



The use of molecular techniques in Brazil

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Aim

• To identify the genetic profile of protected plant varieties by developing specific molecular markers.

✓ PVP in Brazil: Breeding Testing System



Mapa SNPC

Identification of Genetic Profiles

- SSR Markers
- DNA fingerprints: PBR and NLi varieties
- Support for Identification of varieties
- Enforcement of Seed Law
- Seed Certification
- Comparison of "new" and "old" varieties
- Post control



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Legal Basis

- Normative Instruction n° 58/2009 established:
 - ✓ Procedures for the delivery of genomic DNA samples of protected varieties;
 - ✓DNA extraction methodology, date and results of qualitative and quantitative analysis of the samples.





Accredited Laboratory

- The Lab analyzes according to an agreed procedure and monitored by a PVP Office
- Control samples
- SNPC provided coded samples
- Doubled samples from SNPC
- Negative Control / Positive Control
- 2 DNA extractions (bulks of 50 young leaves)
- Genetic Analysis by 2 different staff in different days – minimize human error



Molecular Markers

- SSR Markers
- Highly informative
- Extensively validated in scientific literature
- Minimal or no environmental influence
- High discrimination power
- Database for commercial protection



Species

- Soybean
- Rice
- Cotton
- Eucalyptus
- Pearl millet
- Sugarcane, Cowpea, Grape (under construction)

Species

Crops	Samples	SSR Loci
Soybean	881	15
Rice	95	16
Eucaliptus	66	25
Cotton	103	15
Sugarcane	135	20
Cowpea	10	20
Sugarcane	8	15
Grape	22	15

Practical Example

- Soybean (556 Samples x 15 SSR loci)
- Selected markers differentiate samples
- Genetic Similarity: based on the multiloci genetic profiles
 - \checkmark 525 samples \rightarrow Unique genetic profiles (94,77%)
 - \checkmark 21 samples \rightarrow Undistinguishable proflies (5,23%)
- Further side by side field comparison with similar varieties
 - ✓ All varieties were phenotypically distincted (hilum color, flower color, time of flowering, pubescence color).
 - ✓ On a case by case assessment the SNPC verified EDVs, trangenesys, siblings, etc.



Conclusions

- Molecular markers are useful to:
 - ✓ Investigate possible PBR infringements;
 - ✓ PVP enforcement and Seed Inspection;
 - ✓ Identify mixture in seed samples;
 - ✓ Check purity of DNA samples;
 - ✓ Check declared parental information;
 - ✓ Organize official trials;
 - ✓ Improve the effectiveness and efficiency of the DUS testing process;
- Nowadays in Brazil molecular markers are just used to identify varieties, not to analyze distinctness.





Thank you for your attention!



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