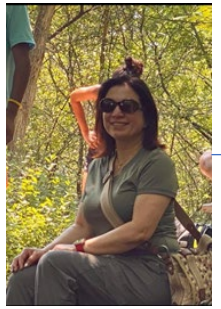
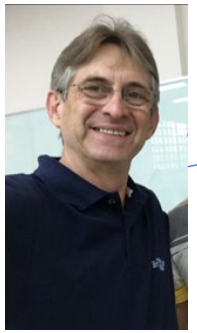
Tetrazolium Committee Report

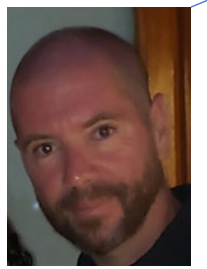
PhD Sergio Pasquini, TEZ Chair

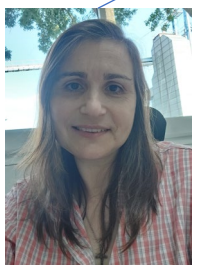
Disclaimer: I have no actual or potential conflict of interest in relation to this presentation.

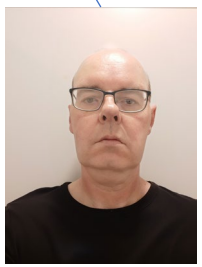
	Committee composition		
1	Chair: Sergio Pasquini		Italy
2	Vice-Chair: Steffi Krämer		Germany
3	Ignacio Aranciaga		Argentina
4	Maria Belen Aranguren		Argentina
5	Alessandra Arioli		Italy
6	Jose de Barros Franca-Neto		Brazil
7	Sunita BH		India
8	Valerie Blouin		France
9	Edith Daboue		Burkina Faso
10	Veena Gouda		India
11	Augusto Martinelli		Argentina
12	Shaminder Miranpuri		USA
13	Eoin O'Connor		Ireland
14	Tauhid Parvez		Canada
15	Asia Roberts-Yalland		United Kingdom

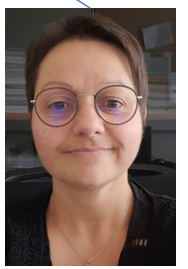


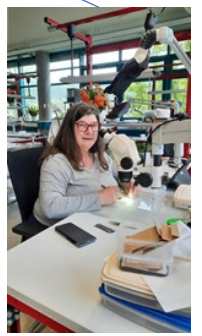


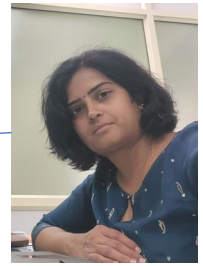


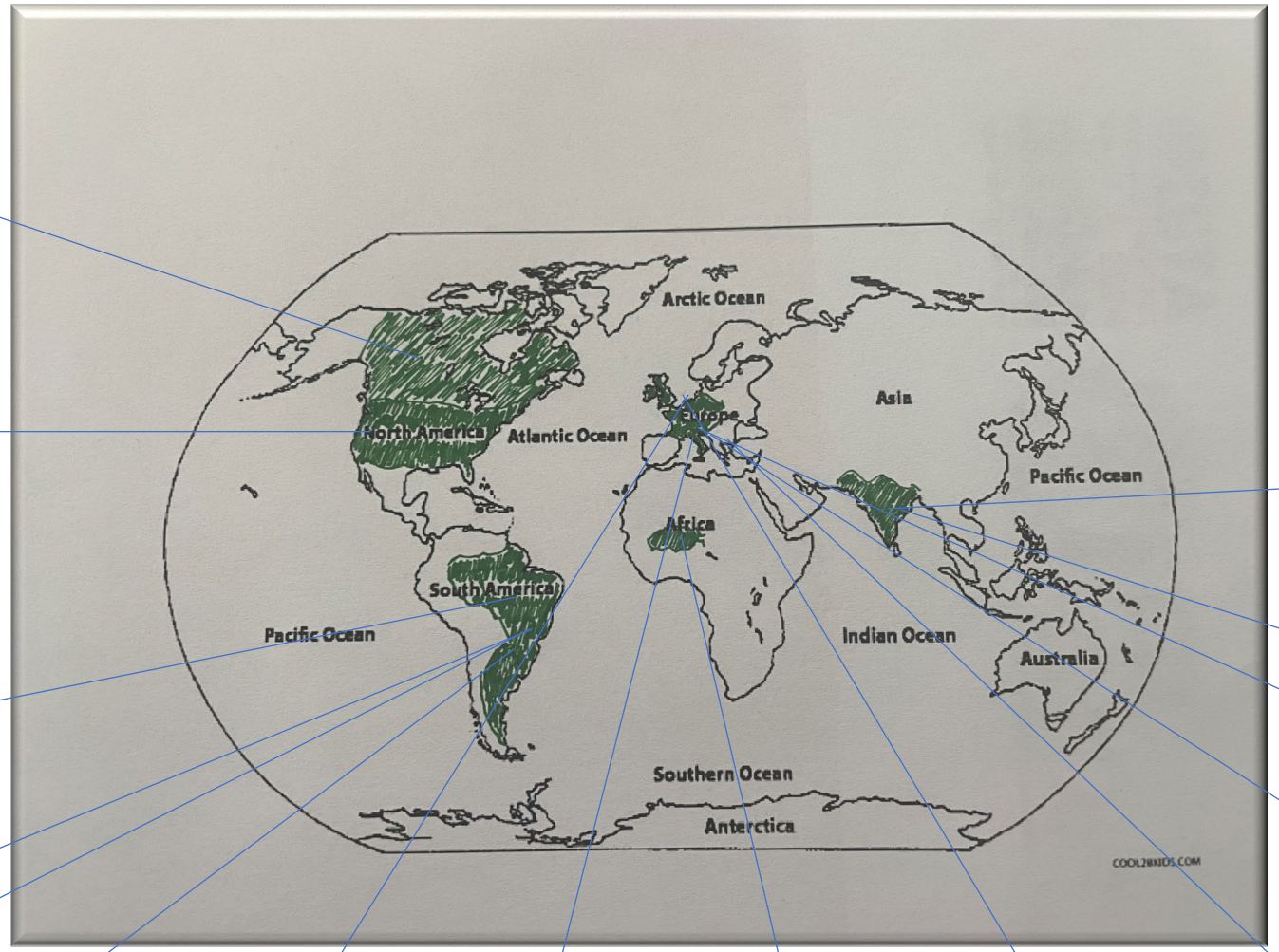












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Aims of Tetrazolium Committee:

Developing new tetrazolium tests to determine the viability of seeds for species not yet included in the ISTA Rules and re-evaluate existing methods

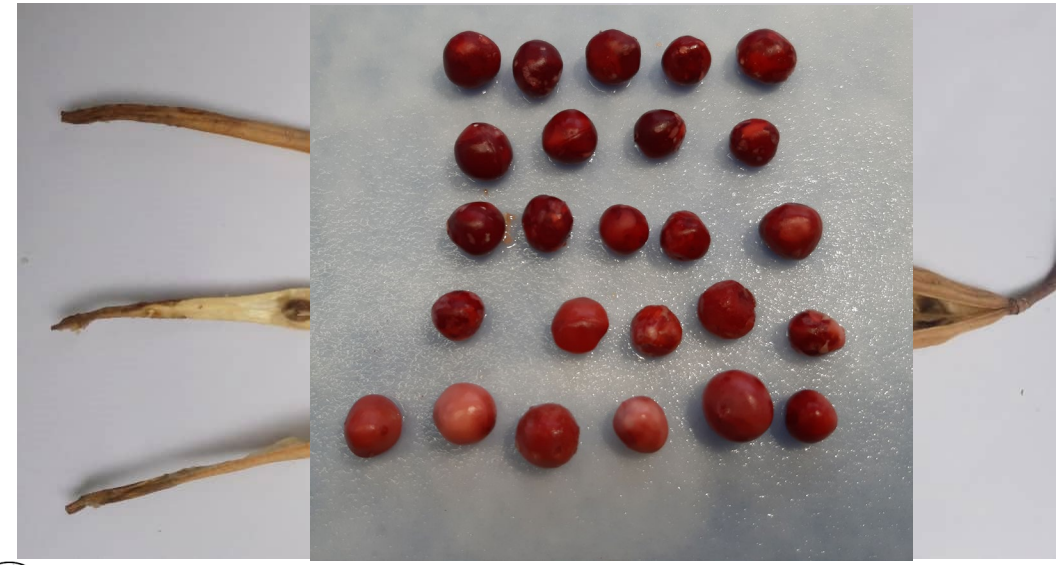
Providing theoretical and hands-on training on tetrazolium testing during workshops

Creating tetrazolium working sheets for many species with detailed and standardized descriptions to conduct and evaluate tetrazolium tests

Answering to the questions received by others about the TEZ test

Next steps…(from Activity Report 2023 – 2024)

	A2. Introduction of New Species	
	Species and test	proposed finalisation
	Glycine max L. Merr.	Jun 2025
	Moringa oleifera Lam.	Jun 2025



1. Objectives

The objectives of this comparative tetrazolium test are:

1. To validate *Moringa oleifera* Lam. for introduction into Chapter 6.
2. To confirm that the protocol used will meet objective 1, and identify any amendments necessary.

New species to test

Prepare a method to test the seeds

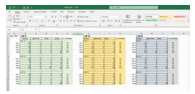
Do you want to add this species to ISTA Rules?

NO → Job is done!

YES

Keep the seeds possibly from different seedlots and find at least two friends!

Prepare the Method Validation Test Plan and do it!



Do the tests and KEEP THE RECORDS!

Prepare a Rule Proposal to be voted at AM

Validation test on *Glycine max L.*			
Test Leader	Ignacio Aranciaga		
Participants			
ISTA accredited labs	Argentina	Ignacio Aranciaga	ARDL01
	Canada	Tauhid Parvez	CAML12
	France	Valerie Blouin	FRDL02
	Germany	Stefanie Kraemer	DEDL04
	Ireland	Eoin O'Connor	IEDL01
	Italy	Alessandra Arioli	ITDL03
Not accredited labs	Argentina	Maria Belen Aranguren	Corteva
	Brazil	Jose B. Franca-Neto	Embrapa

Method Validation Test Plan on *Moringa oleifera* Lam.			
Test Leader	Steffi Kraemer		
Participants			
ISTA accredited labs	Canada	Tauhid Parvez	CAML12
	France	Valerie Blouin	FRDL02
	Germany	Stefanie Kraemer	DEDL04
	India	Veena M Gouda	INML07
	Italy	Sergio Pasquini	ITML06
Not accredited labs	Burkina Faso	Edith Daboue	National Forestry Seed Testing Centre
	India	Sunitha BH	Sakata Seeds
	USA	Shaminder Miranpuri	University of Wisconsin

A3. Introduction of Rules Changes

1 Consequential changes if C.5.1 is accepted of paragraphs: 6.5.2.1.2, 6.5.4 and 6.7

Consequential change if C.5.1 is accepted:

CURRENT VERSION	PROPOSED VERSION
6.5.2.1.2 Soaking in water … If the percentage of hard seeds of ~~the Fabaceae~~ is to be determined for the purpose of issuing an ISTA Certificate, the seed should be soaked in water at 20 °C for 22 h. Other procedures may lead to excessive variability in results.	**6.5.2.1.2 Soaking in water** … If the percentage of hard seeds, of those species listed in 5.2.10.1, is to be determined for the purpose of issuing an ISTA Certificate, the seed should be soaked in water at 20 °C for 22 h. Other procedures may lead to excessive variability in results.
6.5.4 Evaluation … Hard seeds are seeds with water-	**6.5.4 Evaluation** … Hard seeds are seeds with water-

OGM Document
ECOM Approved?

6/25/2024
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Rules Proposals for the International Rules for Seed Testing 2024 Edition

impermeable seed coats (~~e.g. Fabaceae~~) and remain hard even after premoistening	impermeable seed coats (e.g. see 5.2.10.1) and remain hard even after premoistening
6.7 Reporting results … In addition, in the case of species ~~of Fabaceae~~, one of the following, and only one, must be reported:	**6.7 Reporting results** … In addition, in the case of species with hard seeds, one of the following, and only one, must be reported:

	B. PUBLICATIONS		
	B1. Publications to accompany the Rules	Proposed finalisation	In collaboration with
1	Tetrazolium Handbook – General part – Second Edition	Dec 2024	ISTA Secretariat

3.4 PREPARATION

In addition to moistening, most kinds of ning.

3.4.2 Preparation met

Size and sha e embryo and other seed cl d for seed of a given spe- ble, desired accuracy of results ng factors. Different methods can of- paration methods include:

paration (e.g. small-seeded legumes with water perme-

tening without additional preparation (e.g. large-seeded legumes with eable seed coats);

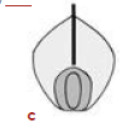
ongitudinal bisection through embryo and approximately ¾ of the endosperm of cereals and grass seeds (figure a).


a

d Transverse cut of Avena spp. and grass seeds (figure b);


b

e Longitudinal cut through distal part of the endosperm of grass seeds (figure c);


c

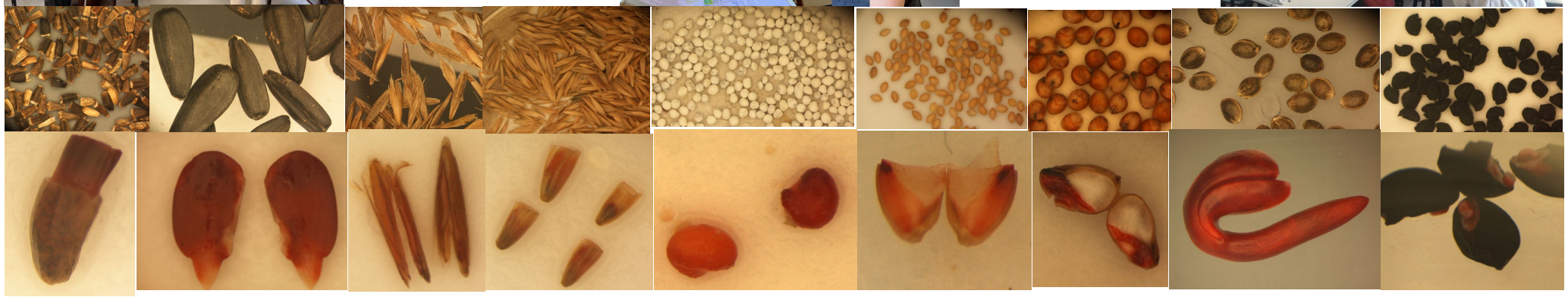
A

	Training publications on specific seed testing topics		
	Publication title	proposed	collaboration
		finalisation	
1	Short videos on TEZ methods	2024	
2	TEZ working sheets Vol. 1 electronic version	On going	
3	TEZ Working sheets new edition and revision	On going	

	C. Workshops and Seminars		
			collaboration
1	ISTA Tetrazolium test in different crop groups: A hands-on workshop (held in Spanish)	Montevideo (Uruguay) July/August 2024	INASE
2	ISTA Workshop on Germination and Tetrazolium Seed Testing	Lincoln (New Zealand) November 2024	GER TCOM Kimihia Research Centre Laboratory

	C. Workshops and Seminars		
			collaboration
1	ISTA Tetrazolium test in different crop groups: A hands-on workshop (held in Spanish)	Montevideo **31th July – 2nd August 2024**	INASE URUGUAY



The workshop, aims to improve the knowledge and technical skills of seed analysts performing tetrazolium determination as part of their daily work

	C. Workshops and Seminars		
			collaboration
2	ISTA Workshop on Germination and Viability Tests	Lincoln **4th - 8th November 2024**	GER TCOM PGG Wrightson Seeds Kimihia Research Centre Laboratory

From Germination Test to Tetrazolium Test through different Crops, tested according to the current ISTA Rules

	D. PROFICIENCY TESTS		
	Proficiency test subject	Scheduled in	In collaboration with
1	*Triticum aestivum L.* subsp. *aestivum* (PT 24-1)	2024	PTC
2	*Festuca rubra* L. (PT 25-1)	2025	PTC
3	*Oryza sativa* L. (PT 25-2)	2025	PTC
4	*Pinus sylvestris* L. (PT25-3)	2025	PTC




As Member of an Accredited ISTA Laboratory, within the **TEZ PT Programme** you have the possibility to check your performance in the TEZ Test. Furthermore, you could have the possibility to collaborate in a **Working Group** participating in validation programmes regarding tetrazolium tests, which will allow you to acquire experience in this matter, demonstrate your competence on new species and get in contact with other colleagues who are also interested in sharing their knowledge.

This would allow a joint improvement with open possibilities of:

- proposing and working on a **new species** (still not included in Table 6A of the ISTA Rules) in order to study it;
- proposing the **method** so that a new species (still not included in Table 6A of the ISTA Rules) could be studied;
- proposing that a new species already included in Table 6A of the ISTA Rules could be studied again in case you consider the **current methodology** proposed is not the most adequate.

Anyway everybody can contact TEZ TCOM for questions, clarifications, proposals,…

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Thank you