academicJournals

Vol. 9(12), pp. 1052-1060, 27 March, 2014
DOI: 10.5897/AJAR2013. 8500
Article Number: 979E3C647001
ISSN 1991-637X
Copyright ©2014
Author(s) retain the copyright of this article
http://www.academicjournals.org/AJAR

African Journal of Agricultural Research

Full Length Research Paper

Genetic relationship between sesame (*Sesamum indicum* L.) and related wild species based on chromosome counts and isozyme markers

Benson Ouma Nyongesa[1]*, Beatrice Ang'iyo Were[1], Samuel Gudu[2,3], Otto George Dangasuk[1] and Augustino Osoro Onkware[1]

[1]Department of Biological Sciences, University of Eldoret, P. O. Box 1125-30100, Eldoret, Kenya.
[2]Department of Botany, Moi University, P. O. Box 3900-30100, Eldoret, Kenya.
[3]Rongo University College, P. O. Box 103-40404, Rongo, Kenya.

Received 12 January, 2014: Accepted 20 February, 2014

Sesame is an orphan crop with little research attention in Kenya. Genetic relationship between cultivated sesame and related wild species in Kenya is not well known. The objective of this study was to determine genetic relationship between traditional landraces of sesame and related wild species using somatic chromosome counts and isozyme markers. Somatic chromosome counts of four wild species revealed a consistent chromosome number of 2n = 32, which differed from that of the cultivated sesame (2n = 26), indicating genetic variation in chromosome counts. Only esterase exhibited significant variation and accession-specific esterase bands were identified. Three cathodic and eight anodic bands were observed and the variable bands ranged from 2 to 6 per accession. Cathodic bands with varied relative migration were observed in wild species only, whereas anodic bands were observed for all the accessions. Accessions of cultivated sesame were more genetically diverse compared to wild species. Morogoro, 107UG, Indian-1 and Indian-2 recorded the highest number of esterase bands, while 103w had the lowest number of bands. Few common bands were found between cultivated sesame and related wild species indicating a distant genetic relationship. Few gene markers are available in sesame and related wild species, therefore, esterase isozymes can contribute to studies in the breeding and genetics of sesame.

Key words: Genetic variation, isozyme, chromosome counts, wild species, sesame.

INTRODUCTION

The genus *Sesamum* belongs to the family Pedaliaceae and consist about 20 species, most of which are indigenous to tropical Africa, with two disjunct sections found in India (Ashri, 1998; Bedigian, 2011; Bedigian, 2003). However, many wild species in Africa remain unknown, their chromosome numbers have not been determined and distribution areas have not been fully described (Agnew and Agnew, 1994; Ashri, 1998;

*Corresponding author. E-mail: bengesa79@yahoo.co.uk, Tel: +254-20-800-8143,Fax: +254-53-203-1299.
Author(s) agree that this article remain permanently open access under the terms of the Creative Commons Attribution License 4.0 International License

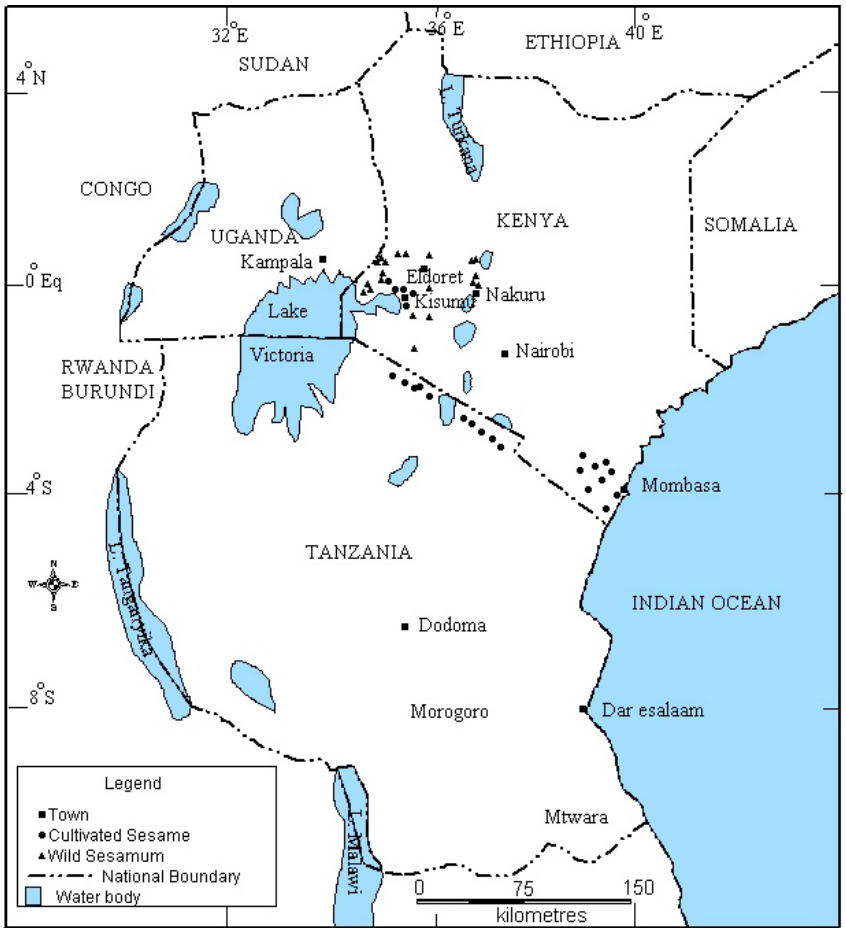

Figure 1. Map of East Africa showing the collection sites for sesame landraces and related wild species studied.

Grubben and Denton, 2004). *Sesamum indicum* L. is the most commonly cultivated due to edible and useful properties of its oil. Related wild species including *Sesamum angustifolium* L. and *Sesamum angolense* L. have been cultivated in Africa providing source of leafy vegetable during famine period and as traditional medicine (Ashri, 1998; Maundu et al., 1999).

Sesame is an orphan crop with little research attention in Kenya. For example, no international agricultural research institute is mandated to conserve and improve sesame (Ashri, 1998). Genetic relationship between cultivated sesame and related wild species in Kenya is not well known. In order to enhance the genetic potential of this crop, there is need to understand genetic relationship that exists between traditional landraces of sesame and related wild species in Kenya. This may enable the broadening of the genetic base of traditional landraces of sesame grown by farmers in Kenya as well as conserving the gene pool of related wild species. Somatic chromosome counts and isozyme markers have the potential to contribute to advances in breeding, genetics and conservation of biodiversity in sesame and related wild species (Diaz et al., 1999; Isshiki and Umezaki, 1997; Kobayashi, 1981; Martinello and Schifino-Wittmann, 2003; Nanthakumar et al., 2000; Parani et al., 1997). Chromosome counts and isozyme were used because they are cost-effective for scientists in developing countries like Kenya.

The aim of this study was to determine genetic relationship between traditional landraces of sesame and related wild species in order to broaden the genetic base of traditional landraces of sesame and conserve the gene pool of related wild species in Kenya.

MATERIALS AND METHODS

Plant materials

Forty-six accessions of sesame representing different regions in Kenya, Tanzania and Uganda were used in this study (Figure 1 and Table 1). These materials comprised 34 landraces, 3 commercial varieties, and 9 accessions of 4 wild species. Seeds of related wild species were collected from different parts of Kenya including Sega, Kitale, Busia, Marigat, Kabarnet and Kabimoi centre (Figure 1). These sites were selected because they form natural habitat for most wild *Sesamum* species and some of these sites were not adequately sampled during earlier work by Agnew and Agnew

Table 1. List of sesame accessions and place of collection.

S/N	Accession	Seed source/Locality
	***S. indicum* L.**	
1	Morogoro	Kenya-Tanzania border
2	107UG	Nambale, Kenya
3	Tan 3	Kenya-Tanzania border
4	102UG	Webuye, Kenya
5	Ug 7	Kenya-Uganda border
6	109UG	Adungosi, Kenya
7	Mtwara-1	Mtwara, Tanzania
8	304BU	Ahero, Kenya
9	302TZ	Kisumu, Kenya
10	110UG	Bumala, Kenya
11	Sik114	Breeder
12	105BU	Mumias, Kenya
13	Ug2	Kenya-Uganda border
14	Ug4	Kenya-Uganda border
15	103UG	Kanduyi, Kenya
16	308KE	Ugenya, Kenya
17	Msambweni	Mombasa, Kenya
18	301TZ	Kisumu, Kenya
19	Mtwara-2	Mtwara, Tanzania
20	Tan7	Kenya-Tanzania border
21	Tan6	Kenya-Tanzania border
22	Ug5	Kenya-Uganda border
23	303BU	Kisii, Kenya
24	306BU	Kisumu, Kenya
25	Ug3	Kenya-Uganda border
26	Ug1	Kenya-Uganda border
27	111BU	Luanda, Kenya
28	307KE-a	Ugenya, Kenya
29	307KE-b	Ugenya, Kenya
30	101UG	Kitale, Kenya
31	108BU	Busia, Kenya
32	Stewa	Shimo la Tewa, Kenya
33	113BU	Koyonzo, Kenya
34	Indian-1	India
35	Indian-2	India
36	Lungalunga	Mombasa, Kenya
37	Majengo	Kisumu, Kenya
	S. angolense	
38	105w-a	Kapropita (kabarnet), Kenya
39	202w	Kabarnet, Kenya
40	105w	Kabarnet , Kenya
41	108w	Busia air strip, Kenya
42	109w	Kabimoi, Eldama Ravine Rd, Kenya
	***Sesamum* spp**	
43	103w	Kitale-Kapenguria Rd, Kenya
	S. latifolium	
44	104w-a	Kambi ya Samaki (Marigat), Kenya

Table 1. Contd.

45	104w	Loboi, Marigat, Kenya
	S. calycinum	
46	101w	Sega, Kenya

(1994) and Grubben and Denton (2004). Herbarium specimens representing wild species were prepared and used to identify species as described in taxonomic literature (Agnew and Agnew, 1994; Grubben and Denton, 2004). Photographs of wild species were also taken using a digital camera (Sony DSC-130) to aid in species identification.

Somatic chromosome counts of wild species

For somatic chromosome counts, seeds were germinated in Petri dishes with moistened filter paper and 1 to 1.5 mm long roots were fixed in 3:1 ethanol-acetic acid for 24 h, and stored in 70% ethanol below 0°C until required. For slide preparation, roots were hydrolyzed in 1 M HCl at 60°C for 10 min as described by Martinello and Schifino-Wittmann (2003) with modification in staining regime. The roots were stained in 2% aceto-orcein and semi-permanent slides were examined by light microscopy. Only intact and well-spread cells were analyzed and each root tip was considered as one individual. Chromosome numbers were determined from cells at mitotic metaphase using a Zeiss phase contrast microscope. Photomicrographs were taken using Sony DSC-W130 under ×1000 magnification.

Isozyme analysis

Enzyme extraction, electrophoresis and staining

Enzymes were extracted from young leaves of 2-week-old seedlings and separated electrophoretically on 11% horizontal starch slab gels following the procedure described by Tanksley and Orton (1983) and Wendel (1983). Ten enzyme systems [acid phosphatase (ACP; EC 3.1.3.2), alcohol dehydrogenase (ADH; EC 1.1.1.1), peroxidase (PER; EC 1.11.1.7), esterase (EST), glumate dehydrogenase (GDH; EC 1.4.1.30), aspartate aminotransferase (AAT; EC 2.6.1.1), glucose-6-phosphate isomerase (GPI; EC 5.3.1.9), isocitrate dehydrogenase (IDH; EC 1.1.1.42), malate dehydrogenase (MDH; EC 1.1.1.37), and Phosphogluconate dehydrogenase (PGD; EC 1.1.1.43)] were stained following standard procedures as described by Vallejos (1983).

Statistical data analysis

The esterase bands were scored for their presence (1) or absence (0) in each accession. Data entry was done into a binary matrix as discrete variables. Jaccard's similarity matrix was used to compute pair-wise genetic similarity values (Digby and Kempton, 1994). A dendrogram was generated based on similarity coefficients using un-weighted pair group method with arithmetic mean (UPGMA). The computer package NTSYS-pc, version 2.1 was used for cluster analysis (Rohlf, 2000).

RESULTS

Chromosome counts of *Sesamum* species

All the 153 cells of the 63 analyzed individuals from the 9 accessions representing 4 wild *Sesamum* species had 2n = 32 chromosomes (Figure 2 and Table 2). The cultivated sesame had 2n = 26 chromosomes. Thus, since the base chromosome number in the *Sesamum* genus is x = 8 and x = 13 (Kobayashi, 1981), these species belong to tetraploid group of *Sesamum* sp (2n = 4x = 32). Accessions from Marigat (104w) and Kambi ya Samaki (104w-a) had less number of individuals analyzed because of poor germination.

Isozyme variation

Out of 10 enzyme systems assayed, only esterase showed significant variations among the examined accessions. Therefore, we focused on the isozymes of esterase in the present study. Genetic variation between traditional landraces of sesame and related wild species was observed for the number of bands and banding intensity. Esterase bands were designated as C_1, C_2, C_3 for cathodic and A_1, A_2, A_3, A_4, A_5, A_7, A_8 and A_9 for anodic according to their relative migration. Three cathodic and eight anodic bands were observed and the variable bands ranged from 2 to 6 per accession (Table 3). Cathodic bands with varied relative migration were observed in wild species only, whereas anodic bands were observed for all the accessions. Four accessions namely, Morogoro, 107UG, Indian-1 and Indian-2 recorded the highest number of esterase bands, while 103w had the lowest number of bands. Bands C_1 and A_1 were specific to accession 101w (*Sesamum calycinum)* in both α and β-esterases (Figure 4 and Table 3). Band A_8 occurred in all the accessions but stained more intensely in the wild species than traditional landraces of sesame (Figure 3 and Table 3). Band A_9 was recorded only in traditional landraces of sesame (Figure 3 and Table 3). Accessions of traditional landraces of sesame and related wild species formed separate clusters at 0.44 similarity level (Figure 5). Genetic distances among the 4 wild species were greater than distance among landraces and cultivars.

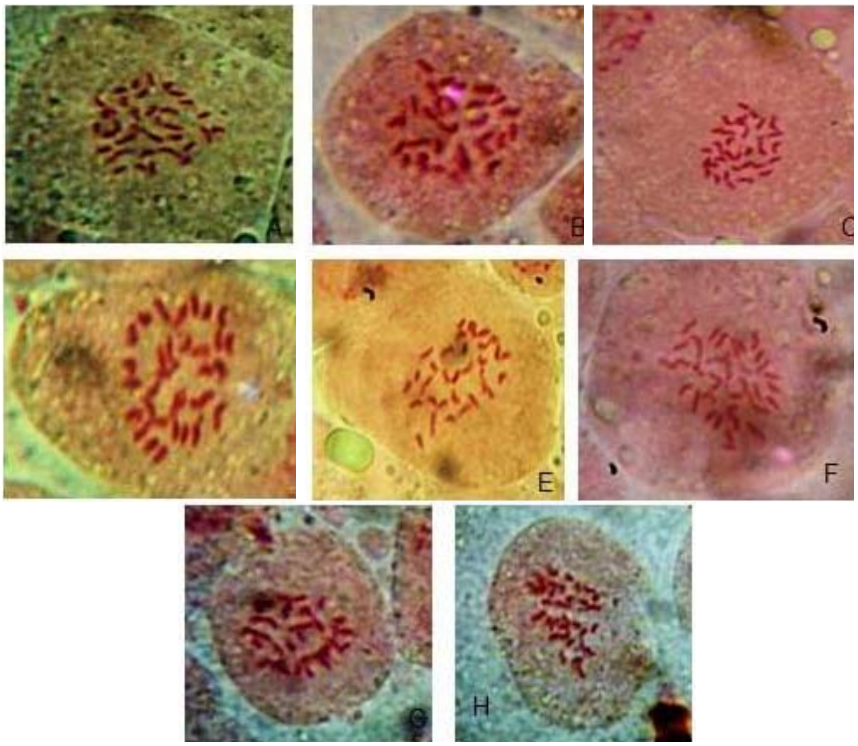

Figure 2. Somatic chromosome spread of wild *Sesamum* species. (A): (101w), (B): (104w-a and 104w), (C): (103), (D-H): (105w-a, 105w, 108w, 109w, 2002w).

Table 2. Somatic chromosome counts of wild *Sesamum* species.

Accessions	Number of individuals	Number of somatic cells	Somatic chromosomes (2n)
S. calycinum			
101w	10	28	32
Sesamum spp			
103w	10	21	32
S.latifolium			
104w-a	5	12	32
104w	4	11	32
S. angolense			
105w-a	5	12	32
105w	7	13	32
108w	6	21	32
109w	7	18	32
202w	9	17	32
Total	63	153	

DISCUSSION

Chromosome counts

Somatic chromosome counts of 4 wild *Sesamum* species namely *S. calycinum*, *S. angolense*, *Sesamum latifolium* and *Sesamum* spp. revealed a consistent chromosome number of 2n = 32, which differed from that of cultivated sesame (2n = 26), indicating genetic variation in chromosome counts. Variation in chromosome counts between cultivated sesame and related wild species has been reported in previous studies (Ashri, 1998; Grubben and Denton, 2004; Kobayashi, 1981; Maundu et al., 1999; Prabakaran, 1996). Chrosomose count result

Table 3. α-esterase bands and banding intensity in the 46 sesame accessions based on migration distance of the bands from origin.

Band number	Anodal bands								Cathodal bands		
Accessions	**A_1**	**A_2**	**A_3**	**A_4**	**A_5**	**A_7**	**A_8**	**A_9**	**C_1**	**C_2**	**C_3**
Cultivated sesame											
107UG		D	M	M	M	M	M	D			
Tan 3			M	M		L	M	D			
102UG						M	M	D			
Ug 7			M	M		L	M	D			
109UG		D	M		L		M	D			
Mtwara-1			M	M		L	M	D			
304BU		D	M	L			M	D			
302TZ		D	M				M	M			
110UG		D	M	L		L	M	D			
Sik114						L	M	D			
105BU			M	M		L	M	D			
Ug2			M	L		L	M	D			
Ug4			M	L		M	M	D			
103UG		D	M	M	L	L	M	D			
308KE		D	M	M			M	D			
Msambweni		D	M	M			M	D			
301TZ		D	M	M	L	M	M	D			
Mtwara-2		D	M	M	L	M	M	D			
Tan7			M	L		M	M	D			
Tan6			M	M		L	M	D			
Ug5		D	M	L	L		M	D			
303BU		D	M	M	L	M	M	D			
306BU		D	M	L	L	M	M	D			
Ug3		D	M	L	L	M	M	D			
Ug1		D	M	L	M	L	M	D			
111BU						L	M	D			
307KE-a		D	M	L			M	D			
307KE-b		D	M	M		M	M	D			
101UG			M	L		L	M	D			
108BU						L	M	D			
Stewa		D	M	M		M	M	D			
113BU		D	M	L		L	M	D			
New Indian		D	M	L	L	M	M	D			
Indian		D	L	M	M	M	M	D			
Lungalunga		D	M	M		M	M	D			
Majengo			M	M		L	M	D			
Wild species											
105w-a				M	L		D			D	
105w				M	M		D			D	
108w					M		D			D	
109w					M		D			D	
202w					M		D			D	
104w-a					M	M	D				D
104w					M	M	D				D
103w							D			D	
101w	D	D	M				D		D		

Key: D - Dark staining; M - Medium staining; L - Light staining.

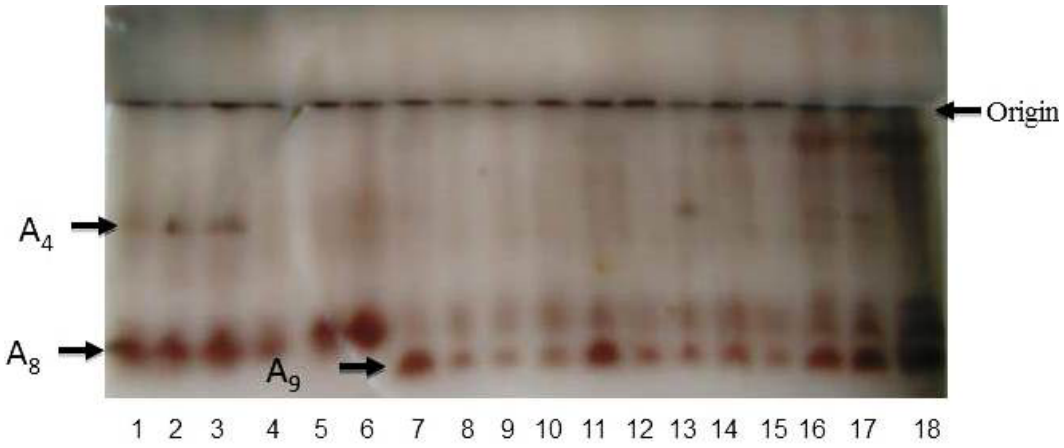

Figure 3. α-esterase isozyme patterns of *Sesamum* species. Lane: 1-3 belong to (*Sesamum* spp); 4-6 (*S. angolense*); 7-18 (*S. indicum*) respectively. Variable anodal bands are shown by the arrows.

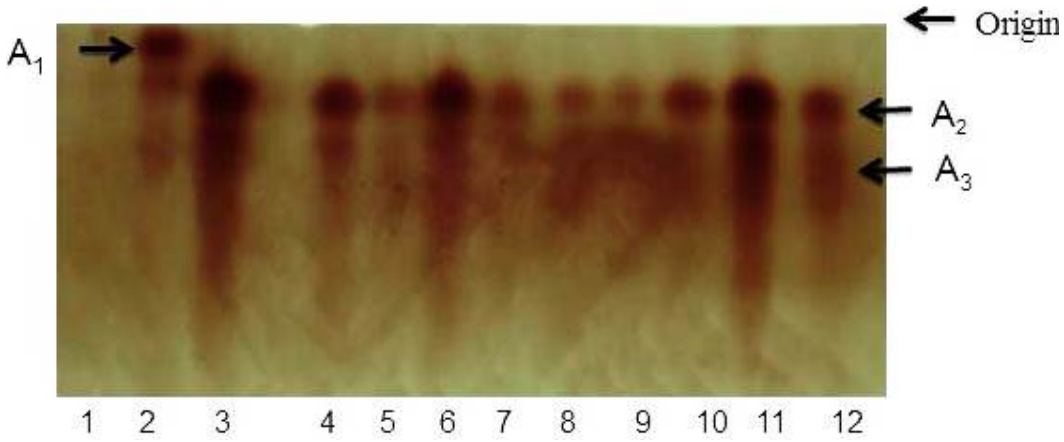

Figure 4. β-esterase isozyme patterns of *Sesamum* species. Lane: 1-2 (*S. calycinum*) and 3-12 (*S.indicum*) respectively. Variable anodal bands are shown by the arrows.

suggests a distant genetic relationship between cultivated sesame and related wild species.

Isozyme variation

Esterase exhibited only significant genetic variation among sesame accessions in terms of number of bands and staining intensities. These findings are supported by previous studies where one or few isozyme markers contributed to variation among different sesame genotypes (Isshiki and Umezaki, 1997; Diaz et al., 1999; Parani et al., 1997). These results suggest that few gene markers are available in sesame and related wild species. A comparable pattern of isozyme variation was also reported in other plant species such as *Elymus* (Yan et al., 2000), *Phalaris* (Matus and Hucl, 1999) and *Trifolium* (Lange and Schifino-Wittmann, 2000) in terms of number of bands and staining intensities. Although cultivated sesame is self-pollinated, up to 60% levels of out-crossing has been reported (Yermanos, 1980). Out-crossing plants tend to present between 10 to 20% of the genetic variation between populations (Hamrick and Godt, 1989). Therefore, out-crossing among neighbouring fields may explain isozyme variation observed in this study. According to Hamrick and Godt (1997), cultivated crops have more allozyme diversity and allele frequency differences are much greater as whole than other seed plant species, although the mean genetic diversity partitioned within population of crop species is similar to that of other plant species. These results are in agreement with earlier studies which reported a high genetic diversity among accessions of cultivated sesame in East Africa based on morphological traits (Pham et al., 2010; Were et al., 2001, 2006).

The wild species had a lower number of isozyme bands

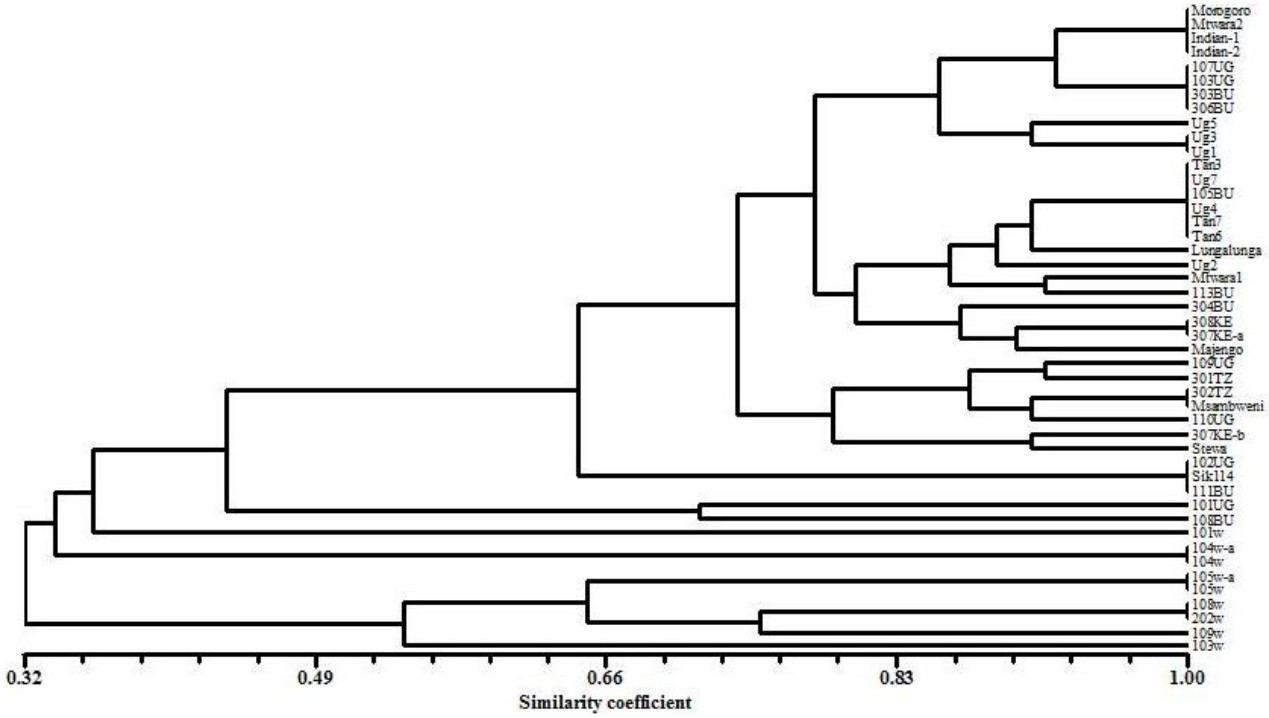

Figure 5. Dendrogram showing relationships between cultivated sesame and related wild species.

compared to traditional landraces of sesame indicating low levels of genetic variation in the wild *Sesamum* species. Wild *Sesamum* species in Kenya occurred along the roadsides and in disturbed environments. Most of these species have been destroyed to pave way for road construction, leaving only a few scattered individuals. Disturbance occasioned by human activities may have a great effect on population diversity by removing a proportion of reproductive plant populations (Chamberlain, 1998). This could result in the loss of alleles, either temporarily within one reproductive cycle, or permanently depending on the extent of damage. Esterase bands observed in wild species stained more intensely than traditional landraces of sesame indicating high enzyme activity. This might be attributed to the effect of chromosome numbers on the plant species. The increase in chromosome number has been reported to affect gene expression and it is attributed to gene dosage effect (Olng'otie, 1991). Peroxidase activity per cell in fern plants was reported to increase in direct proportion to increase in the genome (DeMaggio and Lambrukus, 1974). High enzyme activity may also be attributed to the influence of both biotic and abiotic factors on these species. Significant levels of esterase were observed in diseased wild species of sesame in India (Lubaina and Murugan, 2013). Plants have been reported to alter their gene expression patterns in relation to environmental changes such as temperature, water availability or the presence of deleterious levels of ions (Hazen et al., 2004).

The accessions of sesame and related wild species formed separate groups indicating a distant genetic relationship. The distinctness observed in this study could be attributed to different modes of evolution coupled with sexual incompatibility barriers that exist in some *Seamum* species (Kobayashi, 1981). A high number of chromosome (2n = 32) observed in wild species in this study could also explain the genetic divergent between cultivated sesame and related wild species. Genetic divergence between cultivated and related wild species has been reported in a number of plant species including sesame (Lubaina and Murugan, 2013; Nanthakumar et al., 2000; Prabakaran, 1996), sunflower (Cronn et al., 1997) and Trifolium species (Lange and Schifino-Wittmann, 2000).

Conclusion

Generally, accessions of cultivated sesame were genetically diverse compared to wild species. Morogoro, 107UG, Indian-1 and Indian-2 recorded the highest number of esterase bands, while 103w had the lowest number of bands. Genetic distances among the four wild species were greater than distance among landraces and cultivars, indicating that no cross pollination with wild species occurred during sesame domestication. Few gene markers are available in sesame and related wild species, therefore, esterase isozymes can contribute to studies in the breeding and genetics of sesame.

ACKNOWLEDGEMENTS

The authors acknowledge the Department of Biological Sciences, University of Eldoret for providing financial support towards this research. We thank Biotechnology Centre, University of Eldoret for providing laboratory space. We are also grateful to Mr. Bernard Wanjohi for identifying wild *Sesamum* species.

Conflict of Interests

The author(s) have not declared any conflict of interests.

REFERENCES

Agnew ADQ, Agnew S (1994). Pedaliaceae. In: Upland Kenya wild flowers: A flora of the ferns and herbaceous flowering plants of upland Kenya. 2nd edition. East Africa Natural history Society, Nairobi, Kenya. pp. 264-265.

Ashri A (1998). Sesame breeding. Plant Breed. 16:179-228.

Bedigian D (2011). Cultivated sesame and wild relatives in the genus Sesamum LIn: D. Bedigian (ed). Sesame: the genus Sesamum. Medicinal and aromatic plants-industrial profiles series. CRC press, Taylor and Francis Group, Boca Raton, FL. pp. 33-77.

Bedigian D (2003). Evolution of sesame revisited: domestication, diversity and prospects. Genet. Resour. Crop Evol. 50:779-787.
http://dx.doi.org/10.1023/A:1025029903549

Chamberlain JR (1998). Isozyme variation in *Calliandra calothyrus* (Leguminosae): Its implication for species delimitation and conservation. Am. J. Bot. 85:37-47.http://dx.doi.org/10.2307/2446552 PMid:21684878

Cronn R, Brothers M, Klier K, Bretting PK, Wendel JF (1997). Allozyme variations in domesticated sunflower and its wild relatives. Theor. Appl. Genet. 95:532-545.http://dx.doi.org/10.1007/s001220050594

DeMaggio AE, Lambrukus J (1974). Polyploidy and gene dosage effects on peroxidase activity in ferns. Biochem. Genet. 12:429-440.http://dx.doi.org/10.1007/BF00486060 PMid:4458656

Diaz AJP, Layrisse A, Pugh TM (1999). Analisis de la diversidad genetic en el ajan joli mediante electrophoresis de isoenzimes (Abstract in English). Trop. Agron. 49:169-186.

Digby PGN, Kempton RA (1994). Multivariate analysis of ecological communities. New York: Chapman and Hall.PMid:7802229

Grubben GJH, Denton OA (2004). Plant Resources of Tropical Africa 2.Vegetables. PROTA Foundation, Wageningen, Netherlands/ Backhuys Publishers, Leiden, Netherlands/CTA, Wageningen, Netherlands, pp. 174-467.

Hamrick JL, Godt MJW (1997). Allozyme diversity in cultivated crops. Crop Sci. 37:26-30. http://dx.doi.org/10.2135/cropsci1997.0011183X003700010004x

Hamrick JL, Godt MJW (1989). Allozyme diversity in plants species, pp. 43-63. In: AHD Brown, MT Clegg, AL Kahler and BS Weir (eds). Plant population genetics, breeding and genetics resources. Sinauer Associates Inc. Publishers, Sunderland, Mass.

Hazen SP, Wu Y, Kreps JA (2004). Gene expression profiling of plant response to abiotic stress. Funct. Integr. Genomic 3(3):105-111.http://dx.doi.org/10.1007/s10142-003-0088-4 PMid:12827524

Isshiki S, Umezaki T (1997). Genetic variations of isozymes in cultivated sesame (*Sesamum indicum* L.). Euphytica 93:375-377. http://dx.doi.org/10.1023/A:1002985625087

Kobayashi T (1981). The wild and cultivated species in the genus Sesamum. In: Sesame: Status and Improvement. Proceeding of Expert Consultation, 8-12 December 1980, FAO, Rome, Italy, 1981.

Lange O, Schifino-Wittmann MT (2000). Isozyme variation in wild and cultivated species of the genus Trifolium L. (Leguminosae). Ann. Bot. 86:339-345.http://dx.doi.org/10.1006/anbo.2000.1190

Lubaina AS, Murugan K (2013). Ultrastructural changes and oxidative stress markers in wild and cultivar *Sesamum orientale* L. following Alternaria sesame (Kawamura) Mohanty and Behera. inoculation. Indian J. Exp. Biol. 51:670-680.PMid:24228391

Maundu PM, Ngugi GW, Kabuye CHS (1999). Traditional food plants of Kenya. Kenya resource center for indigenous knowledge, National Museum of Kenya, Narobi, Kenya, pp. 208-209.

Martinello GE, Schifino-Wittmann MT (2003). Chromosome numbers of Bromus auleticus Trin.ex Nees (Poaceae). Genet. Mol. Bio. 26(3):369-371.http://dx.doi.org/10.1590/S1415-47572003000300024

Matus M, Hucl P (1999). Isozyme variation within and among Accessions of Annual Phalaris species in North American Germplasm collections. Crop Sci. 39(4):1222-1228. http://dx.doi.org/10.2135/cropsci1999.0011183X003900040044x

Nanthakumar G, Vaidyanathan P, Singh KN (2000). Relationships between cultivated sesame (Sesamum sp.) and the wild relatives based on morphological characters, isozyme and RAPD markers. Genet. Breed. 54(1):5-12.

Olng'otie PAS (1991). Acacia tortilis (Forsk) Hayne: A study of genetic structure and breeding systems. PhD Thesis. Oxford University, Green College, Oxford.

Parani M, Singh KN, Rangasamy S, Ramalingam RS (1997). Identification of Sesamum alatum × Sesamum indicum hybrid using protein, isozyme and RAPD markers. Indian J. Genet. Plant Breed. 57(4):381-388.

Pham TD, Nguyen TDT, Carlsson AS, Bui TM (2010). Morphological evaluation of sesame (*Sesamum indicum* L.) varieties from different origins. Austr. J. Crop Sci. 4:498-504.

Prabakaran AJ (1996). Genetic diversity of wild sesame from Southern India. School of Genetics, TNAU, Coimbatore, India.

Rohlf FJ (2000). NTSYS-pc: Numerical Taxonomy and Multivariate Analysis System, Version 2.11X: Applied Biostatistics Inc.

Tanksley DS, Orton TJ (1983). Isozymes in Plant Genetics and Breeding, Part A. Elsevier Science Publishers B.V, Amsterdam, pp. 444-468.

Vallejos CE (1983). Enzyme Activity Staining. In: DS Tanksley and JT Orton (Eds.). Isozymes in Plant Genetics and Breeding. Part A. Elsevier, Amsterdam. pp. 469-513.

Wendel JF (1983). Electrophoretic analysis of genetic variation in wild and cultivated Camellia japonica L. PhD Thesis, University of North Carolina, Chapel Hill.

Were BA, Onkware OA, Gudu S, Welander M, Carlsson AS (2006). Seed oil content and fatty acid composition in East African sesame (Sesamum indicum L.) accessions evaluated over 3 years. Field Crop Res. 97:254-260.http://dx.doi.org/10.1016/j.fcr.2005.10.009

Were BA, Lee M, Stymne S (2001). Variation in seed oil content and fatty acid composition of Sesamum indicum L. and its wild relatives in Kenya. Swedish Seed Association 4:178-183.

Yan X, Guo Y, Zhou H, Wang K (2000). Isozyme variability among Elymus species indigenous to the Tibetan and Inner Mongolian Plateaus. Grassland Sci. 53(2):91-96.http://dx.doi.org/10.1111/j.1744-697X.2007.00078.x

Yermanos DM (1980). Sesame. In: WR Fehr and HH Hadley (eds). Hybridization of crop plants. American Society of Agronomy, Madison.