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Genetic Diversity and Structure of *Acacia senegal* (L.) Willd. in Uganda

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Abstract: The study assessed the genetic variation and population structure of *Acacia senegal* in Uganda. Based on 129 mature individuals, representing eight populations, the genetic variation and population structure of *Acacia senegal* in Uganda was analysed at four microsatellite loci. All four loci were highly variable, with the number of alleles per locus ranging from 8-14 (mean = 10.5). Substantial levels of genetic diversity were found (mean expected heterozygosity, H_e = 0.479, range 0.245-0.846; Information Index, I = 0.927, range 0.646-1.206). Analysis of molecular variance demonstrated moderate genetic differentiation among populations [F_{st} = 0.100, $p \leq 0.001$], comparable to similar tropical species. Isolation by distance, based on Mantel Test showed a positive and significant correlation [R_{xy} = 0.197, $p \leq 0.001$]. Based on population assignment, pair-wise population comparisons and PCA, four populations emerged; one on the eastern and the other on the western side of lake Kyoga, suggesting isolation due to a water body barrier; two other populations emerged in Karamoja. The observed southern-northern flowering pattern across the species distribution range appears central to the species differentiation. The four populations, therefore, form the target for conservation and sustainable utilization of the species genetic variability in Uganda.

Key words: *Acacia senegal*, cross-species amplification, genetic diversity, population structure, Uganda

INTRODUCTION

Acacia senegal (L.) Willd. is a leguminous species whose importance cuts across ecological, cultural and traditional, pharmaceutical, confectionary and other industrial spheres of utilization (Fagg and Stewart, 1994; Wickens *et al*., 1995; Fagg and Allison, 2004). Its unmatched gum (Gum arabic) enjoys substantial utilization in the food and pharmaceutical industry due to absence of toxicity (Fagg and Allison, 2004). The future of its utilization, however, will very much depend on the ability to increase production to stable commercial scale. To achieve this, selection and promotion of promising progenies is inevitable. Unfortunately, continuing destruction of natural ecosystems places both *in-situ* conservation and future commercial utilization of *A. senegal* in serious jeopardy. In order to support conservation efforts, either *in situ* or *ex situ* and enable realization of sustainable utilization of this species, a proper understanding of variability, across the range of the species is becoming critical.

Acacia senegal is a very variable taxon presenting difficulties and contradictions (Ross, 1979; Brenan, 1983; Hassan and Styles, 1990; Mulumba and Kakudidi, 2009, 2011). However, apart from the few limited studies undertaken with allozymes (Chevallier *et al*., 1994) and the more recent equally limited studies of some Kenyan and Sudanese populations with RAPDs, ISSRs and microsatellites (Josiah *et al*., 2008; Rami and Eisa, 2009; Omondi *et al*., 2010), the genetic diversity and structure of the species across its range have not been thoroughly elucidated. In Uganda, no such studies have been undertaken before on *A. senegal*. It is thus, still not possible to delineate credible relationships between the highly variable phenotypic expression or gum quality and the genetic diversity or structure of the species. Apart from resolving phenetic, ecological and phylogenetic questions, genetic diversity and structure studies facilitate identification of unique populations/genotypes for conservation and/or development.

Though use of several nuclear DNA markers in population genetic studies has resulted in resolving complex taxonomic relationships (Voigt *et al*., 1995; Comincini *et al*., 1996; Cottrell *et al*., 1997; Wolff and Morgan-Richards, 1999), microsatellites have proved to be more reliable and reproducible (Byrne *et al*., 1996;

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Brondani *et al*., 1998; Devey *et al*., 1999; Ni *et al*., 2002; Belaj *et al*., 2003; Nybom, 2004) and were therefore chosen for this study. At the time of this study, however, no microsatellite marker development work had been done on *Acacia senegal*. The earliest work on isolation and characterization of microsatellites markers for *Acacia senegal* was undertaken by Assoumane *et al*. (2009). Previous work had focused on other species in the genus, particularly on *Acacia mangium* (Butcher *et al*., 2000) *Acacia nilotica* (Wardill *et al*., 2004), *Acacia auriculiformis* (Ng *et al*., 2005) and *Acacia brevispica* (Otero-Arnaiz *et al*., 2005). Cross-species amplification of microsatellite loci is considered a cost-effective approach for assembling microsatellite markers for related species that have not yet had species-specific markers isolated. This study was therefore based on previously reported success in cross-species amplification.

The objective of the study was to quantify the genetic variation, diversity and structure of *A. senegal* in Uganda, using transferred microsatellite markers.

MATERIALS AND METHODS

Study area: The study was undertaken during the period January 2007 to December 2009. Eight locations representing the entire range of the species occurrence in the country were sampled. Twenty to twenty nine mature individuals were sampled per location, with a distance of 50-800 m between individuals, giving a total of 129 individuals. The eight locations were assumed to represent fairly continuously distributed populations. Out of the eight populations, five namely; P1, P3, P4, P5 and P 8 were within the greater lake Kyoga basin while three namely; P10, P11 and P12 were from Karamoja region (Fig. 1).

Plant material and DNA isolation: Tender young leaves were collected from each individual and kept separately in an airtight ziplock bag. The leaves were dried using Silica gel and stored at -20°C until the time for DNA extraction. The CTAB modified protocol of Gawel and Jarret (1991) was used to extract DNA. Preliminary DNA quantification and sizing was done by comparing with known concentrations of a DNA standard (Lambda DNA) in Ethidium bromide-stained, 1% agarose gels. Final quantification was done using the Picofluor Fluorometer and with Molecular Probes' PicoGreen dsDNA Quantitation Reagent.

Primer pre-testing and microsatellite analysis: Five primers developed for *A. nilotica* (Wardill *et al*., 2004) and

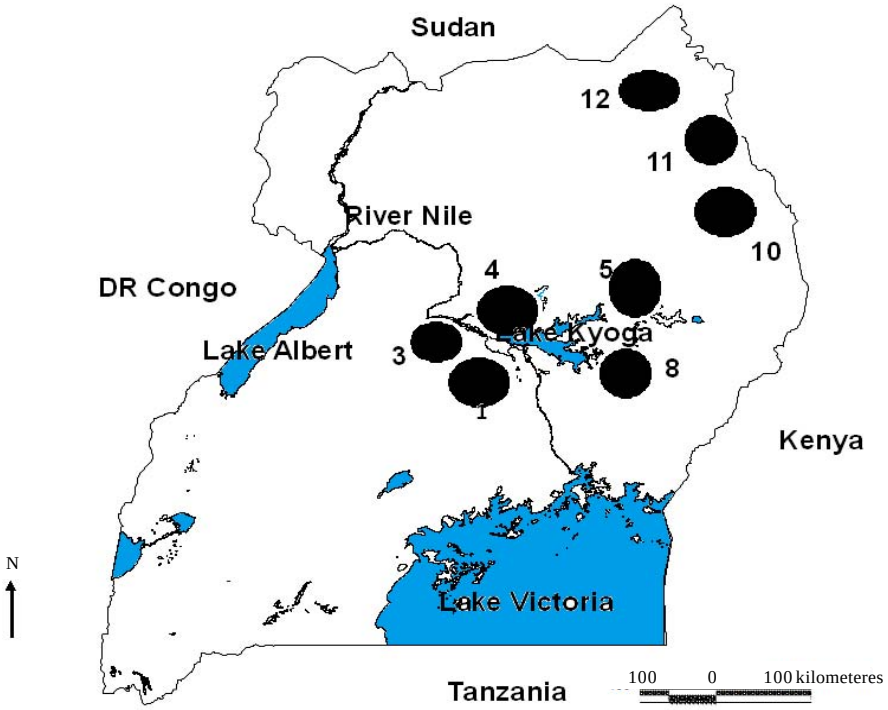

Fig. 1: Locations of the eight *Acacia senegal* populations sampled in Uganda

ten for *A. brevispica* (Otero-Arnaiz *et al*., 2005) were screened for possible amplification of DNA, as well as for polymorphism. The four best performing primers (Primer names: AN17, AN18, Ab05, Ab06) were carried forward for the study. All the visualization for the pre-testing was done, based on electrophoresis using Ethidium bromide stained 2% agarose gels. Final PCR amplification was run on the Thermal Scientific Matrix PlateMate plus for high throughput liquid handling (pipetting) and a Kbiosystems Super Duncan thermal cycler, based on the following PCR reaction mix: Buffer 1x, primer 0.64 pmol μL^{-1}, $MgCl_2$ 3 mM, *Taq* 0.08 units μL^{-1}, BSA 0.08x, DNA 10 ng μL^{-1}; with the following thermocycler profile: 94°C for 2 min; 40 cycles of (94°C for 20 sec, 50°C for 40 sec, 72°C for 60 sec); 72°C for 10 min. Employing applied biosystems Liz 500 bp size standard for sizing, fragment analysis was done with the Applied Biosystems (ABI Prism) 3730 genetic analyser and was finally cross-checked and edited by manual visualization of peaks, using PeakScanner software. Scoring of errors was mitigated, following the procedures as described by Dewoody *et al*. (2006) including; screening at least twenty five individuals for each marker to understand the peak pattern, the levels of allelic variation and identifying alleles producing inconsistent patterns; visual screening of electrophoretic patterns and re-amplification and scoring of samples.

Data analysis: To estimate overall levels of genetic diversity, data were analysed using GenAlEx ver. 6.3 software (Peakall and Smouse, 2006). We calculated allelic frequencies, mean number of alleles per locus (A), percentage of polymorphic loci (p), mean observed (H_o) and unbiased expected heterozygosity (H_e) within populations. A hierarchical analysis of molecular variance (AMOVA) was implemented, following the methods of Excoffier *et al*. (1992), Peakall *et al*. (1995) and Michalakis and Excoffier (1996), to estimate F-statistics, with 9999 permutations for both testing of population divergence in general and for pairwise population comparisons. We chose F-statistics, instead of R-statistics (Slatkin, 1995), because the former is more conservative and robust, when dealing with small numbers of loci and small number of individuals (Gaggiotti *et al*., 1999; Musoli *et al*., 2009). The information index:

$$I = \Sigma p_i * \ln(p_i)$$

where, ln is the natural logarithm and p_i is the frequency of the ith allele (equivalent to Shannon Index, more familiar in ecology) was also used to estimate the level of genetic diversity within and among populations (Weising *et al*., 2005). The deviations from Hardy-Weinberg expectations were estimated using the Program Arlequin ver. 3.0 (Excoffier *et al*., 2005) following the procedure of Guo and Thompson (1992), based on MCMC techniques (1,000,000 steps and 100,000 de memorisation steps), at a significance level of 0.05.

Isolation by distance based on correlation between the genetic and the geographical distances between populations using a Mantel test was estimated based on the methods of Smouse *et al*. (1986) and Smouse and Long (1992). Population assignment based on 'self' or 'other' population with 'leave one out' option (Paetkau and Strobeek, 1995) was undertaken. This assignment test assumes Hardy-Weinberg equilibrium, but it is known to be fairly robust to violation of this assumption (Cornuet *et al*., 1999), unlike Structure which in addition; assumes all potential source populations have been sampled, requires a larger number of loci and a larger number of individuals (Pritchard *et al*., 2000). Principal Coordinate Analysis (PCA), based on conversion of a genetic distance matrix to a covariance matrix, was undertaken.

RESULTS

Cross-species amplification: All the four markers were successfully amplified using a lower annealing temperature (50°C) than one used in the source species. Except for one marker (AN18), the rest exhibited a wider size range (bp) than expected.

Genetic diversity: Three of the four loci namely, AB05, AN17 and AN18 had their commonest alleles occurring at frequencies of less than 85% across the eight populations, while one locus (AB06) had the commonest allele occurring at a frequency of 97%. The four microsatellite loci in the final analysis were highly polymorphic yielding a total of 42 alleles. The mean number of alleles per locus was 10.5, across the whole study, with a range of 8-14 alleles (Table 1). On a single population basis, the mean number of alleles observed per locus was only 4.3 (range = 3.5 -5.5) whereas the mean number of effective alleles was 2.3 (range = 1.5-3.1). Each population had two private alleles with the exception of Populations 4 and 8 which had one and population 10 which had three. Private alleles were found in only 12% of the individuals studied. The private allelic frequency, averaged across loci and populations, ranged from 2.8-5.9%, except for one AN18 allele in population 10, with a frequency of 10%. Based on number of alleles detected, the *A. senegal* loci show higher levels of variability (up to 10 and 14 alleles) than these same loci in their own source species, *A. nilotica* (Wardill *et al*., 2004) and *A. brevispica* (Otero-Arnaiz *et al*., 2005), respectively.

Table 1: Summary statistics of genetic variation at four SSR loci in eight populations of *Acacia senegal* from Uganda

	Population								Grand
Locus	P1	P3	P4	P5	P8	P10	P11	P12	Total
AB06									
N	18	17	19	17	17	17	11	12	128
A	4	5	3	7	4	2	2	5	10
A_e	1.600	1.656	1.112	1.680	1.550	1.061	1.095	1.714	-
I	0.746	0.831	0.243	0.953	0.731	0.133	0.185	0.873	-
H_o	0.333	0.353	0.105	0.471	0.294	0.059	0.091	0.500	0.276
H_e	0.375	0.396	0.101	0.405	0.355	0.057	0.087	0.417	0.274
F_{is}	0.111	0.109	-0.041	-0.162	0.171	-0.030	-0.048	-0.200	-0.006
AN17									
N	17	13	18	15	16	15	9	11	114
A	4	4	5	2	2	4	3	4	8
A_e	2.688	2.299	2.204	1.557	1.679	2.093	1.588	1.793	-
I	1.155	1.003	0.979	0.543	0.594	0.936	0.684	0.862	-
H_o	0.765	0.692	0.611	0.467	0.313	0.600	0.333	0.364	0.518
H_e	0.628	0.565	0.546	0.358	0.404	0.522	0.370	0.442	0.479
F_{is}	-0.218	-0.225	-0.119	-0.304	0.227	-0.149	0.100	0.178	-0.080
AN18									
N	15	17	17	14	13	15	10	13	114
A	5	5	4	2	4	4	5	4	10
A_e	3.629	1.789	1.360	1.324	2.099	3.543	2.941	2.432	-
I	1.377	0.940	0.573	0.410	0.912	1.310	1.257	1.069	-
H_o	0.400	0.176	0.059	0.000	0.077	0.600	1.000	0.385	0.337
H_e	0.724	0.441	0.265	0.245	0.524	0.718	0.660	0.589	0.512
F_{is}	0.448	0.600	0.778	1.000	0.853	0.164	-0.515	0.347	0.353
AB05									
N	17	17	19	17	16	17	11	13	127
A	4	7	6	3	4	7	4	9	14
A_e	1.871	3.400	2.579	1.615	2.738	4.129	3.667	6.500	-
I	0.883	1.495	1.203	0.678	1.126	1.616	1.342	2.021	-
H_o	0.059	0.647	0.158	0.000	0.063	0.647	0.000	0.308	0.236
H_e	0.465	0.706	0.612	0.381	0.635	0.758	0.727	0.846	0.641
F_{is}	0.874	0.083	0.742	1.000	0.902	0.146	1.000	0.636	0.633
A (total)	17.000	21.000	18.000	14.000	14.000	17.000	14.000	14.000	42.000
I (average)	1.04	1.067	0.749	0.646	0.841	0.999	0.867	1.206	0.927
H_e(average)	0.548	0.527	0.381	0.347	0.479	0.514	0.461	0.573	0.479
F_{IS}(average)	0.304	0.142	0.34	0.383	0.538	0.033	0.134	0.24	0.264

N: Number of individuals genotyped, A: Number of alleles detected; A_e: Number of effective alleles, I: Shannon information index, H_o: observed heterozygosity, H_e: Expected heterozygosity, F_{IS}: fixation index

Departures from panmixia: The level of allelic polymorphism observed was also evident in the mean expected heterozygosity per population (H_e = 0.479, Table 1), with an average observed heterozygosity of H_o = 0.342. The observed heterozygosity was less than expected within population heterozygosity for loci AB05 and AN18 (H_o<<H_e, F_{IS}>0), but AB06 and AN17 had a small excess of heterozygotes (H_0>H_e, F_{IS}<0) (Table 1), resulting into a positive mean (F_{IS} = 0.264>0), averaged over loci. This indicates an overall excess of homozygosity (deficiency of heterozygosity) within populations, compared with that expected under random mating which in this case is more likely to be due to presence of null alleles rather than an expression of in-breeding. The two loci with the largest F_{IS}-values (AN18 and AB05) also had the highest F_{ST}-values (0.259 and 0.152, respectively, compared with 0.051 and 0.039 for loci AB06 and AN17, respectively), indicating that they were more divergent among populations. Expected heterozygosity across the populations ranged from 0.347 and 0.573 with four populations (Pop 1, 3, 10 and 12) having over 0.500 indicating a higher diversity than the rest. The Shannon information index ranged from 0.6 to 1.2 across the populations with an average of 0.927. Of the 32 tests of conformity to HW proportions, fifteen showed a significant departure from expected proportions at the 5%. Fourteen of them arose from loci AN18 and AB05. Based on the estimates of F_{is}, particularly for loci AN18 and AB05 (Table 1) all the significant deviations were due to a deficit in heterozygotes.

Population differentiation and structure: Table 2 presents the AMOVA results. The results indicated that 10% of the variation was explained by the eight populations differentiation [F_{ST} = 0.100, p≥0.001], whereas 39% and 51% was explained by variation among and within individuals, respectively. Similar results were obtained with a regional-level AMOVA (results not shown), with only 5% of the variation explained by the regions.

Table 2: AMOVA partition and F-statistics, based on eight populations of *Acacia senegal* from Uganda; probability (p-value) for F_{ST}, F_{IS} and F_{ST} is based on 999 permutation across the full data set

Source	df	SS	MS	Est. Var.	%	F-statistics	p-value
Among population	7	38.408	5.487	0.122	10	F_{ST} = 0.100	0.001
Among individual	121	190.337	1.573	0.473	39	F_{IS} = 0.429	0.001
Within individual	129	81.000	0.628	0.628	51	F_{IT} = 0.486	0.001
Total	257	309.744	-----	1.222	100	-----	-----

Table 3: Pairwise population F_{ST} values of eight populations of *Acacia senegal* from Uganda

	P10	P11	P12	P1	P3	P4	P5	P8
P10	**0.000**	0.001	0.003	0.000	0.000	0.000	0.000	0.000
P11	0.068	**0.000**	0.000	0.000	0.002	0.001	0.000	0.000
P12	0.052	0.098	**0.000**	0.001	0.000	0.000	0.000	0.050
P1	0.106	0.092	0.059	**0.000**	0.000	0.000	0.002	0.091
P3	0.122	0.068	0.126	0.067	**0.000**	0.367	0.000	0.000
P4	0.162	0.080	0.194	0.087	0.002	**0.000**	0.000	0.000
P5	0.207	0.181	0.090	0.055	0.169	0.204	**0.000**	0.261
P8	0.110	0.086	0.026	0.017	0.090	0.109	0.007	**0.000**

F_{st} values below bold diagonal values, Probability values based on 9999 permutations shown above diagonal

Table 4: Assignment test results showing percentage proportions of *Acacia senegal* individuals assigned to each of the eight populations from Uganda

	P10	P11	P12	P1	P3	P4	P5	P8
P10	**88**	6	6					
P11	18	**73**				9		
P12	38		**8**	23				21
P1	6	6		**44**	6	18	12	6
P3		6		6	**53**	29	6	
P4		16		10	16	**58**		
P5				6	6	6	**65**	18
P8		6	6		12	12	35	**24**

Correct assignments to populations of origin are shown along the bold diagonal values

Significant differentiation between populations; P12 and P3, P12 and P4, P10 and P5, P11 and P5 and P 4 and P5 were detected based on Fst values (Table 3). While no significant differentiation was observed between P3 and P4 and P5 P8. Figure 2 shows the relationship between genetic and geographical distances. The results indicate a positive, correlation [Rxy = 0.197, P(rand> = data) = 0.001].

Population assignment outcomes to 'self' or 'other' population with leave one out option (Table 4) show that Population 12 and P8 had particularly very low self assignment with most of their individuals (up to 50%) of those assigned to other population assigned to P10 and P5 respectively. The table also shows that the majority of the few individuals of P10 and P5 that were assigned elsewhere were respectively assigned to P12 and P8 further emphasizing the close genetic resemblance observed between these populations as detected by the pairwise Fst values (Table 3). First three axes of the PCA explained 67% of the variation with the first and second explaining 26 and 23% respectively. The eight populations appeared to merge into four clusters (Figure not shown) characteristically reflecting the findings of the population assignment (Table 4). Cluster 1 is dominated by individuals from populations 10 and 12; cluster 2 has a mixed composition of individuals most likely an indication of level of overlap among the populations; cluster 3 is made up of P 11 while cluster 4 is dominated by P3 and P4.

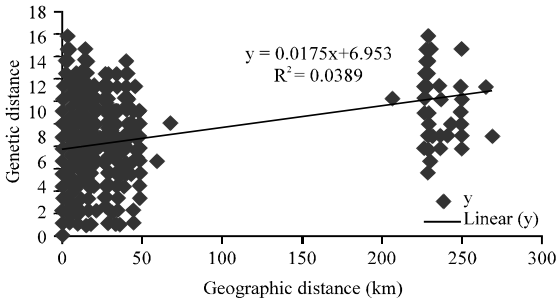

Fig. 2: Mantel test results of the relationship between genetic and geographical distances (Isolation by distance) of *Acacia senegal* populations from Uganda

DISCUSSION

Cross-species amplification: This study was based on the assumption that microsatellite primers developed for loci of related species can be successfully employed in *Acacia senegal*. Although from a different subgenus (*Acacia*), the two source species for the primers were amplifiable in a species from subgenus *Aculeiferum*. The amplification success rate appears higher with *A. nilotica* primers than with *A. brevispic*a (two of five vs two of the ten, respectively). Otero-Arnaiz *et al.* (2005) reported considerable variation in success rates among these

same *A. brevispica* loci, using DNA of the source species (*A. brevispica*) which partly explains the lower success rate for the *A. brevispica* primers. A review of cross-species amplification of microsatellite loci in legumes (Peakall *et al*., 1998) revealed a moderate to complete primer conservation (50-100%) within genera, with the level of polymorphism ranging from as low as 20 to 100%. The present study portrays similar trends.

Genetic diversity of Acacia senegal in Uganda: Acacia senegal occurs naturally as a dominant or as an associate in deciduous bushland and thicket climax vegetation (Langdale-Brown *et al*., 1964; White, 1983). In Uganda, the species may occur at densities of 741 to almost pure stands of 1449 trees per hectare in the Karamoja region, where it is most abundant (Obua *et al*., 2006). The species is also known to be almost exclusively outcrossed (Fagg and Allison, 2004) and self-incompatible (Tandon *et al*., 2001). These factors would favour high genetic variation, as observed in this and previous studies of tropical tree species (Dayanandan *et al*., 1999; Lacerda *et al*., 2001; Lemes *et al*., 2003; Omondi *et al*., 2010). Previous studies on genetic variation in *Acacia senegal*, based on ALFP, RAPD and ISSR markers (Josiah *et al*., 2008), have shown much lower genetic diversity than in this study (i.e., H_e = 0.283 vs 0.479). Omondi *et al*. (2010), using microsatellite markers, showed higher diversity in *Acacia senegal* var. *kerensis* (H_e = 0.677) than previously obtained based on other markers. This can be explained by the findings of comparative studies of RFLPs and microsatellite loci which have shown that microsatellite loci are more variable particularly in trees (Byrne *et al*., 1996; Devey *et al*., 1999) and present high levels of polymorphisms and greater information content (Belaj *et al*., 2003). Microsatellite markers also have the advantage of being co-dominant (Brondani *et al*., 1998; Ni *et al*., 2002) and randomly distributed throughout the genomes (Brondani *et al*., 1998). Coupled with the fact that they are multiallelic and hyper-variable, microsatellites have proved to be highly informative in intra-specific studies of genetic and evolutionary relationships (Buchanan *et al*., 1994; Nybom, 2004). The level of expected heterozygosity, based on microsatellites from *Acacia nilotica,* is similar to that observed in the source species (Wardill *et al*., 2004) whereas that based on microsatellites from *A. brevispica* is slightly lower (Otero-Arnaiz *et al*., 2005). The slightly lower He observed in this study, compared to other tropical tree species (Dayanandan *et al*., 1999; Lemes *et al*., 2003) and that of primer source species for the microsatellites could be explained by the high levels of null alleles alluded to by Wardill *et al*. (2004) and Otero-Arnaiz *et al*. (2005), as well as detected in this study. This may indeed limit the versatility of these loci.

The populations P1 and P3 (with high genetic variation) were from Masindi Port area and Nakasongola. In our previous diversity and distribution-based study (Mulumba *et al*., 2011), the same areas exhibited the highest infraspecific richness in the country based on phenotypic variation. This indicates that the wide phenetic variation observed may indeed have a qualitative genetic basis. Considering the small number of individuals per population, as well as the high level of observed Hardy-Weinberg disequilibrium due to null alleles, the Shannon information index and AMOVA were important for this study in determining the genetic diversity and structure since the two measures are not based on the Hardy-Weinberg assumption (Fritzch and Rieseberg, 1996; Lacerda *et al*., 2001).

Population differentiation: The moderate among population differentiation (F_{ST} = 0.100, $p \leq 0.001$), agreed with regional AMOVA findings where regional separation only explained 5% of the variation. This may not negate some restriction on gene flow among populations. The genetic distances based on pairwise population comparisons (F_{ST}, see Table 3), coupled with the population assignment outcome and the principal component analysis reveal four population clusters. The first cluster comprises mainly P10 and P 12 which happen to come from the same region (Karamoja). The second cluster is the most mixed but predominantly made of P5 and P8, both from eastern and north eastern of L. Kyoga basin. The third cluster comprises mainly individuals from P3 and P4 both from the western side of the L. Kyoga basin while cluster 4 is predominantly P11, a Karamoja population.

Due to the weak, though significant, correlation (r_{xy}= 0.197 and $p \leq 0.001$) between geographical distances and the genetic variation observed, isolation by distance may not be the main factor contributing to the observed genetic differences. Field observations noted that there was a clear phenological pattern (flowering pattern) across the species range in Uganda. The individuals in the southern part of the study area (around lake Kyoga basin) flowered over two weeks earlier than the northern part (Karamoja) and a gradual pattern was evident. But even in one region, Karamoja (along Moroto-Kotido route), flowering time was not fully synchronized; one large population was observed with just a few flower buds, while the rest were approaching floral senescence. This may explain the mixed nature of cluster 2, in which P5, the intermediate population between the L. Kyoga

basin populations and the Karamoja populations, has clustered, implying that it shares pollen with the early flowering as well as the late flowering populations. The observed flowering phenomenon also explains to a great extent the two population clusters that have emerged in Karamoja region. A thorough phenological study is recommended to quantify the contribution of the observed flowering phenomenon to genetic variation and population structuring as well as to known great species morphological variability.

Implications for crop development and species conservation: Previous studies have indicated that Uganda has potential for utilization of *Acacia senegal* in dryland agroforestry and gum arabic production (Obua *et al*., 2006). Variation, though, has been reported in quality, molecular composition and molecular weight of gum arabic of *Acacia senegal* (Fagg and Allison, 2004; Motlagh *et al*., 2006; Al-Assaf *et al*., 2007) and it has been attributed to diversity of the geographical source origins. Until recently (Josiah *et al*., 2008; Rami and Eisa, 2009) no significant amount of work has been done to understand the relationship between the genetic variability in the species and the gum properties. Lack of this important information limits quality progeny selection and crop improvement programs. The wide variation within and among populations observed in this study calls for careful collection of germplasm, paying particular attention to the patterns of genetic divergence across geographic space. It will be desirable to assemble a wide array of different genotypes from many localities and to undertake "common garden" studies of gum quality and other important agronomic features of the phenotype, allowing dissection of genotypic and environment influence. Earlier diversity and distribution studies (Mulumba *et al*., 2011) based on phenotypic variation of the species in Uganda identified the lake Kyoga area to be most important for *in situ* conservation purposes complemented by parts of Moroto and Kotido. Based on the population clustering as well as the genetic variability observed this current study is in agreement with the previous one. This emphasizes the need to undertake ecological studies in the forest reserves within these areas as listed and recommended by Mulumba *et al*. (2011) to understand the status of *Acacia senegal*. The pressure on these forest reserves, however, due to population pressure related activities (NEMA, 2005; NFA, 2008; Obua *et al*., 2010) as well as the predicted climate change impacts on natural ecosystems and species, calls for alternatives to preserving the *Acacia senegal* diversity to be sought. Collecting and conserving the diversity in seed GenBanks would be one of them but even more challenging would be to undertake studies that can predict the locations that are likely to be suitable for the species in future climate change scenarios. Plans can then be undertaken for establishment of arboreta or even more innovative ecosystem management approaches.

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