

“Further strengthening of capacities of phytosanitary sector in the fields of plant protection products, plant health and seeds and seedlings, including phytosanitary laboratories and phytosanitary inspections”

(TWINNING BA/12/IB/AG 01)

Component 3: Seeds and propagation materials

ISTA RULES FOR PURITY ANALYSIS

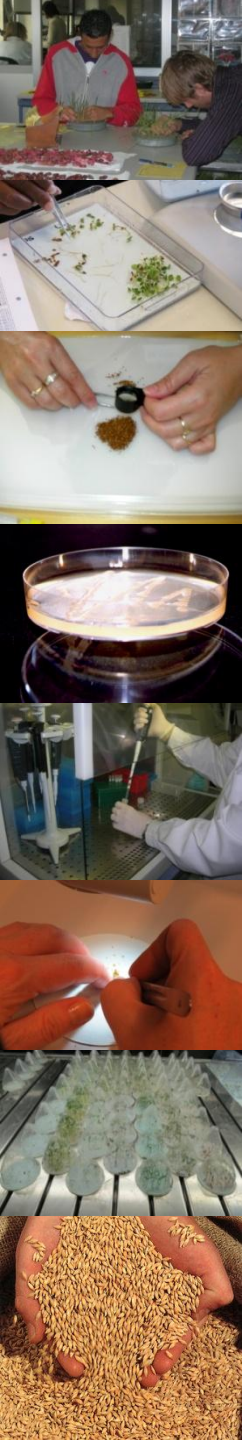
Rita Zecchinelli

CHAPTER 3: THE PURITY ANALYSIS

Object

The object is to

- determine the % composition by weight of the sample (→ of the seed lot)
- identify other species and inert matter retrieved



PURITY ANALYSIS

General Principles

- The working sample is separated in three components parts:
 - *pure seed*
 - *other seeds*
 - *inert matter*
- The percentage of each part is determined by weight.
- All species of seed and each kind of inert matter shall be identified.
- If required, the percentage by weight of specified species and/or each kind of inert matter shall be determined.

DEFINITION

Pure seed

- it refers to the species stated by the applicant (or found to predominate)
- it includes all varieties of that species
- it includes:
 - intact seed units (as defined in the relevant PSD)
 - pieces of seed units larger than one-half their original size

even if immature,
undersized, diseased (..)

... with some exceptions

e.g. Seed units of some families (*Fabaceae*, *Cupressaceae*, *Pinaceae*, *Taxaceae*, *Taxodiaceae*) with seed coats entirely removed shall be regarded as inert matter.

Poaceae (*Poa pratensis*, *Poa trivialis*, *Dactylis glomerata*): caryopsis of the stated species contained in the light fraction obtained by uniform blowing procedure must be regarded as inert matter



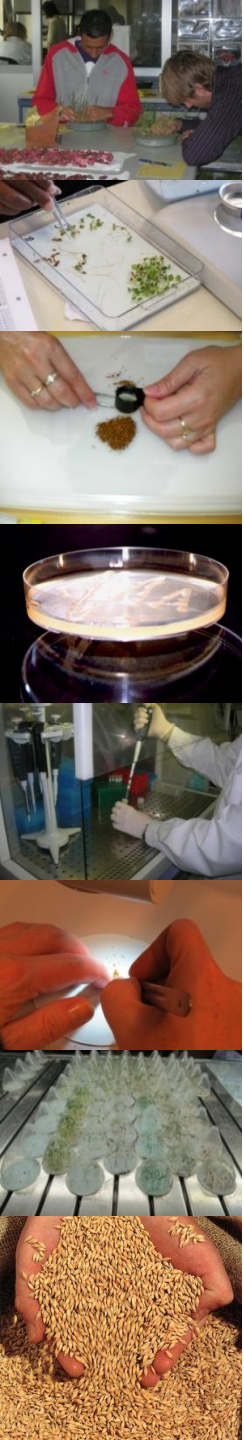
DEFINITION

Other seeds

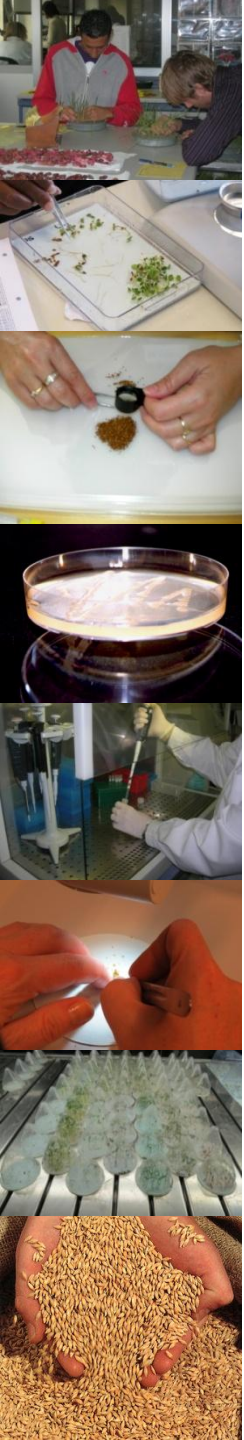
- shall include seed units of any plant species other than that of pure seed
- the classification as other seed or inert matter is based on the relevant PSD
- if a PSD is not available, the general definition of pure seed shall be applied
- except for certain species (see PSDs), capsules, pods (..) are opened and non seed material are placed in the inert matter

... with some specific provisions

e.g. *Cuscuta* spp. seed units which are fragile or ashen grey to creamy white in colour are classified as inert matter.



ISTA RULES FOR PURITY ANALYSIS



DEFINITION

Inert matter

- shall include seed units and all other matter not defined as pure seed or other seed

E.g.

- seed units in which no true seed is present
- pieces of broken seed units half or less than half the original size
- appendages not classed as being part of the pure seed in the relevant PSD
- seeds of *Fabaceae*, *Cupressaceae*, *Pinaceae*, *Taxaceae*, *Taxodiaceae* without seed coats
- non seed matter, as soil, stones, stems, leaves, chaff, bark, cone scales (...)

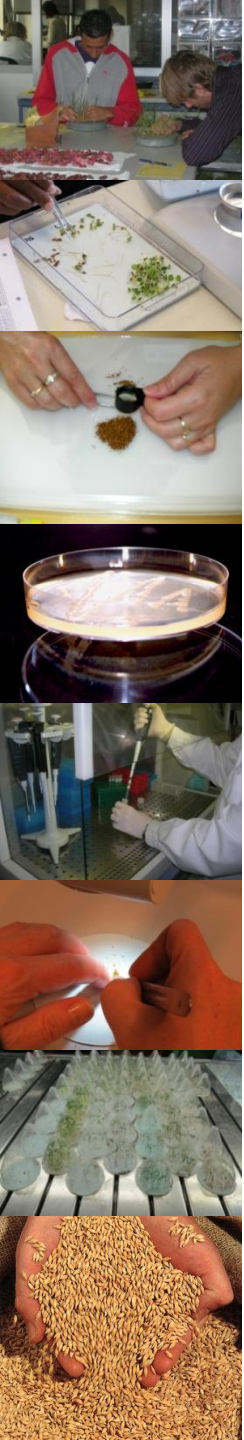
ISTA RULES FOR PURITY ANALYSIS

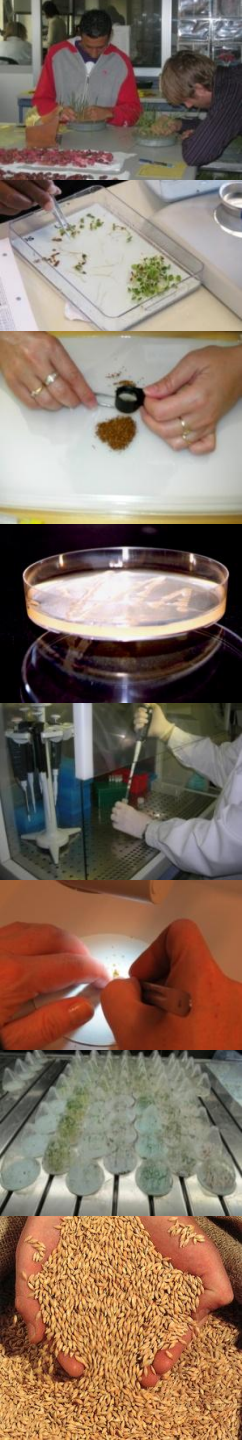
EXAMPLES

Inert matter in samples of cereals



Inert matter in samples of tree species





ISTA RULES - 3.2.1.A Pure Seed Definitions

≈ 1000 species in the ISTA Rules – 63 PSDs

Table 3B Part 1: PSD number, chaffiness (by genus)

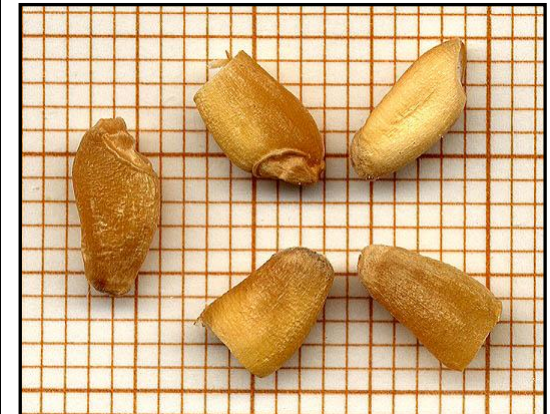
Table 3B Part 2: numbered PSD (description)

PSD 40

Poaceae (e.g. *Triticum* excluding *T. spelta*, *T. dicoccon*)

Caryopsis

Pieces of caryopsis larger than one-half the original size



Triticum durum

PSD 11

Fabaceae (e.g. *Medicago*, *Phaseolus*, *Vicia*)

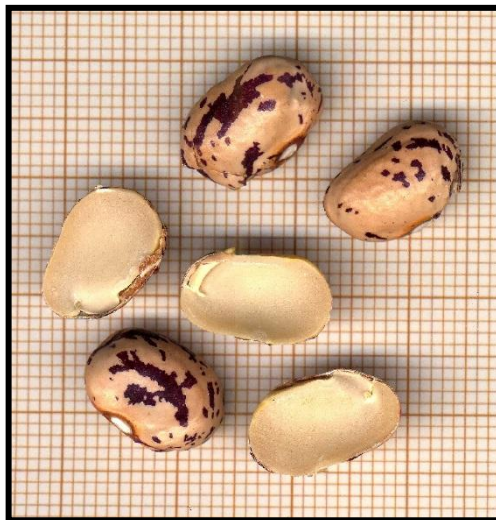
Seed, provided a portion of the testa is attached. Piece of seed larger than one half the original size, provided a portion of the testa is attached.

Fabaceae: cotyledons that are broken apart but held together within the testa.

Seeds and pieces of seeds entirely without testa are regarded as inert matter.

Fabaceae: separated cotyledons are regarded as inert matter, irrespective of whether the radicle-plumule axis and/or more than half of the testa is attached.

Separated cotyledons of *Phaseolus vulgaris*



Seeds of *Vicia sativa* with and without testa



ISTA RULES FOR PURITY ANALYSIS

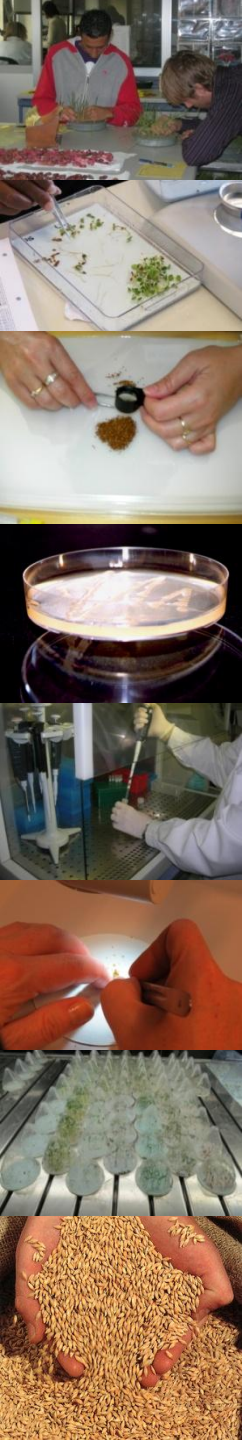
PSD 47

Pinaceae (Picea, Pinus II).

Seed, without wing or integument, provided a portion of the testa is attached.

Piece of seed larger than one half the original size, without wing or integument, provided a portion of the testa is attached.

Picea abies



ISTA RULES FOR PURITY ANALYSIS

PSD 51

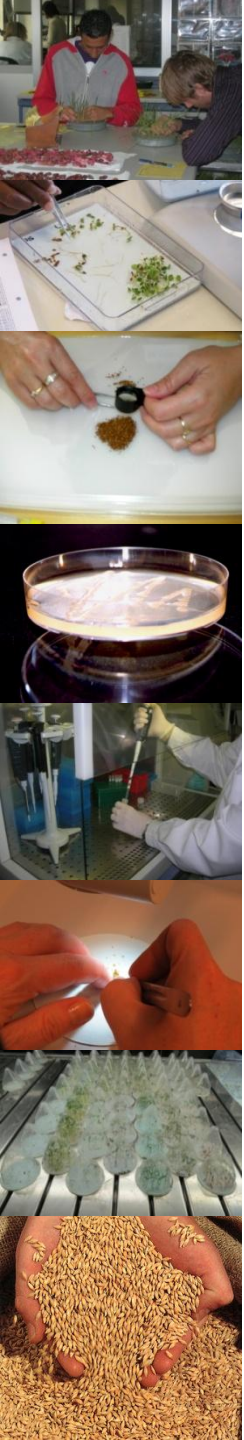
Pinaceae (Abies, Cedrus, Larix, Pinus I, Pseudotsuga, Tsuga).

Seed, without wing, with (but occasionally without) integument, provided a part of the testa is attached.

Piece of seed larger than one half the original size, without wing, with (but occasionally without) integument, provided a portion of the testa is attached.



Abies alba



ISTA RULES - TABLE 2A Part 1-2-3

- Maximum weight of lot (kg)
- Minimum sample weights (g)
 - Submitted sample
 - **Working sample for purity analysis**
 - Working sample for other seed determination (OSD)



From 0,5 g (e.g. *Nicotiana tabacum*, *Origanum vulgare*)

.... 5 g (e.g. *Trifolium pratense*) ... 120 g (e.g. *Triticum durum*) ...

to 1.000 g (e.g. *Phaseolus coccineus*, *Vicia faba*)



ISTA RULES FOR PURITY ANALYSIS

PROCEDURE

Working sample

- The purity analyses may be made on

{	one working sample (full weight)
	two independent sub-samples (half weight)
- The working sample (or each sub-sample) shall be weighed in grams to a minimum number of decimal places prescribed by the ISTA Rules.



Weight (g)
Less than 1.0000
1.000 to 9.999
10.00 to 99.99
100.0 to 999.9
1000 or more

Number of decimal places
4
3
2
1
0

PROCEDURE

Separation → pure seed - other seeds - inert matter

Principle: in general, separation is based on an examination of each particle; pure seed separated on the basis of visible seed characteristics, using pressure or mechanical aids, without impairing the capacity of germination.

Apparatus: aids may be useful for separating the sample into its component parts. *E.g. hand lenses, binocular microscopes, reflected light, sieves.*

Final test: the final weight (sum of weights of the components) is to be compared with the original weight.

Tolerances: when two half working sample are analyzed, the difference between their results shall not be in excess of the tolerance given by the ISTA Rules (Table 3.C)

Calculation, expression, reporting of results



ISTA RULES FOR PURITY ANALYSIS

Calculation and expression of results

ONE WHOLE WORKING SAMPLE

- Calculate % on the sum of the weights of the components, to one decimal place.
- Components of less than 0.05% shall be reported as TR (for 'Trace').
- If the sum of all components ('Trace' excluded) is $\neq 100.0$, add/subtract 0.1% from the largest value (rounding procedure).

CHECK: COMPARE WITH THE ORIGINAL WEIGHT
(TOLERANCE: MAX DIFFERENCE: 5%)

	Weight (g)	% by weight
Pure seeds	51,27	95,3
Other seeds	1,35	2,5
Inert matter	1,19	2,2
	-----	-----
	53,81	100,0

Calculation and expression of results

TWO HALF WORKING SAMPLES

Checking against tolerances:

- Calculate % on the sum of the weights of the components of each sub-sample (at least two decimal places); calculate the average %; check table 3.C.

	A	
	Weight (g)	%
Pure seeds	26,26	91,79
Other seeds	1,15	4,02
Inert matter	1,20	4,19
	<hr/>	
	28,61	100,00
	B	
	Weight (g)	%
Pure seeds	25,98	91,61
Other seeds	1,13	3,98
Inert matter	1,25	4,41
	<hr/>	
	28,36	100,00

CHECK: COMPARE WITH THE ORIGINAL WEIGHT
(TOLERANCE: MAX DIFFERENCE: 5%)

Calculation and expression of the final results:

- Add the weights of the corresponding fractions; calculate % to one decimal place.
- Components of less than 0,05% shall be reported as TR (for 'Trace').
- If the sum of all components ('Trace' excluded) is $\neq 100,0$, add/subtract 0,1% from the largest value (rounding procedure).

	Weight (g)	% by weight
Pure seeds	52,24	91,7
Other seeds	2,28	4,0
Inert matter	2,45	4,3
	<hr/>	
	56,97	100,0

Pure seeds: AVERAGE = 91,70 ▶ TOLERANCE = 2,74
(difference: 91,79-91,61=0,18)

ISTA RULES FOR PURITY ANALYSIS

Reporting results on an ISTA certificate

- When the weight of the working sample tested deviates from that prescribed, the actual weight examined shall be reported on the certificate.
- The results shall be given to one decimal places.
- The percentage of all components must total 100.0.
- Components of less than 0.05% shall be reported as TR (for 'Trace').
- If the result for a component is nil, show it as '0.0'.
- The scientific name of the species of pure seed must be reported (if identification at species level is difficult: report the genus. E.g. *Lolium* sp.).
- The kind of inert matter must be reported.
- The scientific name of other seeds must be reported
(ISTA List of Stabilised Plant Name).
- For PSD 47 and 51, the percentage of winged seed must be reported.
- Upon request, other information can be reported



THANK YOU FOR YOUR ATTENTION!

