ASSESSMENT OF PCR-BASED SIMPLE SEQUENCE REPEATS TO FINGERPRINT NORTH AMERICAN POTATO CULTIVARS

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Abstract

Seven primer pairs flanking di- and tri-nucleotide repeat sequences, identified from previously sequenced regions of the potato genome, were examined for their potential use in DNA-fingerprinting of thirty-nine *Solanum tuberosum* subsp. *tuberosum* cultivars (released between 1861 and 1988) and one diploid *S. phureja* breeding line. Of the simple sequence repeats (SSRs), the primers for six SSRs amplified DNA sequences within the potato genome between cultivars for a total of 14 bands. The polymerase chain reaction (PCR)-based amplification products generated from each primer pair consisted of 1 to 2 bands per cultivar but band variation among cultivars demonstrated up to 4 bands per SSR. A similarity matrix generated from five SSRs was able to distinguish 24 of the 40 cultivars. However, when the potato cultivars were grouped by tuber type (round white-skinned, long white-skinned, russet-skinned, red-skinned, and yellow flesh) only five pairs of cultivars remained indistinguishable: Atlantic/Katahdin, Belchip/Wauseon, Red LaSoda/Bliss Triumph, Red Pontiac/Norland, and Burbank/Spunta. Although SSRs did not generate unique fingerprints for all of the North American genotypes examined, the potential to discriminate most cultivars should increase as additional SSRs are identified in potato.

Introduction

A method for cultivar identification that is rapid, reliable, and efficient has continually been sought to distinguish potato (*Solanum tuberosum* subsp. *tuberosum*) cultivars (3, 5, 7, 17, 22, 38, 4 8, 12, 20, 17, 32, 31). Traditionally, potato cultivars have been separated by morphological characteristics such as flower color, growth habit, leaf type, disease reaction, and sprout and tuber type. However, limitations to this type of identification exist since many of these characteristics are subjective and often influenced by the environment (4). Most importantly, these morphological traits cannot be scored simultaneously nor can cultivar identification be performed quickly and efficiently (31).

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Recently, simple sequence repeats (SSRs), a polymerase chain reaction (PCR)-based marker adopted from human genetics, has been employed to differentiate anther-derived potato lines (35). SSRs are DNA regions of short, tandem repeated units which demonstrate polymorphism due to differences in the number of repeated units within a length of DNA. The most frequently observed repeat in plant species is $(AT)_n$ which has been observed to be randomly distributed across the genome (19, 36). Tri- and tetra-nucleotide repeats are also abundant within plant species (36). Simple sequence repeats have demonstrated high levels of polymorphism in many plant species including soybean (19), barley (28), tomato (27, 24), rice (13), maize (29), brassica (25), pine (30), and wild yam (34).

Wang *et al.* (36) performed a series of database searches on known sequences within several plant species. With mono-nucleotide sequences excluded, they estimated one SSR every 27.8 kb. In potato, they found eight repeats (3 (AT), 1 (AG), 1 (AAG), 1 (AATT), and 2 (ACTC)), and estimated eight SSRs every centimorgan. Using the primers developed by Veilleux *et al.* (35), our objective was to determine: 1) if SSRs will generate polymorphism between North American potato cultivars and 2) whether SSRs will be an efficient, rapid, and reliable technique for DNA-fingerprinting potato cultivars.

Materials and Methods

DNA from 39 commercial potato varieties (Table 1) and one diploid *S. phureja* (Juz. et Buk.) species selection was extracted according to Doyle and Doyle (6). Young leaves from greenhouse-grown plants were the source material for DNA extraction. The DNA samples were quantified by fluorometer (Hoefer TKO100, Hoefer Scientific, San Francisco, CA) and a subsample was diluted to 50 ng ul^{-1} for PCR.

Seven sets of primer sequences were obtained from Veilleux *et al.* (35) by EMBL + GenBank searches of known potato DNA sequences. The source genes, number of alleles for each set, and SSR repeat are shown in Table 2. The 20 to 24 bp SSR primers were synthesized by Genosys Biotechnologies, Inc. (Woodland, Texas) and diluted to 10 ng ul^{-1}. Using 30 ng of each flanking primer, DNA from each of the 40 potato samples was amplified in a 25 µl reaction consisting of 10mM tris-HCL (pH 8.3); 10 mM KCL; 200 uM each of dATP, dTTP, dCTP, and dGTP; 0.5 units of AmpliTaq DNA polymerase (GIBCO/BRL, Gaithersburg, MD); 100 ng of template DNA; and a single drop of mineral oil. The $MgCl_2$ concentration for this reaction was optimized for each primer set (Table 3). Amplification was performed using a Perkin Elmer Cetus (Norwalk, CT) 480 thermal cycler and the protocol was: 5 min at 95 C; 30 cycles of 45 sec at 95 C, 60 sec at "T" C, 60 sec at 72 C; and one cycle of 10

TABLE 1.—*Potato cultivars used in SSR analysis, year of introduction and tuber type.*

Cultivar	Year of Introduction	Type
Alaska Frostless	1967	RW
Atlantic	1976	RW
Beauty of Hebron	1878	RW
Belchip	1979	RW
Bintje	1910	Y
Bliss Triumph	1878	R
Burbank	1876	L
Butte	1978	RU
Chieftain	1966	R
Chipbelle	1981	RW
Desiree	1967	R
Early Rose	1861	L
Early Ohio	1871	RW
Frontier Russet	1990	RU
Garnet Chili	1853	R
Green Mtn.	1885	L
Irish Cobbler	1876	RW
Katahdin	1932	RW
Kennebec	1948	RW
Krantz	1985	RU
Lemhi Russet	1981	RU
Nooksack	1973	RU
Norchip	1968	RW
Norgold Russet	1964	RU
Norland	1957	R
Onaway	1956	RW
Ontario	1946	RW
Red Pontiac	1945	R
Red LaSoda	1953	R
Rosa	1981	RW
Rural New Yorker	1888	RW
Russet Burbank	1914	RU
Russet Norkotah	1987	RU
Saginaw Gold	1988	Y
Spunta	1968	L
Superior	1961	RW
Wauseon	1967	RW
White Rose	1893	L
Yukon Gold	1980	Y

[1]L=long white-skinned; R=red-skinned; RW=round white-skinned; Y=yellow flesh; and RU=russet-skinned.

TABLE 2.—*SSR name, predominant repeat, product size and number of alleles. Specific sequences primer sets can be found in Senior and Heun (1993).*

Name	Source Gene	Primer Sequence (5 to 3)	SSRs	Product Size (bp)	No. of Alleles
SSR1	*actin* gene	TTTCTATTGAAAACCTTGAGAGGG CATCTCTACATTCACGAGCATTG	(TA)13	228	0
SSR2	*proteinase inhibitor* gene	CTTGCAACTTGTTAGTACCCCC AAATCCTTTGTGACCTCCCC	$(TC)_{12}$ & $(TA)_{18}$	180	3
SSR3	*waxy* gene	TGATTCTCTTGCCTACTGTAATCG AGTCAGAGTATGGTTCCTGAGTCC	$(CTT)_4$	246	2
SSR4	*granule bound starch synthase* gene	TTCTGATTTCATGCATGTTTCC ATGTGTGGTCTACAAAAAGGGG	$(AAG)_8$	236	2
SSR5	mRNA for leaf type α-*starch phosphorylase*	TTCAGAGACATCATGGCAACTT ATCCTTCATCAGAGGAAGAATCC	$(AGA)_5$	220	1
SSR6	*waxy gene*	CCCATAATACTGTCGATGAGCA GAATGTAGGGAAACATGCATGA	$(ACTC)_5$	223	2
SSR7	*patatin* pseudogene	CAACCAACAAGGTAAATGGTACC TGGTCTGGTGCATTAGAAAAAA	$(AATT)_5$	386	4

min at 72 C. The "T" temperature for elongation was also optimized by primer set as shown in Table 3. The completed reaction products were held at 4 C until electrophoresis.

The PCR products were separated on a 3% Metaphor (FMC Bioproducts, Rockland, ME) agarose gel stained with ethidium bromide (0.15mg ml^{-1}) in 1X TAE (40mM tris-acetate and 1mM EDTA) or TBE (89mM tris-borate, 89mM boric acid and 2mM EDTA) buffer. The gels were run at 100 V for 4 hours at room temperature and the products were viewed by ultraviolet light (254 nm) and photographed for permanent record. At least two PCR amplifications with each primer set were performed on all cultivars to test the reliability of each banding pattern.

The SSRs were scored as 1 for presence and a 0 for absence for each band observed among the 40 cultivars with band number 1 being the largest fragment. The data was entered into NTSYS (26) to generate a similarity matrix (21) after which an unweighted group pair means analysis was performed to construct a dendrogram of the cultivars. The cluster analysis was repeated after tuber trait data was added to the SSR data matrix.

Results

Of the seven sets of primers, six amplified DNA fragments of sizes ranging from 180 to 386 bp. SSR1 did not amplify any fragments whereas SSR5 was monomorphic. The rest of the SSR primer sets amplified consistently with the number of bands ranging from 2 to 4 for a total of 14 bands (Table 2). The band polymorphism for each SSR is shown in Figure 1. Each cultivar had no more than one or two of the bands for each SSR, although bands per SSR ranged from one to four. Comparisons between two data sets generated from each PCR reaction revealed conflicting band patterns in only 15 of 520 data points. The major difference in patterns was due to the poorer resolution of the TAE buffer system in the initial electrophoretic runs. The TBE buffer proved to be a more consistent buffer system for separating and resolving bands.

With the five polymorphic SSRs, we were able to discriminate certain cultivars, however, some groupings of cultivars did occur. Cultivars with identical SSR banding patterns were as follows: Atlantic/Katahdin, Norland/Red Pontiac/Beauty of Hebron/Early Rose, Bliss Triumph/Kennebec/Norgold/Red LaSoda, Burbank/Russet Burbank/Spunta, and Saginaw Gold/Belchip/Wauseon. The dendrogram for the SSR cluster analysis is shown in Figure 2.

When tuber type data (round white-skinned, long white-skinned, russet-skinned, red-skinned, and yellow flesh) was combined with SSR data, the power of the SSRs for separating cultivars was improved (Figure 3). Five pairs of cul-

TABLE 3.—*Amplification optimization conditions for $MgCl_2$ concentration and elongation ("T") temperature for each primer set.*

SSR	[$MgCl_2$] (mM)	"T" (C)
SSR2†	2.0	55
SSR3	2.0	55
SSR4	1.5	58
SSR5	2.0	55
SSR6	2.5	55
SSR7	2.0	55

†SSR1 did not amplify and was not used for this study.

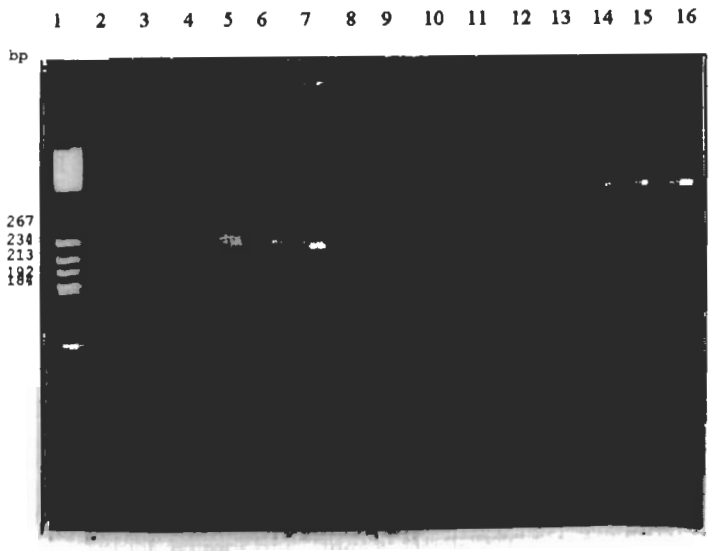

FIG. 1 Metaphore agarose gel depicting polymorphisms using SSR technology. Lanes 2, 5, 8, 11, and 14 are cultivar Red LaSoda; lanes 3, 6, 9, 12, and 15 are cultivar Superior; and lanes 4, 7, 10, 13, and 16 are cultivar Nooksack. Products in lanes 2, 3, and 4 were amplified using the SSR2 primer set; products in lanes 5, 6, and 7 were amplified using the SSR3 primer set; products in lanes 8, 9, and 10 were amplified using the SSR4 primer set; products in lanes 11, 12, and 13 were amplified using the SSR6 primer set; and products in lanes 14, 15, and 16 were amplified using the SSR7 primer set. Lane 1 is plasmid pBR322 cut with Hae III.

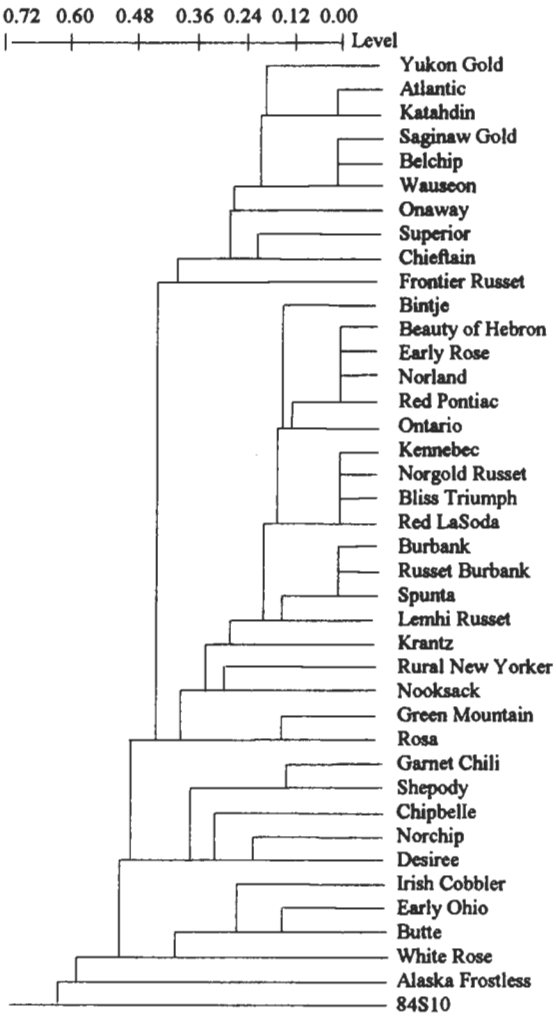

FIG. 2. Similarity matrix of cultivars based on fingerprinting using five SSRs (SSR2, SSR3, SSR4, SSR6, and SSR7). Cultivars at above level 0.00 can be distingushed by this method.

tivars remained indistinguishable from each other: Atlantic/Katahdin, Belchip/Wauseon, Red LaSoda/Bliss Triumph, Red Pontiac/Norland, and Burbank/Spunta.

Discussion

SSRs are tandemly-repeated short oligonucleotide sequences that are flanked by conserved DNA sequences. SSRs are also referred to as short tan-

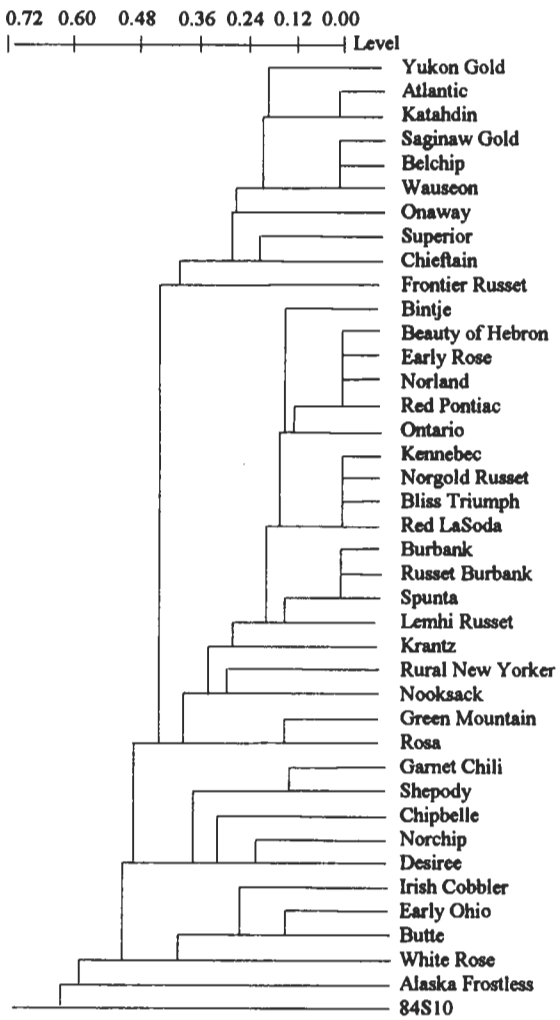

FIG. 3. Similarity matrix of cultivars based on fingerprinting using five SSRs (SSR2, SSR3, SSR4, SSR6, and SSR7) and five tuber type data (round white, red-skinned, long white-skinned, russet-skinned, and yellow flesh). Cultivars at above level 0.00 can be separated by this method.

dem repeats or microsatellites. This class of DNA markers is attractive because of their abundance and relatively simple technical requirements (28, 36). Complementary primers which flank the SSR can be used in combination with PCR to amplify the locus. Variation in the SSR region results in a variable number of core sequence repeats which are resolved on either a polyacrylamide or agarose gel. SSRs were first studied in humans and shown to be abundant and

dispersed throughout the genome (10, 11). More recently, SSRs have been studied in several monocot and dicot plant species (18, 36, 29, 16, 35). SSRs are the marker of choice for species with low levels of genetic variation as detected by RAPD and RFLP markers (16).

In this study, we demonstrated that variability exists for five of the seven SSRs tested within potato cultivars. On average, 2.3 band variants were observed per amplified SSR. A similar level of variation was observed by Veilleux, *et al.* (35) using DNA from anther-derived monoploids and diploids of potato. This level of polymorphism is substantially below that observed for other crop species. Maughan *et al.* (16) studied genetic variation of soybean SSRs. Among 94 accessions of wild and cultivated soybean, 79 variants were found using five SSRs. Alleles per locus ranged from 5 to 21. A total of 32 alleles were found in the cultivated soybean accessions. For barley, 17.7 alleles per locus were observed (28). Yang *et al.* (37) identified nine alleles per locus in rice. The disparity in the level of SSR variation found within heterozygous potato cultivars and highly inbred self-pollinated crops could be due to the limited sample of loci available in potato and the lower power of resolution using agarose versus polyacrylamide gels. If SSR variation is a result of mismatch and unequal crossing-over, the reduced number of meiotic events in the breeding and maintenance of a vegetatively propagated crop may explain the lower allelic diversity compared to highly inbred, seed propagated crops. Until more loci are examined in potato and SSR variation is studied in more crop species, we cannot fully explain the disparity in polymorphism levels.

Electrophoretic techniques to elucidate enzyme variation has been an effective, non-destructive and rapid technique to distinguish potato cultivars (32, 3, 22, 4). However, differences between sports and line selections were not observed using these biochemical markers. Although isozymes are currently being used commercially for potato fingerprinting, the number of scorable loci is limited. As the number of cultivars increases, the probability of having cultivars with matching isozyme patterns could also increase.

To circumvent the limitations of isozyme fingerprinting, DNA-based markers have been applied to fingerprinting. Restriction fragment length polymorphisms (RFLPs) are the most widely used DNA-based marker in plant species and have been used to effectively separate potato cultivars (7, 8; 5). Although RFLPs generate many more polymorphisms than isozymes, they are more expensive, and labor intensive (33, 20). Random Amplified Polymorphic DNA (RAPDs), a PCR-based technique using arbitrary primers, is more cost effective, technically simple, and rapid and requires minute amounts of DNA. Furthermore, RAPDs have demonstrated a greater intra-specific level of polymorphism than RFLPs in tomato and common bean (15, 9). Complete potato cultivar discrimination has been achieved with RAPDs (2, 20, 12, 31). Unfortunately, the repeatability of amplified bands by this method has been debated bringing into question the credibility of this technique (14).

For practical purposes, highly informative markers should allow cultivar discrimination in a single step (8). The limitation of other marker-based identification systems are described above. SSRs provide many positive attributes to DNA fingerprinting. They can be highly polymorphic demonstrating codominant banding patterns and are easily generated by PCR amplification. The primers are developed from gene sequence data which can be easily exchanged between laboratories. At this time, the limitation of SSRs for fingerprinting is the identification and availability of this marker in potato. The development of additional SSRs involves radioactive probing of genomic libraries and DNA sequencing. Panaud *et al.* (23) studied the relative frequencies of 13 microsatellite motifs in the rice genome. Their results suggest that SSRs would occur approximately every 80 kb. At this level of abundance, a map of highly-polymorphic PCR-based markers could be developed. Similar efforts should be made in potato to develop more SSRs. After the preliminary step, SSRs can be easily maintained and shared among laboratories through the sharing of published sequence data/primer pairs.

In this study, 24 of 40 potato lines were discriminated using five polymorphic SSRs, while the addition of easily identifiable tuber traits reduces the number of common fingerprint patterns to four pairs. The common pairing of Atlantic/Katahdin, Belchip/Wauseon, and Red LaSoda/Bliss Triumph is expected due to the genetic similarity based upon pedigrees. The development of additional polymorphic SSRs would probably discriminate the five cultivar pairs. SSR discrimination was based upon band presence and absence. Due to the heterozygous nature of potato cultivars, we would expect to observe multiple-banded patterns for many of the SSR/cultivar combinations. Because SSRs are codominant DNA markers, we attempted to examine dosage differences, but the resolution of the bands on the agarose gel limited our ability to score dosage effects. The use of polyacrylamide gels (and/or radioactivity) may improve band resolution and may result in greater cultivar discrimination.

Genetic fingerprints are useful for the protection of plant breeders' rights, monitoring seed production, and marketing. Limitation of fingerprint analyses will occur when a cultivar is a sport or somatic mutation (line selection) of a previously-released cultivar. The same limitation would be found if a new cultivar was a result of genetic engineering. Isozyme and RFLP markers have not been effective in discriminating sports or line selections; however, RAPDs have revealed some differences among some line selections. This array of SSRs was not able to discriminate Burbank from its russet sport. As more SSRs are developed, this issue can be more fully addressed.

Bailey (1) lists basic criteria for a molecular marker preferred for cultivar identification. These include distinguishable intercultivar variation, minimal intra-cultivar variation, environmental stability, and experimental reproducibility. SSRs should satisfy all these criteria. Our study shows that SSR variation does exist among potato cultivars. Secondly, potato cultivars are suited to fingerprinting because each one is a unique genotype reproduced vegetatively

and we expect limited intra-cultivar variation. By definition, DNA markers should be independent of the environment. With the use of longer primers (24-base versus 10-base) compared to those used for RAPDs, the reproducibility of amplifying bands among labs should be greater. These characteristics, combined with the relatively simpler technology of PCR amplification versus DNA hybridization, makes these SSRs the DNA marker of choice. Although SSRs did not generate unique fingerprints for all of the North American genotypes examined, the potential to discriminate most cultivars should increase as additional SSRs are identified in potato.

Literature Cited

Bailey, D.C. 1983. Iozymic variation and plant breeder's rights. *In:* Tanksley, S.D. and T.J. Orton (eds.) Isozymes in plant genetics and breeding, part A. Elsevier. Amsterdam. pp. 425-440.

Demeke, T., L.M. Kawchuk, and D.R. Lynch. 1993. Identification of potato cultivars and clonal variants by random amplified polymorphic DNA analysis. Am Potato J 70:561-570.

Desborough, S.L. and S.J. Peloquin. 1968. Potato variety identification of electrophoretic patterns of tuber proteins and enzymes. Am Potato J 45:220-229.

Douches D.S. and K. Ludlam. 1991. Electrophoretic characterization of North American potato cultivars. Am Potato J 68:766-780.

Douches, D.S., R. Freyre, and K. Hicks. 1990. Use of RFLPs to fingerprint North American potato varieties. *In:* First Planning Conference for the Use of Molecular Techniques in Potato Germplasm Enhancement. CIP pp. 166-175.

Doyle, J.J. and J.L. Doyle. 1987. A rapid DNA isolation procedure for small quantities of fresh leaf tissue. Phytochem Bull 19:11-15.

Gebhardt, C., C. Blomendahl, U. Schachtschabel, T. Debener, F. Salamini, and E. Ritter. 1989. Identification of 2n breeding lines and 4n varieties of potato (*Solanum tuberosum* ssp. *tuberosum*) with RFLP finger prints. Theor Appl Genet 78:16-22.

Gorg, R., U. Schachtschabel, E. Ritter, F. Salamini, and C. Gebhardt. 1992. Discrimination among 136 tetraploid potato varieties by fingerprints using highly polymorphic DNA markers. Crop Sci 32:815-819.

Haley, S.D., P.N. Miklas, L. Afanador, and J.D. Kelly. 1994. DNA Markers and Plant Breeding Programs. Advances in Agronomy 55:265-344.

Hamada, H. and T. Kakunaga. 1982. Potential Z-DNA forming sequences are highly dispersed in the human genome. Nature (London) 298:396-398.

Hamada, H., M.G. Petrino, and T. Kakunaga. 1982. A novel repeated element with Z-DNA-forming potential is widely found in evolutionarily diverse eukaryotic genomes. Proc Natl Acad Sci USA 79:6465-6469.

Hosaka, K., M. Mori, and K. Ogawa. 1994. Genetic relationships of Japanese potato cultivars assessed by RAPD analysis. Am Potato J 71:535-546.

Kun-Sheng, W. and S.D. Tanksley. 1993. Abundance, polymorphism and genetic mapping of microsatellites in rice. Mol Gen Genet 241:225-235.

Lee, M. 1995. Random amplified polymorphic DNA marker variability between and within gene pools of common bean. J Amer Soc Hort Sci 119:122-125.

Martin, B., J. Nienhuis, G. King, and A. Schaefer. 1989. Restriction fragment length polymorphisms associated with water use efficiency in tomato. Science 243:1725-1728.

Maughan, P.J., M.A. Saghai Maroof, and G.R. Buss. 1995. Microsatellite and amplified sequence length polymorphisms in cultivated and wild soybean. Genome 38:715-723.

May, B., J.E. Staub, and L.J. Kuhns. 1982. Potato cultivars: genetic variation within putative clones. Am Potato J 59:179-187.

Morgante, M. and A.M. Olivieri. 1993. PCR-amplified microsatellites as markers in plant genetics. Plant J 3:175-182.

Morgante, M., A. Rafalski, P. Biddle, S. Tingey, and A.M. Olivieri. 1994. Genetic mapping and variability of seven soybean simple sequence repeat loci. Genome 37:763-769.

Mori, M., K. Hosaka, Y. Umemura, and C. Kaneda. 1993. Rapid identification of Japanese potato cultivars by RAPDs. Japanese J Genetics 68:167-174.

Nei, M. 1972. Genetic distances between populations. Amer Naturalist 106:283-292.

Olivier, J.L and J.M. Martinez-Zapater. 1985. A genetic classification of potato cultivars grown in the United States. HortScience 9:328-330.

Panaud, O., X. Chen, and S.R. McCouch. 1995. Frequency of microsatellite sequences in rice (*Oryza sativa* L.). Genome 38:1170-1176.

Phillips, W.J., C.G.D. Chapman, and P.L. Jack. 1994. Molecular cloning and analysis of one member of a polymorphic family of GACA-hybridising DNA repeats in tomato. Theor Appl Genet 88:845-851.

Poulsen, G.B., G. Kahl, and K. Weising. 1993. Abundance and polymorphism of simple repetitive DNA sequences in *Brassica napus* L. Theor Appl Genet 85:994-1000.

Rohlf, F.J. 1992. NTSYS-pc. Numerical taxonomy and multivariate analysis system, version 1.70. Exeter Software, New York.

Rus-Kortekaas, W., M.J.M. Smulders, P. Arens, and B. Vosman. 1994. Direct comparison of levels of genetic variation in tomato detected by a GACA-containing microsatellite probe and by random amplified polymorphic DNA. Genome 37:375-381.

Saghai Maroof, M.A., R.M. Biyashev, G.P. Yang, Q. Zhang, and R.W. Allard. 1994. Extraordinarily polymorphic microsatellite DNA in barley: Species diversity, chromosomal locations and population dynamics. Proc Natl Acad Sci 91:5466-5470.

Senior, M. L. and M. Heun. 1993. Mapping maize microsatellites and polymerase chain reaction confirmation of the targeted repeats using a CT primer. Genome 36:884-889.

Smith, D.N. and M.E. Devey. 1994. Occurrence and inheritance of microsatellites in *Pinus radiata*. Genome 37:977-983.

Sosinski B. and D.S. Douches. 1996. Using PCR-based DNA amplification to fingerprint North American potato cultivars. HortScience 31(1):130-133.

Stegemann, H. and V. Loeschke. 1977. Das europäische kartoffelsortiment und seine indexierung. Potato Res 20:101-110.

Tanksley, S.D. 1983. Molecular markers in plant breeding. Plant Mol Biol Rep 1:3-8.

Terauchi, R. and A. Konuma. 1994. Microsatellite polymorphism in *Dioscorea tokoro*, a wild yam species. Genome 37:794-801.

Veilleux, R.E., L. Yin Shen, and M.M. Paz. 1995. Analysis of the genetic composition of anther-derived potato by randomly amplified polymorphic DNA and simple sequence repeats. Genome 38:1153-1162.

Wang, Z., J.L. Weber, G. Zhong, and S.D. Tanksley. 1994. Survey of plant short tandem DNA repeats. Theor Appl Genet 88:1-6.

Yang, G., M.A. Saghai Maroof, C.G. Xu, Q. Zhang, and R.M. Biyashev. 1994. Comparative analysis of microsatellite DNA polymorphism in landraces and cultivars of rice. Mol Gen Genet 245:187-194.

Zwartz, J.A. 1966. Potato varieties and their protein electropherogram characteristics. Eur Potato J 9:111-128.