Annual Meeting 2023

Variety Committee report 2022-2023

Presenter: Marie-Claude Gagnon

Location: Verona, Italy

Date: May, 2023

Agenda

Members

Update on the development on new markers for detection of annual types in perennial rye grass varieties
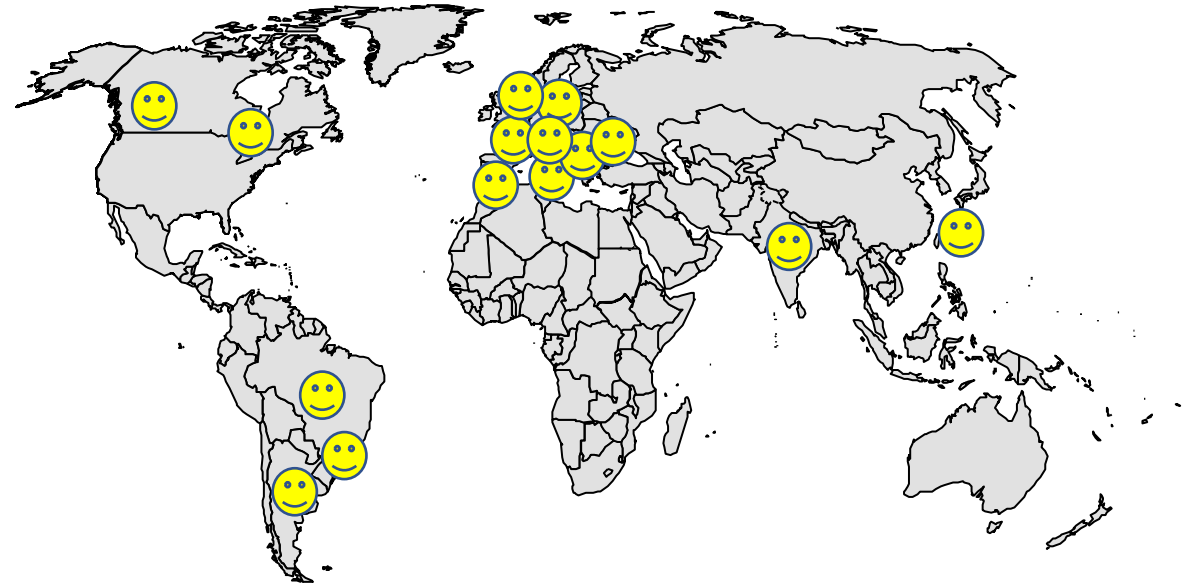
Proficiency Test (wheat) PT "1" – Results

Artificial Intelligence based markers

Other topics

ISTA VARCOM members

		Member Since
1	Chair: Ana Laura Vicario AR	2007
2	Vice-Chair: Marie Claude Gagnon CA	2020
3	Anne Bernole FR	2016
4	Berta Killermann GER	2004
5	Chiara Delogu IT	2007
6	Kae-Kang Hwu TW	2007
7	Ksenija Markovic RS	2013
8	KsenijaTaski-Ajdukovic RS	2010
9	Keshavulu Kunusoth IN	2010
10	Mariana Menoni UY	2021
11	Ana Patricia Fernández Getino SP	2021
12	Sean Walkowiak CA	2022
13	.	
14	.	
15	.	

Project leader: Giovanny López
Collaboration with Shaun Bushman from USDA who is developing the markers and Daniel Curry from Oregon State University who is providing seeds samples and technical support.

- Next-Generation sequencing of the genomes.

- Selection of a set of 8 to 10 SNP markers.

- Development of the primers and probes. Preliminary test for selecting a subset of primers.

- Oregon Seed Lab, Utah Lab and CFIA lab tested 3 KASP markers with 6 variety combinations (PRG varieties spiked with AGR varieties) on seed samples to define the sensitivity of the method (1/50, 2/50, 3/50).

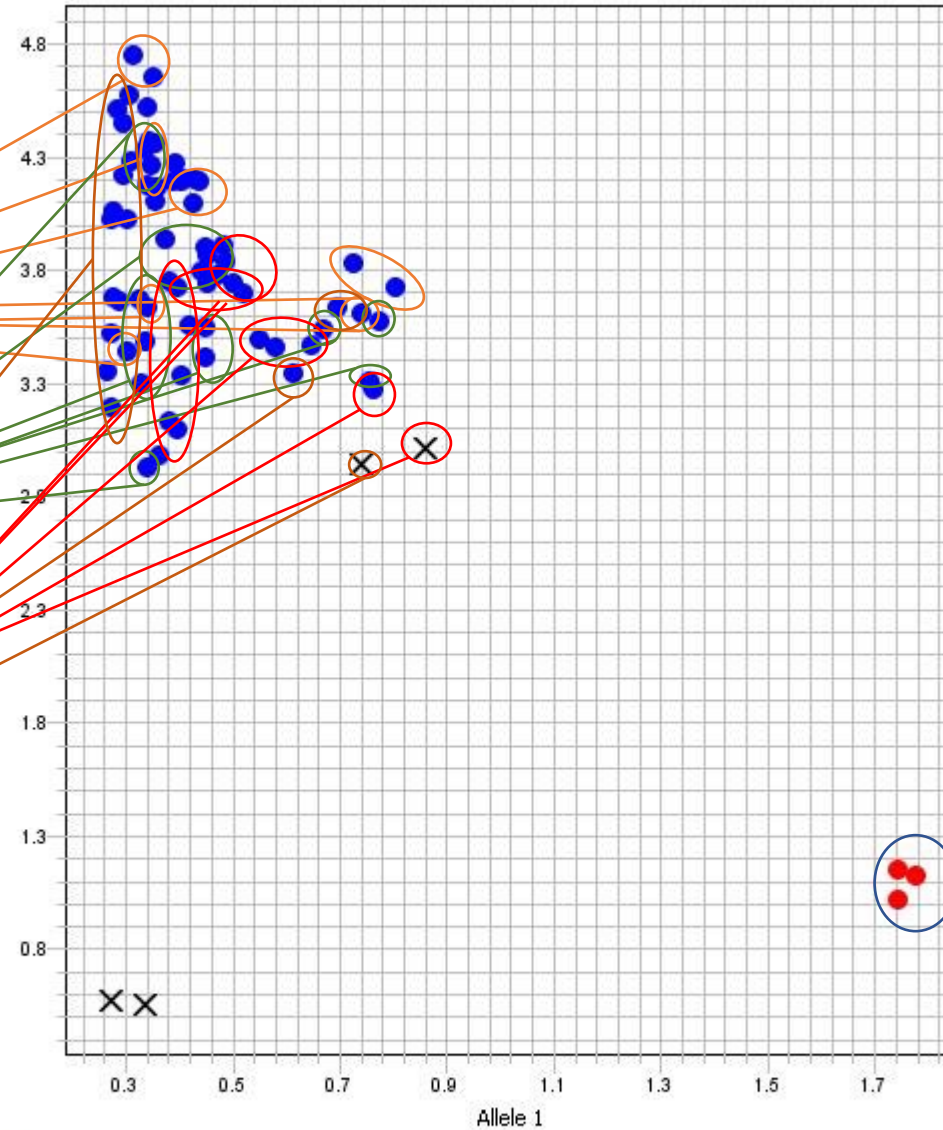Example of the mixtures and spiking levels:

Panterra V ARG

CO1 primers

Allelic Discrimination Plot

1 Panterra, 49 perennial

2 Panterra, 48 perennial

3 Panterra, 47 perennial

Perennial

Panterra V

Legend
- Homozygous Allele 1/Allele 1
- Homozygous Allele 2/Allele 2
- Heterozygous Allele 1/Allele 2
- X Undetermined

CO1 primers

- When examining the whole group with endpoint analysis, CO1 provided better separation between pure PRGs and PRGs spiked with contaminated ARG seed.

- When removing some samples to make specific comparisons, separation was consistent enough for separation at the 6% contamination (3/47 spike) level.

Proposal: examining the CO1 gene with new quantitative PCR markers.

PT "1" already finalized and evaluated

5 participating laboratories: 3 accredited and 2 willing to evaluate their performance.
Mandatory for accredited laboratories.
Set of 6 varieties, 8 SSR wheat markers, 2 varieties sent as control and also their SSR profiles.

Only 1 lab could be rated properly. For the others, some problems were found:

Some labs reported allele sizes without calibrating the results with the standard.
Some labs reported results for other markers.
Some labs reported results for less makers than those stated in the Rules.
Some labs reported on a different format than the excel sheet for reporting results.

A meeting will be organized with the participating labs to reorganize the data and make it fit the Rating Matrix.

Samples were prepared and shipped by the CGC (Canadian Grain Commission).

VARCOM is preparing a special project together with ATC (Advanced Technology Committee).

The software is able to discriminate varieties within a "universe" of varieties already learned by the software.

The equipment consist of a scanner (for taking the seed image) and a PC (for image analysis and results output).

The software learns, through convolutional neural networks, the morphological profile of thousands of seeds from the same variety that gather most variability as possible (i.a. harvesting years and climate and soil conditions).

The outcome of the software in an excel file indicating the % of identity of the sample with a variety or the whole set of varieties learned by the software.

STACOM is analyzing data obtained for barley and wheat to design a test for ISTA validation.
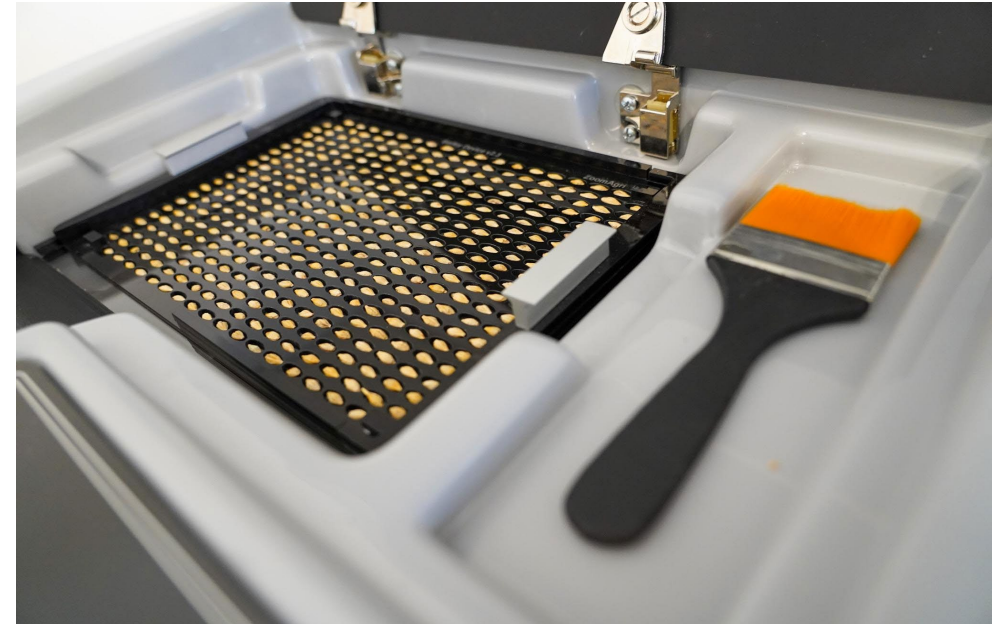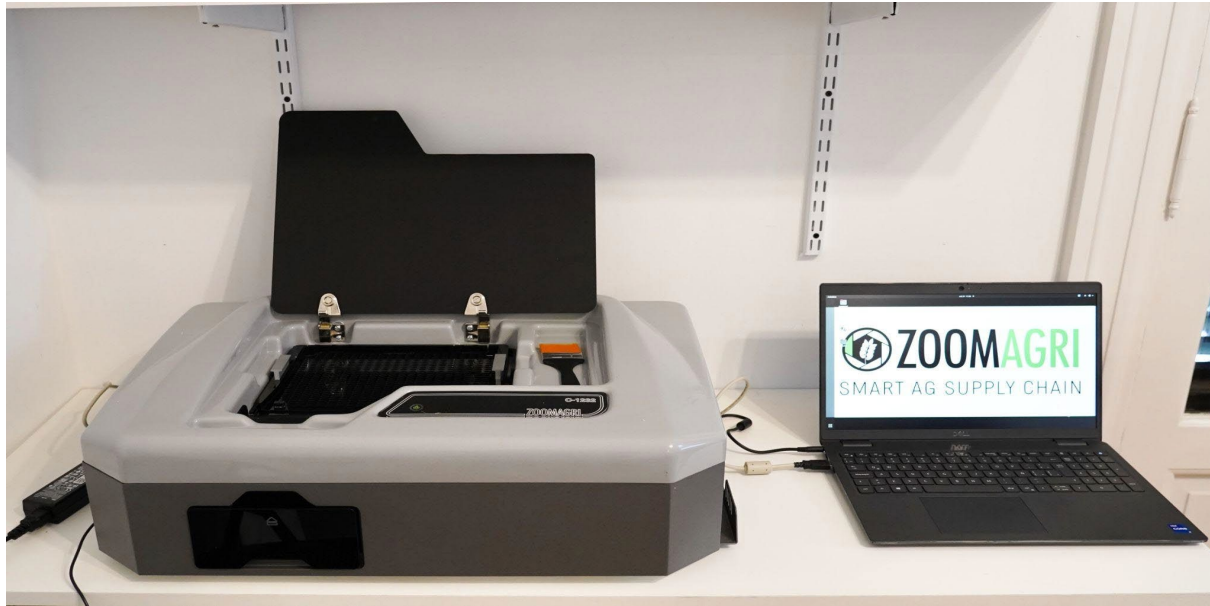
Inclusion of statistical considerations in chapter 8: include criteria for estimating sample size for purity, adventitious presence and sample identification.

We are open to new crops and markers.

Thanks to members, *Lolium* Project leader, PT Leader and chair, for the great work done so far to achieve VARCOM and ISTA goals.

To STACOM chair and vice-chair for data analysis and help in thinking.

Thanks to Hedwich Teunissen from the Netherlands who left the Committee last April. We wish her good luck in her new position.

Thank you!

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