



BMT-TWA/Maize/1/1

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INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS
GENEVA

**AD HOC CROP SUBGROUP ON MOLECULAR TECHNIQUES
FOR MAIZE**

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STATE OF THE ART OF MOLECULAR TECHNIQUES IN MAIZE

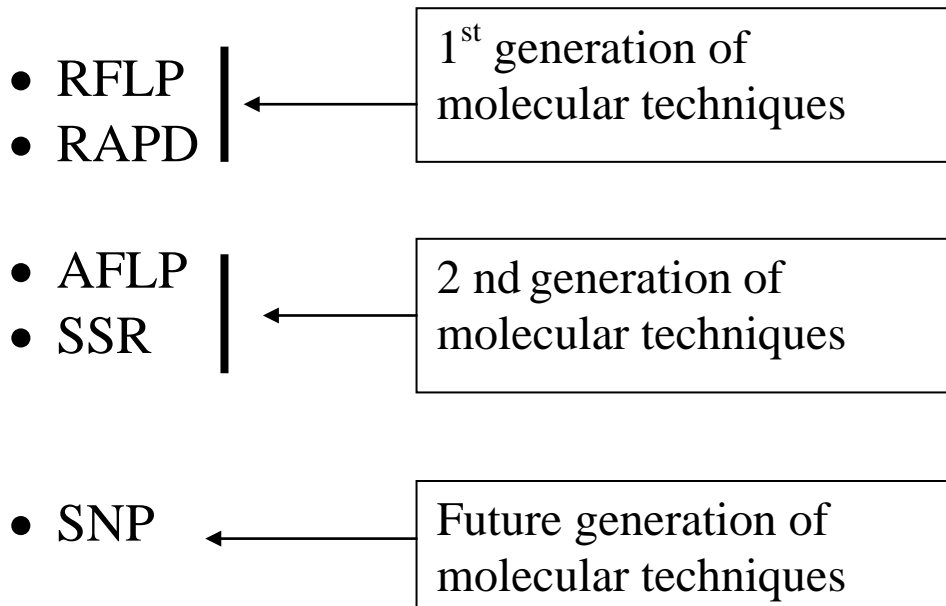
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STATE OF THE ART OF MOLECULAR TECHNIQUES IN MAIZE

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All work on maize !



Are all the techniques available and suitable for DUS ?

<u>Criteria of performance</u> Set by UPOV	Techniques			
	RAPD	RFLP	AFLP	SSR
a). Polymorphism	++	+++	+++	+++
b). Dev. cost	+	+++	+	+++
c). Cost & accessibility				
-Accessibility of technology	Yes	Yes	No	Yes
-Cost of analysis (per locus)	++	+++	+	++
-Cost of equipment	+	++	++	+ or ++
-Safety issues	No	Yes or No	Yes or No	No
d). Repeatability	+	+++	++	+++
e). Reliability & agreement between lab and with different equipment	+	+++	+	+++
f). Means of improving repeatability	No	Yes	Yes	Yes

Are all the techniques available and suitable for DUS ?

<u>Additional criteria</u> of performance	Techniques			
	RAPD	RFLP	AFLP	SSR
g). Dominance	D.	Cod.	D	Cod.
h). Possibility of establishing DB	+	++	+	+++
i). Easiness for protocol standardization	+	++	+	+++
j). Possibility of mapping	+	++	++	+++
k). Random distribution in the genome	++	+++	+	+++
Suitability for DUS	 	 	 	✓

About SSR in maize

- 1679 public SSR primer pairs in MaizeDB (update in Oct 2000) (<http://www.missouri.edu/ssr.html>)
- 1623 SSR loci mapped
- Polymorphism level:
 - 6.8 alleles/locus (Pejic et al., TAG (1998) 97: 1298-1255)
 - PIC = 0.62 (Smith J.S.C., BMT/4/2)

About SNP (Single Nucleotide Polymorphism) in maize

- New generation of molecular markers
- We are in a stage of development for their identification & detection
- The most abundant molecular markers
- High frequency of SNP in maize ([Rafalski et al. W149](#); [Useche et al., P333](#); 2001, [PAG9](#), San Diego):
 - 1 SNP / 49 bp in non-coding regions
 - 1 SNP / 138 bp in coding regions
 - 1 SNP / 70 bp on average in maize
 - ⇒ 35 000 000 SNPs in maize genome

For comparison:

- Human
 - 1 SNP / 1000 bp ⇒ 3 000 000 SNPs in human genome
- Soybean (Perry Cregan's team, [P337](#) & [P338](#), [PAG9](#), San Diego, 2001)
 - 1 SNP / 206 bp in non-coding regions
 - 1 SNP / 610 bp in coding regions

CONCLUSION

SSR are molecular markers of choice for DUS for:

- Polymorphism
- Availability
- Reliability
- Codominance (possibility of detection of heterozygote locus)
- Easiness for automatization of analysis and for standardization of protocol
- Low cost (by multiplex in PCR as well as for gel electrophoresis)
- Easiness for establishing a DB for interlab change

Additional references

Berenardo, R., et al., 1997. RFLP-based estimates of parental contribution to F2- and BC1 – derived maize inbreds. *Theor Appl Genet*, 94: 652-656.

Berenardo, R., et al., 2000. Parental contribution and coefficient of coancestry among maize inbreds: pedigree, RFLP, and SSR data. *Theor Appl Genet*, 100: 552-556.

Jones, C.J., et al., 1997. Reproducibility testing of RAPD, AFLP and SSR markers in plants by a network of European laboratories. *Molecular Breeding* 3: 381-390.

Pejic I., et al., 1998. Coparative analysis of genetic sililarity among maize inbred lines detected by RFLPs, RAPDs, SSRs, and AFLPs. *Theor Appl Genet*, 97: 1248-1255.

Pejic I., et al., 2000. Further characterization of AFLP data as a tool in genetic diversity assessments among maize (*Zea mays* L.) inbred lines. *Molecular breeding* 6: 265-276.

Rafalski, A., et al., 2001. SNP markers in maize: discovery and applications. *Plant & Animal Genome IX*, San Diego, Workshop abstract 149, page 44.

Taramino G. and Tingey S., 1996. Simple sequence repeats for germplam analysis and mapping in maize. *Genome*, 39: 277-287.

Useche, F., et al., 2001. Computer detection of Single Nucleotide Polymorphisms (SNPs) in maize ESTs. *Plant & Animal Genome IX*, San Diego, Poster abstract 333, page 144.

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