

## Molecular identification of genetically distinct accessions in the USDA chickpea core collection

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### Summary

Knowledge of the molecular genetic variation of the accessions of core collections will be important for their efficient use in breeding programs, and for conservation purposes. The present study was undertaken for genotyping the part of the USDA chickpea core collection (Hannan et al 1994) with 20 microsatellite or simple sequence repeat (SSR) markers. In addition to understand the molecular diversity in the core collection, the genetic relationship was studied. A total of 376 accessions from the USDA chickpea core collection were genotyped. Twenty SSR markers revealed a total of 388 alleles among the 376 accessions. In the USDA core collection, the shared allele frequency (SAF) varied from 7.5% to 47.5% with an average of 21.6%. In the present study, the structure of the population was determined by using  $K=4$  based on model-based (Bayesian) clustering algorithm.

Understanding of the molecular diversity in germplasm collections has several applications and advantages. These include genebank management issues and association mapping studies. Genebank management uses of molecular diversity information include maintaining genetic diversity, increasing diversity through knowledge-based acquisition, reducing redundancy and creating association mapping studies populations. Association mapping studies have increased the possible ways of utilizing the genetic diversity of the 6,193 accessions in the USDA chickpea germplasm collection. The initial step for using the chickpea core collection for these studies is determining the underlying population structure to improve the likelihood of finding true associations between a gene and trait.

A total of 376 accessions from the USDA chickpea core collection were used and DNA was extracted from bulks of ten plants per accession. Twenty SSRs were used to genotype the collection (Table 1). Standard SSR PCR followed by capillary electrophoresis was used to collect the genotypes. A dendrogram based on similarities was generated using NTSYS pc software (Rolf 2000). The genetic diversity structure of the collection was determined by using the clustering algorithm of STRUCTURE software (Pritchard et al. 2000).

**Table 1. Microsatellites used to genotype the USDA chickpea core collection from Huttel et al 1999 and Winter et al. 1999.**

CaSTMS15	TA113	TA200	TA72
CaSTMS2	TA118	TA206	TR29
CaSTMS21	TA130	TA22	TR31
NCPGR19	TA135	TA64	TR7
NCGRP6	TA14	TA71	TS84

Twenty SSR markers revealed a total of 388 alleles among the 376 accessions. The shared allele frequency varied from 7.5% to 47.5% with an average of 21.6%. These results suggest a high level of genetic diversity present in the germplasm investigated. Our preliminary determination of the population structure is  $K=4$

**Table 2. The accessions that were the most distant from the rest of the core are listed. All but one accession are from the USDA Regional Pulse Improvement Program conducted in Iran and India in the 1960's and early 70's.**

Accession	RPIP	Origin	Accession	RPIP	Origin
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PI 360108	12-069-06278	India	PI 360641	12-071-06533	Iran
PI 426536	K202 (not RPIP)	Pakistan	PI 360680	12-069-06208	India
PI 360662	12-075-00858	Italy	PI 450658	12-069-00434	India
PI 360687	12-139-00922	Spain	PI 450669	12-069-0475	India
PI 450843	12-069-01490	India	PI 450717	12-069-00711	India
PI 451671	12-153-06965	Turkey	PI 451597	12-071-07051	Iran

Table 3. Thirty-seven sets of accessions that were indistinct based on 20 SSRs indicated by same cell location in this table.

PI 359009	PI 359805	PI 439831	PI 450852	PI 359607	PI 450654
PI 451501	PI 359815	PI 439858	PI 450902	PI 359968	PI 451394
PI 360660	PI 426536	PI 359245	PI 359186	PI 193487	PI 207470
PI 360664	PI 439834	PI 462168	PI 502991	PI 343016	PI 451085
PI 359481	PI 214311	PI 360133	PI 360342	PI 451032	PI 359260
PI 359913	PI 426571	PI 360328	PI 360574	Annigeri	PI 450772
PI 359489	PI 360672	PI 368485	PI 360358	PI 360348	PI 426195
PI 509156	PI 426546	PI 451664	PI 450585	PI 450622	PI 503010
PI 360078	PI 215588	PI 219728	PI 359228	PI 359372	PI 273879
PI 451127	PI 360609	PI 359363	PI 359316	PI 359544	PI 359213
	PI 360630	PI 359969	PI 450930	PI 451054	PI 450975
PI 358916	PI 360418	PI 257584	PI 359374	PI 359801	PI 359007
PI 360063	PI 360470	PI 315813	ICCV2	PI 360292	PI 359555
PI 451688	PI 451622	PI 358930	PI 359560	PI 360315	PI 359673
		PI 360194	PI 462176	PI 426193	PI 360599
PI 359595, PI 360230, PI 360304, PI 360344, PI 360425, PI 360456					

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