & Kunkel (Figure 7), in conjunction with the 1.00 mm internal sieve, all material passing to the collecting tube meets the ISTA sieving specification as shown in Figure 8.

Virtually no material was retained on a 1.00 mm sieve and more than 60% passed through a sieve of 0.5 mm. However when opening the grinder to clean it after grinding it is clear that the internal sieve is selectively sorting the ground material with husks and course material being retained in the drum of
the grinder (Figure 9a).

In addition to being retained in the drum the husks have a tendency to block the holes of the internal sieve (Figure 9b) and this increases the time required to obtain a ground sample of the required weight. It also increases the time that the material is subjected to hammering within the drum. The temperature of the drum and its contents increases, due to friction, and the temperature of the ground material obtained in the collecting tube also increases to temperature greater than 35°C (Figure 9c and d).

If the 1.00 mm internal sieve in the Janke & Kunkel is replaced by a 2.00 mm sieve there is not a large increase in temperature and the sieve does not block thereby avoiding a build up of material in the drum. However, as shown in Figure 10 the ground material obtained does not meet the fineness specification required by the ISTA Rules for most samples.

Figure 9a: Husks and course particles are retained in the drum of the Janke & Kunkel hammer mill.

Figure 9b: The Janke & Kunkel grinder can become blocked with husks obstructing holes in the internal sieve.

Figure 9c: When grinding starts with the Janke & Kunkel the temperature of the ground material is just above ambient.

Figure 9d: After grinding one oat replicate, the temperature of the second replicate to be ground is nearly 40°C.

Figure 10: Performance of Janke & Kunkel grinder with 2.00 mm sieve.
Conclusion

The effect grinding to the specification required by ISTA Rules

Grinding to the specification required by ISTA Rules can cause a significant increase in the temperature of ground material obtained. This temperature increase is more pronounced when large numbers of samples have to be ground and if chaffy species, which block the holes internal sieves in hammer mills, have to be ground. In addition hammer mills with internal sieves do not give a representative sample of material with chaff being retained in the drum of the mill.

This temperature increase is related to a loss of moisture by the ground material and can lead to out of tolerance replicate results where independent replicates are ground.

By grinding to the specification required by the current ISTA Rules an accurate determination of the moisture content of a sample may not be obtained. When large numbers of samples are being processed it is possible that the moisture contents reported, particularly of the last samples to be processed, will be lower than they should be due to heating of the ground material and a consequential loss of moisture.

At the OSTS for Scotland we use the Regent-Maskin mill or the Janke & Kunkel with a 2.00 mm internal sieve for chaffy samples such as oats.

Table 2 indicates that adopting such a strategy provides results which appear to be more accurate particularly when they are compared to moisture contents of the same samples determined using calibrated moisture meters.

The present specification for grinding in the ISTA Rules can not be satisfied using plate mills such as the Regent-Maskin that are common in a large number of ISTA Laboratories. Although hammer mills can produce ground material of the required specification they do not give representative samples, with chaffy material being retained within the mill. They can also significantly increase the temperature of the sample, which can lead to a reduction in the moisture content in the ground material.

Recommendation

It is recommended that the current ISTA specification for grinding is changed. It would be preferable to adopt a requirement similar to that contained in the Institute of Brewing Rules, i.e. that the ground material for moisture determinations should be sufficiently fine. Adopting such a change to the rules would allow laboratories to set their own specification for fineness and produce their own validation evidence to satisfy auditors. If such a "vague" specification is not acceptable we would recommend a specification that could be satisfied using a plate mill such as the Regent-Maskin. An example of such a specification would be:

100% passing through a 2.0 mm sieve, at least 20% passing through a 1.0 mm sieve and at least 20% passing through a 0.5 mm sieve.

It would be possible to tighten this specification for non chaffy cereal.

Table 2:
Moisture content results obtained on oat samples using oven method after grinding samples with different mills and using calibrated moisture meters.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Oven MC Janke &amp; Kunkel mill 1.00 mm sieve</th>
<th>Oven MC Regent-Maskin mill</th>
<th>GAC Meter</th>
<th>Protimeter</th>
<th>Sinar Meter</th>
</tr>
</thead>
<tbody>
<tr>
<td>Winston Oat1</td>
<td>12.3</td>
<td>13.4</td>
<td>13.6</td>
<td>13.0</td>
<td>12.6</td>
</tr>
<tr>
<td>Winston Oat2</td>
<td>14.4</td>
<td>16.1</td>
<td>16.2</td>
<td>16.5</td>
<td>16.0</td>
</tr>
<tr>
<td>Argyle Oat1</td>
<td>12.4</td>
<td>13.3</td>
<td>13.1</td>
<td>12.8</td>
<td>12.0</td>
</tr>
<tr>
<td>Argyle Oat2</td>
<td>14.1</td>
<td>15.6</td>
<td>14.9</td>
<td>15.4</td>
<td>15.0</td>
</tr>
</tbody>
</table>
At the occasion of the 2006 Annual Meeting of the Association the auditors gathered in the late afternoon after the official meeting programme of June 27. A total of fourteen auditors discussed a range of technical issues that had been raised during the year. Most of these questions had been forwarded to Technical Committees and ISTA expert panels and could be clarified by these. Martina Rösch, who chaired the meeting in her capacity as the Head of Accreditation provided an overview of last year’s activities and an outline for the coming year. In her positive overall summary she emphasised the important input from the technical auditors in the routine audit work and in the many non-routine interactions that helped to provide effective, harmonised, technically valid and meaningful auditing services to the Association.

A lively discussion arose from the proposal on how to define a laboratory’s scope of accreditation. The Rules as a collection of technical protocols have a long tradition of providing technical guidance for solving analytical problems and making sure that laboratories all over the world are able to work in a way that their results are comparable; some of the requirements of a formal accreditation system are relatively new and may not be addressed in an ideal way by the design of the Rules. This concerns particularly the single elements of laboratory accreditation and the way they are defined, namely the methods and their respective field of application. A laboratory’s scope of accreditation is composed of methods and the species to which these methods are applied. Traditionally, this is expressed by referring to Rules Chapters and a range of species for each of the applicable chapters.

Due to the complexity of some of the chapters, particularly since the option of accreditation has been extended to performance approved methods, this representation no longer provides unambiguous information on what precisely is covered in a laboratory’s scope of accreditation. The proposal discussed during the meeting was a first step in the process of achieving more clarity and the auditors agreed to contribute to developing an approach that could accommodate the needs of the Proficiency Test Programme and of Accreditation.

After the technical discussions the auditors took a bus and drove to a forest a few kilometers north of Zurich. Despite the early afternoon rain showers and thanks to the support by the Swiss Laboratory and Silvia Zanetti providing transport, there was an improvised barbecue on a public fire place. We all enjoyed this outing very much, especially as the technical auditors hardly ever meet personally. During audits there is mostly one technical auditor and one system auditor and this was one of the rare occasions where we all came together. Silvia, Martina and Gerhard, with all benefits of being locals, managed to organise things at short notice. Ana, who has the burden of organising the audit trips also joined and eventually met people in person she only knew from e-mail till then.

Time flew and it was just before midnight when we returned to the hotel after a few hours in enjoyable company sitting by the fire with roasted sausages and Swiss potato salad.
It is for almost two years that the accreditation database has been used to record audit observations from ISTA accreditation assessments. An evaluation of non-conformities on the level of the Association as done for the ISTA Extraordinary Meeting in Santa Cruz in 2002 is now relatively easy to perform for any period after 2004.

More than half of the 37 audits for which records are kept in the data base were third (re-accreditation) audits, almost one quarter were first audits. The remainder were second and a few were fourth audits (Figure 1). The number of non-conformities in a single laboratory vary from one to 48. No distinction between substantial and non-substantial non-conformities is made for the purpose of this overview. During accreditation audits substantial non-conformities have to be addressed by the laboratory before a recommendation for accreditation by the audit team is put to the ISTA Executive Committee. Non-substantial non-conformities have to be addressed at latest before the next accreditation assessment takes place.

The non-conformities are grouped according to the checklist sections representing the different chapters of the ISTA Seed Testing Laboratory Accreditation Standard (Figure 2). As these chapters contain different requirements, both in number and nature, the potential for having non-conformities is not the same in all the sections. In addition some non-conformities cut across several sections and their classification may carry an element of arbitranness. For example, the section ‘records’ attracted few observations, particularly as record keeping was hardly ever considered an overall weak spot in a laboratory. If record related observations were made, these were often limited to the specific area in which they occurred and classified accordingly.

Only the areas with a higher number of non-conformities will be presented in this summary. The complete presentation can be viewed at the ISTA website in the ‘meeting presentations’ section (https://www.seedtest.org/upload/cms/user/Presentation of the Accreditation Department.pdf)

**Equipment and Environment**

Non-conformities in this area were mostly related to single pieces of equipment that were not appropriately checked for being within specification, or for which specifications were not defined in a way that these checks could be performed and used for systematic verifications.

**Calibration, reference and testing material**

In this section missing control limits, inadequate reference materials or inappropriate handling of reference materials were observed (Figure 4).

**Sampling**

Sampling is an activity often taking place at locations where the immediate influence of the laboratory is rather limited and by staff operating with less supervision than analysts. Inappropriate equipment for drawing, manipulating or transporting samples, including insufficient validation of automatic sampling arrangements were the major causes for audit observations (Figure 5).

**Quality assurance system**

Within the documented quality systems implemented in laboratories seeking accreditation, document control has attracted the highest number of observations. Outdated, uncontrolled and not regularly reviewed documents gave raise to non-conformity statements. Others were management reviews and internal audits that were not comprehensive enough or not performed according to the documented plan. In this particular area, non-conformities were often related to the lack of suitable records (Figure 7).

**Methods and procedures**

Deviations from the Rules’ prescriptions in the laboratories’ technical protocols accounted for more than half of the non-conformities in this section. They also include that Rules changes were not implemented and that work instructions were incomplete.

**Conclusion**

On-site assessments help to detect inconsistencies in applying the ISTA Rules and contribute to establish and maintain uniformity in seed testing. Even for experienced laboratories that renew accreditation for the first, second or third time, opportunities for improvement and continual benefits may be accomplished.

Technical Committees can use data from evaluating non-conformities for improving the clarity of the Rules’ text and identifying areas of priority for Rules changes.
Evaluation of Non-conformities Observed During ISTA Accreditation Audits

Figure 4: Calibration, Reference and Testing Material

Figure 5: Sampling

Figure 6: Methods and Procedures

Figure 7: Quality Assurance System

New ISTA Publications

ISTA Handbook on Flower Seed Testing
1st Edition

By the ISTA Flower Seed Committee
Editor: Z. Ripka

Price: CHF 264.00*
(approx. US$ 195.00/EUR 169.00)


By the ISTA Germination Committee
Editor: R. Don

Price: CHF 264.00*
(approx. US$ 195.00/EUR 169.00)

Amendments 2006
Price: CHF 74.00*
(approx. US$ 61.00/EUR 47.00)


By the ISTA Moisture Committee
Editors: H. Nijenstein, J. Nydam and R. Don

This handbook provides additional help to Chapter 9: Moisture of the International Rules for Seed Testing. Its detailed instructions, interpretations of Rules sections, and examples of how to calculate results in detail should prove to be of value to those concerned with Seed Moisture Testing.

Price: CHF 264.00*
(approx. US$ 195.00/EUR 169.00)

CHF = Swiss France
* (50% off for ISTA Members) plus shipping costs

To order, contact the ISTA Secretariat or visit www.seedtest.org
6th ISTA Proficiency Test on GMO Testing on *Brassica napus* L.

1. Aim
The aim of the proficiency test is to check the ability of individual laboratories to detect the presence or absence of GM seeds and to quantify their presence in samples of conventional seed of oil rape seed, *Brassica napus*.

2. Experimental Design
Samples were either negative, i.e. did not contain any transgenic events, or positive, i.e. contained the transgenic event GT73 (synonym RT73). When preparing the positive samples different quantities of GT73 seeds were mixed with non-GM seeds. The genetic purity was tested prior to the sample preparation.

The GT73 seeds in the samples have the figwort mosaic virus (FMV) 35S promoter, the CP4 EPSPS and gox247 genes. The CP4 EPSPS gene encoding the CP4 EPSPS protein (5-enolpyruvylshikimate-3-phosphate synthase) confers tolerance to the glyphosate herbicide (the active ingredient in Roundup Ready®). The gox247 gene encoding a modified version of glyphosate oxidase, a bacterial enzyme from *Ochrobactrum anthropi* improves the affinity of the enzyme for glyphosate.

Each participating laboratory received a set of 10 oil rape seed samples each containing about 3300 seeds. The positive samples were made positive by adding a defined number of seeds from the GT73 seed lot to the negative seeds. For each sample, the non-GM seeds were weighted and the GM seeds were counted and the weight determined. Three samples were negative and seven samples were positive. Two samples contained 0.3% GT73 seeds, three samples contained 0.6% and two samples contained 1.2% GT73 seeds. The choice of the method used for testing was at the laboratory’s discretion.

3. Results
Fifty laboratories received samples. Forty-nine submitted their results. Fourteen laboratories submitted only qualitative results. Twelve laboratories performed the quantification using the sub-sampling strategy. Twenty-three laboratories reported quantitative results performing a quantitative test (e.g. RT-PCR). One laboratory did not report data.

The identity of the individual laboratories is kept confidential.

3.1 Descriptive Statistics of the Qualitative Results
Each laboratory reported for the individual sample whether this is a negative sample or a positive sample. This could be either derived from the quantitative test result, or from a separate test on the sample. For a given sample, the result reported by the laboratory can be either correct or false (Table 1).

Out of the 49 laboratories:
- Forty-five laboratories classified all ten tested samples correctly. These are 91.8% of the laboratories.
- 97.6% of the 489 samples were reported correctly by the 49 laboratories.
- In total, four laboratories reported results falsely reporting both, false positives and false negatives. These are 8.2% of the laboratories.
- The four laboratories reported false positives (between one (1/3) and three (3/3) out of the three negative samples) with a total number of eight out of 147 negative samples tested. These are 5.4% of the negative samples.

Table 1: Number and percentage of samples for which false results were reported.

<table>
<thead>
<tr>
<th></th>
<th># of samples tested</th>
<th># of false results</th>
<th>% of false results</th>
</tr>
</thead>
<tbody>
<tr>
<td>All samples</td>
<td>489</td>
<td>12</td>
<td>2.4</td>
</tr>
<tr>
<td>Negative samples</td>
<td>147</td>
<td>8</td>
<td>5.4</td>
</tr>
<tr>
<td>Positive samples</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.3% GT73 content</td>
<td>97</td>
<td>2</td>
<td>2.0</td>
</tr>
<tr>
<td>0.6% GT73 content</td>
<td>147</td>
<td>1</td>
<td>0.7</td>
</tr>
<tr>
<td>1.2% GT73 content</td>
<td>98</td>
<td>1</td>
<td>1.0</td>
</tr>
</tbody>
</table>

by Bettina Kahlert
ISTA GMO Task Force Proficiency Test Working Group Leader
ISTA Secretariat, Switzerland
bettina.kahlert@ista.ch

Seed Testing International No. 132 October 2006 31
The four laboratories reported false negatives (1/7) with a total number of four out of 3421 positive samples tested. These are 1.2% of the positive samples. Two laboratories reported false negatives for the samples with a spiking level of 0.3%. Both classified 1/3 samples falsely as negative with a total number of two samples out of 97. These are 2.0% of the samples. One laboratory reported false negatives for the samples with a spiking level of 0.6%. It classified 1/3 samples falsely as negative. This is one sample out of 147 or 0.7% of the 0.6% GM seed samples. One laboratory reported false negatives for the samples with a spiking level of 1.2%. It classified 1/3 samples falsely as negative. This is one sample out of 98 or 1.0% of the 1.2% GM seed samples.

3.2 The Quantitative Results
Twelve laboratory reported the number of sub-samples tested, the size of the sub-samples (number of seeds) and the number of positive sub-samples per sample (see Table 2). These elements were used by the laboratories to compute the estimate of the percentage of GM seeds (Figure 1).

The seedcalc6 programme was recommended to use for designing the testing plan and to perform the computation (freely available on the ISTA Website).

This quantitative test was for checking the ability of the laboratories to quantify the GM seeds in a sample. The laboratories could use the method they thought appropriate. The results were given in percentage of GM seeds in the sample.

Twenty-three laboratories performed the quantitative test and reported for the individual test sample the estimated value of the GM content as the percentage GM seeds in number of seeds, percentage mass of GM seeds or percentage GMO DNA copies (Figure 2).

Table 3 shows the overall performance of the laboratory regarding the different spiking levels: The (overall) mean of the quantitative and sub-sampling test results for each spiking level, the standard deviation, the variation coefficient and the relative error among the samples within each spiking level. The variation coefficient (% variation coefficient = standard deviation/mean*100) shows the inter-sample variability. The results show the lowest variation for the spiking level 0.6% and similar variation for 0.3% and 1.2%. These variation coefficients are similar to the ones of previous test rounds. The relative error (% relative error = [reported value – true value]/true value*100) shows the closeness of agreement between the reported value (test result) and the true value. There is no significant difference between the relative errors.

3.3 Summary
The percentage of laboratories reporting correct qualitative results for all samples was higher as in the previous tests, i.e. >90% of labs made no misclassification, and >97% of samples were reported correctly.

The test plan selected by the laboratory for the sub-sampling quantification had a bigger influence on the results, i.e. test plans with a lower number of sub-samples led to results with a higher variation and distance from the true value and to missing values for the higher spiking level of 1.2%.

The quantitative results showed similar variation for the spiking levels (VC~50%) and there was a high number of laboratories overestimating the true value.

---

**CALL FOR REGISTRATION**

**8th and 9th ISTA Proficiency Test on GMO Testing**

Since GMO testing has been included in the ISTA Accreditation Programme, the participation in the ISTA Proficiency Tests on GMO Testing is compulsory for those laboratories which have GMO testing methods in their scope of accreditation. The ISTA GMO Proficiency Test Programme on GMO Testing is also open to all laboratories involved in GM seed testing. Your laboratory can select the method appropriate to detect the presence or absence of GM seeds, to quantify and to identify their presence in samples of conventional seeds.

**8th ISTA Proficiency Test on GMO Testing on Glycine max (L.) Merr.**

Your laboratory will receive 14 soybean test samples each of 3000 seeds, either containing GM seeds or not.

**Registration deadline: December 31, 2006**

**9th ISTA Proficiency Test on GMO Testing on Zea mays L.**

Your laboratory will receive maize samples, either containing GM seeds or not.

**Registration deadline: April 30, 2007**

Registration forms and further details on proficiency tests can be found on the ISTA Website at [www.seedtest.org](http://www.seedtest.org)

Laboratories interested in participating should contact the:

ISTA Secretariat:

Zürichstrasse 50, PO Box 308

8303 Bassersdorf

CH – Switzerland

Phone +41 44 838 6000

Fax +41 44 838 6001

E-mail ista.office@ista.ch
6th ISTA Proficiency Test on GMO Testing on *Brassica napus* L.

Table 1: Quantitative test – The (overall) mean of the quantitative test results for each spiking level, the standard deviation, the variation coefficient and the relative error among the samples within each spiking level. The standard deviation, the variation coefficient and the relative error in this table are related to the single results per sample and not to the laboratories’ means.

<table>
<thead>
<tr>
<th>Spiking level (%)</th>
<th>Replicates</th>
<th>Mean ± SD (%)</th>
<th>Variation coefficient</th>
<th>Relative error (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.3/0.33</td>
<td>65</td>
<td>0.38 ± 0.22</td>
<td>59.3</td>
<td>17.9</td>
</tr>
<tr>
<td>0.6/0.67</td>
<td>94</td>
<td>0.80 ± 0.40</td>
<td>49.6</td>
<td>19.0</td>
</tr>
<tr>
<td>1.2/1.35</td>
<td>61</td>
<td>1.57 ± 0.90</td>
<td>57.9</td>
<td>20.0</td>
</tr>
</tbody>
</table>

Figure 1: The results of the sub-sampling quantification: Estimates of the percentage of GM seeds reported by the laboratory (circles) for each sample of the different spiking levels: 0.3% (a), 0.6% (b) and 1.2% (c) GT73 seeds and the mean of these estimates for each laboratory (black short line). The mean is only shown in case that at least three estimates were reported as values. The red line shows the spiking level.

Figure 2: The results of the quantification: Estimates of the percentage of GM seeds reported by the laboratory (circles) for each sample of the spiking levels 0.6% (0.67% GT73 seeds, and the mean of these estimates for each laboratory and the standard deviation.

Table 2: The table shows testing plans used by the laboratories (lab #), i.e. number of sub-samples and the size of sub-samples (in number of seeds).

<table>
<thead>
<tr>
<th>Lab #</th>
<th># of sub-samples</th>
<th>Size of sub-samples</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>~300</td>
<td>10</td>
</tr>
<tr>
<td>2</td>
<td>~90</td>
<td>30</td>
</tr>
<tr>
<td>3, 4</td>
<td>~30</td>
<td>~110</td>
</tr>
<tr>
<td>5, 6</td>
<td>20-25</td>
<td>~150</td>
</tr>
<tr>
<td>7, 8</td>
<td>15</td>
<td>210-220</td>
</tr>
<tr>
<td>9, 10, 11</td>
<td>5-10</td>
<td>330-550</td>
</tr>
<tr>
<td>12</td>
<td>2-3</td>
<td></td>
</tr>
</tbody>
</table>

Table 3: Quantitative test – The (overall) mean of the quantitative test results for each spiking level, the standard deviation, the variation coefficient and the relative error among the samples within each spiking level. The standard deviation, the variation coefficient and the relative error in this table are related to the single results per sample and not to the laboratories’ means.

<table>
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<tr>
<th>Spiking level (%)</th>
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<td>61</td>
<td>1.57 ± 0.90</td>
<td>57.9</td>
<td>20.0</td>
</tr>
</tbody>
</table>
The GMO Task Force (GMO TF) had the opportunity to discuss the ISTA Proficiency Tests on GMO Testing in more detail at the ISTA Annual Meeting, Zurich, Switzerland. Also the participants of the ISTA Proficiency Tests on GMO Testing (PTs) were invited to join this informal meeting to discuss technical and theoretical aspects of GM seed testing. Twenty-five participants attended the meeting.

The aim of the meeting was to exchange ideas following the presentations given in the main meeting (see http://www.seedtest.org/en/content---1--1205.html) and to discuss specific questions coming from the group or submitted by PT participants by email prior to the meeting.

In the following section you can find extracts from the discussion.

**ISTA Proficiency Tests on GMO Testing (PTs)**

The meeting started with discussing if the PTs are only designed in such a way that they are primary adequate for using of PCR methods rather than other methods like bioassay or protein methods. Because bioassays are only appropriate for some events, for example, Roundup Ready, but not for other traits which were used in some PTs. This was the case in PT round 05 were the event A2704 was included.

Independent of the method type, it is important that laboratories use a test that is fit for purpose, so they need to constantly review the test schemes. Further, methods should be evaluated, approved and implemented, i.e. shown that they produce reliable and reproducible results. The methods are not specified by ISTA but ISTA supports the use of all methods which follow the above criteria. Concerning the organisation of PTs, ISTA aims at reflecting the real world. For example, this could mean to include an event with two copies of the promoter which is difficult to quantify or an event where a spray test is not appropriate.

The Exchange of Information Working Group of ISDA will take into consideration to add information on bioassays to the specified trait area of the ISTA website and therefore, will ask for contributions in this area from experienced people currently using these methods.

**Rating of PTs**

The discussion continued on how laboratories are to be graded. It was explained that the grading is based on a six-round average. For example, a laboratory can have four Bs and two BMPS and will still achieve an overall C. On the question if the system is stringent enough, it was answered that the use of grades needs to reflect ‘state of the art’ in testing. It can be changed every year and adjusted as needs. Concerning the organisation of PTs, ISTA aims at reflecting the real world. For example, this could mean to include an event with two copies of the promoter which is difficult to quantify or an event where a spray test is not appropriate.

The Exchange of Information Working Group of ISDA will take into consideration to add information on bioassays to the specified trait area of the ISTA website and therefore, will ask for contributions in this area from experienced people currently using these methods.

**Accreditation**

There were few questions regarding the laboratory accreditation for determination of specified traits in seed lots. In this context it was important for the participants to know the consequences of failing in PTs for traits they do not test for or they are not accredited for. Since testing of seed with specified traits has been included in the ISTA Accreditation Programme, the participation in the ISTA Proficiency Tests on GMO Testing is compulsory for those laboratories accredited or currently in the process of accreditation for testing of seed with specified traits. If the scope of the PT round does not correspond to the laboratory’s scope of accreditation it shall seek advice from the GMO Proficiency Test Working Group Leader. A consequence could be that a laboratory is exempt from participating in a PT or testing of samples on species or traits that are not included in its scope of accreditation. Voluntary participation is still possible.

**ΔCt versus dilution series for standard curve in real-time PCR**

It was not possible to come to a conclusion if it is better to use the ΔCt method or a dilution series to build the calibration curve for quantification. Experience shows that both methods can work, and that appropriate curves can be obtained in both ways. However, there might be other reasons why to opt for the one or the other standard curve, regulative ones or the cost considerations. For example, in some countries regulations determine how to report results. Reporting results affects the decision on how to define the standard curve. A decision can be also determined by costs,
Conductivity Testing for Phaseolus vulgaris

By Alison A. Powell
ISTA Vigour Committee Chair
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The conductivity test, validated as a vigour test for Pisum sativum, has also been shown to identify differences in vigour in other grain legumes, such as Phaseolus vulgaris, soyabean (Glycine max), chickpea (Cicer ciceris) and cowpea (Vigna unguiculata). This is not surprising since the test measures solute leakage from the seed, which is one of the consequences of the two main causes of vigour differences in all grain legume species, namely seed ageing and imbibition damage (Matthews and Powell, 2006). One of the aims of the Vigour Committee is therefore to extend the application of the validated test to other grain legumes. One step towards this aim is to establish that the test can be repeated and reproduced when applied to these other species.

A series of comparative tests were therefore set up in June 2005 to examine the application of the conductivity test to Phaseolus vulgaris. Samples of six seed lots were sent to five laboratories (AUS, FR, GB, IT, NZ) where the conductivity test was carried out on three separate occasions. The tests were completed following the same procedure as the validated test for peas in the ISTA Rules, with 4 replicates of 50 seeds, each soaked in 250 ml water for 24 h at 20°C.

The data for the replicates of each seed lot tested within each laboratory, and for the mean conductivity for each lot from the five laboratories, were compared on the basis of the tolerance values that have been established for pea in the ISTA Rules. In all cases the replicates within the laboratories and the mean values obtained for each lot in different laboratories were within the tolerance range established for peas. While there are no tolerance values for conductivity readings for Phaseolus at this stage, this does indicate that both the repeatability and reproducibility of the test is good for Phaseolus.

Two-way analysis of variance was com-
Conductivity Testing for *Phaseolus vulgaris*

Completed for the data (Tables 1–3). The mean conductivity values for the six lots in the three repeat runs of the test (Table 1) ranged from 27.0 μS cm⁻¹ g⁻¹ to 38.3 μS cm⁻¹ g⁻¹ in Run 1, 25.5 to 40.2 μS cm⁻¹ g⁻¹ in Run 2 and 25.3 to 39.0 μS cm⁻¹ g⁻¹ in Run 3. There were clear significant differences between the seed lots in each run (Table 1). Lot C had the highest conductivity, suggesting the lowest vigour. This was followed by lot D, lots E and F (similar conductivity readings), lot A, then B with the lowest conductivity and highest vigour. The ranking order of the seed lots was the same in each of the three runs of the test.

In each run there were small, but significant differences in the overall mean conductivity readings obtained in the five laboratories (Table 2), with 2 and 3 tending to have slightly higher values than the other three laboratories. This was confirmed by the analysis of the overall means (Table 3).

Comparison of the means of the three runs for each seed lot from each laboratory (Table 3) further confirmed the differences in readings between the lots (conductivity of lot C>D=E and F>A>B). The ranking of the seed lots from low to high vigour was completely consistent between laboratories (Table 3).

Calculation of the Coefficient of Variation (CV) for the whole experiment gave a value of 4.3%, indicating that there was very little variability in the data. Indeed, the good repeatability (little variability) within each laboratory, both within and between runs, explains why the small differences in conductivity between laboratories were significant.

**Conclusion**
The conductivity test can be carried out for *Phaseolus*, following the same procedure as for *Pisum sativum* in the ISTA Rules, producing both repeatable and reproducible results. This data provides the first step forwards in extending the species base for the conductivity test.

**Acknowledgements**
Thank you to Karen Hill (AUS), Marie-Hélène Wagner (FR), Gillian McLaren (GB), Emanuela Casarini (IT) and Barbara Brunton (NZ) for participating in these comparative tests. We are grateful to Dr Anthony Biddle and Mark White of the Processors and Growers Research Organisation, Peterborough, UK for providing the seeds.

**Reference**

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**Table 1: Comparison of seed lot means in each of three test runs.**

<table>
<thead>
<tr>
<th>Lot</th>
<th>Run 1</th>
<th>Run 2</th>
<th>Run 3</th>
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<tr>
<td>A</td>
<td>27.0d</td>
<td>27.2d</td>
<td>26.7d</td>
</tr>
<tr>
<td>B</td>
<td>25.7c</td>
<td>25.5c</td>
<td>25.3c</td>
</tr>
<tr>
<td>C</td>
<td>38.3a</td>
<td>40.2a</td>
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<tr>
<td>D</td>
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<td>34.9b</td>
<td>34.4b</td>
</tr>
<tr>
<td>E</td>
<td>29.9c</td>
<td>29.9c</td>
<td>30.4c</td>
</tr>
<tr>
<td>F</td>
<td>29.7c</td>
<td></td>
<td>29.7c</td>
</tr>
</tbody>
</table>

In each column, values followed by different letters are significantly different p≤ 0.05

**Table 2: Comparison of mean conductivity from five laboratories in each of three test runs.**

<table>
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<tr>
<th>Laboratory</th>
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<tr>
<td>1</td>
<td>29.2d</td>
<td>29.8d</td>
<td>28.3c</td>
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<td>2</td>
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</table>

In each column, values followed by different letters are significantly different p≤ 0.05

**Table 3: Comparison of seed lots and laboratory mean conductivity readings.**

In each column the number in parentheses is the rank order of the seed lot as determined by that laboratory, with 1 = highest conductivity reading (lowest vigour) and 5 = lowest conductivity reading (highest vigour).

<table>
<thead>
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<th>Laboratory</th>
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<tr>
<td></td>
<td>1</td>
</tr>
<tr>
<td>A</td>
<td>25.1 (5)</td>
</tr>
<tr>
<td>B</td>
<td>24.2 (6)</td>
</tr>
<tr>
<td>C</td>
<td>36.2 (1)</td>
</tr>
<tr>
<td>D</td>
<td>32.2 (2)</td>
</tr>
<tr>
<td>E</td>
<td>28.1 (4)</td>
</tr>
<tr>
<td>F</td>
<td>28.4 (3)</td>
</tr>
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</table>

Mean 29.1b 32.8b 33.8b 29.0c 30.1c

In a column, values followed by different letters are significantly different p≤ 0.05.

In a row, values followed by different upper case letters are significantly different p≥ 0.05.
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<th>Country</th>
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<th>Fax</th>
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<td>IN09</td>
<td>New accredited ISTA Member Laboratories</td>
<td>Seed Testing Laboratory</td>
<td>Monsanto, Bellary</td>
<td>+91 839 291005</td>
<td>+91 839 291008</td>
<td><a href="mailto:poornachandra.rao@monsanto.com">poornachandra.rao@monsanto.com</a></td>
</tr>
<tr>
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<td>VN01</td>
<td>New accredited ISTA Member Laboratories</td>
<td>NCVESC</td>
<td>Seed Testing Laboratory Hanoi</td>
<td>+84 4 821 3453</td>
<td>+84 4 971 2054</td>
<td><a href="mailto:ncvesc-dung@fpt.vn">ncvesc-dung@fpt.vn</a></td>
</tr>
<tr>
<td>BE - Belgium</td>
<td>BE02</td>
<td>Re-accredited ISTA Member Laboratories</td>
<td>Laboratorium voor Zaadontleding</td>
<td>Burgemeester van Gansberghelaan 109</td>
<td>+32 9 272 2703</td>
<td>+32 9 272 2711</td>
<td><a href="mailto:kristine.rooms@ewbl.vlaanderen.be">kristine.rooms@ewbl.vlaanderen.be</a></td>
</tr>
<tr>
<td>CH - Switzerland</td>
<td>CH01</td>
<td>New accredited ISTA Member Laboratories</td>
<td>Forschungsanstalt Agroscope Reckenholz-Tänikon ART Saatgutprüfung</td>
<td>Reckenholzstrasse 191</td>
<td>+41 44 3777111</td>
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<td><a href="mailto:silvia.zanetti@art.admin.ch">silvia.zanetti@art.admin.ch</a></td>
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<td>DE - Germany</td>
<td>DE04</td>
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<td>Staatliche Landwirtschaftliche Untersuchungs- und Forschungsanstalt Augustenberg Referat 3.1: Saatgutuntersuchung und angewandte Botanik</td>
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</tr>
<tr>
<td>NL - Netherlands, The</td>
<td>NL05</td>
<td>New accredited ISTA Member Laboratories</td>
<td>Advanta Seeds B.V.</td>
<td>Dijkwelsestraat 70</td>
<td>+31 113 347911</td>
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<td>NZ - New Zealand</td>
<td>NZ02</td>
<td>New accredited ISTA Member Laboratories</td>
<td>Centre for Plant Reproduction &amp; Seed Technology</td>
<td>Institute of Natural Resources Massey University P.B. 11 222 Palmerston North</td>
<td>+64 6 3505790</td>
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<td>Agri Seed Testing, Inc.</td>
<td>1930 Dacov Court SE Salem, Oregon 97302</td>
<td>+1 503 585 1440</td>
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What does seed quality mean in terms of germination?
Seed germination is subjected to very precise regulation, the complexity of which originates both in the action of various external factors (temperature, oxygen, light, water potential of the medium) and in characteristics within the seeds themselves. Germination of a seed lot results from the functioning of the genome, but is also largely dependent on numerous factors which intervene throughout the seed life, from its development on the mother plant up to sowing. A seed population is therefore heterogeneous, which leads to a lack of uniform performance especially when environmental conditions at sowing are not optimal. Seed quality includes genetic purity, health quality and seed germination and vigour. Successful stand establishment requires high quality seeds, i.e. seeds that

- germinate completely,
- germinate quickly and simultaneously,
- produce normal and vigorous seedlings, and
- have germination which shows little sensitivity to external factors enabling them to germinate in a wide range of agro-climatic conditions.

To these criteria can be added storability of the seeds which can also be added. Plant breeding and advances in seed technology such as priming have led to improved seed quality.

What is seed priming?
Seed priming is widely used for enhancing seed performance by improving the rate and uniformity of germination and decreasing seed sensitivity to external factors. This technique is based on the progress of germination in three phases: imbibition (phase I), germination stricto sensu (the true germination process) (phase II), and growth (phase III). In particular, water uptake follows this triphasic pattern (Figure 1) with an initial rapid imbibition phase (phase I), followed by a lag period (phase II) referred to as germination stricto sensu, and finally a second water uptake phase associated with radicle growth (phase III) (Côme, 1980; Bewley 1997). Imbibition results in the resumption of respiratory activity and protein synthesis using extant mRNAs. Phase II, the most important phase, is associated with various cellular and biochemical events including mitochondria repair and synthesis, protein synthesis relying on the translation of new RNA, changes in soluble sugars, etc (Bray, 1995; Bewley, 1997). The objective of seed priming is to allow a controlled water uptake by the seeds up to the end of phase II, before the radicle protrudes from the seed coat. Since most seeds are desiccation tolerant up to this stage, the germination process can be arrested by drying. Phase II is more sensitive to external factors than phase III (Côme and Thévenot, 1982), therefore seeds that have passed through this phase in the priming process germinate in a wider range of environmental factors than control, non-primed seeds.

Several methods are used for controlling seed water uptake: priming with an osmotically active agent (Figure 1), usually a salt or polyethylene glycol (osmopriming) (Bradford, 1986; Khan, 1992), with a water-absorbing carrier (solid matrix priming or matric conditioning) (Khan, 1992), with pure water (prehydration in water) or with water vapour (drum priming) (Rowse, 1996). The major problem encountered in seed priming is to either (a) control seed imbibition to a level permitting the achievement of the germination stricto sensu but preventing radicle emergence, or (b) to stop the imbibition of seeds in free water before phase III starts.

Factors affecting seed priming
The stimulatory effect of priming depends on the conditions (temperature, seed water content, oxygen availability) and the duration of the treatment. Water potential for osmopriming is usually between -0.5 and -2.0 MPa depending on the species, but generally seed moisture content achieved during priming is maintained at around 40% (fresh weight basis), which is lower than the moisture content that would permit radicle elongation (Bradford, 1995). The beneficial effect of priming on subsequent germination increases with the duration of the treatment and usually becomes optimal after about 5-7 days. The ranges of temperatures and oxygen concentrations which are effective during priming are similar to those which allow germination of unprimed seeds, which suggests that priming might correspond to the realization of germination stricto sensu (phase II of the germination process). For example, the optimal temperature for priming and germination

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**Figure 1.** Changes in water uptake during germination in the presence of water (1) or an osmotically active agent (2). Phase I: imbibition; phase II: germination stricto sensu; phase III: growth.
of unprimed seeds is around 25°C for tomato (Özbingöl, Corbineau and Côme, 1998), 20-25°C for sunflower (Smok, Chojnowski, Corbineau and Côme, 1993) and 10-15°C for leek (Corbineau, unpublished data). To be efficient on subsequent germination, the priming treatment requires more than 5% oxygen in the atmosphere (Özbingöl, Corbineau and Côme, 1998; Capron, Corbineau, Dacher, Job, Côme and Job, 2000), indicating that metabolic processes are necessary for syntheses associated with priming (Bray, 1995).

**Beneficial effects of priming**

The germination of primed seeds is more rapid and uniform than that of unprimed ones since germination stricto sensu (phase II) has occurred in all the seed population during the treatment. Priming encourages better germination in a wider range of temperatures and in hypoxia. For example, osmoprimed tomato seeds germinate to higher percentages at 35°C than unprimed seeds and are able to germinate at 10°C whereas control seeds do not germinate at this low temperature (Figure 2A). Leek seeds do likewise (Figure 2B). Primed seeds are also less sensitive to oxygen deprivation (Corbineau, Picard and Côme, 1994; Özbingöl et al., 1998). This technique has been demonstrated to improve germination of seeds of numerous vegetable species (e.g. tomato, leek, carrot, celery, onion, lettuce, cabbage, cauliflower, endive, cucumber, water melon, melon) and flowers (e.g. pansy, begonia, petunia, primula, salvia, verbena) (McDonald, 2000). Such treatment also activates the germination of other crop species such as sweetcorn, sunflower and sugarbeet.

Experiments in the field have shown that priming increases the percentage of seedling emergence in suboptimal conditions of sowing, and results in an increase in mean plant weight.

An added advantage of priming is improvement of the germination of low vigour or aged seeds of various species. This improving effect is observed as long as seeds remain viable. However, the more the seeds are aged the longer they must be primed for restoring their original germination ability. Bailly, Benamar, Corbineau and Côme (1998) have shown that recovery of the germination performance of aged sunflower seeds is associated with a decrease in lipid peroxidation linked to the recovery of detoxifying enzyme (superoxide dismutase, catalase and glutathione reductase) activities occurring during the priming treatment.

**Risks of priming**

For priming to be of any practical interest its beneficial effects must be maintained after drying back the seeds to their original moisture content and during storage. The stimulatory effect of priming generally persists after drying back, but drying must be performed slowly and at relatively low temperatures. Conflicting results have been obtained concerning the influence of storage of primed seeds upon their subsequent longevity (Bray, 1995; McDonald, 2000). Various reports have shown that it is possible to store primed seeds without losing the beneficial effect of priming, but other findings have demonstrated that primed seeds often deteriorate faster during storage than untreated ones.

**Markers of priming**

There is a strong interest in the characterization of biochemical or molecular markers for use by the seed industry in the design of priming protocols because, at present, optimization of these treatments relies solely on carrying out germination assays which can only yield a posteriori indications on the priming conditions (i.e. duration, water potential and availability, temperature, oxygen availability). The beneficial effects of priming are associated with various metabolic, biochemical, cellular and molecular events including synthesis of proteins, RNA and DNA (Bray, 1995), and cell cycle processes (β-tubulin accumulation, percentage of 4C DNA nuclei) (Lanteri, Saracco, Kraak and Bino, 1994; De Castro, Zheng, Bergervoet, De Vos and Bino, 1995; Özbingöl, Corbineau, Groot, Bino and Côme, 1999). Figure 3 shows that for tomato seeds primed at temperatures ranging from 10 to 25°C, a positive linear correlation exists between the efficiency of osmopriming, evaluated by the reciprocal of time to obtain 50% germination at 15°C, and the 4C/2C values in radicle cells. Priming also results in a decrease in oligosaccharide content and in an increase in sucrose, the content of which is clearly correlated with the efficiency of the treatment in carrot (Figure 4) and fennel seeds (Corbineau, unpublished data). There is also a linear relationship between the extent of 11S globulin B-subunit and the advancement of priming in sugar beet seeds (Job, Kersulec, Ravasio, Chareyre, Pépin and Job, 1997), and accumulation of endo-b-mannanase is a good indicator of priming effect in tomato (Still, Dahal and Bradford, 1997). Bailly, Benamar, Corbineau and Côme (1997) demonstrated that catalase activity is also sub-linearly correlated with germination rate of primed sunflower seeds. In addition, the ability of sunflower and carrot seeds to convert 1-amino-1-cyclopropane-1-carboxylic acid (ACC) into ethylene is well correlated with the efficiency of priming (Chojnowski, Corbineau and Côme, 1997; Corbineau, unpublished data). Thus, ethylene ACC-dependent synthesis is a good indicator of mem-

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**Figure 2:** Effects of temperature on the germination percentages obtained after 7 days with unprimed seeds (1) and primed seeds (2) of tomato (A) and leek (B). Seeds were primed for 7 days at 15°C on a PEG-8000 solution at -1 MPa (tomato) or -1.5 MPa (leek) and then germinated in water. Means of 4 replicates.
brane properties, since it is mediated by ACC oxidase, the in vivo activity of which depends on membrane integrity.

Conclusion

Priming appears to be an effective physiological treatment for improving seed performance. It results in a better homogeneity of germination and emergence especially when the environmental conditions are not optimal, since primed seeds are less sensitive to temperature and oxygen deprivation. Although priming increases seed vigour according to criteria such as increase in rate of germination and decrease in sensitivity to external factors, conflicting results concern its effect on seed resistance to ageing. Seed companies have to take into consideration this risk of deterioration during drying and storage.

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Identification of Region of Provenance for a Rationale Management of Common Ash (Fraxinus excelsior L.) Seeds

By Piero Belletti¹, Diana Ferrazzini¹, Ignazio Monteleone¹, Beti Piotto²

Abstract
The identification of Regions of Provenance is a fundamental aspect for a rational management of activities involved in forest trees propagation, with particular emphasis on seed collection. The aim of the study was the definition of areas within the Italian distribution of common ash (Fraxinus excelsior L.) that are homogeneous both for ecological aspects and genetic characteristics of the populations and useful to delineate seed zones for this species.

Thirty-one natural ash populations were sampled and genetic variability distribution was evaluated through analysis of neutral DNA markers (microsatellites). In the meantime, the ecological characteristics of the collection sites were analysed and homogeneous regions were defined.

Sampled populations showed a large genetic variability, although its distribution is only partially related with geographical distances between populations. Furthermore, genetic differentiation among populations is low, due to the lack of barrier to gene flow. The soil does not seem to influence the distribution of genetic variability, while the climatic conditions have a major effect. Notwithstanding the low genetic differentiation it was possible to identify six Regions of Provenance, sufficiently homogeneous both for ecological and genetic conditions.

Introduction
Common ash is a forest tree widespread in most of Europe, from the Atlantic ocean to the Don rive and Kaukas mountain, and from the southern part of Scandinavia to the Mediterranean Sea. It is a pioneer species that occurs in mixed deciduous forest of plain and hilly regions, as well as in mountain areas up to 1.500 m above sea level. The species is increasing its diffusion, mainly in areas cultivated in the past, but that now are abandoned.

Forest management of common ash in Europe has shown increasing interest in the last 40 years, mainly due to the high demand for its quality timber. Furthermore, the species has been recognized as a prominent element of forest ecosystems, which provide substantial benefits for human society. Despite the increasing interest, only a few European countries have gene conservation or tree breeding programmes in place for common ash (Pliùra and Heuertz, 2003).

Usually, common ash is propagated through natural regeneration. Sometimes, however, reforestation programmes are carried out using vegetative material produced in nurseries. In the latter case, the genetic quality of the used material is of utmost importance, both to guarantee good chance of success to the afforestation and to preserve natural genetic variability of the species (Bosher and Ama-ral, 2004). In fact, forest trees are non-mobile and long-lived organisms, which grow under environmental conditions that are heterogeneous in time and space. Moreover, they are exposed to many stress factors, most of which are due to human activities: pollution, climate change, and habitat fragmentation. In order to survive these threats, and to persist over time, a high adaptive potential is need-ed: this is mainly determined by the within-species genetic diversity. Programmes aimed at the conservation of forest genetic resources should address the issue of maintenance of this diversity (Namkoong, 1998).

European Union faced with this problem, issuing the Council Directive 1999/105, that concerns the marketing of forest reproductive material. One of the most important feature of the act is the definition of Region of Provenance, that is “...the area or group of areas subjected to sufficiently uniform ecological conditions in which stands or seed sources showing similar phenotypic or genetic characters are found...”. The identification of regions of provenance is therefore a basic aspect for a rational management of activities linked with forest trees propagation, including afforestation and in situ genetic preservation. Regions of provenance should be defined according to adaptative features of the populations. However, the scoring of such characters is very difficult, due to the high demand in time and space of the needed cultivation trials.

The purpose of the study was the evaluation of neutral DNA markers as a tool to study genetic variability distribution of common ash in Italy, and to group populations according to their genetic similarity, so defining Regions of Provenance and creating basis to delineate seed zones for this species.

Materials and methods
Pedo-climatic characteristics of the study area were inferred from existing cartography, namely the Soil Regions of Europe developed by the European Commission (1999). This document reports the climate types that are present in Europe according to the CLIMWAT database, and joins regions according to their climate, geology and pedology characteristics.

Thirty-one natural populations of Fraxinus excelsior were chosen within the natural area of diffusion of the species in Italy (northern part of the country). All the populations belong to mixed forests, in which common ash is never the dominant species, although a certain variation among stands according to ecological conditions could be detected. Buds or young leaves were sampled from about 30 non-adjacent trees in each population, randomly chosen over a 5 to 10 ha area. After collection, buds and young leaves were frozen at –20°C until DNA extraction. Six primers pairs of microsatellite loci were used for the polymerase chain reactions and for the estimation of genetic parameters, namely mean number of alleles per locus, average observed heterozygososity and average gene diversity. Moreover, the total variance was partitioned into components due to differences among populations within pedo-climatic regions, among regions and within populations.

Results and Discussion
The area where material was sampled was divided in regions that are homogeneous as regards their geological, pedological and climatic characteristics. It was possible to
define the following 5 regions: Po Valley, Ligurian Mountains, Alps with crystalline soil, Alps with calcareous soil, Apennines.

Genetic diversity assessed with DNA markers was considerable and comparable with those of an analogous study carried on common ash populations in Bulgaria, that is an important refuge for plant species, including common ash, during the Quaternary glacial period. In this region species are therefore expected to preserve high level of intraspecific biodiversity. Italian populations thus appear to constitute an important gene reservoir, not limited to the Apennine region (another possible refuge area), where indeed the level of genetic variation is slightly lower than in other areas.

Genetic differentiation among populations was quite low: only about 5% of the total genetic variability could be assigned to inter-populational variability. In spite of this low value, it was still possible to observe a pattern of population genetic differentiation. In particular, populations from Ligurian Mountain, and the majority of those belonging to Po Valley and Apennines showed a clear tendency to group together, so largely confirming the results of ecological characterization. On the other hand, no clear structure was observed for populations from the two regions in which the Alpine area was divided according to soil characteristics.

On the basis of the former statements and joining ecological and genetic data, the area under study can be divided into the following sic sectors (Figure 1), which are sufficiently homogeneous and that are therefore proposed as Regions of Provenance for common ash:

- Po Valley (populations n. 1, 2, 3, 4);
- Alps with calcareous soil (populations n. 10, 11, 14, 17, 19, 20, 21, 22, 23, 24, 25);
- Alps with crystalline soil (populations n. 6, 7, 12, 13, 15, 16);
- Hilly regions of Piedmont;
- Ligurian mountains (populations n. 5, 8, 9);
- Apennines (populations n. 26, 27, 28, 29, 30, 31).

The subdivision of Alps in two Regions of Provenance is based mainly on soil differences: in fact, neither climatic data nor genetic characteristics of populations have shown significant divergence. Therefore the identification of two regions should be considered as a precaution, with the aim to avoid movement of germplasm in locations characterized by different soil conditions. The hilly Region of Piedmont is defined only on ecological data, since no population was sampled within this Region.

The results of the study contribute to a better understanding of our knowledge of genetic variation of common ash in Italy, so making for more efficient programmes aimed at the preservation of the biodiversity. Furthermore results give useful indications of how to plan for more rational planning of the management of reproductive material, for instance in selecting the stands where seeds can be collected and in defining the areas where they should be utilised, according to the Region of Provenance definition.

Acknowledgements
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Figure 1: Provenance Regions for common ash in Northern Italy. A, Po Valley; B, Alps with calcareous soil; C, Alps with crystalline soil; D, Hilly regions of Piedmont; E, Ligurian mountains; F, Apennines.
Water Concentration Considerations in Recalcitrant/Non-orthodox Seeds

By Patricia Berjak, Deon Erdey and Norman Pammenter

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Introduction

Seeds categorised as recalcitrant are shed at high water concentrations (g H₂O g⁻¹ dry mass) and cannot withstand the loss of more than a small proportion of water without deleterious – or even lethal – consequences: such seeds are desiccation-sensitive. Hence, as originally recorded by Roberts (1973), recalcitrant seeds cannot be stored at low water concentrations (‘moisture contents’), i.e. under low relative humidity (RH) conditions, as are ideal for desiccation-tolerant orthodox seeds. In fact, storage quality and lifespan of recalcitrant seeds of several species have been found to be diminished by even a slight degree of dehydration (Corbineau and Come, 1988; Drew et al., 2000; Eggers et al., 2006).

While seed recalcitrance is generally perceived to be a characteristic of tree species, the phenomenon is certainly not restricted to woody plants. For example, many species of amaryllids, which are herbaceous geophytes, produce recalcitrant seeds (Figure 1 [Ser-

Figure 1. Fruit and seeds of Amaryllis belladonna. The variation in size of seeds from a single harvest is clearly indicated.

shen Naidoo, 2006]), as do Zizania spp., and other members of the Poaceae (Probert and Longley, 1989; Kovach and Bradford, 1992; Vertucci et al., 1995). Furthermore, although seed recalcitrance is more common in tropical tree species (e.g. Sacandé et al., 2004), there are a number of tree species of temperate provenance such as Quercus spp. (e.g. Finch-Savage and Blake, 1994; Connor and Bonner, 1996) and Aesculus hippocastanum (e.g. Tompsett and Pritchard, 1993; 1998) that also produce recalcitrant seeds. Other non-orthodox seeds, broadly categorised as showing intermediate post-harvest behaviour, are those that will withstand considerable dehydration, but cannot be dried down to the low water concentrations tolerated by orthodox types. Such seeds are generally relatively short-lived, and those of tropical provenance may be intolerant of chilling temperatures (Hong and Ellis, 1996).

As an aside, the amount of water in an orthodox seed is frequently expressed as a percentage calculated on a fresh mass basis (that is, the proportion of hydrated or partially dehydrated seed that is water), although the amount of water present can also be expressed on a dry mass basis. At the low water concentration suitable for storage of orthodox seeds it makes very little difference if data are expressed on a fresh or dry mass, but in the case of recalcitrant seeds that contain considerable amounts of water, there are marked differences in values when expressed on a fresh or dry mass basis. We prefer data to be expressed relative to dry mass. In this case the basis to which values are being normalised does not change as the amount of water changes, and the proportional change in ‘water content’ reflects the proportional change in the amount of water in the tissue; if the water concentration changes from 1.0 to 0.5 g water g⁻¹ dry mass, the tissue has lost half its water. If the data are expressed on fresh mass basis, for tissue at 1.0 g water per gram dry mass that loses half its water, water content on a fresh mass basis changes from 50% to 33.3%. However, since the deleterious processes occurring in recalcitrant seeds as they dry are influenced by the free energy of the tissue water, ideally the water status of recalcitrant seeds should be expressed in terms of their water potential.

Desiccation sensitivity of recalcitrant seeds is the outcome of two major, but inter-related features: firstly, the suite of mechanisms and processes that collectively confer the property of desiccation tolerance in orthodox seeds may be absent, or incompletely expressed, in recalcitrant types; and secondly, the seeds remain actively metabolic, grading virtually imperceptibly from development into germination (Pammenter and Berjak, 1999; Berjak and Pammenter, 2004). This second feature is at least partly the outcome of the non-occurrence of two processes of the suite of events conferring desiccation tolerance, viz. intracellular de differentiation and metabolic ‘switch-off’ (e.g. Farrant et al., 1997). The expression of recalcitrant traits, however, may well also be influenced by the degree of seed development at shedding (Finch-Savage and Blake, 1994; Daws et al., 2004b; 2005).

Variability is a key feature among recalcitrant seeds

Recalcitrance is not an all-or-nothing situation (Berjak and Pammenter, 1994). From a perusal of the literature up to 1988, Farrant and co-workers had already loosely categorised desiccation-sensitive seeds as being minimally, medially and maximally recalcitrant in terms of the proportion of water loss tolerated, responses to chilling (above zero) temperatures and storability. Since then, however, it has become apparent that variability among recalcitrant seeds of different species – and within the same species – is far more complex.

Rate of dehydration

Drying rate has been shown to be an important factor in the degree of dehydration that recalcitrant seeds or excised axes will withstand. Although there is the occasional recorded exception (e.g. for the relatively large axes of Theobroma cacao [Liang and Sun, 2000]), in general, the more rapidly dehydration is achieved, the greater the water loss that will be tolerated without irreversible injury (Normah et al., 1986; Pammenter et al., 1991; 1998; Pritchard, 1991; Kundu and Karachi, 2000; Potts and Lumpkin, 2000). Additionally, seeds of different species will lose water at different rates under the same
conditions. In a study of three unrelated species dehydrated under identical conditions, Farrant et al. (1989) showed that seeds of the species that dried most rapidly, survived to the lowest water concentration – although this level was still high. The effect of drying rate is attributed to the fact that at water concentrations still sufficiently high to permit metabolism, the damage that is accrued is a consequence of this metabolism (which is aqueous based), becoming disturbed. Slow drying permits the accumulation of considerable metabolism-linked damage, while rapid loss of water limits the time during which seeds can accumulate damage as a consequence of such aqueous-based disruption of metabolism (Pammenter et al., 1998; Walters et al., 2001). Nevertheless, there is a limit to the dehydration that recalcitrant material will tolerate before the onset of desiccation damage – sensu stricto (Pammenter and Berjak, 1999; Walters et al., 2001).

Desiccation-sensitivity
A further complicating factor is that the desiccation sensitivity of recalcitrant seeds within a species increases as germinative metabolism progresses, before there is macroscopical evidence of germination (Farrant et al., 1986; Ellis and Hong, 1991; Berjak et al., 1992; 1993; Finch-Savage et al., 1996; Tomsett and Pritchard, 1998). In the early stages of ontogeny also, the seeds are markedly more desiccation-sensitive than immediately prior to shedding at putative maturity, indicating that the more heightened the metabolic status, the greater the desiccation sensitivity (Berjak and Pammenter, 2004). Those authors also review the evidence that marked intra- as well as inter-seasonal differences in characteristics, including desiccation sensitivity, have been recorded for recalcitrant seeds of individual species.

Provenance and developmental status
While such variability has, until recently, been largely inexplicable, the data of Daws et al. (2004b; 2005) have provided elucidation. An analysis of seed characteristics, comprising fresh and dry mass, axis water concentration and solute potential, relative desiccation sensitivity and germination performance at a range of temperatures, was carried out for Aesculus hippocastanum from five defined latitudinal provenances between 57°10’ N 2°04’ W in Scotland and 38°10’ N 23°45’ E in Greece (Daws et al., 2004b). The seeds from the southernmost provenance were significantly larger (fresh and dry mass), and the water concentrations of both axes and whole seeds were significantly lower, as was the axis osmotic (solute) potential, than those from the northern extreme. These characteristics showed a gradation that was latitudinally related, across the provenances. A comparison of the A. hippocastanum seeds from the southernmost provenance in Greece with those from Scotland, the most northerly provenance, showed that those from Greece were less desiccation sensitive and germinated at lower temperatures. Principal component analysis carried out by Daws et al. (2004b) showed that a strong correlation existed between the seed characteristics and the heat sum, \( \Sigma \) [temperature at logging interval \( \times \) logging interval \( h \)]/24, along the latitudinal gradient. This led those authors to conclude that temperature had a marked influence on the rate of development of the A. hippocastanum seeds, such that those from the northern, cooler sites were less developed.

In a subsequent study, Daws et al. (2006) showed that the characteristics of Acer pseudoplatanus seeds could similarly be related to heat sum during development. Seeds of A. pseudoplatanus from nine provenances over a 21° latitudinal range (from Scotland to Italy) were assessed, with those from the northernmost provenance being of significantly lower dry mass, higher embryo water concentration and water potential, and greater desiccation sensitivity. In fact, although previous work on A. pseudoplatanus seeds from provenances in England resulted in their being categorised as recalcitrant (Hong and Ellis, 1990; Dickie et al., 1991; Greggain et al., 2000), from the study of Daws et al. (2006), those from the most southerly provenances in France and Italy were sufficiently different to lead to the suggestion that, from these latitudes, they showed characteristics of intermediate behaviour.

Those experiments (Daws et al., 2004b; 2006), in which geographical provenance, and hence heat sum, were shown to be strongly correlated with seed characteristics and responses, go far in explaining aspects of the variability – particularly that of water concentrations and desiccation sensitivity – that have been reported for recalcitrant seeds within species. Nevertheless, there usually is also some variability in any one sample, among seeds produced at individual sites (Berjak and Pammenter, 2004; Daws et al. 2004b).

There are further characteristics of recalcitrant seeds that need to be highlighted, before a consideration of the inherent difficulties of formulating rules for testing such seeds. That is, the marked difference in water concentration between axis and storage tissue (cotyledons or endosperm) that has been recorded across species by many authors (reviewed by Berjak and Pammenter, 2004; Daws et al., 2004a; Erdey and McGill, 2006). In general, axes, which make up only a small volume or mass fraction of the whole seed (Figure 2), are at considerably higher water concentrations than are the storage tissues. Hence, water concentrations of whole seeds generally reflect those of the storage tissues, while it is the characteristics of the axes that are of critical importance to the ability for seedling establishment. Additionally, when recalcitrant seeds are dried experimentally, the axes and storage tissues are likely to dry at different rates, thus measuring whole seed water concentration could obscure changes in the axes.

In addition, many tree (and other) species produce seeds that are enclosed by closely-associated fruit-derived structures which contribute significantly to the mass of the propagule at shedding. Such structures may be hard and water impervious (e.g. in the case the surrounding structures of the nuts of Carya and Juglans, and the sclerotesta of Encephalartos
spp.), or fibrous and hygroscopic (e.g. the endocarp of *Sclerocarya birrea*). In such cases, water concentration determination on a whole seed basis only (by cutting or grinding), tends to be markedly influenced by that of the fruit-derived structures. The propagatory unit of *S. birrea* is dominated by the endocarp (Gaméné et al., 1994), from which the seed cannot be separated (Figure 3). When this unit is extracted from the surrounding fruit tissues, the water concentration of the endocarp is considerably higher than that of the seed tissues. Thus water concentration determined on a whole propagule basis, is misleadingly high for the seed itself. Upon drying, however, the endocarp loses water more readily than do the seed tissues, and consequently water concentration determinations reflect a misleadingly low value for the seed tissues (Gaméné et al., 1994).

The current ISTA Rules prescribe bulk sampling, processing (cutting or grinding) and sub-sampling of large seeds of both trees and other plant forms. For the reasons discussed above, we consider that such an approach is unsuitable for water concentration assessments of non-orthodox seed material. In contrast to the ISTA approach, the practice adopted by many investigators to measure water concentration (or potential) and assess responses to desiccation, involves relatively small numbers of seeds (IPGRI-DFSC, 2004; Pritchard et al., 2004). For water concentration determination the seeds are not bulked, but are considered as single units. Furthermore, each seed is divided into its component parts, for which water concentrations are assessed individually. As a consequence, especially for recalcitrant seeds, instead of the results for bulked samples, the individual measurements of water concentrations facilitate appreciation of the variation around the mean, which can be considerable. Thus we recommend that uniformity in recalcitrant/non-orthodox seed testing, should be on the basis of the component parts of individual seeds, with data being expressed on a dry mass-specific basis (g H2O g⁻¹ dry mass).

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ISTA GMO Workshop
Methods for GM Seed Detection and Statistical Aspects
Ege University, City of Izmir, Turkey, 27–30 March 2007

ORGANISERS
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Center for Seed Technology and Department of Horticulture,
Ege University, Turkey

LOCATION
Faculty of Agriculture, Ege University, Izmir, Turkey

CONTENT
Part 1: PCR based Methods for GMO Detection
• DNA extraction, Measurement of DNA concentration (photometry)
• PCR for qualitative GMO detection (end-point PCR):
  – CaMV 35S promoter (screening)
  – NOS terminator (screening)
  – Lectin (endogenous gene for soya)
  – HMG (endogenous gene for maize)
  – RR-soya (epsp gene, event specific, nested PCR)
  – Bt11 (cryIA gene, event specific, nested PCR)
  – Bt176 (cryIA gene, event specific, nested PCR)
  – Agarose gel electrophoresis of amplified DNA and detection of the DNA bands by UV-light
  – Use of ImmunoStrips for quantitative GMO detection

Part 2: Statistical Aspects of GMO Detection
• Refresher on statistical tests
• Distributions usually found in living material, use in computations
• Determination of appropriate testing plans to thresholds or quality levels
• Introduction to Seedcalc, exercises with Seedcalc
• Getting an estimate of % GM of seed
• Repeatability reproducibility using ISO 5725 (quoted in ISO 17025)
• ISTA proficiency test and performance data evaluation.

CRITERIA
Participants should be technicians with basic knowledge/experience in the field of GMO detection and statistics. Ability to understand English well.

LECTURERS
Christoph Haldemann
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Registration form also available on: http://www.seedtest.org
Report: ISTA Seed Testing Technology Seminar
Zhejiang University, Hangzhou China May 16 –17, 2006
By Jin Hu and Norbert Leist

Around 50 Chinese participants from universities, institutes, seed testing stations, seed companies, governmental authorities and visiting scholars for Asia gathered together to discuss Methods for Sampling and Testing Seeds according the newest ISTA Rules and their correct application according to the ISTA Accreditation Standard as well as GMO testing aspects. Besides this new techniques and the abilities of their application in seed testing, with the availability of the computer tomography, determination of germination and vigor was made possible. The seminar was led by Prof. Dr. Norbert Leist, past President of ISTA and the local organizer was Prof. Dr. Jin Hu, head of Seed Science Center, Zhejiang University.

The seminar was categorized under these 13 subjects and each was followed by a question and answer session with intensive discussions:
- The Seed World and the International Seed Testing Association, ISTA
- The ISTA Accreditation System and principles of Quality Management
- Sampling the seed lot, sampling in the laboratory, the precondition for reliable results
- Purity testing, seed identification, the general principle; examples Avena, Rumex, Silene, Vicia
- Germination of seeds, biology and techniques, evaluation of seedlings: Oryza, Zea, Vigna, Glycine, Cucumis on examples using the new ISTA Handbook for Seedling evaluation
- Viability determination by the Topographical Tetrazolium Method: Zea, Helianthus, Brassica, Lycopersicum
- Vigour testing
- X-ray testing the new challenge
- Seed health testing with respect of seedborne fungi
- Variety testing in general and the use of protein electrophoresis in detail
- GMO – the Performance Based Approach of ISTA; GMO – results from the ISTA Proficiency Test; GMO – and Thresholds.

The English presentations were translated into Chinese by Jin Hu. Besides this the questions on how to become an ISTA member were intensively discussed amongst the participants. The information brochures delivered from the ISTA Secretariat supported this topic in a splendid way. Once again, Branislava Opra proofed her high organizational skills as the responsible person from the secretariat, which was very much appreciated by all the attendees.

Printed copies and additional hints for literature were handed out to participants to take home. The teaching faculty members from universities or institutes received information in the form of CDs and memory sticks for their personal use.

The representatives of the University were highly appreciative of the seminar. Earlier, they welcomed the participants at the opening ceremony. Being among the top three major universities in China, their main priorities are exchange of knowledge, active support of teaching techniques and seed science.

The logistics facilities and perfect organization were highly rated by all participants. Jin Hu and his colleagues had done a wonderful job at arranging the accommodation for all the participants at the Campus of the University. They also included a visit to Hefangjie, the famous night market of Hangzhou on the first evening and at the end of the seminar a fine supper finalized a splendid seminar which had lead to new connections and hopefully established longlasting friendships between the participants.

Norbert Leist finished the seminar with the wish for closer ties and cooperation between the Chinese colleagues with ISTA, be it by personal memberships, or by university memberships, and personal involvements in Technical Committees. Finally, he offered the invaluable opportunity of exchange of experts and laboratory training courses to improve skills in special areas of seed sampling and testing.
Report: APSA/ISTA Seed Health Training Course
Kasetsart University, Bangkok, Thailand, 18–22 July, 2006

By Valerie Cockerell, ISTA Seed Health Committee Chair
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The APSA/ISTA Seed Health Training Course in Bangkok was hosted by the Asia-Pacific Seed Association (APSA), Asian Vegetable Research and Development Center (AVRDC) Asian Regional Center and Kasetsart University. There were 19 participants from seven countries (Australia, India, Japan, Philippines, South Korea, Thailand and Vietnam). The majority of participants were from seed companies and the remainder from research institutes. The instructors were Mrs Valerie Cockerell, ISTA Seed Health Chair, Dr. Steve Roberts, a former member of the ISTA Seed Health Committee and Dr. Som-siri Sangchote, Associate Professor of the Department of Plant Pathology at Kasetsart University in Bangkok.

The training course was designed as an introduction to seed health testing for fungi, bacteria and viruses. Lectures were restricted to 1-1½ hours with practical being the main focus of the workshop. Lecture topics included, Introduction to ISTA, Sampling, Statistics and Interpretation of results, Method Validation and ISTA Accreditation and Quality Assurance, Molecular Techniques in Seed Health Testing and Serological Techniques in Seed Health Testing. Dr. Campiranon, Deputy Director of APSA made a presentation on Regional Issues which provided a good stimulus for discussion with the participants. The participants worked very hard at the practical sessions switching between mycology, bacteriology and virology, with productive discussions throughout the course. Mid week a visit had been planned to the Quarantine Offices of Kasetsart University but unfortunately had to be cancelled at the last minute. However at very short notice Efren Altoveros arranged for us to visit the Grand Palace in Bangkok.

It was a great time to visit the Grand Palace as many renovations had been completed in time for the 60th anniversary of the King of Thailand, to celebrate his 60 years as monarch. The week finished with the awarding of certificates to the participants by Dr. Campiranon, Dr. Ooi, Regional Director, AVRDC-ARC and Mrs Cockerell and a lovely tea party at the AVRDC offices. Many sweet dishes and fruits were on offer, the durian in particular was quite memorable. If you get past the aroma seemingly the taste grows on you!

There is a tremendous amount of work goes into this type of workshop and it is important to thank all of those who contributed to its success, and in particular Steve for stepping in at short notice, Efren for his organisation ensuring the smooth running of the course and to Dr. Sangchote and his staff for preparing all the samples and media and equipment.
Report: ISTA/APSA Training Course in Seed Testing  The Philippines, 12–16 September 2005

By Maribel M Querijero, Senior Agriculturist
NSQCS, Bureau of Plant Industry

On September 12-16, 2005 the Training Course in Seed Testing was hosted by the National Seed Quality Control Services (NSQCS), Bureau of Plant Industry, Quezon City, Philippines, through the initiative and support of the International Seed Testing Association (ISTA) and the Asia and Pacific Seed Association (APSA). The National Seed Quality Control Services, Bureau of Plant Industry is ISTA Member Laboratory involved in seed testing and certification and one of the participants in the ISTA Proficiency Test Programme.

Topic of the Training Course was Sampling, Purity, Germination, Tetrazolium, Seed Health Testing and ISTA Accreditation. There were 20 participants from nine countries in the Asia Pacific region namely, China (1), Hongkong (1), India (3), Japan (2), Korea (1), Philippines (7), Taiwan (1), Thailand (3) and Vietnam (1) from both government and private seed companies and institutions engaged in seed production, trade and seed quality control.

The International Seed Testing Association had an invaluable contribution in supporting the training course through the expertise of ISTA lecturers, Prof. Dr. Norbert Leist and Ms. Andrea, coming from the LUFA, Augustenberg, Germany, Department of Agrobiology; Seed Testing and Applied Botany.

The opening ceremony was held at the Sulo Hotel in Quezon City, Philippines. Mr. Lealyn A. Ramos and Mr. Dante V. Fidel, directors of The Bureau of Plant Industry, Mr. Henry T. Carpiso, NSQCS chief, Dr. Francisco Malabanan, GMA Rice Program director and Mr. Willy, APSA Co-President greeted welcomed participants at the opening ceremony.

The lectures and practicals were conducted at the NSQCS laboratory. Mr. Henry Carpiso and his staff were able to prepare interesting seed samples that opened ground for discussion and exchange of ideas and experience from respective laboratories. The technical knowledge and wide experience of the ISTA experts encouraged participants to learn more of the evaluation procedures following the ISTA rules and standards. Prof. Dr. Leist emphasized the importance of uniformity in seed testing and the establishment of standards as well as ensuring the qualified implementation of procedures that are comparable worldwide.

“We shall work in many aspects of seeds and seed life and the knowledge shall increase like the seeds grow.”

Participants had the chance to increase their knowledge:
• of the seed sampling, as the most important step in the laboratory for obtaining the representative sample of the entire seedlot
• of the significance of the purity test prior to any further test that is to shall be conducted
• that the germination test has to be conducted with the appropriate media, temperature and light requirement for each seed type
• of the ISTA accreditation and the significance of the quality management in seed testing laboratories.

Participants also had the chance to visit the International Rice Research Institute (IRRI), Los Banos, Laguna., which It was very informative and enabled participants to see the IRRI genebank and seed testing facilities as well as have a glimpse of the Philippines rice farming area.

The five day training was not only work.

Even with the very intensive schedule and interesting practicals and lectures, the participants were given the chance to explore the city life in the Philippines, to sample some Filipino cuisine and to experience some of the Philippine local culture.

“All the flowers of today are the seeds of tommorow.”

The most important major output of the training course is the new knowledge and skills gained from the ISTA experts as well as the experience exchange with other colleagues, which is are all brought back in to their respective countries. With this kind of capacity building trainings, the seed industry can become more productive and successful in the trade of the quality planting materials, particularly the SEED.
The International Seed Testing Association (ISTA) in collaboration with the Seeds and Plant Genetic Resources Services of the Food and Agriculture Organisation (FAO) and the Ministry of Food and Agriculture (MOFA) in Ghana organised a training course on Seed Quality Testing for selected African countries.

The course was held at the National Seed Testing Station, Pokoase, Ghana from March 27–31, 2006 by a total of 21 participants from the French as well as from the English speaking countries coming from Tanzania, Uganda, Kenya, Zambia from East Africa, South Africa from Southern Africa and Benin, Ghana, Nigeria, Togo, Burkina Faso, Cote D’Ivoire, Gambia, Guinea Bissau, Guinea, Mali, Niger, Senegal and Sierra Leone from West Africa. Out of this number, only three came from the private sector.

Although this course had an major objective to train participants on seed quality testing, a second objective emerged which is to introduce participants to the principles, objectives and practices of field inspection as an integral part of seed quality assurance.

By its mission statement the Association of African Seed Testing Laboratories (AASTEL) is to complement the roles of ISTA through training and accreditation of some laboratories in Africa which would conduct training, workshops, research and finally accelerate seed trade in Africa and the rest of the world. The formation of AASTEL was conceived with the view that the seed industry in Africa is facing serious challenges in relation to the task of providing good seeds for an ever expanding agricultural sector which will in turn stabilise high yields and anchor food security in the region.

After an impressive inaugural session chaired by the Ministry of Agriculture on March 27, 2006, the training course started with lectures and practical work in seed testing. The lectures were scheduled in two phases, with the first two days taken by Professor Dr. Norbert Leist, the third day was taken by Dr. Michael Larinde and the last two days by Norbert Leist. The practicals were handled by Dr. Andrea Jonitz. There were a total of 26 lectures of which 10 were practical and the rest 16 theoretical. The course covered different aspects of seed testing like seed sampling, purity estimation, identification of seeds, seed collection, use of the GRIN system, germination testing, seedling evaluation and tetrazolium testing. Seeds tested represented the main field crops of the region namely rice (Oryza sativa), maize (Zea mays), cowpea (Vigna unguiculata), groundnut (Arachis hypogaea), pepper (Capsicum spp.), soybean (Glycine max), tomato (Lycopersicon lycopersicum) and sorghum (Sorghum bicolor).

Specials of this workshop were the eclipse on Wednesday morning as well as the official dinner on top of the hotel under a fine African moon.

At the excursion to a prominent pineapple plant production company nearby the laboratory in vitro plant production, the nursery and the distribution of the you pineapple plants have been presented. The already highly impressed participants confirmed the high quality of the produced plants by tasting some fresh pineapple fruits of different varieties.

Thanks worthy to the special support of the FAO all the lectures and instructions for the practicals as well as the handouts have been translated in French language to optimize the understanding for the French speaking participants. Printed versions as well as CD containing all information on presentations and lecture notes were made available to the participants. Copies of ISTA Rules for Seed Testing, ISTA Handbook on Seedling Evaluation and ISTA Handbook on Seed Sampling were also distributed.

After an evaluation at the end of the course, the results proved positive as the participants indicated that they have benefited a lot from presentations, lectures and practicals and would make use of their training when they return to their various countries. Results from informal discussions of workshop participants also gave a positive feedback. In terms of the venue participants were unanimous in accepting that it was conducive and met their aspirations in the provision of logistics. However, majority of the participants also complained that the duration of the course was too short and would need further extension in future at least for two weeks.

We would wish to thank the following team of operators and mentors, Dr. Michael Larinde of Plant Genetics Resources Services, FAO headquarters, Rome, Mr. Josiah Wobil, FAO headquarters, Rome and ISTA Secretariat, in particular Ms. Branislava Opra for their hard work in making it possible for this course to take place in Ghana. This obviously is an honour done to the nation as well as to the West African sub region.

Thanks are also due to Professor Dr. Norbert Leist and Dr. Andrea Jonitz, the ISTA resource persons, for taking the effort to come to Ghana for leading this training course and also to plan and arrange the practicals perfectly.
The 2nd ISTA Vigour Workshop was held from 10 – 12 May 2006 in Beaucouzé, France at the kind invitation of GEVES-SNES. The 21 participants came from 15 different countries from as far as Japan, Chile, South Africa and India, as well as a good representation from within Europe. Approximately half (10 people) of the participants came from seed testing laboratories, while seven were from seed companies and four from research institutes or universities. For this workshop Dennis TeKrony, Stan Matthews and Alison Powell were joined in the presentation of the vigour tests by Sylvie Ducournau and Marie-Helene Wagner from SNES. We soon realised we had a lively and interesting group of participants, when, after the introductory video and lecture, the questions started and they kept coming throughout the workshop.

Our programme was a typical ISTA workshop mix of lectures and practical work. Our main aim was to describe the physiological background to vigour and its implications and to describe the validated vigour tests, Accelerated Ageing and Conductivity. Other ISTA recommended vigour tests presented were the Cold and Controlled Deterioration tests. The participants were able to complete the Conductivity test and to carry out aspects of all other tests. In most cases, the vigour test results are only obtained after a germination test. However, staff at SNES had worked hard to provide us with completed tests for a number of seed lots that illustrated the concept of vigour differences very well.

A new aspect to this workshop reflected a growing interest in the rate of germination as an assessment of vigour. Sylvie Ducournau described the methods and conditions for assessing germination rate, including an image analysis method under development at SNES. She illustrated how germination rate reveals physiological changes due to ageing, priming and phytochemical treatment, while Marie-Helene Wagner described collaborative work done on germination rate and other vigour tests in maize by SNES, Vilmorin and Aberdeen University.

All the domestic arrangements of travel, visual aids, coffees and French lunches were admirably arranged and executed and particular thanks were due to Veronique Binoit for all she did on our behalf. Our hosts also ensured that all our time was not completely full of vigour testing. All participants toured the facilities available in SNES in the germination, pathology and physical analysis laboratories and in the sampling department. One evening we had an interesting tour of the Cointreau Museum in Angers in which we learnt of the history of the company, viewed the large, shapely copper stills for distillation and finally were instructed in how to make several cocktails. The sampling of Cointreau cocktails, for which we needed no instruction, was most enjoyable!

The next evening our workshop dinner was preceded by a visit to the Chateau d’Angers. Here we were the testing ground for our guide as we were the first tour that he had conducted in English. He did not have to worry, we were a sympathetic and appreciative audience. First, our guide gave us a detailed description of the Apocalypse Tapestry, the oldest and largest tapestry in the world, and then, after the educational part, came the exercise when we climbed one of the towers. This was the vigour test for the participants; the photo was taken after we had got our breath back! After a walk though the peaceful gardens we finished in another tower where English prisoners had been held - two of us kept quiet! On leaving the chateau our dinner was held in a local restaurant, where our meal was all that we might expect from a delicious meal in France, well supported by good wine and conversation with friends.
The 11th Tetrazolium Workshop 2006 held from 24 – 28 July at NAK in Emmeloord, The Netherlands was attended by 14 seed analysts from nine different countries namely: Australia, Switzerland, Germany, Spain, Latvia, Denmark, Belgium, Sweden and the Netherlands.

At the opening session the Director of the General Dutch Inspection Service (NAK) and ISTA President Pieter Oosterveld was present and in his speech he emphasized the importance of organizing seed testing workshops on different topics in different countries.

Resource persons involved were Prof. Dr. Norbert Leist, former ISTA President and lecturer and Mrs. Stefanie Krämer, Chairperson of the Tetrazolium Committee and lecturer who also prepared the lectures.

NAK instructors supporting the practical work were Hillie Post, Sigrid Heyman and Gerarda de Boer. The organisation was in the hands of myself with the help of Ingrid Kuijer. There were various assistants from NAK involved providing the needs for the participants and to ensure that the workshop was a success.

The program of the workshop was compiled as a result of previous Tetrazolium workshop experiences. Prior to the event, the application form had the suggested species stated beforehand. Analysts could indicate further other additional species that might be of interest for the workshop. All additional species were handled during the workshop.

The participants were staff of commercial seed companies and state seed testing laboratories. The Workshop program was a mix of lectures and practical classes. When testing a species, Mrs. Stefanie Krämer held an introduction and a final discussion was held by Prof. Dr. Norbert Leist and Mrs. Stefanie Krämer.

Species covered during the workshop were: *Agrostis*, *Lycopersicon esculentum*, *Zea mays*, *Lactuca sativa*, *Beta vulgaris*, *Allium cepa*, *Brassica*, *Chicorium*, *Linum*, *Lolium*, *Helianthus*, *Phaseolus*, *Daucus* and *Trifolium*.

During the workshop time was also reserved for a guided tour of the premises of the NAK seed- and potato laboratory. For the practical work the excursion room of NAK was transformed into a laboratory, complete with a binocular for each participant and video demonstration facilities.

The workshop focussed on using the TEZ handbook for judging the viability according to the ISTA Handbook for Tetrazolium Testing and to improve further the participants’ knowledge in relation to Tetrazolium Testing.

Mrs. Stefanie Krämer presented the work of the ISTA Tetrazolium Committee, the ISTA TEZ Handbook and Working Sheets by drawing up a list of future activities.

For the scientific background to TEZ testing Prof. Dr. Norbert Leist held lectures on:
- History of development of biochemical viability determination in seeds
- Theory of biochemical viability determination in seeds by the Topographical Tetrazolium test
- Seed Vigour determination by means of the Topographical Tetrazolium test
- Tolerances and Tetrazolium Test
- Quality Assurance and Tetrazolium
- Use of Tetrazolium for evaluation of fresh, ungerminated seeds

A Group Picture
continued from page 54

The participants lodging in the hotel ‘t Voorhuys were transported to and from the workshop and transport was also provided for cultural and social events. The Workshop dinner on Tuesday was held on Urk. Prior to the start of the Workshop dinner a walking tour on Urk was organized. Urk is a small fishing village near Emmeloord and used to be an island in the past but is now connected to the main land. The dinner was an opportunity to meet in a more informal way and supported discussion and friendship between participants. Pieter Oosterveld and Ineke Mastenbroek (Technical Director of NAK) were also present at the Workshop dinner. On Wednesday the participants were invited for a drink, with cakes prepared by Hillie Post, in the beautiful garden of Gerarda de Boer. On Thursday afternoon we set out for a trip to Schokland. Thereafter the bus went on to Blokzijl, where a walking tour was held. On the route back Giethoorn was visited, here we were treated to a boat tour through the canals of what we call the “Venice” of Holland. After a refreshments in an outdoor café in Giethoorn we travelled back to Emmeloord.

After five days of hard work and discussion on Tetrazolium Testing every participant received an ISTA Certificate of Participation. A CD with the lectures and pictures, was also presented to the participants. Besides that, each participant received a map with a hard copy of the presentations and a NAK CD on which the work of the NAK is explained. Hard work for the participants and the lecturers. It was indeed not only due to the workshop program but also the weather. During the workshop, Holland was experiencing an unusual heatwave, with temperatures soaring over 35°C on some days.

Special thanks were expressed to the lecturers by presenting them Orchids from Holland. The lecturers also brought presents for the organisers of the NAK, special wines from Germany.

Finally I like to thank Pieter Oosterveld, director of NAK and president of ISTA for the support before and during the workshop which made it possible to host this ISTA Tetrazolium Workshop.

Report of ISTA Purity Workshops 2006
Zurich (Switzerland, June 22–23) and Nakuru (Kenya, July 13–14)

By Maria Rosaria Mannino1, ISTA Purity Committee Chairperson, GEVES-SNES, Angers, France
Joseph Ahenda2, KEPHIS, Nakuru, Kenya
Silvia Zanetti3, ART, Zurich, Switzerland

One of the main objectives of the Purity Committee work for the period 2004-2007 is the development of workshops. With this aim, two purity workshops were held earlier this year (2006): the first one at Agroscope in Zurich, Switzerland in June, some days before the ISTA Annual Open Meeting, and the second one at KEPHIS in Nakuru, Kenya in July.

The working group on Workshop Development and all the Purity Committee Members have actively participated to the workshop organisation. The different technical subjects handled during the two meetings have been chosen on the basis of the suggestions given by both the members of the Purity Committee and the applicants. By this means, we achieved two main objectives of the workshops: to make progress on the committee working programme and to improve methods and method standardisation, responding to the needs coming from the seed analysis laboratories.

Purity Workshop at Agroscope, Zurich (Switzerland), June 22-23, 2006

This workshop was planned with the objectives of presenting the Purity Committee activity and progressing in its work, improving technical skills and method standardisation for purity analysis, with particular attention to grass species.

The meetings attained the maximum number of participants in relation with the local facilities: 26 participants from 15 different countries of Europe, Africa and North America (Belgium, Canada, Denmark, Estonia, Germany, Great Britain, Italy, Latvia, Lithuania, Norway, Spain, South Africa, Sweden, Switzerland and the Netherlands).

Six presenters animated the technical sessions and the discussions: Maria Rosaria Mannino, ISTA Purity Committee Chair, and Corinne Sahuguède from GEVES (France), Silvia Zanetti and Sandra Reinhard from ART (Switzerland), Tove Due from Danish Plant Direktorate (Denmark) and John Wiersema, ISTA Nomenclature Committee Chair from the United States Department of Agriculture (USA). The team of the hosting

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3 ART Agroscope Reckenholz-Tänikon, Reckenholzstrasse 191, 8046 Zürich (Switzerland)
Purity Workshop at Kephis, Nakuru (Kenya), July 13–14, 2006

At the last ISTA Congress in Budapest 2004, the need for more workshops on species of tropical and sub-tropical countries had been pointed out by ISTA and ISTA Members from these regions to enhance seed testing knowledge. An important number of tropical crops could be introduced in ISTA Rules. Also methods and methods standardisation could be adapted to respond to economically important crops in these regions.

The workshop in Nakuru was planned to give to participants the opportunity to develop and exchange technical skills in seed science and technology and also to plan the future work to do together for ISTA methods improvement with regard to tropical and sub-tropical species.

The meeting was well-attended by seed analysts: 36 participants came from 10 different African countries: Angola, Congo, Ethiopia, Kenya, Rwanda, South Africa, South Sudan Tanzania, Uganda and Zambia.

Six presenters animated technical sessions and discussion: Maria Rosaria Mannino, ISTA Purity Committee Chair from GEVES (France), Tove Due from Danish Plant Direktorate (Denmark), David Okebiro from the National Museum of Nairobi (Kenya), Reuben M. Muasya from the Moi University of Eldoret (Kenya), Joseph Ahenda and Gladys Maina from KEPHIS (Kenya). The staff of KEPHIS also contributed to the organisation and actively participated in the technical workshop programme.

Some sessions presented the ISTA Purity Committee and its work: committee composition and organisation, technical programme, proposal of ISTA rules changes concerning purity analysis, the Universal List of Weeds...
and Crops. Other sessions covered different items related to the purity analysis: presentation of the ISTA Rules Chapter 3, 4 and Annex, purity analysis of mixtures, blower calibration, and seed identification. A part of the programme was reserved for items related to species of tropical and sub-tropical countries: use of Pure Seed Definitions relevant for species of these regions (PSD 36 for Setaria sphacelata, 42 for Chloris gayana and 43 for Pennisetum glaucum), purity analysis of Tanacetum cinerariifolium, purity analysis of Brassica rapa, results of the ISTA questionnaire on Tropical and Sub-Tropical species, introduction of new species in ISTA Rules, needs for ring tests and proficiency tests.

During the meeting, some other different issues were discussed:

• The optimal number of analysts for a seed-testing laboratory to be eligible for accreditation;
• The link between seed research sections and seed testing laboratories;
• The interest of the collaboration between universities and seed testing laboratories in research and development of seed technology;
• The budget difficulty for laboratories to support analysts to attend seed courses and workshops and the possibilities of sponsoring.

The participants appreciated the ISTA literature on seed testing and were advised on how to purchase publications from the secretariat.

As the workshop concluded, the participants agreed on the following actions:

• Work on tropical crops whose seed testing methods are yet to be developed or validated for inclusion into ISTA Rules;
• Communicate with each other continuously on various seed testing issues;
• Disseminate the knowledge they acquired from the workshop to other seed analysts in their respective countries;
• Work together to improve participation and contribution towards ISTA at technical and policy level;
• African seed testing laboratories which are already established to help the young upcoming laboratories to develop technical competence, to join ISTA and to achieve accreditation;
• African seed testing laboratories to develop a ring test program amongst themselves using the available facilities and compare their performance;
• More training courses and workshops to be organized by established African seed testing laboratories in areas such as seed sampling, germination, documentation and quality management.

The meeting has been the occasion for analysts of African countries to come together, identify the challenges and devise an approach to addressing them. It has also allowed participants to know better the technical work of the Purity Committee and foresee their possible contributions with the aim of improving analysis methods and methods standardisation for tropical and sub-tropical species.

Conclusions
Participants of the workshops thanked ISTA Secretariat, Purity Committee and management of hosting organisations for the support in making the workshops possible. They also expressed gratitude for the Handbook on Seed Identification by NIAB with pictures of seeds and to the sponsors who contributed to the workshops.

The issues of these two workshops show clearly the interest of ISTA workshop organisation for ISTA and seed laboratories: these technical meetings are indispensable to make progress on seed testing and method standardisation.

The Purity Committee will work in the later part of the year 2006 towards organising two purity workshops in 2007. Preliminary contacts have been made during the Annual Meeting in Zurich and places and dates will be soon announced.
Introduction

Seed is the basic propagation material for the agriculture and plays an important role in agricultural production. With globalization, increase of international trade and introduction of biotechnology into seed industry, seed testing became more and more important to agriculture. Thus uniform testing procedures are needed to ensure a certain quality level in the worldwide seed trade. Variety verification and GM seed detection in seed lots are the most important seed quality parameters. A number of advanced techniques are available for seed testing. However, their applications require appropriate technical competence and laboratory infrastructures. The use of reliable and economical methodology for variety verification and detection of GM seeds are becoming more important for all countries especially for those exposed to international seed trade.

The International Seed Testing Association (ISTA) plays an important role by assisting seed quality control agencies throughout the world with rules and procedures for seed testing. Additionally, ISTA is conducting training courses at regional and sub-regional level to improve and upgrade seed testing skills and level of testing technicians in collaboration with Food and Agriculture Organization of United Nations (FAO).

Some notes from the Workshop in Turkey

Between 20th and 24th of February, 2006, Ege University in Izmir, Turkey had the pleasure of hosting the 8th training course on “Electrophoretic Methods and PCR-Techniques for Variety Verification and GMO Detection” organized by FAO and ISTA. As the local organizers, the Horticulture Department of Agricultural Faculty and Seed Technology Center, arranged the workshop by providing their laboratory facilities and lecture room, transportation and accommodation. Besides FAO, The Turkish Research and Scientific Council (TUBITAK) also supported the workshop by contributing to the accommodations for lecturers and participants.

The aim of the Workshop was to improve and exchange the knowledge on methods for variety identification, hybrid purity determination as well as quantitative and qualitative GM seed detection. These factors are important for food safety besides increasing the seed quality and develop links between seed analysts and researchers in Balkan, Caucasian and Mid-East Countries where the seed industry has been rapidly growing.

The topics were so attractive that 30 nominations were received from different countries apart from the 25 nominations from Turkey alone. It was very difficult to select participants amongst them because all were related directly to both subjects. Participants were selected based on the consideration of at least one participant from each country in order to strengthen cooperation and information exchange in the target region. Including Turkish participants, there was a total of 20 participants from 12 different countries as Slovenia, Bulgaria, Macedonia, Greece, Romania, Bosnia and Herzegovina, Albania, Azerbaijan, Iran, Syria, Egypt and India attended the workshop in Izmir. Participants were researchers with different experiences on seed testing, molecular biology and biotechnology. Also present were directors and seed technicians from research organizations, universities and seed companies.

The workshop started with the opening ceremony, and the Vice Rector and Dean welcomed the participants and lecturers to Ege University Campus. On behalf of ISTA, Prof. Dr. Norbert Leist emphasized the role of ISTA by assisting seed quality control agencies throughout the world with rules and procedures for seed testing as well organizing training workshops in order to improve and upgrade seed testing skills and level of testing technicians. Dr. Thomas Osborne and Melek Cakmak as representatives of FAO, pointed out the importance of variety verification and GM seed determination in word seed trade, and the regulations and bio-safety protocols related to GMO. The head of the Horticulture Department, Prof. Dr. Uygun Aksoy and director of Seed Technology Center, Prof. Dr. Benian Eser introduced briefly the department and seed technology center and emphasized the importance of such training workshops in order to enhance knowledge exchange and to start networking on seed quality testing.

The workshop gathered different cultures as well as providing knowledge exchange consisted of two parts: In the first part, practical and theoretical lectures on electrophoretic methods for variety verification took place. After Prof. Dr. Norbert Leist (Staatl. Landw. Untersuchungs und Forschungsanstalt Augustenber, Karlsruhe; Germany and ISTA GMO Task Force Chairman) gave an overview on objectives and programs of the workshop, he introduced the methods for
culivar identification by emphasizing the advantages/disadvantages of these methods and why electrophoretic methods based on seed proteins became more important in variety verification and testing hybrid seed purity. The principle and applications of electrophoretic methods such as PAGE, SDS-PAGE, isoenzyme and IEF were presented. In this segment, the practical work was led by Mr. Rainer Knoblauch. He led participants by showing whole procedures and giving “provided tricks of the trade” on the IEF of seed storage proteins. In his part Here, the participants enjoyed making gels, extraction of seed proteins from Cucumis sativa, Oryza sativa, Zea mays and Triticum aestivum, loading the samples into gels, staining by using chemicals of Sinus Company, looking forward to the results outcome. The staining results of Comassie blue (Sinus Company) and Rothy Blue (Carl Roth Company) were evaluated. They gained experience on how the bands on the gel are evaluated for variety verification or hybrid purity and how they decide on proper extraction buffer. During the workshop, the IEF method was tested for Cucumis melon to differentiate some varieties.

In the second part of the workshop, PCR based methods for GMO detection, led by Dr. Christoph Haldemann (Swiss Federal Research Station for Animal Production and Dairy Products, ALP and ISTA GMO Task Force Member), included a theoretical and a practical part. The requirements among the participants clearly showed that there was a strong demand on the practical aspects on GMO detection. He outlined what transgenic plants are and how they are developed, which methods can be used for detection, identification and quantification for GMO as well giving general information about GM crop species and their production areas and quantities. The PCR based methods for GMO detection were introduced following the general information on DNA properties. Dr. Haldemann stressed the quality control in GMO analysis by using validated methods and assay control. The quantitative PCR approaches, competitive- and real-time(rt)-PCR techniques were introduced and the factors affecting rt-PCR quantification were pointed out. He presented a very demonstrative movie on how lateral flow strips for GMO detection are working. In the practical part, participants isolated DNA from seeds of soybean and maize samples and from CRM materials and quantified DNA of samples by spectrophotometer. The PCR of 35S promoter as screening method, nested-PCR for RR-Soybean, Bt 11 and Bt 176 as gene specific methods were practiced to determine/identify GMO’s. Quantification based on determination of 35S promoter was performed on Real-time PCR machine which was provided from the Gastroenterology Department of the Medical School at Ege University. The participants were highly interested in the immunostrip technology too, which were provided from the two US companies Agdia Inc. and EnviroLogix Inc. Dr. Haldemann also pointed out sub sampling strategies for GMO quantification. Finally, the objective and progress of ISTA GMO Task Force was presented by Prof. Dr. Norbert Leist.

The lecturers were not the only presenters at this workshop. The participants also showcased their country reports focusing on seed production and certification system and national strategies for transgenic crops and introduced their research institutes and labs. At the end of the first day of workshop, a welcome dinner was organized in Cumba Restaurant which was located on a hill and had a nice view of Izmir City and delicious Turkish cuisine. The participants and lecturers had some time to visit Izmir and walked along the seaside, enjoying the sunny weather.

At the closing session all participants received ISTA/FAO certificates of attendance and a CD with lectures notes, laboratory protocols, publications etc. An opportunity to evaluate the workshop was offered too. The participants were all satisfied with the workshop in terms of knowledge given and initiation of a collaboration which would be helpful in their future work.

Acknowledgements and thanks
Prof. Dr. Norbert Leist, Dr. Christoph Haldemann and Mr. Rainer Knoblauch for participation to this workshop and sharing their knowledge and experiences with us.

Dr. Kakoli Ghosh for her guidance us in getting the opportunity to host the workshop.

Ms. Branislava Oprà from ISTA, Mr. Larinde, Ms. Lazerini and Mr. Osborne from FAO for their assistance in the organisation of this event.

SINUS company from Germany that supplied all chemicals required for protein electrophoresis and CARL ROTH company from Germany for gel staining solution, the US companies, Agdia Incorporated and EnviroLogix Inc., providing the workshop with ImmunoStrips / lateral flow strips:

FAO and TUBITAK for their financial support

Prof. Dr. Ulus Akarca and Ms. Hulya Yilmaz from the Gastroenterology Department of the Medical Faculty for their technical assistance and supplying rt-PCR machine.

Finally I would like to thank all participants for their full attention given to the lectures and practical works as well as for the team spirit and the friendship built during time that we spent together.
The ISTA Quality Management Training Course was kindly hosted by Dr. Silvia Zanetti, head of the Swiss ISTA member laboratory at Agroscope Reckenholz-Tänikon Research Station ART. There were a total of 24 participants from 17 different countries.

The aim of this workshop was to increase comprehension of ISTA Accreditation Standard requirements and how they may be implemented in a laboratory’s quality management system. The workshop content focused on quality system aspects. The workshop was organised in such a way that the theory was conveyed through presentations and lectures which were then put in practice by the participants during group work.

Topics that were presented:
• Introduction QM principles and definitions
  Presentation and moderation techniques
• Interpretation of the ISTA Accreditation Standard
• Q-documentation: general principles of documenting a QM system and keeping of records, flow charts
• Introduction to audits: audit types, audit principles
• Audit preparation: planning, preparation of checklists
• Conducting of an audit: course of an audit, communication techniques, asking questions, acceptance and motivation, psychology
• Audit follow-up: Audit report, follow-up corrective action

The tasks the different groups were given were to sketch a procedure of one of the following topics:
• Purchasing services and supplies
• Contract review
• Handling of non-conformities and corrective action

The groups presented their results which were then discussed among the participants.

A large portion of workshop time was dedicated to internal audit, starting with some theory on audits, how they are to be prepared, conducted and followed-up. An extensive part was spent on practical work and Silvia Zanetti and her team volunteered to serve as guinea pigs for the internal auditors in training.

ISTA Workshops are not only work but also a good opportunity to meet people and hence a tour on Sunday was organised also to give the participants the chance to see a bit of Switzerland. We visited Lucerne, climbed up the mountain Pilatus by cogwheel railway and had some breathtaking views on famous peaks such as Eiger or Jungfrau and the Lake Lucerne. The weather was awesome and everybody enjoyed this day very much before the workshop went on with a mock internal audit on Monday.

A big Thank You goes to Silvia Zanetti and her team who provided excellent facilities and made everybody feel at ease and which made this workshop a great success and a very pleasant experience.
January
14–21 5th International Workshop on Desiccation Sensitivity and Tolerance in Seeds and Vegetative Plant Tissues (Drakensberg, South Africa)

February
19–23 ISTA/APSA/FAO Training Course on Seed Health Testing (Manila, Philippines)

March
6–9 AFSTA Congress (Livingstone, Zambia)
27–30 ISTA GMO Workshop: Methods for detection and Statistical Aspects (Izmir, Turkey)

April
25–27 ISTA Variety Workshop (Pelotas, Brazil)
30 April–3 May ISTA Tetrazolium and Germination Workshop on Tropical and Subtropical Seed (Curitiba, Brazil)

May
1–3 ISTA Workshop on Statistical Aspects of GMO Detection (Iguassu, Brazil)
1–3 ISTA Seed Health Workshop and Method Validation Aspect (Iguassu, Brazil)
1–3 ISTA Vigour Testing Workshop (Cascauel, Brazil)
5–11 28th ISTA Congress (Iguassu Falls, Brazil)
21–23 ISF Congress (Christchurch, New Zealand)

September
9–13 Seed Ecology II Conference (Perth, Australia)

2008
May
26–28 ISF Congress (Prague, Czech Republic)

June
16–19 ISTA Annual Meeting (Bologna, Italy)