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Full Length Research Paper

Variability, heritability and genetic advance in upland cotton (*Gossypium hirsutum* L.)

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The analysis of variance study indicated the presence of significant difference among all the traits in *Gossypium hirsutum* accessions. The highest phenotypic coefficient of variation (GCV) and genotypic coefficient of variation (GCV) were recorded by seed index, plant height, lint index and boll weight. Genotypic co-efficient of variation had a similar trend as PCV. High heritability along with high genetic advance was observed in traits viz., number of sympodia per plant, single plant yields, seed index and micronaire value. The combinations of high heritability with high genetic advance will provide a clear base on the reliability of that particular character in selection of variable entries. Based on *per se* performance, the accessions MCU5, TCH1715, TCH1716 and G cot 16 were identified as potential donors for single plant yield (g), number of bolls per plant, 2.5% span length (mm) and bundle strength (g/tex). So these accessions may be utilized for crossing programme to improve a particular character in crop improvement.

Key words: Genetic variability, heritability, genetic advance, upland cotton.

INTRODUCTION

Cotton is an important fibre crop of global importance which is grown in tropical and subtropical regions of more than 60 countries of the world. Despite threat from synthetic fibre or manmade fibre, cotton retains its reputation as “queen of the fibre plants””. For multiple uses of lint and by-products, cotton is also referred to as “white gold”. In any crop improvement programme, knowledge on nature of gene action and inheritance of traits is essential so as to choose a suitable breeding methodology in crop improvement (Vineela et al., 2013). Development of an effective breeding programme depends on the existence of genetic variability for various economic characters in the gene pool. Selection is effective only when there is enough magnitude of variability in the breeding population. An understanding of precise magnitude of variability present in a population is important in formulating the most appropriate breeding technique for improvement of various characters. The present investigation was carried out with 54 *Gossypium hirsutum* lines of diverse origin to estimate their *per se* performance, variability, heritability and genetic advance on the genetic architecture of 16 yield, yield components and fibre quality traits.

MATERIALS AND METHODS

The study was conducted in the Department of Cotton, Tamil Nadu Agricultural University, Coimbatore during winter 2010. Fifty four *G. hirsutum* cotton genotypes were planted in randomized block design with two replications. Uniform spacing of 90 x 45 cm and all the recommended field operations were carried out. In each replication five competitive plants were randomly selected and observations were recorded for 16 characters viz., days to 50% flowering, plant height (cm), internode length (cm), number of sympodia/plant, number of ovules/plant, number of bolls/plant, boll weight (g), number of seeds/plant, seed setting percentage, seed cotton yield/plant, lint index, seed index, ginning outturn (%), 2.5% span length (mm), bundle strength (g/tex) and fibre fineness.

Analysis of variance was carried out statistically utilizing the mean values (Panse and Sukhtame, 1995). The phenotypic and genotypic coefficient of variation was estimated using the formula suggested by Burton (1952) and expressed in percentage. The phenotypic and genotypic variances were calculated by utilizing the

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Table 1. Mean and range performance for different characters among G. *hirsutum* accessions.

Characters	Mean	Minimum	Maximum
Days to 50% flowering	56.6	50.0	65.0
Plant height (cm)	108.8	78.0	147.0
Inter node length(cm)	5.4	4.0	6.5
No of Sympodia per plant	19.6	14.0	27.0
No of bolls per plant	23.6	19.5	38.5
Boll weight (g)	4.3	12.4	5.5
No of seeds per boll	27.4	23.0	35.5
No of ovules per flower	30.7	24.0	38.0
Seed setting percentage	89.6	71.5	100.0
Single plant yield (g / plant)	57.6	31.7	91.6
Lint index	5.7	4.0	7.9
Seed index	9.6	6.4	16.3
Ginning outturn (%)	37.3	30.8	46.1
2.5 % span length (mm)	28.4	24.3	34.0
Bundle strength (g / tex)	20.4	16.2	23.7
Micronaire value	4.3	3.5	5.2

respective mean sum of square from variance table (Lush, 1940). Heritability, expected genetic advance and genetic gain in the broad sense was calculated according to the formula suggested by Johanson et al. (1955).

RESULTS AND DISCUSSION

The *per se* performance of yield and fibre quality characters were recorded on 54 germplasm accessions and the range of variations observed in respect of all the 16 traits studied are presented in the Table 1. Based on *per se* performance, the accessions such as MCU 5, TCH1715, TCH1716 and G cot 16 in *G. hirsutum* were identified as potential donors which recorded highest mean values for single plant yield, number of bolls per plant, 2.5% span length (mm) and bundle strength (g/tex). These accessions may be exploited for further improvement of the above traits by breeding programmes. Regarding the identification of donor for specific trait, the highest performer of that particular trait can be considered. Among the accessions, TCH 1715 (5.5 g of boll weight), RB 488 (46.1% of GOT) were expressed the highest *per se* for that particular traits. The culture TCH 1710 (34 mm, 23.1g/tex) recorded the highest 2.5% span length. So these accessions may be utilized for crossing programme to improve that particular character in crop improvement. Anjali was found to be compact type which can be utilized in development of genotype suitable for high density planting.

The analysis of variance showed highly significant differences among genotypes for all the characters studied (Table 2) and infers existence of considerable genetic diversity among genotypes. Phenotypic variance, genotypic variance, phenotypic coefficient of variation, genotypic coefficient of variation, heritability in broad sense and genetic advance as percentage of mean which were estimated for 16 characters are shown in Table 3. The knowledge of nature and magnitude of variability available in the genotypes for different characters is an important prerequisite for making simultaneous selection over more number of characters to bring remarkable improvement in cotton. The analysis of variance study indicated the presence of significant difference among all the traits in the accessions. The heritable (genotypic) variation is usually masked by non-heritable variation creating

Table 2. Analysis of variance for the different characters among the germplasm accessions of *G. hirsutum*.

Source of variation	Degrees of freedom	Days to 50% flowering	Plant height (cm)	nter node length (cm)	Number of sympodia / plant	Number of bolls / plant	Boll weight (g)	Number of sees / boll	Number of ovule / flower	Seed setting percentage	Single plant yield (g)	Lint index	Seed index	Ginning out turn (%)	2.5 % span length (mm)	Bundle strength(g /tex)	Micronaire value
Genotypes	53.0	10.4**	466.6**	0.7**	14.1**	18.3**	0.6*	10.8**	17.1**	90.4**	441.9**	1.3**	4.6**	21.1**	10.2**	6.1**	0.3**
Error	53.0	2.7	218.6	0.4	2.8	5.0	0.3	5.50	6.5	40.1	2.1	0.5	1.2	10.9	0.2	0.3	0.1

*,** Significant at 5 and 1% levels, respectively.

difficulty in exercising selection. Hence it becomes necessary to partition overall variability into heritable and non-heritable components to enable the breeders to plan for proper breeding programme. The plant height recorded the highest value for phenotypic variance (342.6) and the single plant yield recorded highest genotypic variance (219.9). The micronaire value exhibited the lowest phenol-typic variance (0.2) and in case of genotypic variance, the traits *viz*., inter node length, boll weight and micronaire value had recorded the lowest value (0.1). The co-efficient of phenotypic and genotypic variance were calculated for all the characters under study. The PCV ranged from 4.5 (days to 50% flowering) to 25.9% (single plant yield). The highest PCV was followed by seed index (17.7%), plant height (17.0%), lint index (17.0%) and boll weight (15.4%). Genotypic co-efficient of variation had a similar trend as PCV. The range varied from 3.5 (Days to fifty percent flowering) to 25.8 (Single plant yield). The maximum genotypic coefficient of variation (GCV) was observed for single plant yield (25.8) followed by seed index (13.5), number of sympodia per plant (12.1), lint index (11.2) and number of bolls per plant (10.9), plant height (10.2). The lowest PCV (4.5%) and GCV (3.5) values was observed in days to 50% flowering. In the present study, there was a close correspond-dence between phenotypic and genotypic variance for days to 50% flowering, inter node length, boll weight, single plant yield, lint index, micronaire value and 2.5% span length indicating less environmental influence. But plant height, number of sympodia per plant, number of bolls per plant, number of seeds per boll, seed setting percentage and ginning outturn showed higher variation indicating the influence of environment on these characters. Since the variations are influenced by the magnitude of the units of measurements of different traits, a measure of coefficient of variation which is independent of the unit of measurement is more useful in comparing between populations. In *G. hirsutum* accessions PCV was higher than the GCV for all characters. From this, we can understand that the environment plays a major role on expression of all these traits leading to increase in the PCV more than GCV. The highest PCV and GCV estimates were recorded for single plant yield indicating the presence of significant genetic variability in this character. Selection pressure can be applied on this character to isolate promising genotypes. Similar observations in cotton was reported by Dheva and Potdukhe (2002) and Preetha and Raveendran (2007).

Moderate PCV and GCV estimates were noticed for the characters such as plant height, number of sympodia per plant, number of bolls per plant, lint index and seed index. Girase and Mehetne (2002) and Harshal (2010) also reported the moderate PCV and GCV in various traits with the suggestion that these characters can be improved though rigorous selection. The characters such as days to 50% flowering, 2.5% span length and micronaire value exhibited low PCV and GCV which indicated that the breeds should go for source of high variability for these characters to make improvement. Similar suggestion were given by Kowsalya and Raveendran (1996), Do Thi Ha An et al. ((2006) in their conclusion. In a population, the observed variability is a

Table 3. Components of variance for yield and fibre quality characters of *G. hirsutum* accessions.

Characters	Phenotypic variance	Genotypic variance	PCV (%)	GCV (%)	h^2 (%)	A	GA as percentage of mean
Days to 50% flowering	6.6	3.9	4.5	3.5	58.5	4.0	7.0
Plant height (cm)	342.6	124.0	17.0	10.2	36.2	17.7	16.3
Inter node length(cm)	0.5	0.1	13.5	6.6	23.5	0.5	8.4
No of sympodia per plant	8.5	5.7	14.8	12.1	66.9	5.1	26.2
No of bolls per plant	11.7	6.7	14.4	10.9	57.2	5.2	21.8
Days to 50% flowering	6.6	3.9	4.5	3.5	58.5	4.0	7.0
Plant height (cm)	342.6	124.0	17.0	10.2	36.2	17.7	16.3
Boll weight (g)	0.4	0.1	15.4	8.4	29.8	0.5	12.1
No of seeds per boll	8.2	2.6	10.4	6.0	32.5	2.5	9.0
No of ovules per flower	11.8	5.3	11.2	7.5	44.5	4.0	13.1
Seed setting percentage	65.3	25.1	9.0	5.6	38.5	8.2	9.2
Single plant yield (g / plant)	222.0	219.9	25.9	25.8	89.1	39.0	27.3
Lint index	0.9	0.4	17.0	11.2	43.9	1.1	19.6
Seed index	2.9	1.7	17.7	13.5	58.0	2.6	27.1
Ginning outturn (%)	16.0	5.1	10.7	6.0	31.6	3.3	9.0
2.5 % span length (mm)	5.2	5.0	8.0	7.9	96.2	5.8	20.4

combined measure of genetic and environment causes, where as the genetic variability is the only estimate heritable from generation to generation. However, a measure of heritability alone does not give an idea about the expected gain in the next generation but it has to be considered in conjunction with genetic advance. The traits which expressed high heritability and high genetic advance as percentage of mean could be used as a powerful tool in selection process. According to Panes and Sukhatme (1995) such characters were found to be governed by additive genes and had minimum environment influence. The heritabi-lity estimates ranged from 23.5 (internode length) to 89.1% (single plant yield). The high heritability estimates of 89.1% were recorded by single plant yield followed by 2.5% span length (96.2 %) and bundle strength (91.8%) where as the low heritability was observed in inter node length (23.5%).

The high heritability was registered in the traits *viz.,* number of sympodia per plant, single plant yield, seed index, 2.5% span length and bundle strength. The inter node length exhibited low heritability in this investigation. For efficient selection, we cannot completely depend on heritability alone. The combinations of high heritability with high genetic advance will provide a clear base on the reliability of that particular character in selection of variable entries. The genotypic advance as percent of mean for 16 traits ranged from 7.0 to 27.3%. The higher genetic advance as percent of mean was recorded by single plant yield (27.3%) followed by seed index (27.1 %), number of sympodia per plant (26.2 %), number of bolls per plant (21.8 %). The lowest value of 7.0% was observed in the trait days to 50% flowering. High heritability along with high genetic advance was observed in traits *viz.,* number of sympodia per plant, single plant yields, seed index and micronaire value in *G. hirsutum* (Do Thi Ha An et al., 2008). These traits are highly reliable during selection. High heritability combined with moderate genetic advance was found in the 2.5% span length. It was in accordance with of Muhammad et al. (2004). Among the study materials some of the accessions were identified as potential donors for the improvement of different characters (Table 4). The accessions with high mean performance are generally preferred for all the traits except days to 50% flowering, since earliness is the preferred attribute and early flowering was taken into consideration. From the results of the present study, it can be concluded that direct selection can be done for most of the yield attributing traits since it exhibited high genetic variability and high range of variation. A high PCV over GCV for the characters studied indicated that environment influences the expression of these characters under study. High genetic advance, genetic gain and heritability were recorded for number of sympodia per plant, single plant yield, seed index, micronaire value and 2.5% span length indicated that selection can be resorted for the improvement of these characters in the future crop improvement programmes.

Table 4. Potential donors for yield and fibre quality traits.

S/N	Characters	Potential accessions
1	Days to 50% flowering	MCU-7, SVPR-3, TCH-1716, SVPR-2
2	Plant Height	SCS 102, T CH 1715, Okra narrow
3	Inter node length	MCU 5, RAC9740, SCS 102, F-1946
4	No. of Sympodia	PSCL VII, Okra narrow, RHC 1694, SCS 102
5	No. of ovules	NDLH 1588, TCH 1608
6	No. of Bolls	Okra Narrow, G cot-16, F-1946
7	Boll weight(g)	TCH 1715, MCU 13, LRA 5166
8	No. of seeds per boll	NDLH 1588, SVPR 4
9	Seed setting %	MCU 12, MCU 13, KC 3, TCH 1710
10	Single plant yield	MCU 5, MCU 12, Surabhi, SVPR 2, SVPR 4, KC 2, TCH 1715, TCH 1716, TCH 1732, TCH 1734, TCH 1744, G. COT-16, CCH 2117, Pusa 953, ARB 2001, RHC 1694, LH 1961, RB 488, GISV201, MHIS-5, MHIS-7, Sara 2,Giza 1461, Sumangala
11	Lint index	GISV 201, RAC 9544
12	Seed index	SVPR 4, L-752, TCH 1715
13	Ginning Out Turn	RB 488, TCH 1705
14	2.5 % span length	TCH 1608, CNH 152, TCH 1705, RAC 9740, TCH 1732, MCU 5, MCU 13, TCH 1710, TCH 1715, TCH 1716
15	Bundle strength	TCH 1710, TCH 1715, MCU 5, TCH 1732, TCH 1734, TCH 1744
16	Micronaire value	MHIS-7, MCU 7, CCH 2117, NDLH 1588

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African Journal of Plant Science

Full Length Research Paper

Properties, users' assessment and applicability of nine types of taxonomic keys in diagnosing some Nigerian species of *Ocimum* L., *Hyptis* Jacq. and *Ficus* L.

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Nine types of taxonomic keys have been listed from the literature. These are the dichotomous, numerical, multi-access table of identification, punched-card, columnar-diagram, graphical, flow-chart, pictorial-diagram and circular-diagram keys. In this study, each type of key was observed at two levels namely, its format (which is its peculiarity, depicting the general plan, arrangement or organization of its elements) and style (which is one-to-several structural variants in which a key format may be presented in printable form). The format of each type of key along with its representative styles (which determines whether a key is single-access or multiple-access) is illustrated using anatomical features of six Nigerian species of *Ocimum* L., four of *Hyptis* Jacq., and 12 of *Ficus* L. The systematic mode of application and spectrum of usage of each key are discussed and the users' opinions on its usability, efficiency, prospects, users' familiarity and general acceptability are presented. The strengths and limitations of the key formats are also evaluated and discussed on the basis of which taxonomists are charged to consider research efforts towards improving upon the qualities, and devising new key formats with the prospect for better performance.

Key words: Artificial key, diagnostic key, *Ficus,* leaf epidermis, plant identification, taxonomic key, wood anatomy, *Hyptis, Ocimum*.

INTRODUCTION

There are at least 1.4 million species of plants, animals and microorganisms which share the planet earth with man (Asthana and Asthana, 2012). In other to retrieve, utilize, communicate and accumulate information about these organisms, it is necessary for man to identify, name, describe as well as place them into groups that reflect his current knowledge of their evolutionary relationships. These activities make up the discipline of taxonomy (Sivarajan, 2005; Judd et al., 2007). Identification is particularly important since the correct name of an organism is taken as the basic requirement towards having access to its literature. There are two approaches to identify and classify plants. These are the traditional approach and the scientific or formalized approach. The traditional approach (or folk taxonomy) applies the considerable unwritten knowledge of plants in particular localities for plant diagnosis. These unwritten facts have been unconsciously harmonized into a system of ordering plant life so that there is a system of naming them and of referencing particular plants (Olorode, 1984).

In the formalized approach, three options are available to the taxonomist to identify his plant(s) of interest: one,

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by comparing the unknown plant specimen with already identified plants using herbarium specimens and botanical/horticultural gardens; two, by comparing the unknown plant with appropriate photographs, drawings, and/or paintings (Mbuya et al., 1994; Akobundu and Agyakwa, 1997; Nyanayo, 2006); and three, by the use of descriptions and 'keys', which are available in the floras, manuals or any other taxonomic publications relating to the plants of a particular region (Hutchinson and Dalziel, 1963-1972; Keay et al., 1964; Lowe and Stanfield, 1974; Keay, 1989). These devices are called 'artificial' or diagnostic keys because the choice of characters is limited to those that are found to be most reliable and convenient to use, and which are readily available for evaluation. Hence, the arrangement is solely for the convenience of identification (Pankhurst, 1991). These are at variance to the 'natural' or synoptic keys, each characterized by many features of the plants which may not be easily observable in the field such as chromosome features, chemical characters, etc., and whose main thrust is to reflect as close as possible, the scientific classification of the organisms (Pankhurst, 1991).

Every taxonomic key has an easily recognizable starting point that serves as the first notable step in its application. This starting point delimits the scope and coverage of a key, that is, it defines the extent to which it can be used for the purpose of identification. A key also carries a title, which should normally give the user enough information for him to easily select specimens or objects that fall within the scope of the key.

The literature is rich with much, but fragmented pieces of information on types and applicability of artificial keys. Some authors have also variously pointed out the merits and demerits of using some of the available key formats in plant identification, but information on objective comparative assessment of these tools is lacking. This study was therefore meant to collate information about the different types of taxonomic keys with two goals in mind, which were to generate practical tools and a readily available source of information with wider taxonomic and teaching implications from the existing but dispersed and highly essential materials; and to do a users' comparative assessment of the qualities and potentials of these tools for possible refinement towards enhancing the practice of plant identification.

MATERIALS AND METHODS

Literature search

A literature search was conducted on the basics or formats, the styles of presentation and modes of application of the commonly used artificial key systems (Clark, 1938a, b; Herms and Gray, 1944; Osborne, 1963; Keay et al., 1964; Hopkins and Stanfield, 1966; Lowe and Stanfield, 1974; Saldanha and Rao, 1975; Olorode, 1984; Keay, 1989; Jones et al., 1998). The format of a type of key is its general plan, arrangement or organization of its elements that distinguishes it from any other type, e.g. paired contrasting statements, tabulated statements, diagrammatic or numeric representations (Saupe, 2009). The style is one-to-several structural variants in which a known key format may be presented in printable form. The mode of application refers to the prescribed route(s) and means for navigating an artificial key arrangement for an effective identification of a plant specimen.

Attempts were made to trace the origin of diagnostic keys from available information in the literature. The formats, styles and modes of application of the artificial key types encountered in the literature were then described and illustrated using epidermal and wood anatomical features from some species of *Ocimum*, *Hyptis* and *Ficus* in Nigeria. Details of voucher information, data collection and character definition for this purpose are contained in Ogunkunle (1989, 2013) and Ogunkunle and Oladele (1997, 2000, 2008). Some properties common to all the keys were identified and used for their comparative assessment.

Assessment of types of taxonomic keys

A questionnaire was designed to seek the opinions of academic and research workers in systematic botany and related disciplines on the qualities and potentials of some artificial key arrangements. Details about the respondents in the Nigerian Universities and Research Institutes used for the study are contained in Ogunkunle (2006). There were three parts in the questionnaire. The first part gave some background information to the respondents. This included the names of the nine types of artificial keys for consideration, a short description of each format and a brief explanation of its mode of navigation. In addition, the respondents were introduced to six other general qualities of taxonomic keys namely, spectrum of usage (the range of target users), usability, efficiency, weaknesses (or demerits), strengths (or merits) and prospects (or potentials) to enable them respond adequately to the succeeding questions or statements.

The spectrum of usage of a key is here defined as the range of environments in which a key is meant to be effective for the purpose of identification, e.g. field, herbarium, laboratory, etc. Usability is the ease or difficulty with which a key can be navigated for identification by professionals and/or non-professionals. Usability is a function of two factors, namely, the prescribed mode of applying the key and the degree of prior knowledge that is expected in its navigation. Efficiency of key means the ability to accomplish or fulfill the act of identification within a reasonable time limit with the use of the key. The weaknesses or demerits, pertaining to a key, are those pertinent questions surrounding the determination of a plant's identity to which the answers provided by the key are not on the favourable side, e.g. can the key be used for quick confirmation of a suspected identity? is the usage restricted to only a class of workers? Is the format difficult or easy to construct? and so on. If on the other hand, the answers provided to such questions are on the favourable side, we speak of the strengths or merits of the key. The prospects refer to all other possible applications or potentialities of a taxonomic key in plant sciences apart from identification for which it is primarily meant, e.g. the possibility of recognizing relationships among individuals or groups from the key.

The second part of the questionnaire was devoted to seeking personal information about the respondents. This included their places of work, areas of specialization, years of experience, whether they consulted experts or personally handled identification, and how often they required a taxonomic key for the purpose of identification. The third and the final part of the questionnaire asked the respondents to assess the qualities of the nine artificial key formats under investigation. There were two sections in this part of the questionnaire. Section 'A' required the respondents to give their opinions on four qualities of the types of keys over a four-point scale in decreasing order of magnitude as follows: users' extent of

Table 1. Enumeration of the guided statements used to evaluate the properties of some artificial key formats*.

SN.	Guided statement
1	The key shows relationship or affinities among individuals or groups that are included in it (A).
2	It is simple and easy to construct even when the number of specimens involved are many (B).
3	It takes care of realities in the world of plants with provisions made for missing parts; seasonal variations, geographical variations *et cetera* (C).
4	Confirmation of suspected identity can be undertaken by the use of the key within a short time (D).
5	The key has a wide Spectrum of users that is, it is applicable in many environments (E).
6	It is easy to navigate with minimal error as the mode of use is not intricate (F).
7	There is the possibility of a quick retreat once an error is detected (G).
8	The key is concise and handy that is, less voluminous relative to the number of specimens involved; sometimes represented by a single chart (H).
9	Plant Specimen needs not be on ground for its identification to be effected (I).
10	The key can be electronically programmed and made interactive for greater efficiency (J).
11	User is free to choose any character in any order/sequence, thus avoiding the usual rigid (single-entry) format (K).
12	Number of characters applicable is not usually limited, thus embracing the principle of natural classification (L).
13	The key is not cumbersome for storage and handling as no loss of parts may render the key invalid (M).
14	Amount of prior knowledge or vocabulary required for navigating the key is not high such that it may be handled by professionals and non-professionals alike (N).
15	It is applicable for both situations in which there are small and large number of choices in characters and specimens (O).

*The list emanated from the author's conception of expected properties of an ideal key format. The alphabets in parentheses are the codes assigned to the guided statements during the investigation.

familiarity (high, medium, low, nil); usability (very difficult, difficult, easy, unaware); efficiency (high, low, very low, unaware); and acceptability to the user (very high, high, low, very low). Section 'B' of the third part of the questionnaire presented the respondents with fifteen guided statements for evaluating the merit level of each of the key formats. The statements were all drawn out of the investigator's conception of an ideal tool for plant identification, each being a desirable comment on one definite task (Table 1).

The respondents were made to react to each statement over a two-point scale, 'agree' or 'disagree'. The questions and/or statements in both sections 'A' and 'B' were independent of one another. As such, a respondent who, for instance was not familiar with a type of key, going by his response in section 'A' might still assess its efficiency or usability from the knowledge of the background information given about the key in the introductory part of the questionnaire. Moreover, such a respondent could adequately assess the merit level of the key format by responding to the guided statements in section 'B'. A total of 56 questionnaires were administered and analyzed.

Data analysis

The four responses to each of the questions in Part IIIA were assigned weight values 4, 3, 2 and 1, respectively. The mean of all responses to a question was then computed and taken as the representative opinion of all the respondents in that question. With regards to the analysis of the data in Part IIIB, the percent number of respondents in agreement with each of the fifteen statements for all the key types was first computed and recorded. These percent values were then ranked into five class intervals, each with a size of 20 and assigned a weight value of points in increasing order of magnitude as follows: 1-20% = 1 point; 21-40% = 2 points; 41 -60% = 3 points; 61-80% = 4 points; and 81 – 100% = 5 points. Thereafter, for each type of key, the weighted mean of the percent affirmative responses was computed using the formula adapted from Spiegel (1992) as follows:

$$\text{Weighted mean} = \frac{\sum_{i=1}^{n} PiWi}{\sum_{i=1}^{n} Wi} \quad (1)$$

Where, P_i = percent affirmative response to a statement of merit assessment; W_i = weight value of points attached to each % response; n = number of guided statements adopted for assessment of a key.

The weighted mean of percent responses was taken as the index of merit for each type of key and the key format with the highest index was taken to have satisfied a substantial number of expectations in an ideal artificial key format.

RESULTS AND DISCUSSION

Origin and types of taxonomic keys

There are strong indications from the literature that taxonomic keys (albeit unpublished) have been in use for several hundreds of years, the two basic types being the single-access and the multiple- or multi-access keys (Pankhurst, 1991). These two basic types have however undergone some radical transformations in the hands of taxonomists, evolving into nine different kinds currently in use, that is, dichotomous (Osborne, 1963; Fahn, 1974; Olorode, 1984; Sharma,1993), numerical (Keay et al., 1964; Lowe and Stanfield, 1974; Olorode, 1984), multi-access table of identification (Jones et al., 1998), punched-card (Clarke, 1938a, b; Saldanha and Rao,

Table 2. Dichotomous (bracketed) key to some Nigerian species of *Ocimum* based on leaf and stem stomatal characteristics.

1a	Diacytic, anisocytic and amphidiacytic stomatal complex types found on the lamina; petiole and stem epidermises are also with various types of stomatal complexes……..2
1b	Only diacytic and amphidiacytic stomatal complex types are found on the lamina; petiole and stem epidermises lack stomata/ stomatal complexes……*O. Suave*
2a	Staurocytic and anisocytic stomatal complex types are observable on the stem epidermis along with other types; anisocytic type also occurs on the petiole……3
2b	Diacytic, paracytic and paradiacytic stomatal complex types found on stem epidermis, never staurocytic nor anisocytic……*O. lamiifolium*
3a	Paradiacytic stomatal complexes occur on the stem epidermis; and also on the petiole unless the leaf is sessile ……4
3b	Staurocytic, anisocytic and cyclocytic complexes are observable on the stem, never paradiacytic; if leaf is petiolate, only diacytic, anisocytic and staurocytic stomatal complex types are found……*O. gratissimum*
4a	Stem epidermis, with paracytic stomatal complex; staurocytic stomatal complex, absent on the petiole……*O. basilicum*
4b	Stem epidermis, without paracytic stomatal complex; petiole if present, has staurocytic stomata……5
5a	Diacytic, anisocytic, paradiacytic and staurocytic stomatal complexes are observable on the petiole……*O. canum*
5b	Leaves sessile (that is, without petiole)……*O. Irvinei*

Table 3. Dichotomous (bracketed) key to some Nigerian Species of *Hyptis* based on leaf and stem epidermal characters*.

1a.		Leaf adaxial epidermal cell walls are straight but those of abaxial surface are wavy……2
1b.		Leaf adaxial epidermal cell walls are not straight; rather curved or wavy……3
	2a.	Surfaces of lamina bear only diacytic and amphidiacytic stomatal complexes; petioles bear diacytic, anisocytic and staurocytic stomatal omplexes……*H. lanceolata*
	2b.	Surfaces of lamina bear anisocytic stomatal complexes along with diacytic and amphidiacytic types; petioles also bear paradiacytic complex in addition to diacytic, anisocytic and staurocytic types……..*H. spicigera*
	3a (1).	Leaf adaxial epidermal cell walls are curved (not wavy); stems bear diacytic, anisocytic, staurocytic and paradiacytic complexes ……*H. pectinata*
	3b.	Leaf adaxial epidermal cell walls are wavy; paracytic stomatal complex type are found on stems along with diacytic, anisocytic, staurocytic and paradiacytic complexes…….. ……*H. suaveolens*

*Number in parentheses immediately after couplet number indicates the couplet that sent user to the current couplet.

1975), columnar diagram (Herms and Gray, 1944), graphical (Dawkins, 1951; Hopkins and Stanfield, 1966), flow- chart (Jones et al., 1998; Monteith, 2000), pictorial diagram (Harris and Harris, 2001) and circular diagram (Herms and Gray, 1944) keys, which are illustrated in Tables 2 to 8 and Figures 1 to 7. Preceding these transformations and publications, identification tables were said to have been presented by Richard Waller in 1689 to his assembled colleagues at a meeting of the Royal Society in the form of image-based dichotomous keys for the herbs of Britain (Griffing, 2011).

The single-access key, also called sequential or analytical key is strictly speaking, a family of types of taxonomic keys, each characterized by the sequence and structure of identification steps that are fixed by the author of the key. If the entire key consists of exactly two choices at each branching point, the key is called dichotomous, and is polytomous if there are more than two choices, the former being the more commonly applied format. Some of the other key formats that have evolved from the single-access key arrangement include columnar diagram, graphical, flow-chart, pictorial-diagram and circular-diagram keys (Figures 3 to 7).

There is evidence to show that the origin of single-access keys, and perhaps, of any taxonomic key, could be in the age-long decision tree or tree diagram used in operations research to identify a strategy most likely to reach a goal; a decision support tool that uses a tree-like graph of decisions and their possible consequences including chance, event outcomes, resource costs and utility (Yuan and Shaw, 1995). The dichotomous key gained popularity in botanical (and zoological) works, a form in which single-access keys were first published in 1672 by Morison in his *Plantarum Umbelliferarum*

Table 4. Leaf epidermis-based dichotomous key to some Nigerian species of *Ficus* using an integral hierarchical format*.

1a.	Prickle-like hairs present on both adaxial and abaxial leaf surfaces	……Subgenus *Ficus*
1b.	Prickle-like hairs, absent on both adaxial and abaxial leaf surfaces	……2
2a.	Adaxial epidermal cells, variable in shape and intermixed, including isodiametric, tabular, square and irregular	……...Subgenus *Urostigma*
2b.	Adaxial epidermal cells, isodiametric in shape	……..Subgenus *Sycomorus* (3)
3a(2).	Bulbous glands, unicellular and circular, found in-between and on the veins of abaxial leaf surface; polycytic stomatal complexes with 4 and with 5 subsidiary cells, found on abaxial surface	……...4
3b.	Bulbous glands absent; instead, flask-shaped glands are present in-between and on the veins of abaxial surface; polycytic stomatal complexes with 6, with 7, and with 8 subsidiary cells, found on abaxial surface; those with 3, 4 and with 5 subsidiary cells are absent	……*F.vallis-chondae*
4a (3).	Abaxial epidermal cells, isodiametric in shape, with curved anticlinal walls; stomata located in-between the veins only; stomatal complex types with 4 and with 5 subsidiary cells are found; mean stomatal density/unit of veins, less than 100	……..*F. mucuso*
4b.	Abaxial epidermal cells, irregular in shape, with wavy anticlinal walls; stomata located in-between and on the veins; anisocytic stomatal complexes (that is, with 3 subsidiary cells) are found along with those with 4 and with 5 cells; mean stomatal density/unit of veins, about 168	….....*F. Sur*

*Number in parentheses immediately after couplet number indicates the couplet that sent user to the current couplet, while that after the couplet statement indicates the next couplet to which the user is directed.

Table 5. Wood and leaf anatomy-based dichotomous key to some Nigerian species of *Ficus* using separate hierarchical (or multi-part) format.

Part A:	Subgenera of *Ficus*	
1a.	The characteristic ray type observable in wood TLS is convex type II, occurring in two forms that is, the thicker and the thinner; in the thicker rays, the cells are round, relatively thin-walled, conspicuously tiny and numerous per ray, with few of the cells enlarged; in the thinner rays, the cells are variable in shape but usually angular or round and thin-walled with none being enlarged	……..Subgenus *Ficus*
1b.	The characteristic ray type combination of the convex type II in wood TLS as described in 1a is not applicable	……2
2a.	Convex type I ray, observable in wood TLS	……Subgenus *Sycomorus*
2b.	Convex type I ray, absent in wood TLS	……Subgenus *Urostigma*
Part B:	Sections of the subgenus *Urostigma*	
1a.	Wood substance, variable in colour but frequently whitish or yellowish- white; heartwood/sapwood boundary is distinct; mean density of epidermal cells on abaxial leaf surface, relatively low (between 300 and 400/mm^2), never up to 600/mm^2	……Section *Galoglychia*
1b.	Wood substance, pinkish-white; heartwood/sapwood boundary indistinct; mean density of epidermal cells on abaxial leaf surface, very high (up to 800/mm^2)	……Section *Urostigma*
Part C:	Subsections of the section *Galoglychia*	
1a.	Glands, present on abaxial leaf surface	……2
1b.	Glands, absent on abaxial leaf surface	……3
2a.	Scales or other sessile glands on abaxial leaf surface are unicellular and circular; rarely oval and if oval, are also unicellular	……Subsection *Caulocarpae*
2b.	Scales or other sessile glands on abaxial leaf surface are multicellular and oval; else, such glands are absent	……Subsection *Chlamydorae*
3a.	Anisocytic stomatal complex type is observable along with the polycytic (4 or more subsidiary cells) types; mean stomatal density/mm^2 of leaf surface is very high (above 200)	……...4
3b.	Anisocytic stomatal complex type, absent on leaf surface; mean stomatal density/mm^2 of leaf surface is low (less than 40)	……Subsection *Crassicostae*
4a.	Epidermal cell walls on abaxial leaf surface are curved; glands occur on adaxial epidermis; usually bulbous and circular	……Subsection *Galoglychia*
4b.	Epidermal cell walls on abaxial leaf surface are straight; glands are absent on adaxial epidermis	……Subsection *Cyathistipulae*

Table 5. Contd.

Part D:	Species of the subsection *Caulocarpae*	
	1a.	Dumb-bell shaped (or constricted) rays are found in wood TLS……...2
	1b.	Dumb-bell shaped rays are absent in wood TLS..……..3
	2a.	Vessels in wood TS occur as solitary units and in radial chains of up to 7; uniseriate rays and giant rays are found in TLS……*F. umbellata*
	2b.	Vessels in wood TS occur as solitary units and in radial chains of maximum of 2; uniseriate and giant rays are absent in TLS……..*F. ovata*
	3a.	Rays in wood TLS occur as heterocellular types only……...*F. ottonifolia*
	3b.	Rays in wood TLS occur as both homocellular and heterocellular types……*F. polita*

Table 6. Leaf epidermis-based dichotomous (indented) key to some Nigeria species of subsection *Caulocarpae* (Section *Galoglychia*, subgenus *Urostigma*, genus *Ficus*).

1a.	Bulbous glands found on one or both of the adaxial and abaxial leaf surfaces……2	
	2a.	Bulbous glands observable on abaxial epidermis only; scales also present on adaxial surface only; number of radiating basal cells of glands on abaxial leaf surface 10-14; types of basal cells of the glands on adaxial surface consist of the unmodified and the radial type II……*F. ottonifolia*
	2b.	Bulbous glands and scales, found on both leaf surfaces; number of radiating basal cells of glands on abaxial leaf surface 7 or 8; types of basal cells of the glands on adaxial surface consist of the unmodified and the radial type I……...*F. polita*
1b.	Bulbous glands, absent on both leaf surfaces……3	
	3a.	Epidermal cell walls on abaxial leaf surface are irregular in shape; scales and club-shaped glands, found on both adaxial and abaxial epidermises; mean stomatal density is high, being 200/mm^2 of leaf surface and 89/unit of the veins……*F. ovata*
	3b.	Epidermal cell walls on abaxial leaf surface are tabular in shape; scales are found only on adaxial while club-shaped glands are found only on abaxial surface; mean stomatal density is low, being 87/mm^2 of leaf surface and 23/unit of the veins……*F. umbellata*

Distributio Nova (Stace, 1991), and in 1778 by Jean Baptiste-Larmark (Saupe, 2009; Griffing, 2011).

As much as possible, a single-access key is made to start with characters that are reliable, convenient and generally available throughout most of the year. But this is often impossible to achieve for all the taxa in a key. Polyclave, random-access or multiple-entry key is the identification key which overcomes this problem along with that which has to do with fixed sequence of identification steps in the single-access keys. The flexibility in multi-access key lets the user undertake character choices in the key according to the state of the specimen being identified and the prevailing circumstances such as seasonal variations, and field or laboratory situations. He is thus free to choose the set of characters that are convenient to evaluate for the plant to be identified. Some of the states or condition which may arise include occurrence of important characters that are difficult to observe, presence of some characters that may likely be misinterpreted, a situation when a single character may be unreliable in isolation, and when a part of a specimen is missing or seems abnormal. The printable forms of multi-access keys include numerical, tabular, matrix, formula styles, the pictorial diagram and the punched- cards (Virtual Field Herbarium, 2000; Key-to-Nature, 2010; Tables 7 to 8; Figures 1 to 2)

Structure and applicability of some types of taxonomic keys

A simplified dichotomous key is illustrated in Tables 2 and 3, both of which refer to the bracketed or linked variant of the key format.

As it is in all single-access keys, there is only one point of entry in each of the two keys in Tables 2 and 3. In order to identify a specimen, there is a step-wise perusal of the contrasting statements in the key, starting from the first pair (or couplet), resulting in the acceptance of one (a lead) and the rejection of the other with respect to the features of a plant specimen on hand. The acceptance of one of the first pair of statements leads to the second pair in the series and so on. This exercise will eventually terminate when, instead of an earlier accepted lead pointing to a further pair of couplet now points at a name. Such name is generally taken as the identity or correct name of the 'unknown' plant (Tables 2 and 3).

The bracketed or linked style in a key makes polytomous statements applicable since the leads within a couplet follow each other contiguously (Table 3). More-

Table 7. Wood anatomy-based numerical key to some Nigerian species of *Ficus.*

Diagnostic characters									
1. Oval-shaped vessels in wood T.S									
2. Tylose in the vessels									
3. Broad rays in TS of wood									
4. Convex rays of Type III in wood TLS									
5. Occurrence of heterocellular rays of type A									
6. Homocellular rays									
7. Uniseriate rays									
8. Biseriate rays									
9. Maximum number of vessels in radial chains in TS is up to 6 or more									
Codes and species									
1	2	3	0	0	0	7	8	0	*F. lutea*
1	0	3	4	0	6	7	8	9	*F. exasperata*
1	0	3	4	0	6	7	8	0	*F. mucuso*
1	0	3	0	5	6	7	8	9	*F. umbellata*
1	0	3	0	5	6	7	8	0	*F. thonningii*
1	0	3	0	5	6	0	0	9	*F. populifolia*
1	0	3	0	0	6	7	8	0	*F. polita*
1	0	3	0	0	6	0	8	0	*F. ovata*
1	0	3	0	0	0	7	8	0	*F. ottonifolia*
1	0	0	0	0	6	7	8	0	*F. ingens*
1	0	0	0	0	0	7	8	0	*F. natalensis*
0	2	3	0	5	6	0	0	0	*F. sur*

Table 8. A wood anatomy-based multi-access table of identification for some Nigerian species of *Ficus*.

Diagnostic character	Species											
	LUT	**NAT**	**MUC**	**OVA**	**ING**	**UMB**	**OTT**	**POL**	**THO**	**EXA**	**SUR**	**POP**
OVV	2+	2+	5+	3+	3+	2+	2+	3+	5+	3+	-	5+
ROV	5+	3+	3+	5+	5+	3+	3+	5+	4+	4+	6+	4+
TYL	3+	-	-	-	-	-	-	-	-	-	4+	-
NAR	5+	6+	4+	4+	6+	5+	5+	3+	3+	3+	2+	5+
BRR	3+	-	4+	4+	-	3+	3+	5+	5+	5+	5+	2+
CRI	-	-	-	-	-	-	-	-	-	-	+	-
CII	+	6+	+	+	+	+	6+	6+	6+	+	+	+
CIII	-	-	+	-	-	-	-	-	-	+	-	-
DBR	3+	-	3+	2+	3+	3+	-	-	-	2+	2+	3+
HCR	-	-	3+	2+	3+	3+	-	3+	3+	3+	3+	3+
HRA	3+	-	-	-	-	3+	-	-	3+	-	5+	4+
HRB	5+	6+	5+	5+	5+	5+	6+	5+	5+	5+	3+	4+
USR	2+	3+	3+	-	3+	5+	3+	3+	2+	2+	-	-
BSR	3+	4+	3+	3+	3+	3+	3+	3+	3+	3+	-	-
MSR	5+	4+	5+	5+	5+	5+	3+	5+	5+	5+	6+	6+
GIR	+	-	-	+	+	-	-	-	-	-	+	+
PSV	73	86	61	78	63	69	68	72	69	53	34	38

LUT = *F. utea*, NAT = *F. natalensis*; MUC = *F. mucuso*; OVA = *F. ovata*; ING = *F. ingens*; UMB = *F. umbellata*; OTT = *F. ottonifolia*; POL = *F. polita*; THO; *F. thonningii*; EXA = *F. exasperata*; SUR = *F. sur*; POP = *F. popufolia*. OVV = oval-shaped vessel in wood TS; ROV = round- shaped vessels; TYL = presence of tyloses in vessels; NAR = rays narrow in wood TS; BRR = rays broad in wood TS; CRI = convex ray type I, CRII = convex rays of type II; CRIII = convex rays of type III; DBR = dumb-bell shaped rays; HCR = homocellular rays; HRB = heterocellular rays of type B; USR = uniseriatte rays; BSR = biseriate rays; MSR = multiseriate rays; GIR = giant rays; PSV = percent (frequency) of solitary vessels in wood TS; = absent/not applicable; + = observable but frequency is unknown; 2+ = 1-9% observation; 3+ = 10-39% observation; 4+ = 40-59% observation; 5+ = 60-99% observation; 6+ = 100% observation.

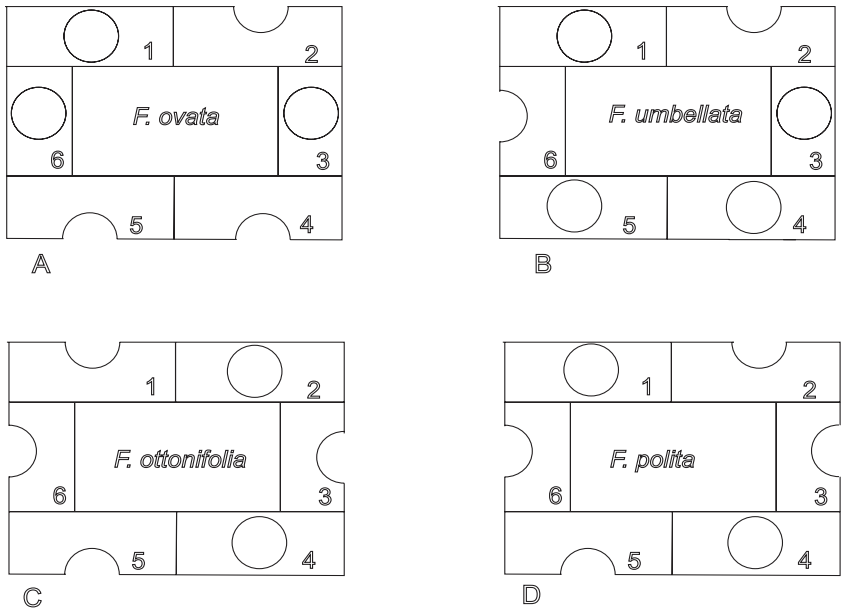

Figure 1. Diagrams of edge-punched cards for identification of some Nigerian species of *Ficus* (subgenus *Urostigma*, section *Galoglychia*, subsection *Caulocarpae*) based on wood anatomy. A, B, C, D represent the taxa; in taxon A (*F. ovata*), characters 1, 3, and 6 are applicable (intact holes) while characters 2, 4 and 5 are not applicable (holes are clipped off). List of the diagnostic characters:1, Homocellular rays found along with heterocellular rays in TLS; 2, Only heterocellular rays are found in wood TLS; 3, Dumb-bell (constricted) rays are present in wood TLS; 4, Uniseriate, biseriate and multiseriate rays are found in wood TLS; 5, Giant rays (more than 20 times taller than wide) are present in TLS; 6, Vessels in TS occur in solitary units and radial chains of maximum of 2.

over, linked dichotomous keys make it possible to have keys for different taxonomic levels possible (for example keys to genera and species) either within a single data matrix (integral hierarchical keys) (Table 4), thus allowing the user to change the level during iden-tification without losing information, or by linking a key to higher categories of taxa with other keys to allow iden-tification to proceed to lower taxonomic level (separate hierarchical keys) (Table 5) (Dallwitz et al., 2009). Another style of presenting dichotomous keys is in the indented (yoked or nested) form (Table 6), in which each successive couplet is indented with an equal distance from the left margin and the indentation increases with increasing couplets. Moreover, all the units in respect of one lead of a couplet are keyed out first before proceed-ing to the other half (Table 6).

Although, most dichotomous keys are simplified, that is, they follow the decision trees or binary search trees but to improve their usability and reliability, they may incur-porate reticulation, changing the tree structure randomly into a directed acyclic graph. Reticulation is a practice whereby different branches of the tree are connected to improve error tolerance and identification success (Osborn, 1963) in such a way that multiple paths lead to the same result. There are two forms of reticulation. The first is terminal reticulation in which a single taxon (or next-level key) is keyed out in several locations in the key after having scored the attributes of the taxa from different perspectives. The second is inner reticulation, in which a couplet with further leads can be reached through more than one path. Reticulations generally improve the usability of a key but may also diminish the overall probability of correct identification averaged over all taxa (Osborn, 1963; Payne, 1977; Payne and Preece, 1977).

Table 7 shows a variant of numerical key. For the purpose of identification, a nine-digit code is normally compiled for the ‘unknown’ specimen with regards to the nine-characters. The resulting code is then compared with the list of code in the key. The user is also free to evaluate and make use of one character at a time, and in the order he prefers for the process of taxa elimination. This arrangement is the simplest form of the numerical key format which assumes that no two characters used are mutually exclusive. In many cases however, a given pair of characters may be mutually exclusive. In that case, the figures representing the two characters can never occur together in a taxon. The figures in the code are usually separated from the neighbouring digits by a period. It is also possible to include more than nine characters. In such a case, those digits representing the first nine characters are separated from the second by a

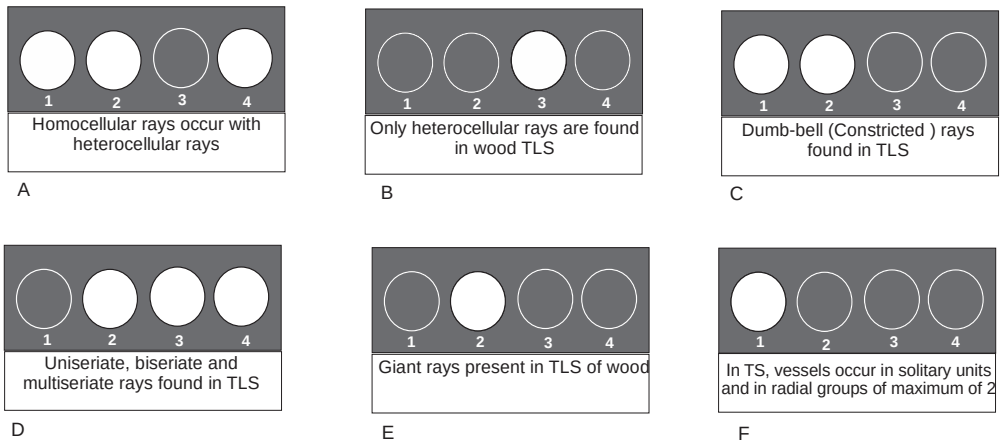

Figure 2. Diagrams of body-punched cards for identification of some Nigerian species of *Ficus* (Subgenus *Urostigma*; Section *Galoglychia*; Subsection *Caulocarpae*) based on wood anatomy. A, B, C, D, E, F = diagnostic characters; 1= *F. ovata*; 2 = *F. umbellata*; 3 = *F. ottonifolia*; 4 = *F. polita*. In Taxon 1 (F.ovata), characters D and E are not applicable (represented by circles) while characters A, B, C and F are applicable (punched into holes).

<table>
<tr><th>O. gratissimum</th><th>O. canum</th><th>O. basilicum</th><th>O. irvinei</th><th>O. lamiifolium</th><th>O. suave</th></tr>
<tr><td>Stalked glands on leaf transection are capitate with peltate head; never club-shaped.</td><td>Stalked glands on leaf transaction are club-shaped (i.e capitate with spherical or oval head)</td><td rowspan="2">Paracytic (Laterocytic) stomatal complex found along with other types on stem epidermis; head of glands on leaf surface with 1, 2 or 4 cells</td><td rowspan="2">Leaf surface cuticle, very thick and conspicuous (up to 5.0µm), head of glandular trichomes, always unicellular</td><td rowspan="2">Pardiacytic stomatal complex absent on the leaf blades; stomata occur at the same level with epidermal cells (i.e neither elevated nor sunken)</td><td rowspan="2">Paradiacytic stomatal complex found on the leaf blades; stomata are elevated or raised above other epidermal cells</td></tr>
<tr><td colspan="2">Staurocytic and anisocytic stomatal complexes are found but paracytic (laterocytic) type is absent on stem epidermis; head of glands on leaf surface with 2 or 4 cells; never unicellular</td></tr>
<tr><td colspan="3">Leaf surface cuticle; thin (less than 2.0µm); number of cells in glandular trichome head sometimes up to 4</td><td colspan="3"></td></tr>
<tr><td colspan="4">Leaves amphistomatic; sessile glands absent on adaxial leaf surface</td><td colspan="2">Leaves hypostomatic; sessile glands present on adaxial leaf surface</td></tr>
</table>

Figure 3. The columnar diagram key to some Nigerian species of *Ocimum* based on leaf and stem epidermal features. The point of entry into the key is the two boxes at the bottom of the columns.

colon, the second nine digits are also separated from the third by another colon, and so on.

Table 8 is a multi-access table of identification. In order to apply this key, the user first evaluates the 'unknown' specimen on the basis of those characters available to him from the list of all the characters in the table, one after the other and in accordance with the definitions attached thereto. Next, with the consideration of each

		Stellate hairs on the costal areas of leaf surface; sessile glands on TS of petiole if present, are normal (not sunken)		No stellate hairs on leaf surface; sessile glands on TS of petiole are sunken in pits	
		Type I stellate hairs on leaf surface	Type II stellate hairs on leaf surface	Stalked glands found on both leaf surface	Stalked glands absent on both leaf surfaces
Anticlinal walls of epidermal cells (Adaxial) are curved or wavy; Stomata on abaxial surface are normal (not raised)	Stomata on stem are scattered; not linearly arranged	*H. pectinata*			
	Stomata on stem are arranged linearly along the stem axis		*H. suaveolens*		
Anticlinal walls of epidermal cells (Adaxial) are straight; Stomata on abaxial surface are elevated (raised) above other epidermal cells	Anisocytic stomata absent on abaxial leaf surface				*H. lanceolata*
	Anisocytic stomatal complex found on abaxial leaf surface			*H. spicigera*	

Figure 4. A graphical key for diagnosing four Nigerian species of *Hyptis.* Type I stellate hair = simple uniseriate hairs occur in tufts; Type II stellate hairs = simple uniseriate hairs occur in tufts with stalked glandular trichomes; anisocytic stomatal complex = stomata with three subsidiary cells.

character in turn, some specimens in the table are eliminated or ignored while the others are selected. Each successive step narrows down the choices of specimens until only one specimen is left, its name being the identity of the 'unknown' plant.

Figures 1 and 2 are the two types of punched-card system commonly used in plant identification. These are named 'species-per-card' and 'character-per-card' types, respectively (Virtual Field Herbarium, 2000). The first step in the use of the edge-punched or 'species-per-card' key (Figure 1) is to align all the cards properly. Considering one character at a time, the user observes the 'unknown' specimen and for each character that is applicable to the specimen, a long needle is inserted through the hole representing that character. The cards are then gently shaken to allow those (cards) that are not applicable to the specimen to fall off the needle (those with clipped hole for the character). This process continues, with successive choice of characters in the desired sequence until only one card remains dangling on the needle. The name of the taxon on this card represents the identity of the plant.

The taxonomist applies the body-punched, that is, 'character-per-card' key (Figure 2) by first identifying and setting aside all the cards whose characters are observed by him on the 'unknown' plant specimen. From these, he selects a few cards at a time, says five, aligns them properly and holds them against a source of light. If more than one hole allows the light to pass through, he selects more of the remaining cards, aligns them with the earlier selected ones and repeats the exercise. At the time when only one of the holes allows the light to pass through, the

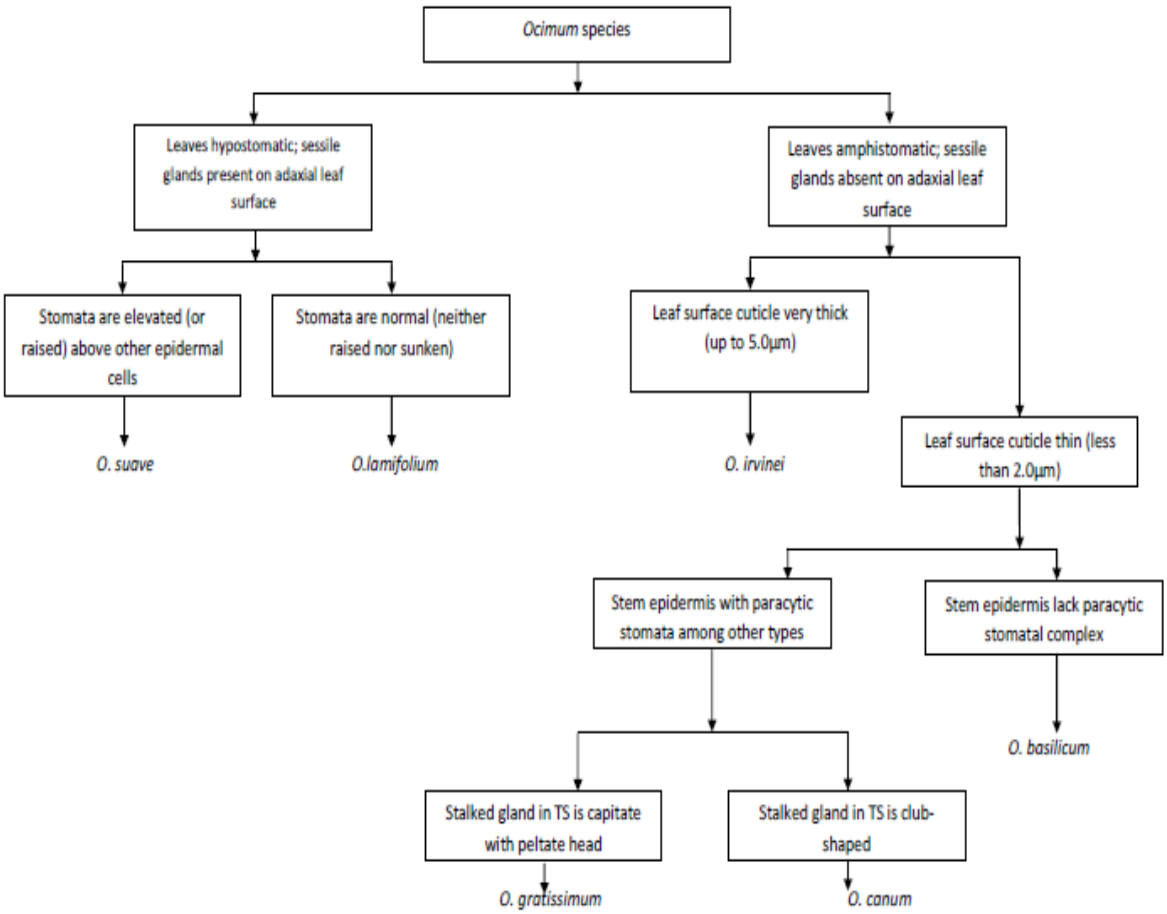

Figure 5. A flow chart key for diagnosing six Nigerian species of *Ocimum.*

identification process is completed. The name of the taxon whose hole allows the light to pass through represents the identity of the plant. The 'species-per-card' type has the advantage that species can be added as time goes on; and additional information in form of notes or images about species can be put on the card.

The punched-card system can be satisfying to use, like a game, especially with large sets of species. One problem however, is that each hole can only represent a 'yes' or a 'no'. So in multistate characters, each state has to be considered as a unit character and assigned a hole. This problem is better appreciated with the observation that only a limited amount of space can be available for characters on a card. Hence, 'species-per-card' packs are best when the number of characters is few. To a certain extent, this problem has been addressed with the use of only one card per character in the method developed by Sinnott (1982) provided there are ten or fewer character states (in a 78 taxa variation) or five or fewer states (in a 156 taxa variation). Another disadvantage is that the loss of a single card may render the pack almost useless, coupled with the requirement that the cards (particularly in the character-per-card arrangement) must be carefully ordered after each use to permit relocation. In order to prevent the pack becoming dispersed, The Virtual Field Herbarium (2000) has recommended some form of loose binding.

Figure 3 shows a columnar diagram key. In order to apply this key format, the user starts by considering first, the statements in the point of entry, that is, the two contiguous bars at the bottom. The acceptance of the statement in one box immediately restricts the user to one of the blocks as the possible provider of the plant's identity. Next, he proceeds to consider the two opposing statements in the two bars on top of the former. Again, this step makes him to drop one and accept the other statement, whose bar serves as a lead to other bars or columns above it with opposing statements. This process continues until only one column is eventually selected. The name at the top of this column gives the identity of the unknown plant.

The graphical key is illustrated in Figure 4. In identifying a plant specimen, one may choose to start with the horizontal or top statement bars, which enable the user to systematically ignore some columns and select others in line with the features of the specimen to be identified.

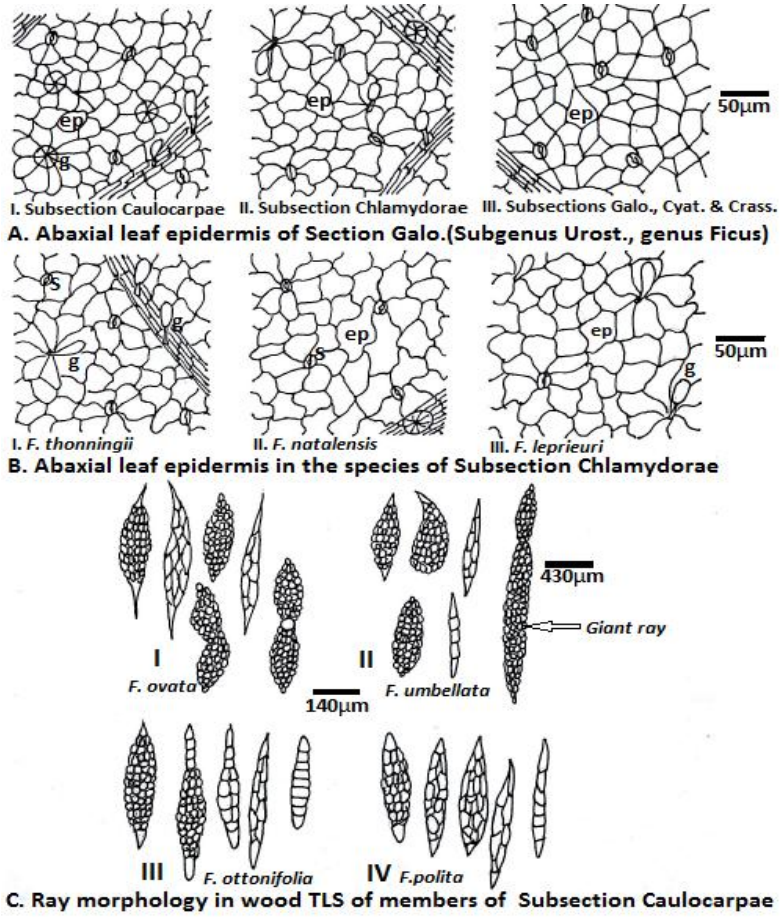

Figure 6. A pictorial diagram key for identification of some Nigerian species of *Ficus*. The key component 'A' distinguishes between the subsections in the section *Galoglychia* [I and II, stomatal complexes (STC) are predominantly with 5, 6, and 7 subsidiary cells (SUC); in I, scales are all in-between the veins; in II, scales, if present, are found on the veins, but other types of glands may occur; III, stomata are predominantly with 3 or 4 subsidiary cells, and glands and scales are absent in-between and on the veins]; Component 'B' separates three species of subsection *Chlamydorae* [I, STC are with 4, 5 and 6 SUC; abaxial epidermal cell walls (ABE) are slightly curved; II, STC also include those with 7 SUC; ABE are wavy; III, STC with 4 SUC are absent; ABE are curved]; and component 'C' diagnoses four species of subsection *Caulocarpae* , in each case the frequency of types of rays shown decreases from left to right.

This procedure eventually leads to a single column of boxes (the possible identities of the specimen). The next task is to pinpoint one out of the selected boxes in which the specimen is located. In order to accomplish this task, the vertical statement bars on the left hand side are followed in a similar way until a single row of boxes is selected. Now the box, which is located at the meeting point of the two axes, should contain the name of the specimen. The graphical key format derives its name from the characteristic vertical and horizontal axes approach for identification.

Figure 5 shows a flow chart key which follows the strict system of two or multiple choices of characters. The choices are laid down in the form of a flow chart, that is, a tree-like scheme of rectangular statement boxes and arrowed branches, which allows easy cross-checking of options. The arrows should be followed strictly after making a choice from the guiding statements. This step normally leads to two or more other statement boxes that require yet another round of choices and so on until eventually the arrow points at a name with which the specimen is identified.

In the pictorial diagram type of key (Figure 6), a set of annotated diagrams or photographs of some observable features of plants are displayed in such a way as to allow two or more choices at a point in time. The choices are

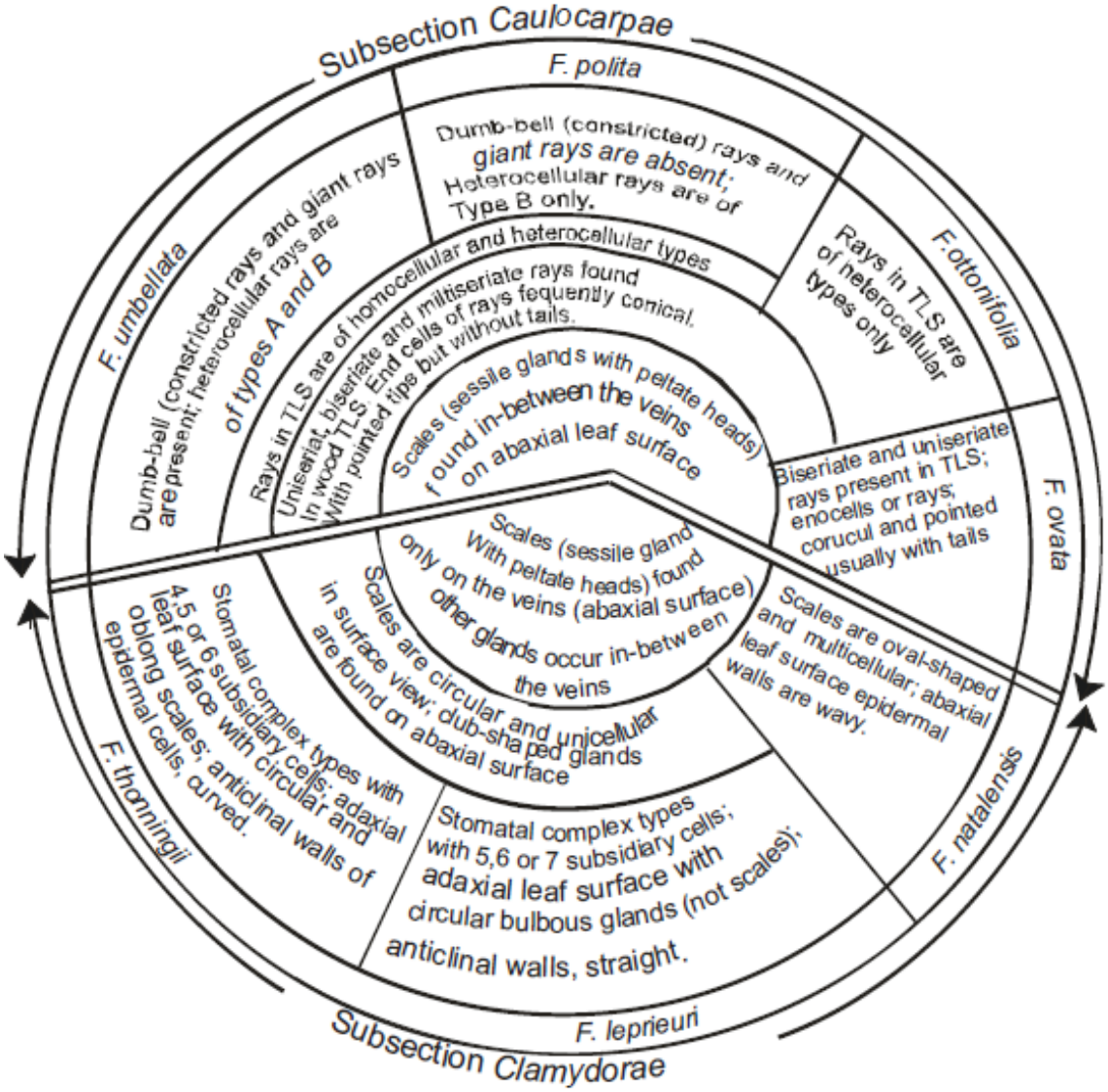

Figure 7. A Circular-diagram key for separating two subsections (*Caulocarpae* and *Chlamydorae*) of the Section *Galoglychia* (Subgenus *Urostigma* of *Ficus*) and for diagnosing some Nigerian species of the two subsections. Types of rays in TLS: heterocellular type A = rays that are pointed at one end; type B = rays that are pointed at both ends; giant ray = ray which is more than twenty times taller than wide.

laid down in a tree-like form for easy comparison with the specimen on hand. The acceptance of one of these illustrations along with its annotation leads the user, with an arrow to one or more other illustrations for yet another round of cross-checking and a choice. As these processes continue, the arrow eventually leads to a diagram or photograph attached with a name (Fishel and Kendig, 2003). This name is usually taken as the identity of the unknown plant specimen. This type of key can be viewed and applied more or less in the same way as the flow chart key, in which the rectangular boxes of statements are replaced by the illustrations (Fishel and Kendig, 2003) (Figures 5 and 6).

A circular diagram key is shown in Figure 7. For the purpose of identification, the user first considers the characters in the two contiguous compartments at the centre of the circle, proceeding outwards, and following the alternative choices.

The adoption of one of the first two statements or compartments restricts the user to a few of the taxa as the possible identities of the unknown plant. Subsequent steps reduce the number of possibilities until only one name is achieved, referring to the identity of the specimen. Moreover, by proceeding inward from the circumference, the distinctive characters of any taxon in the key may by compiled and this makes it possible for one to confirm the identity of a specimen for which a name has been suspected (Figure 7).

Table 9. An artificial key to diagnose the types of uniseriate trichome feet in some Nigerian species of *Ocimum* and *Hyptis.*

1a.		Trichome foot (i.e mode of attachment of a simple trichome to the surface of a plant organ as viewed in transection of the organ) is developed on one or two epidermal cells which are not distinguishable in size, shape or position from the other epidermal cells…….. ……..Unmodified foot (Figure 8 A-C)
1b.		Trichome foot (i.e mode of attachment of a simple trichome to the surface of a plant organ as viewed in transection of the organ) is developed on one or more epidermal cells that have been transformed in a way and distinguished in size, shape and/or position from neighbouring epidermal ells……..Modified foot (2)
	2a.	Tichome is seated on a single epidermal cell that is enlarged more than the other epidermal cells……Modified Type I foot (Figure 8D)
	2b.	Trichome is seated on two or more cells that are raised (or elevated) above the level of the other epidermal cells……3
		3a. Foot consists of two contiguous cells positioned on top of other epidermal cells……Modified Type II foot (Figure 8E)
		3b. Foot consists of some tiers of many contiguous cells positioned on top of other epidermal cells…..Modified Type III foot (Figure 8F)
	2c.	Trichome is seated on a few cells that are radially elongated……4
		4a. Foot consists of elongated cells radiating away from other epidermal cells to the direction of the bottom of the trichome……..Modified Type IV foot (Figure 8 G).
		4b. Foot consists of elongated cells radiating away from the bottom of the trichome to the direction of the other epidermal cells….. ……Modified Type V foot (Figure 8 H).

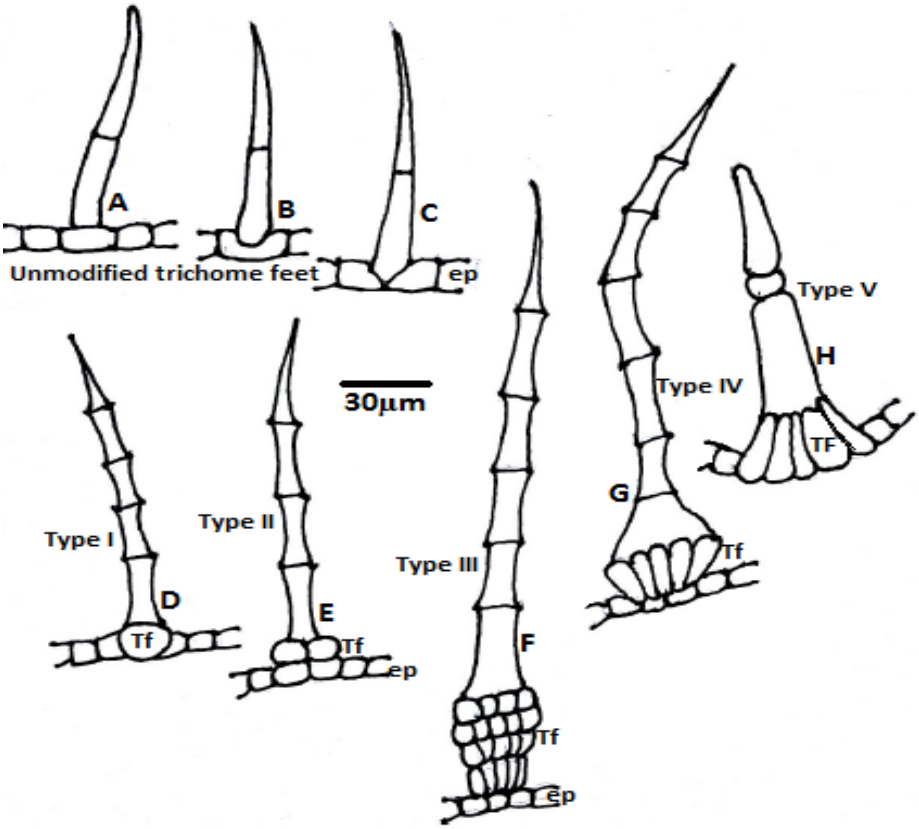

Figure 8. Simple uniseriate trichomes and their feet observed in some Nigerian species of *Ocimum* and *Hyptis*. A-C = unmodified feet in the leaf blades of *O. gratissimum*, *O. basilicum* and *O. canum* respectively; D = Type I foot (leaf blade of *H. suaveolens*), E = Type II foot (leaf blade and petiole of *H. suaveolens*), F = Type III foot (stem of *H. suaveolens*), G = Type IV foot (leaf blade of *H. suaveolens*), and H = Type V foot (leaf blade of *O. lamiifolium*); Tf = trichome foot; ep = epidermal cell.

Table 9. An artificial key to diagnose the types of uniseriate trichome feet in some Nigerian species of *Ocimum* and *Hyptis.*

1a.	Trichome foot (i.e mode of attachment of a simple trichome to the surface of a plant organ as viewed in transection of the organ) is developed on one or two epidermal cells which are not distinguishable in size, shape or position from the other epidermal cells…….. ……..Unmodified foot (Figure 8 A-C)
1b.	Trichome foot (i.e mode of attachment of a simple trichome to the surface of a plant organ as viewed in transection of the organ) is developed on one or more epidermal cells that have been transformed in a way and distinguished in size, shape and/or position from neighbouring epidermal ells……..Modified foot (2)
2a.	Tichome is seated on a single epidermal cell that is enlarged more than the other epidermal cells…….Modified Type I foot (Figure 8D)
2b.	Trichome is seated on two or more cells that are raised (or elevated) above the level of the other epidermal cells……3
3a.	Foot consists of two contiguous cells positioned on top of other epidermal cells……Modified Type II foot (Figure 8E)
3b.	Foot consists of some tiers of many contiguous cells positioned on top of other epidermal cells….....Modified Type III foot (Figure 8F)
2c.	Trichome is seated on a few cells that are radially elongated……4
4a.	Foot consists of elongated cells radiating away from other epidermal cells to the direction of the bottom of the trichome……..Modified Type IV foot (Figure 8 G).
4b.	Foot consists of elongated cells radiating away from the bottom of the trichome to the direction of the other epidermal cells….. ……Modified Type V foot (Figure 8 H).

Table 10. The users' representative opinions about some qualities of nine taxonomic key formats.

S/N	Key format	Users' familiarity	Usability	Efficiency	Users' acceptability
1	Dichotomous	High	Difficult	Low	Low
2	Numerical	Medium	Difficult	Low	High
3	Graphical	Nil	Easy	High	High
4	Circular-diagram	Nil	Easy	Low	Low
5	Columnar-diagram	Medium	Easy	High	High
6	Punched-card	Medium	Easy	Low	Low
7	Multi-access table	Low	Very difficult	Low	Low
8	Flow-chart	Low	Easy	High	High
9	Pictorial-diagram	nil	Easy	High	Low

Enhancement of artificial keys

Taxonomists have devised two common means of introducing additional pieces of information to artificial keys to provide further diagnosis of their specimens. These are by the use of panels and by reference to *tabula.* A panel is a short but diagnostic description of each of a number of plant taxa that have been found to share some common characteristics up to certain level through the use of a key. On the other hand, *tabula* refers to botanical illustrations or representations in the form of drawings, photographs or herbarium/live specimens or even video clips. Although such additional information frequently demand some extra efforts on the part of the key user, they nonetheless assist the taxonomist to identify his specimens with little or no reservation. Moreover the use of these facilities to enhance artificial keys, is an attempt to introduce a visual dimension to the practice of plant identification.

Practically, all the types of key formats earlier enumerated can be enhanced with panels and/or illustrations. The pictorial diagram key format as it were, can be considered as an enhanced form of the flow chart key (Figures 5 and 6). So also, in the numerical key, additional pieces of information are necessary where more than one related taxa e.g. a genus of some species share the same code. The information, usually enclosed in parentheses, could be in the form of a lead or a pointer to one panel. In the panels, names of the related taxa hitherto sharing the same numerical code are listed in alphabetical order. Their features are also described, sometimes along with illustrations, through which the process of elimination arrives at the name of an individual taxon. This is the numerical-to-panel key (Lowe and Stanfield, 1974).

The chart-to-panel key of Hopkins and Stanfield (1966) represents an enhanced form of graphical key. In this case, instead of the pair of selected axes of a graphical key meeting to point at a species name for instance, they may point at a box containing an alphabet or a number which serves as a lead to one panel or the other. Moreover, Table 9 and Figure 8, illustrate how a dichoto-

Table 11. Outcome of the users' merit assessment of nine types of artificial key formats.

S/N	Key format	Percent affirmative responses to the statements of assessment															Index of merit*
		A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	
1	Dichotomous	98	4	41	0	61	82	0	0	0	4	0	94	79	1	65	68.3
2	Numerical	0	89	0	72	59	66	0	4	45	58	0	2	66	20	29	59.1
3	Multi-access Table	0	6	48	1	0	0	0	0	0	0	0	84	71	11	44	55.4
4	Punched-card	0	55	0	60	0	41	0	0	56	44	70	62	0	16	52	54.4
5	Columnar diagram	11	0	0	33	0	20	0	72	0	0	0	50	62	22	31	46.8
6	Graphical	0	2	0	2	0	16	6	58	0	0	0	40	54	18	18	29.9
7	Flow-chart	51	15	0	0	0	65	40	52	0	0	0	31	84	10	12	55.4
8	Pictorial diagram	43	19	0	21	0	42	32	60	0	0	0	4	60	10	6	38.0
9	Circular diagram	15	0	1	19	0	4	0	88	0	0	0	38	66	31	22	48.7

*Index of merit = weighted mean of % of responses. The alphabets A, B, C to O are the codes assigned to the guided statements of assessment of taxonomic key formats as listed in Table 1.

mous or polytomous key can be enhanced with relevant drawings or images (El-Gazzar and Watson, 1970).

Enhancement of taxonomic keys has been epitomized through computerization and development of expert systems for identification of living organisms as recorded by scientists such as Bell (2002), Abdulrahaman et al. (2010) and Gueguim-Kana et al. (2012). These efforts have yielded various forms of computer-aided interactive keys with notable merits over the paper-based keys (Dallwitz et al., 2009). At the moment, there are many computer-based internet-enabled interactive keys with hyperlinks for identification of various groups of plants, animals, microbes and pollen (LPP Foundation, 1999; Monteith, 2000; UNL Nematology Laboratory, 2002; Richter and Dallwitz, 2009; Botany.com, 2010).

Other uses of taxonomic keys

Diagnostic keys are commonly used for identification of plants, animals, microbial organisms, fossils, soils and other biological entities (Marshall, 2000; Soil Survey Staff, 2010). If looked from a general point of view, taxonomic key refers to a way in which classified information is presented (Bauholz, 2013). Going by this definition, a taxonomic key should be found useful in all human endeavors where information is utilized. It can therefore be used to characterize both biological and non-biological entities, or situations alike. It is being adopted, albeit subtly, as a viable tool in organizational decision making process (Gelder, 2010) and in other forms of diagnoses and rational decision making such as in pest control and forensics (Marshall, 2000). Moreover, there is ample evidence that the decision making skills of health diagnosticians (medical doctors, dentists, pathologists e.t.c.) are used to match the facts (or information) of particular case to a diagnostic category (Foucar, 2001; Croskerry and Nimmo, 2011).

The mark of an expert is to seek for precision in each class of things, that is to make better decision and reduce diagnostic error. His ability to make use of diagnostic keys will therefore enhance his thinking skills and ensure rational decision making. In the face of current technological development, training in the construction and use of diagnostic keys can be a good starting point for all professionals for the development of computer-based expert systems to enhance their productivity.

In alpha taxonomy (El-Gazzar and Watson, 1970) descriptive ecology and biodiversity studies, diagnostic keys have been employed for vivid and unambiguous description of observed structures or phenomena; and any type of key format can be used for this purpose. As an illustration, Table 9 and Figure 8 gives a lucid description of the types of uniseriate trichome feet as observed by Ogunkunle and Oladele (2000) in the petiole, leaf blade and stem transections of some Nigerian species of *Ocimum* and *Hyptis*.

Users' assessment of some taxonomic key formats

Table 10 gives the representative opinions of all the respondents about some properties of the nine key formats investigated. The most familiar of the nine was the dichotomous key format while the respondents had no familiarity at all with the graphical, circular diagram, columnar diagram and the pictorial diagram formats (Table 10). According to the respondents, most of these key formats were easy to apply with the exception of the dichotomous, numerical and the multi-access table. Efficiency was acknowledged by the users to be high in the graphical, columnar diagram, flow chart and the pictorial diagram keys but low for the others.

The results of the merit assessment of the nine artificial key formats by the users are shown in Table 11. The key formats can be listed in decreasing order of index of merit as dichotomous (68.3%), numerical (59.1%), multi-access table (55.4%), punched-card (54.4%), circular-diagram (48.7%), columnar-diagram (46.8%), pictorial-diagram (38.0%) and graphical key (29.9%).

This study has revealed that the dichotomous key

format is the most frequently applied and the type to which users of taxonomic keys are most familiar (Table 10). More often than not, a systematist in describing what a taxonomic key is, simply defines a dichotomous key. Sharma (1993), while introducing the topic "Identification with Keys" implicitly described the dichotomous type of key when he defined "a key as an artificial arrangement or analytical device whereby a choice is provided between two contrasting statements resulting in the acceptance of one and the rejection of the other". He further described a single pair of contrasting statements in a key as a couplet and referred to each statement in a couplet as a lead.

Similarly, Olorode (1984) defines a key as a device in which a few characteristics of the plants are so arranged that the features of a known and an unknown plant could be compared in a systematic manner. These submissions are true for the dichotomous key format than for any of the other eight examined in this study. Furthermore, most of those taxonomic publications with keys for the identification of selected groups actually, are catalogues of data presented in the form of dichotomous keys (Hutchinson and Dalziel, 1963-1972; Keay, 1989*).* The foregoing point to the fact that the taxonomic key as hitherto used in botanical literature and circles is synonymous with dichotomous key. This study has however established that other forms of artificial keys also do occur which might equally be usable or more usable than the familiar dichotomous type. It has thus exposed the practicing taxonomist to those available tools from which he can choose to achieve the desired goal.

Of the nine artificial types of keys examined, the widely recognized dichotomous key was among the three that were of low efficiency and tedious to navigate, the other two being the numerical key and multi-access table of identification. In spite of these flaws, the respondents' assessment gave a 68.3% index of merit to the dichotomous key format, putting it over and above all the other eight. This may appear strange, but is understandable; the respondents were not familiar at all with most of the key formats and could expectedly be as critical as possible, only in the assessment of that format to which they have been exposed right from their elementary bio-logy classes (WAEC, 1998-2000). There is little wonder therefore that the dichotomous key arrangement was also the generally acceptable among the supposed users of keys (Table 10).

The results of assessment of the keys have two implications. Firstly, that no single key format is superior to the others; the choice is a function of the availability of the device to the user, the condition of the specimen for identification, the working situation or environment and the exposure level or experience of the user with regards to the key formats. Secondly, the results have brought into limelight those areas of strengths and weaknesses of the taxonomic key types, which are important for consideration in the choice of a format, and which in addition might be helpful towards improving their qualities for better performance. The study therefore affords the taxonomist that opportunity of choosing from a list of instruments for identifying his plants.

Going by these results, the widely accepted dichotomous type of key surpassed the other formats in merit, but then, it is still far from the ideal tool for identification with respect to the expectations in this investigation. The difficulty often encountered in the construction and usage of this key format, especially with the involvement of a large group, is one notable area of its weaknesses. Perhaps this is incidental to the situation that makes people dread the use of keys as indicated by the results of this study. Generally, very few (6.0%) of the respondents frequently used keys while 66% sometimes did. Majority of the latter group often visited herbaria and research institutes where other professionals assisted them in identification.

Another point for noting is that the dichotomous key format is difficult, if not impossible to automate in such a way as for the computer to capture and manipulate generalized information on plant specimens (Okeyinka and Ogunkunle, 2002). If this were readily possible, systematists would be free from laborious process of key construction and tedious character-matching and perusal of volumes that characterize the use of dichotomous keys. On the other hand, multi-access keys, especially in the form of computer-aided interactive devices have advantages over the single-access keys, some of which have been enumerated. With paper-based dichotomous keys, the discovery of a new species renders the key incomplete, while computerized keys are easily updated by adding information for newly discovered species and/or additional diagnostic features, and reposting computer files as appropriate.

CONCLUSIONS AND RECOMMENDATION

This study has collated the hitherto fragmentary pieces of information on nine types of paper-based taxonomic keys usable for plant identification. It has thus generated a handy tool for teaching and research in plant science. The paper has also presented diagnostic data from epidermal and wood anatomical sources on some Nigerian species of *Ocimum*, *Hyptis* and *Ficus* that may be helpful in identifying these potentially useful plants in medicinal, chemical and wood-based industries. Lastly, the paper has reported the outcome of an assessment of the qualities and potentials of the types of keys from the users perspective; and has established that the widely accepted dichotomous key format is still far from an ideal tool for plant identification. Taxonomists should therefore avail themselves of the other available key formats apart from the dichotomous type, and venture into designing new formats that might be more usable. In this direction, those key formats that can readily be electronically pro-

grammed and developed into computer-based interactive applications should be given due priority.

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Review

Coriander (*Coriandrum sativum* L.): Processing, nutritional and functional aspects

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Coriander (*Coriandrum sativum* L.) being an annual herb is most commonly used for seasoning purpose. Its plant seeds, leaves and roots are edible, although they have very distinct flavors and uses. The herb has a light and fresh flavor. Coriander can be used as whole plant and can be processed because of its perishable nature of leaves and to increase the palatability of ripe fruits (seeds) before using it as flavoring agent in different food preparations. Whole plant of coriander mainly fresh leaves and ripe fruits are used for culinary purposes. Coriander leaves have different taste than its seeds, with citrus overtones. Coriander plant is a rich reservoir of micronutrients and nutritional elements which leads us to focus our study on this herb. Coriander is very low in saturated fat however, contains good amount of linoleic acid which is a good source of α-tocopherol and vitamin K. Leaves of plant are rich source of vitamins while seeds are rich in polyphenols and essential oils. Coriander taste is devoted to its essential oil comprising a significant content of linoleic and furanocoumarins (coriandrine and dihydrocoriandrine). Coriander is also well known for its antioxidant, anti-diabetic, anti-mutagenic, anti-anxiety and antimicrobial activity along with analgesic and hormone balancing effect that promotes its use in foods due to numerous health benefits and its protective effect to preserve the food for longer period. The aim of present study was to highlight the processing, nutritional and functional aspects of coriander.

Key words: Coriander, processing, fatty acids, antioxidant content, anti-diabetic, anti-mutagenic, anti-anxiety, diuretic activity, metal detoxification, anti-hyperglycemic activity.

INTRODUCTION

Coriander (*Coriandrum sativum* L.) which belongs to the family Apiaceae (Umbelliferae) is mainly cultivated from its seeds throughout the year (Mhemdi et al., 2011). India is the biggest producer, consumer and exporter of coriander in the world with an annual production of around three lakh tonnes. It is an annual, herbaceous plant which originated from the Mediterranean and Middle Eastern regions and known as medicinal plants. It contains an essential oil (0.03 to 2.6%) (Nadeem et al., 2013). All parts of this herb are in use as flavoring agent and/or as traditional remedies for the treatment of different disorders in the folk medicine systems of different civilizations (Sahib et al., 2012). Coriander closely resembles flat leaf parsley. This resemblance makes many people confused between the two however, coriander has strong fragrance and parsley has mild fragrance. It grows best in dry climates however it can grow in any type of soil like light, well drained, moist, loamy soil, and light to heavy black soil (Verma et al., 2011). Its seeds are almost ovate, globular and have a mild, sweet, slight pungent like citrus flavor with a hint of sage. The most important constituents of its seeds are the essential oil and fatty oil

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(Coskuner and Karababa, 2007).

It is highly reputed ayurvedic medicinal plant commonly known as “Dhanya” in India. This plant is highly aromatic and has multiple uses in food and in other industries. Plants have played a critical role in maintaining human health and civilizing the quality of human life for thousands of years (Dhankar et al., 2011). It is also used to flavor sausages. All parts of plant are edible, fresh leaves can be used for garnishing and are common ingredient in many foods like chutneys and salads. The green herb is also employed for the preparation of either steam-distilled essential oil or the solvent extracted oleoresin (Nadia and Kandi, 2012). Fresh juice of coriander is extremely advantageous in curing many deficiencies related to vitamins and iron. One to two teaspoons of its juice, added to refreshing buttermilk, is incredibly beneficial in curing many diseases. Fresh leaves can be eaten as such because of various health benefits however, if it is not harvested freshly seeds mature and ripen in late summer developing delicate aroma which are then used as dried spice. Moreover, this plant is used to cure diseases like digestive tract disorders, respiratory tract disorders, urinary tract infections. Coriander has been reported to posses many pharmacological activities like antioxidant (Darughe et al., 2012), anti-diabetic (Eidi et al., 2012), anti-mutagenic (Cortes et al., 2004), anti-lipidemic (Sunil et al., 2012), anti-spasmodic (Alison et al., 1999).

Dried coriander fruit is an important ingredient in pickle making. It is sometimes used to mask odd flavors (Parthasarathy et al., 2008). Its fruits contain vegetable oil with a high concentration of monounsaturated fatty acids, especially of petroselinic acid. This oil can be extracted using various techniques; most commonly three different techniques are used: steam distillation, organic solvent extraction (soxhlet), supercritical fluid extraction (Mhemdi et al., 2011). Moreover, coriander oil is used as an antimicrobial agent as it possesses broad spectrum antimicrobial activity (Silva et al., 2011). This oil can be encapsulated in alginates, chitosan etc. so as to enable isolation, protection, transport and release of its active components like vitamins, flavours, peptides, minerals, fatty acids, polyunsaturated fatty acids, antioxidants, enzymes and living cells (Cristian, 2013). Coriander powder and its essential oil are considered as natural food preservatives including antibacterial, antifungal and antioxidant properties (Politeo et al., 2007). The commercial value of its essential oil depends on its physical properties, chemical composition and aroma quality.

Coriander may sometimes be affected by certain diseases and pests like wilt (*Fusarium oxysporum*) and stem gall. The powdery mildew is effectively controlled by spraying sultaf (0.25%). There are no direct control measures for wilt. It is very difficult to control stem-fall (Verma et al., 2011). To check its quality, automatic quality assessment techniques have been developed in Indian spice industries. Quality analysis of its seed was done by image morphological operations (Rohit et al., 2011). It is reported as a non destructive and less time consuming method. With image analysis area, major and minor axis length for counting normal seed, seed having pedestals and foreign element for a given sample were calculated. This technique replaces quality assessment by human sensory methods which is time consuming and less accurate.

PROCESSING

Although coriander is consumed in an unprocessed form; however, its good quantity is also being processed to facilitate the international trade, profitability and palatability. Harvesting is the primary process of collecting the crop and should be done at optimum time period to ensure production of plant material and the best quality of finished spice product (Douglas et al., 2005). It matures in two to three months after sowing. Its smaller-seed requires a longer growing period of approximately 120 days. Depending upon the end use, it can either be harvested green or left to ripe them until fruits turn brown.

Green coriander fruits have sharper and more pronounced flavor than ripe seeds. Mature brown seeds can be used and grind to form powder. Seed heads are cut along with certain inches of stalk or plant may be pulled out with roots and hang in bags upside down, when seeds are fully dry these tend to detach from stalk, collected and store in cool, dry location or ground to obtain a fine powder. In order to remove field heat and prolong shelf life after harvesting, it is necessary to pre-cool harvested coriander before transportation. This can be done by vacuum cooling. Vacuum cooling is achieved through boiling part of the moisture of the product under vacuum conditions. This cooling has a significant effect on amount of vitamin C and shelf life (Apichart et al., 2012).

Coriander may sometimes be irradiated to reduce microbial load before consumption. Irradiation dose of 0.5 kGy of gamma radiations results in reducing aerobic mesophilic count to 99.9%, while the initial total coliform bacteria decreased from 871,000 cfu/g to less than 100 (Cruz-Zaragoza et al., 2011). The microbiological results are lower than the limit levels. Seeds may be also threshed out and dried in sun, winnowed and stored in bags. Usually coriander is sun dried because of cost concerns.

Its seeds after drying may be stored as whole or infused to make delicious vinegar. After harvesting, leaves are properly washed and kept in shade for drying. Drying is followed by powdering in disintegrator and micro pulverizer. Its ground powder is used as a spice in blends. Ground coriander spice loses flavor quickly when stored. This disintegrated product is then standardized, packed and finally distributed. Drying is carried out by number of ways like sun drying, microwave drying, freeze

drying etc. Mainly drying depends on air temperature, greater the air temperature much faster drying will be accomplished. One of the important aspects during drying or thermal processing of its leaves is the loss of chlorophyll content. The most common change that occurs in green vegetables during thermal processing is the conversion of chlorophyll to pheophytins, causing a colour change from bright green to olive-brown, which is undesirable to the consumer. Blanching prior to drying can greatly reduce chlorophyll loss. Loss of chlorophyll content of its foliage can also be prevented by opting microwave drying instead of sun drying. It was found that microwave-dried samples had higher chlorophyll content and the green color was preserved better than for the air-dried and freeze-dried samples (Alibas, 2006). This is because microwave drying of coriander is faster than other conventional drying methods which are gradual and slow to reach the final moisture content. Microwave drying is able to reduce the coriander moisture content to the 12% (wb) target within 21 to 22 min (Shaw et al., 2007).

Ripe seeds of coriander may be sometimes processed to obtain essential oil. This oil is mainly obtained by steam distillation, super critical CO_2 extraction etc. Oil extraction from its seeds was studied with carbon dioxide and propane as solvents, under sub and supercritical conditions. The ratio of solvent to seed (g/g) required to achieve a complete oil extraction was between 20 and 40 using CO_2 at pressures of 200 and 300 bar and temperature of 35°C. A complete oil recovery was attained with propane or propane-rich solvents at 25°C and 50, 80 and 100 bar. The solvating power of propane and propane-containing solvents was proved to be much higher than that of CO_2 (Illes et al., 2000).

Its oil has a characteristic odor of linalool and warm aromatic flavor. This oil is approved for food use by FDA (Food and Drug Administration), FEMA (Foreign Exchange Management Act) and Council of Europe (Silva et al., 2011). In order to preserve the warm aromatic flavor and prevent nutrient loss upto end use, this oil is encapsulated mainly in alginates or chitosans. The aqueous solution of sodium alginate is transformed in gel under the action of calcium ions which form the intermolecular cross-links with the carboxyl groups of guluronate, leading to the well-known “egg-box” structure providing best possible protection to coriander essential oils. The coriander plant is mainly used for making sauces and salsas; on the other hand the fruits are blended into powder for flavouring various products like meat, fish, sodas, pickles, bakery and curry recipes (Ravi et al., 2007).

Its leaves may also be processed to form various products like purees and pastes which are tremendously used nowadays in fast food industries. After harvesting of fresh mature plant, degradation of its components starts. Such degradations may be prevented by processing and converting harvested leaves into product formulation like purees, pastes, sauces salsas etc. Coriander sauce gives an intense flavor and deep green color but is not spicy. In case of powder formation, freshly harvested leaves are blanched at 90°C for at least 2 min so as to inactivate peroxidase enzyme, dried and then ground to uniform size and preserved for further use (Ahmed et al., 2003). Fresh coriander leaves were steam- and water-blanched at 100°C and at 90 and 100°C, respectively, for one to ten minutes, and subsequently comminuted to form a paste. Pasty products obtained from its fruits were processed after water-blanching applying the same time-temperature regimes. Among the eleven phenolics characterized in leaves by high-performance liquid chromatography coupled to mass spectrometric detec-tion, several caffeic acid derivatives, 5-feruloylquinic and 5-p-coumaroylquinic acids were tentatively identified. In fruits, ten phenolics were detected, whereas rutin, a dicaffeic acid derivative and two feruloylquinic and caffeoylquinic acid isomers were newly detected. Upon steam-blanching for one minute, phenolic contents and anti-oxidant capacities remained virtually unchanged. In contrast, water-blanching and extended steam-blanching even yielded increased levels compared to the unheated control, whereas short-time water-blanching resulted in higher values than prolonged heat treatment. Thus, short-time water-blanching was recommended as the initial unit in the processing of coriander leaves and fruits into novel pasty products (Kaiser et al., 2013). A chronological progression for better understanding of different processing aspects of coriander is depicted in Table 1.

NUTRITIONAL ASPECTS

Coriander nutrition is basically due to its green leaves and dried fruits. Like all other green leafy vegetables, its leaves are a rich source of vitamins, minerals and iron. Its leaves contain high amount of vitamin A **(**β-carotene) and vitamin C. The green herbs contain vitamin C upto 160 mg/100 g and vitamin A upto 12 mg/100 g (Girenko, 1982). It is very low in saturated fat and cholesterol and a very good source of thiamine, zinc and dietary fiber. Green coriander contains 84% water.

MAJOR CHEMICAL CONSTITUENTS

Its seeds contain upto 1.8% volatile oil according to origin. The distilled oil (coriander oil BP) contains 65 to 70% of (+)-linalool (coriandrol), depending on the source (Anju et al., 2011).

MINOR CHEMICAL CONSTITUENTS

It includes: Monoterpene hydrocarbons viz α- pinene, β-pinene, limonene, γ-terpinene, ρ-lymene, borneol, citron

Table 1. Chronological progression of different processing aspects of coriander.

Salient feature	Processing method	Researcher
Increased protection of active components of oil	Encapsulation	Cristian et al. (2013)
Higher values of phenolic components and antioxidant activities as compared to prolonged heat treatment	Water-blanching	Kaiser et al. (2013)
Significant effect on amount of vitamin C and shelf life	Vacuum cooling	Apichart et al. (2012)
Moisture content reduction	Drying	Shaw et al. (2007)
Product formulation like sauces, salsas etc. to increase shelf life.	Thermal processing	Ravi et al. (2007)
Prevention of loss of chlorophyll in foliage	Microwave drying	Alibas et al. (2006)
Immature fruit contain higher volatile oil than ripe fruit	Harvesting	Douglas et al. (2005)
Reported to cause inactivation of peroxidase enzyme at 90°C for 2 min.	Blanching	Ahmed et al. (2003)
Greatly reduces chlorophyll losses	Blanching	Ahmed et al. (2001)

wllol, Xmphoe, Geraniol and Geranylacetate; Heterocyclic compounds viz – pyrazine, pyridine, thiazole, furan, tetrahydrofuran derivatives; Isocoumacin viz coriandrin, dihyrocoriandrin, coriandrones A-E, glazonoids; Phthalides viz -neochidilide, Z-digustilide; Phenolic acids and sterols, flavonoids (Wallis, 2005). Carotenoids are of ubiquitous occurrence in all plants with higher concentrations in reproductive organs. In green leafy vegetables, carotenoids, particularly β-carotene is deposited mainly in leaves. Carotenoids can also be processed and used as coloring agents as well as good source of antioxidants. Besides other roles, carotenoids chiefly function as scavengers of the free radicals produced by chlorophylls during photo-oxidation.

Its leaves being good source of β-carotene serve as a precursor of vitamin A. In coriander, β-carotene content, 160 µg/100 g is present whereas total carotenoid content is 1010 µg/100 g (Kandlakunta et al., 2008). Its foliage is used in various types of foods especially in diets of people facing vitamin A deficiency. Green foliage contains anthocyanin (Omidbaigi, 2005). Anthocyanins are bioactive flavonoid compounds that prevent body from various chronic diseases. Anthocyanin in foliage acts as antioxidants which are very useful in improvement of nutritional value as well as maintenance of health and well being (Rahimi et al., 2013).

The various nutrients present in coriander leaf and seeds are shown in Table 2. The characteristic aromatic flavor of its seeds comes from many fatty acids and essential volatile oils. According to USDA (2013), cholesterol content of its seeds is nil. Its seeds are considered as an important source of vitamins, minerals and lipids. Among minerals, potassium is present in high amount (1267 mg/100 g) followed by calcium (709 mg/100 g), phosphorus (409 mg/100 g), magnesium (330 mg /100 g), sodium (35 mg/100 g), zinc (4.70 mg/100 g). The folate content in coriander seed is 200 µg/ 100 g (fresh weight) (Iwatani et al., 2003). Among the various constituents, vitamin C content is present in ample amount (21 mg/100 g) (Table 2). The different vitamins, minerals and phyto-chemicals content in leaf is shown in Table 2.

Coriander contains high amount of essential oils that are very important for growth and for proper functioning of brain. The main essential fatty acids present in coriander include linoleic and linolenic acids. Linoleic acids belong to PUFA (polyunsaturated fatty acid) group. Dietary supplementation of coriander seed greatly affects the lipid composition of carcass by decreasing saturated fatty acid (SFA) contents (palmitic and stearic acids) and by increasing monounsaturated and polyunsaturated fatty acid (MUFA and PUFA) (Ertas et al., 2005).

This plant is a potential source of lipids (rich in petroselinic acid) and an essential oil (high in linalool) isolated from the seeds and the aerial parts (Sahib et al., 2012). The high content of fats and protein in the fruits make distillation residues suitable for animal feed. Coriander fruits yield 5 to 7% of ash, 13% resin, astringent principle, malic acid and alkaloids. Coriander oil contains coriandrol, jireniol and vebriniol (Rao et al., 2012).

FUNCTIONAL ASPECTS

The functional properties of coriander cannot be under estimated. Besides nutritional benefits, it is well known for its health or medicinal benefits as well as for additional benefits like it acts as antimicrobial agent. The type of meat and temperature did not influence the antimicrobial activity of the oil; indicating the potential of coriander oil to serve as a natural antimicrobial compound against *Campylobacter jejuni* in food (Rattanachaikunsopon and Phumkhachorn, 2010). The most important and well characterized functional aspect involves antioxidant activity.

ANTIOXIDANT ACTIVITY

Coriander is a good source of polyphenols and phyto-chemicals due to its high antioxidant activity. Reactive

Table 2. Nutrient composition of coriander leaf and seeds as per USDA (National Nutrition Data base, 2013).

Nutrient	Amount (per 100 g)	
	Coriander leaf	Coriander seed
Water	7.30 g	8.86 g
Energy	279 kcal	298 kcal
Protein	21.93 g	12.37 g
Total lipid (fat)	4.78 g	17.77 g
Carbohydrate, by difference	52.10 g	54.99 g
Fiber, total dietary	10.40 g	41.9 g
Calcium, Ca	1246 mg	709 mg
Iron, Fe	42.46 mg	16.32 mg
Magnesium, Mg	694 mg	330 mg
Phosphorus, P	481 mg	409 mg
Potassium, K	4466 mg	1267 mg
Sodium, Na	211 mg	35 mg
Zinc, Zn	4.72 mg	4.70 mg
Vitamin C, total ascorbic acid	566.7 mg	21.0 mg
Thiamin	1.252 mg	0.239 mg
Riboflavin	1.500 mg	0.290 mg
Niacin	10.707 mg	2.130 mg
Vitamin B-12	0.00 µg	0.00 µg
Vitamin A, RAE	293 µg	0.00 µg
Vitamin A, IU	5850 IU	0 IU
Vitamin D (D2 + D3)	0.00 µg	0.0 µg
Vitamin D	0 IU	0 IU
Fatty acids, total saturated	0.115 g	0.990 g
Fatty acids, total monounsaturated	2.232 g	13.580 g
Fatty acids, total polyunsaturated	0.328 g	1.750 g
Cholesterol	0.00 mg	0 mg

species of oxygen can cause oxidative stress and consequently, the damage of tissues and biomolecules (Barros et al., 2012). Both leaves and seeds of coriander contain antioxidants but leaves contain more amounts of antioxidants than seeds (Wangensteen et al., 2004). Its Antioxidant content is attributed to its high content of pigments particularly carotenoids. The carotenoids of its extract were found to show higher hydroxyl radicals scavenging potential thereby protecting cells from oxidative damage (Peethambaran et al., 2012). Among secondary metabolites, phenolic compounds are considered as one of the most important and largest group. Phenolic groups may be categorized into four main groups depending upon number of phenol rings and structural elements that bind these rings. These groups include: flavonoids (anthocyanins, flavones and isoflavones) tannins, stilbenes and lignans (Balasundram et al., 2006).

In recent years, essential oils have been qualified as natural antioxidants. Coriander essential oils serve as potential antioxidants. Main components of its essential oil are: camphor (44.99%), cyclohexanol acetate (cis-2-tert.butyl-) (14.45%), limonene (7.17%), α-pinene (6.37%). This essential oil at percentage of 0.05, 0.10 and 0.15 is very much effective in inhabiting primary and secondary oxidation products. It was found that at the proportion of 0.02%, its effects were almost equal to BHA (butylated hydroxyanisole) (Darughe et al., 2012).

One of the major problems in high lipid product in food industry is rancidity resulting in undesirable flavor changes and decline in nutrients (vitamins) leading to change in their texture and appearance. Lipid peroxidation causes oxidative stress, resulting in the development of rancidity, unpleasant taste and odors as well as changes in color and losses related to nutritional value. Use of antioxidants reduces oxidative rancidity (Bhanger et al., 2007). Addition of coriander essential oil may greatly serve the purpose. Darughe et al. (2012) studied the antioxidant effects of CEO (coriander essential oils) in cake. It was found that antioxidant effect of CEO may be due to the presence of terpenoid components (camphor, limonene, α-pinene and geraniol). This essential oil due to its radical scavenging activity can be used as natural antioxidant to enhance the shelf

stability of many foods (Ramadan et al., 2003).

ANTI-HYPERGLYCEMIC ACTIVITY

Its seed extract is used as a traditional medicine for diabetic patients. Incorporation of ground coriander seed extract in diet led to marked decline in blood glucose and rise in levels of insulin in diabetic rats. Besides peroxidative damage inhibition, addition of its seed extract reactivated antioxidant enzymes and antioxidant levels in diabetic rats (Deepa and Anuradha, 2011). Gray and Flatt (1999) studied insulin releasing and insulin like activity of coriander. It was observed that its aqueous consumption evoked 1.3-5.7 fold stimulation of insulin secretion from colon B- cell line. An aqueous extract of coriander (1 mg/ml) increased the 2-deoxyglucose transport by 1.6 folds, glucose oxidation by 1.4 folds and incorporation of glucose into glycogen of isolated murine abdominal muscle by 1.7 folds.

HYPOLIPIDEMIC EFFECT

Hyperlipidemia increases the risk for generation of lipid oxidation products, which accumulate in the subendothelial spaces of vasculature and bone. Atherogenic high-fat diets increase serum levels of oxidized lipids, which are known to attenuate osteogenesis in culture and to promote bone loss (Pirih et al., 2012). Lal et al. (2004) studied the hypolipidemic effect of coriander (*Coriandrum sativum* L.). Coriander was given at a dose of 1g/kg to triton induced hyperlipidemic rats. It was found that coriander decreases the uptake and enhances the breakdown of lipids. Results were compared with commercially available herbal drug for hypolipidemia. From these findings it was assumed that coriander can be used as preventive and curative herbal against hyperlipidemia.

ANTI-ANTHELMINTIC ACTIVITY

In vitro anthelmintic activities of crude aqueous and hydro-alcoholic extracts of the seeds of Coriander were investigated on the egg and adult nematode parasite *Haemonchus contortus.* Its aqueous extract was also investigated for *in vivo* anthelmintic activity in sheep infected with *H. contortus*. Both extract types of coriander inhibited hatching of eggs completely at a concentration less than 0.5 mg/ml (Debella et al., 2007).

ANTI-ANXIETY EFFECT

An internal turmoil often accompanied by various diseases like stomach ache, nervous behavior. Anxiety may be sometimes uncontrollable and irritating. Coriander has been used as folk medicine in Iran for treatment of insomnia. Mahendra and Bisht (2011) studied anti-anxiety activity of hydro alcoholic extract of coriander in mice using diazepam as standard. It was observed that the extract of 100 and 200 mg/kg produced anti-anxiety effects similar to diazepam. Its aqueous extract causes anxiolytic effect and was studied by using extract (10, 25, 50, 100 mg/kg) in male albino mice using elevated plus-maze as an animal model of anxiety. It was found that aqueous extract at 50, 100 and 500 mg/kg significantly reduced spontaneous activity and neuromuscular coordination, compared to control group. All these results led to conclusion that coriander extract can be used as potential sedative and muscle relaxant (Masoumeh et al., 2005). A chronological progression of different functional properties of coriander is depicted in Table 3.

DIURETIC ACTIVITY

Jabeen et al. (2009) studied the diuretic activity of the plant extracts on wistar rats of either sex (200 to 250 g). Negative and positive control group comprising of five animals, each received saline and standard diuretic drug: furosemide (10 mg/kg), while rest of the groups with similar number of animals, were given different doses of the plant extracts dissolved in saline (50 ml/kg). The results concluded that the diuretic effect of coriander was confirmed due to significant increase in urine output (diuresis) in rats, similar to furosemide, a standard diuretic. Therefore, diuretic is considered as one of the best choices for the treatment and management of uncomplicated hypertension.

METAL DETOXIFICATION

Coriander can be used as a natural cleansing agent as it has potential to remove toxic metals from body. Chemical compounds present in coriander attach to toxic metals and remove them from cells (Abidhusen, 2012). Arunasagar et al. (2005) observed that this plant is very effective to remove inorganic (Hg^{2+}) and methyl mercury (CH_3Hg^+) from aqueous solutions. This effect was due to the binding effect of carboxylic group to mercury. These results clearly showed that sorbent can be used to remove inorganic and methyl mercury from contaminated water. Kansal et al. (2011) found that coriander led to marked decline in oxidative stress caused by lead nitrate.

ANTI-BACTERIAL ACTIVITY

Aliphatic (2E)-alkenals and alkanals characterized from the fresh leaves of the coriander were found to possess

Table 3. Chronological progression of different functional properties of coriander

Functional activity	Compounds involved	Researcher
Inhibition of oxidative products	BHA	Darughe et al. (2012)
Antianxiety effect	Coriander extract	Mahendra (2011)
Antiglycemic activity	Aqueous seed extracts	Deepa and Anuradha (2011)
Antimicrobial activity	Coriander oil	Rattanachaikunsopon and Phumkhachorn (2010)
Inhibition of oxidative products	Terpenoids	Bhanger et al. (2006)
Oxidative damage protection	Flavonoids (Anthocyanins, tannins, lignins)	Balasundarum et al. (2006)
Metal detoxification	Hydroxyl groups	Arunasagar et al. (2005)
Antioxidant activity	Phenolics	Wangensteen et al. (2004)
Hypolipidemic effect	Constituents present in it due to increase in lipid catabolism and excretion	Lal et al. (2004)

bactericidal activity against *Salmonella choleraesuis* spp. choleraesuis ATCC 35640. (2E)-Dodecenal was most effective against this food-borne bacterium with the minimum bactericidal concentration (MBC) of 6.25 µg/mL (34 µM), followed by (2E) - undecenal (C11) with an MBC of 12.5 µg/mL (74 µM).

ANTI-FUNGAL ACTIVITY

Moisture content and pH of the foodstuffs have been reported as the main biotic factors affecting the fungal deterioration especially in high moisture foods. Common moulds found in cakes and bakery products are *Penicillium expansum, Penicillium stoloniferum, Rhizopus stolonifer, Aspergillus niger, Monilia sitophila* and *species* of *Mucor* and *Geotrichum*. Among these moulds, *Penicillium expansum, Penicillium stoloniferum* and *Mucor* are *mycotoxins* producers. Darughe et al. (2012) studied the antifungal activity of coriander essential oil in cakes having moisture content and pH ranges between 13 to 15.5% and 6.64 to 7.81%, respectively. After 30 days, it was found that percentage of moulds in cakes containing coriander essential oil at 0.05% did not differ from cakes containing 0.01% BHA and both concentration were not able to control mould growth in cakes but increasing concentration of CEO up to 0.15%, antifungal activity of CEO showed better control on mould growth in cakes than control sample and BHA samples.

Despite of these important functional activities, coriander has several other functional properties that promote its use as a preventive and curative herb. Some of these important activities include: antifertility activity due to significant decline in serum progesterone (Al-said et al., 1987). It is used in the preparation of many household medicines to cure bed cold, seasonal fever, nausea, vomiting, stomach disorders and also used as a drug for indigestion, against worms, rheumatism and pain in the joints. Many of its healing properties can be attributed to its exceptional phytonutrients and hence, it is often referred to as store house for bioactive compounds (Ullagaddi and Bondada, 2011). It also has preventive action on gastric mucosal membranes due to many reasons like free radical scavenging activity or due to formation of protective layer (Al-Mofleha, 2006). Its oil can also be used as anti microbial agent. This oil is effective against both gram positive as well as gram negative bacteria and also against pathogenic fungus. Coriander oil also exhibits bactericidal activity with the exception of *Bacillus cereus* and *Enterococcus faecalis* (Silva, 2011).

CONCLUSIONS

Herbs and spices are processed in foods from early times for seasoning as well as to increase shelf life of food and to restore health. Coriander is one of miraculous herb that functions as both, spice as well as herbal medicine. Although plant can be grown throughout the year, it is processed to increase its palatability, profitability and facilitate international trade. The leaves and fruits are highly fragrant and contain nutrients like fat, proteins, vitamins minerals etc. Its health benefits activities ranging from antibacterial to anticancer activities. Most important and well characterized property of coriander is its use as antioxidant. Due to its multifunctional uses and protective and preventive action against various chronic diseases, this herb is righty called as “herb of happiness”. Moreover, processing of fruits and leaves of coriander is the best way to preserve this herb.

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Full Length Research Paper

Hepatoprotective and hepatocurative properties of alcoholic extract of *Carthamus oxyacantha* seeds

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Carthamus oxyacantha **commonly known as "*Poli*" or "*Peeli kandiary*" belongs to family *Asteraceae/Compositae*. Protective and curative effects of alcoholic extract of *C. oxyacantha* seeds were investigated against carbon tetrachloride (CCl_4) induced hepatic damage in rats (Sprague Dawley Strain). Hepatic damage was induced by injecting a single intraperitoneal dose of 4 ml (50%v/v) CCl_4 in olive oil. In protective studies, plant extract (400 mg/kg body weight) was given before hepatic damage while in curative studies; hepatic damage was induced before the application of plant extract. The hepatoprotective and hepatocurative effects were monitored by estimating the activities of the serum glutamate pyruvate transaminase (SGPT), serum glutamate oxalacetate transaminase (SGOT), serum alkaline phosphatase (ALP), total proteins, glutathione level serum bilirubin concentration and histopathological studies. Results show that alcoholic extract of seeds of *C. oxycantha* possessed both hepatoprotective and hepatocurative activity. However, hepatoprotective activity was more pronounced as compared to hepatocurative activity. Histopathological studies also supported the biochemical parameters.**

Key words: *Carthamus oxyacantha,* hepatoprotective, hepatocurative.

INTRODUCTION

Hepatotoxicity is inflammation of the liver. It is generally associated with various drugs used in modern medicine, different chemicals, toxins and viruses (Ravikumar et al., 2005; Stierum et al., 2005). Hepatic problems along with heart problems and diabetes are one of the major causes of human death across the world. Roger et al. (2001) reported that more than two million people die annually from liver related disorders in the world.

One of the known hepatotoxins used in biological research is Carbon tetrachloride (CCl_4). Inside the body, CCl_4 produces trichloromethyl free radicals. These free radicals react with other molecules in the cell and stimulate a series of reactions. End result of this series of reactions is the initiation of peroxidation of membrane lipids (Reinke et al., 1988; Sahu et al., 2005) and hence liver damage. Trichloromethyl radical is believed to be an intermediate product of the reductive dechlorination of CCl_4, catalyzed by certain cytochrome P_{450} enzymes particularly the ethanol inducible isoform of the cytochrome (Reinke et al., 1988). There are many reports which show similarities between CCl_4 induced liver damage and human liver cirrhosis (Halim et al., 1997). That is why CCl_4 induced liver damage is generally used as experimental model for screening of hepatoprotective and hepatocurative drugs. The intensity of hepatic damage is generally accessed by measuring the activities of hepatic cytoplasmic enzymes [serum glutamate pyruvate transaminase (SGPT), serum glutamate oxalacetate transaminase (SGOT), serum alkaline phosphatase (ALP)], serum bilirubin concentration and

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histological studies (Ravikumar et al., 2005; Stierum, 2005). The extent of oxidative stress may be predicted by estimating the serum glutathione level (Sallie et al., 1991; Sahu et al., 2005).

Whereas no reliable cure is available for hepatic disorders in modern medicine practice, herbal treatment has recommended variety of plants for its treatment (Sanmugapriya and Venkataraman, 2006). The efficacies of most of these medicinal plants have not yet been validated. Scientific-based pharmacological data is not available for most of the herbal formulations. Hence, all such herbal formulations cannot be recommended for liver diseases (Stickel and Schuppan, 2007). Thus, investigation of medicinal plants with potential hepatic regenerative activity becomes important.

Carthamus oxyacantha is commonly known as *Poli* or *Peeli Kandiari* in different parts of the Pakistan. It is an annual herb with spiny leaves. Like other spiny plants in the genus *Carthamus*, this species is not used as fodder for livestock.

Plants in genus *Carthamus* are reported to have a mixture of glycerides composed of linoleic and oleic acids. Concentration of these compounds varies in different species. San Feliciana et al. (1982) reported the presence of a sesquiterpene glycoside as the dominant component in hexane extract from aerial parts of *C. lanatus.* This compound seems to be characteristic of genus *Carthamus.* Many plants in family *Asteracceae* / *Compositae* are reported to contain pyrrolizidine alkaloids which are reported to be hepatotoxic (Mattocks, 1990; Borba et al., 2001). Pyrrolizidine alkaloids are a diverse class of natural compounds based on azabicyclo ring, generally occurring as esters of a "necine base" with "necic acids" as mono- or diesters (Mattocks, 1990).

The seeds of *C. oxyacantha* are used by many local *Tabibs* for treatment of diabetes in some parts of the Pakistan. However, scientific data is still not available about the physiological effects of this plant. We hope that this study will help us to evaluate the exact use of this plant in treatment of different diseases.

MATERIALS AND METHODS

Plant collection

C. oxyacantha seeds were collected from fields of different parts of District Attock, Chakwal, Jehlum and Rawalpindi. Plant and it seeds were then brought to the Department of Biochemistry, Faculty of Biological Sciences, Quaid-i-Azam University, Islamabad. Plant was identified at the Department of Plant Sciences (voucher no. 109, dated; 21-8-2006), Faculty of Biological Sciences, Quaid-i-Azam University Islamabad. The seeds were then thoroughly washed with distilled water and dried in an oven at 30°C.

Preparation of plant extract

The dried seeds were powdered mechanically with a China herb grinder. The powder was kept in dry, clean, air tight glass jars and lipids and was filtered. The residue was mixed with 250 ml absolute ethanol (Merck) for 48 h with occasional shaking and was filtered. The filtrate was dried in Petri dishes and concentrated to greenish / brownish residue (4.6±0.07 g/100 g plant material) by evaporation at 4°C under reduced pressure in vacuum drying oven (Toyo Seisakusho co, Japan). The dried alcoholic extract was stored in a refrigerator until used. The extract was dissolved in adequate amount of 50% ethanol just before injection to respective group of rats. The extract was given as intraperitoneal injection as desired.

Experimental animals

Healthy young adult male albino rats of Sprague Dawley strain, weighing between 180-300 g were obtained from the National Institute of Health, Islamabad and kept at the animal house, Department of Biochemistry, Faculty of Biological Sciences, Quaid-i-Azam University, Islamabad. The animals were kept on standard feed purchased from National Institute of Health, Islamabad. The animals were handled according to European Community guidelines (EEC Directive of 1986; 86/609/EEC).

Grouping

25 rats were randomly divided into five groups of five rats each. All rats were maintained on standard feed and water *ad libitum* with natural day light and dark periods. Group 1 was normal control and Group 2 was sham control for hepatoprotective studies. Animals in this group were given a single dose of CCl_4 (50% v/v in olive oil) on the 30th day to cause hepatic damage. Group 3 was sham control for hepatocurative studies. Animals in this group were given a single dose of CCl_4 (50% v/v in olive oil) on the 1st day to cause hepatic damage. Rests of the two groups (4 and 5) were treated. Animals in group 4 were given plant extract 400 mg/kg body weight for 29 days and a single dose of CCl_4 (50% v/v in olive oil) on the 30th day. Animals in group 5 were given a single dose of CCl_4 (50% v/v in olive oil) on the 1st day and thereafter plant extract 400 mg/kg body weight for 30 days. All rats were sacrificed on the 31st day.

Blood collection

Animals were anaesthetized on the 30^{th} day and heart puncture was done with a 5-ml disposable syringe. 2 ml blood was drawn very gently and slowly. Serum was separated by centrifugation at 3000 rpm for 15 minand then preserved in the appendorf tubes at -20°C in the freezer until analyzed.

Serum analysis

Serum samples collected from different groups were analyzed for chemical estimation by using packed kits made by Sigma (USA). The absorption was recorded using a Spectrophotometer UV-240 (Shimadzu). All estimations were made according to the procedure provided with the kit. All values given are mean ±SE of 5. Computer program “Statistica 5.5” was used for statistical analysis. Student’s t-test” and “ANOVA” were applied to find the difference between normal control, sham control and treated groups. A “p” value of 0.01 or < 0.01 was taken as a level of significance.

Histological studies

Lives were removed in respective groups and stored immediately in a solution containing 10% formalin and 0.9% NaCl for histopatholo-

Table 1. Effect of *C. oxyacantha* seeds extract on body weight and liver weight in hepatoprotective and hepatocurative studies.

Group	Initial body weight (g)	Final body weight (g)	Difference in body weight (g)	Body weight gain/loss (%)	Weight of liver (g)	Liver/body weight (%)
Hepatoprotective studies						
Normal control	180.1±10.3	191.3±12.2	11.1±3.7	6.16±1.8[a]	8.4±0.23	4.4±0.4[a]
Sham control	199.3±14.5	212.4±15.4	13.1±2.9	6.5±1.4[a]	7.2±0.45	3.89±0.6[a]
Treated	179.2±16.8	190.9±13.1	11.7±3.2	6.5±1.4[a]	6.4±0.14	3.35±0.2[a]
Hepatocurative studies						
Sham control	220.8±17.2	211.7±14.3	-9.1±3.1	-4.12 ±0.9[b]	5.2±0.31	2.5±0.8[b]
Treated	187.4±20.5	183.8±18.3	-3.6±0.6	-1.92±0.06[c]	5.8±0.2	3.2±1.1[b]

Values given are mean ±SE of 5. Sham control was compared with normal control. The treated was compared with respective sham control. Values with same superscript in a column differ non-significantly while with different superscript differ significantly at $p<0.01$.

gical examination. The tissues were then embedded in paraffin, thinly sectioned using a microtome, stained with haematoxylin and eosin (H&E) and examined under optical microscope.

RESULTS AND DISCUSSION

Hepatoprotective studies are conducted to investigate the protective / preventive effects of the plant extracts against liver damage. Plant extract 400 mg/kg body weight was given prior to the liver damage in hepatoprotective studies. The extract at the doses of 250 and 500 mg/kg were comparable to the effect produced by Liv-52®, a well established plants-based hepatoprotective formulation against hepatotoxins (Arulkumaran et al., 2009).

Hepatocurative studies were conducted to investigate the efficacy of plant extract in curing the liver damage. In these studies, liver damage was induced prior to the application of plant extract. In both the hepatoprotective and hepatocurative studies, liver damage was caused by a single dose of CCl_4.

In hepatoprotective studies, a net body weight gain was observed in all the groups (Table 1) including control (6.16±1.8%), sham control (6.5±1.4%) and treated which is in accordance with previous finding of Smialowicz et al. (1991). However, Shibayama (1989) reported a 6-8% loss in body weight in sham control group. This difference may be due to the difference in experimental designs. In our study, hepatic damage was induced on the 30th day and experiment was run for 31 days.

The percent mean body weight gain in control group (1); sham control group (2) and treated group (4) differ non-significantly. Similarly, no significant difference was observed in percent liver / body weight ratio between control (4.4±0.4%) group and sham control group (3.89±0.6%). This was expected because the experiment was run for one month and liver damage was induced on the second last day (30th). On the very next day (31st day) rats were sacrificed. CCl_4 was not given enough time to affect the metabolism, body weight and percent liver / body weight ratio. Therefore, no significant difference in mean percent liver / body weight ratio in different groups of rats were found.

In hepatocurative studies, a net body weight gain was observed (Table 1) in the control group (6.16±1.8%) as compared to a net body weight loss (4.12±0.9%) in sham control group (3). In the treated group (5) however, a weight gain was observed (5.8±0.2). A body weight loss in sham control group was expected because CCl_4 was given on the 1st day and rats were sacrificed on the 31st day. A complete one month is quite enough time for CCl_4 to severely affect the body metabolism and hence causing a decrease in body weight. Similarly, a significant difference in percent liver / body weight ratio was observed between control group (4.25±0.4%) and sham control group (2.5±0.8%).

The animals in the sham control groups of both hepatoprotective (on 30th day) and hepatocurative (1st week of the experiment) studies were quite sluggish and less active as compared to the control group. The animals in treated groups of hepatocurative studies showed same symptoms. This may be due to the fact that CCl_4 also acts as an anesthetic agent, causing slowness in all reflexes of the animal and other general behavior.

A significant difference in liver marker enzymes that is ALP, SGPT, SGOT and serum glutathione level was observed between control and sham control groups of both hepatoprotective and hepatocurative studies (Table 2 and Figure 1). This shows the hepatic damage caused by CCl_4.

Smialowicz (1991) had reported no notable change in the ALP value in sham control group while Vadiraja et al. (1998) had reported a 5-8 times increase in SGPT and 10-13 times increase in SGOT value in CCl_4 treated group. In the present study, about 4 times increase in SGPT, 5 fold increase in SGOT and 2 fold increase in ALP was observed in sham control groups compared to normal control group. Similarly, a significant decrease in

Table 2. Effect of *C. oxyacantha* seeds extracts on liver marker enzyme activities, bilirubin, glutathione concentration and serum total proteins in hepatoprotective and hepatocurative groups**.**

Group	ALP (U/L)	SGPT (U/L)	SGOT (U/L)	Bilirubin (mg/dl)	Glutathione (mg/dl)	Total protein (g/dl)
Hepatoprotective studies						
Normal control	112.3±10.6[a]	36.2±6.9[a]	38.1±5.8[a]	0.48±0.03[a]	30.4±6.2[a]	7.3±0.4[a]
Sham control	240.4±16.3[b]	163.8±11.5[b]	186.7±12.2[b]	0.49±0.07[a]	23.3± 4.6[b]	7.1±0.7[a]
Treated	140.4±10.7[c]	87.51±6.4[c]	69.78±7.3[c]	0.3±0.05[a]	29.3±4.8[c]	7.2±0.05[a]
Hepatocurative studies						
Sham control	261.4±16.2[b]	177.6±12.7[b]	176.4±14.7[b]	1.3±0.07[b]	14.7±3.7[b]	4.1±0.3[b]
Treated	140.37±9.5[c]	87.51±11.2[c]	109.78±7.5[c]	0.61±0.03[c]	24.8± 2.5[c]	6.8±0.06[c]

Values given are mean ±SE of 5. Sham control was compared with normal control. The treated was compared with respective sham control. Values with same superscript in a column differ non-significantly while with different superscript differ significantly at $p<0.01$.

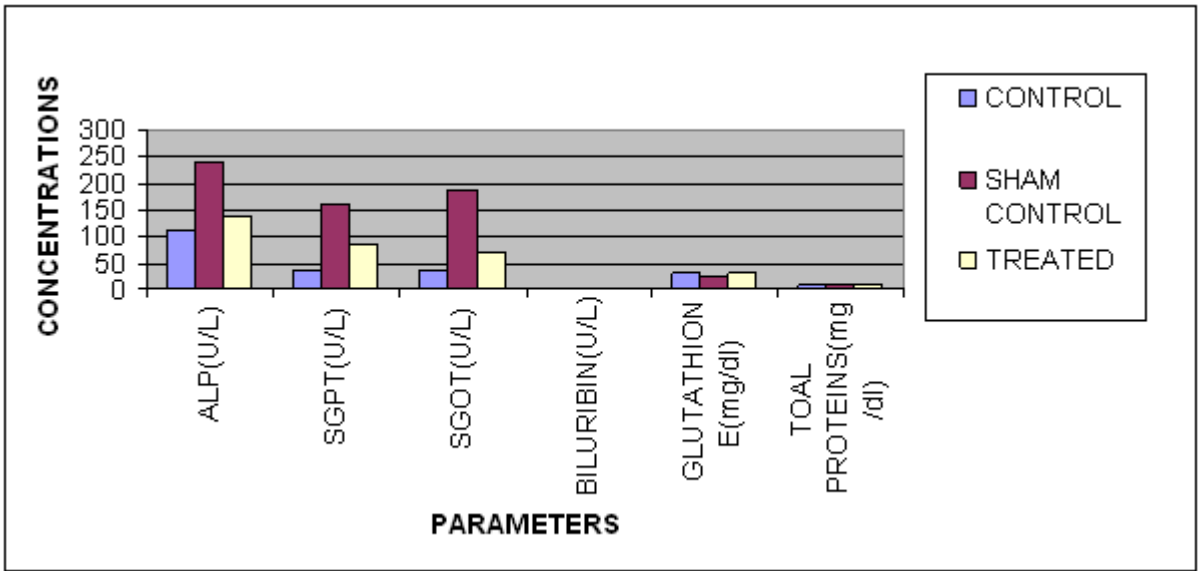

Figure 1. Concentrations of different parameters for different control groups (Hepatoprotective studies).

the serum glutathione level in sham control group (23.3±4.6 mg/dl) was observed as compared to the control group (30.4±6.2 mg/dl) both in hepatoprotective as well hepatocurative studies. In hepatocurative studies, the decrease in glutathione level in sham control group (14.7±3.7 mg/dl) was even more as compared to the control group. This shows an increased use of glutathione which is also characteristic of oxidative stress as a result of hepatic damage due to the CCl_4.

In hepatoprotective studies, no significant difference in the value of serum bilirubin and serum total proteins was observed between control group, sham control group and the treated group (Table 2 Figure 1). This might be due to the fact that CCl_4 could not get enough time (24 h) to effect the bilirubin concentration and serum total proteins level. Perhaps to affect the bilirubin and total proteins more time is required by the CCl_4. However, in hepatocurative studies, a significant difference in bilirubin and total proteins level was observed between control group and sham control group.

Histological studies of the liver also showed severe damage to the hepatocytes. Necrosis of the hepatocytes is quite prominent in rats in sham control group of both hepatoprotective as well as hepatocurative group (Figures 4 and 5) as compared to the control group (Figure 3).

Alcoholic extract of seeds also showed significant hepatoprotective and hepatocurative effects. A significant decrease in liver marker enzymes, bilirubin and a significant increase in glutathione and serum total proteins was observed in treated groups as compared to the sham control group (Table 2 and Figure 2).

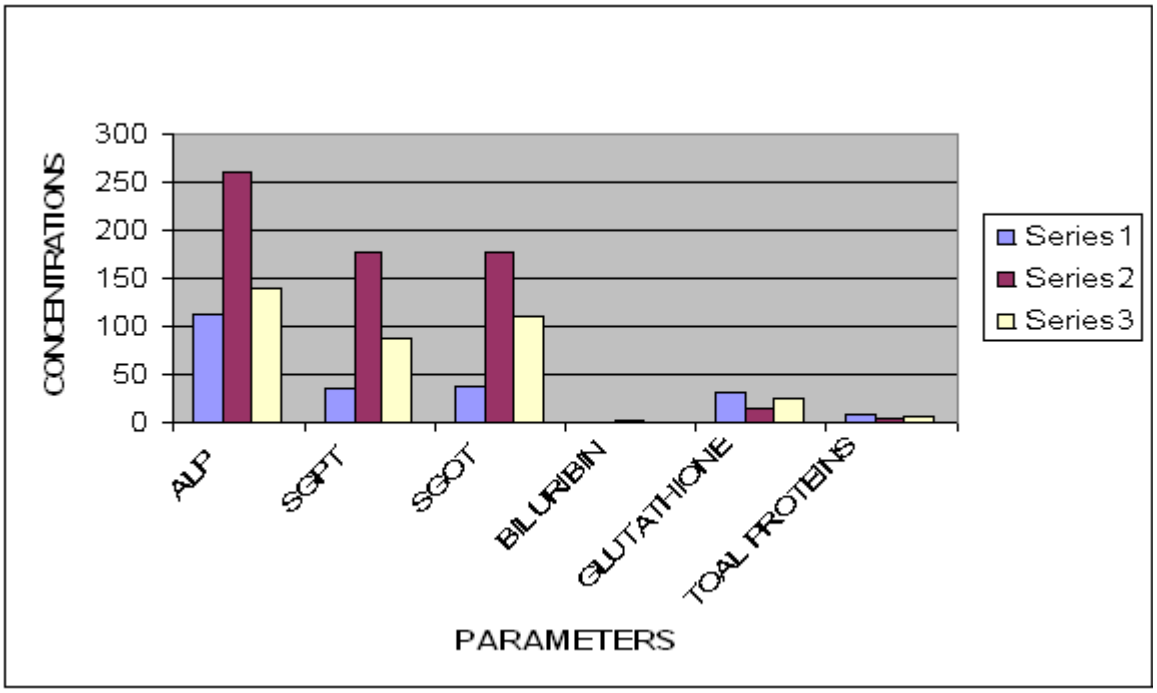

Figure 2. Concentrations of different parameters for different control groups (hepatocurative studies).

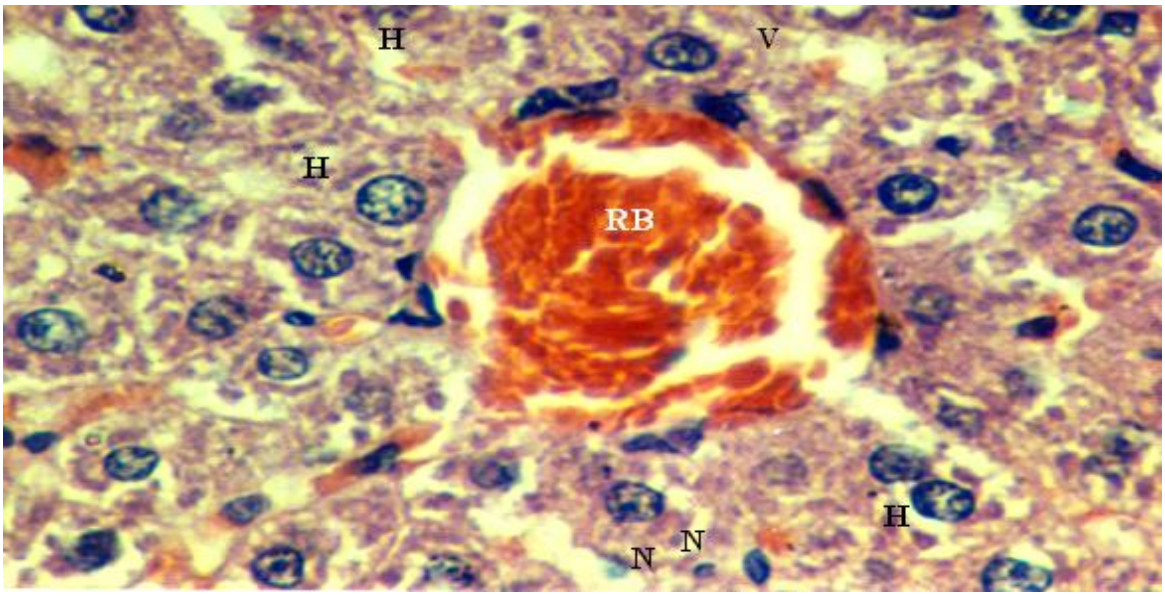

Figure 3. Photograph of the liver of a normal rat at 40X showing normal hepatocytes (H) with prominent nuclei (N), red blood cell (RB) and central vein (CV) (hepatoprotective studies).

Histological studies also show less damage in treated groups as compared to the sham control group (Figures 6 and 7). This reflects the presence of chemicals / compounds like antioxidants which help the hepatocytes to get cured from the damage. *Carthamus tinctorius,* is reported to be having a mixture of glycerides of linoleic and oleic acids in proportions that differ with plant variety. San Feliciana et al. (1982) reported that a sesquiterpene glycoside is the major component of the hexane extract from aerial parts of *C. lanatus* and that this type of compound seems to be characteristic of genus *Carthamus.*

CCl_4 can cause damage to many tissues in the body. However, the most important primary target organ for

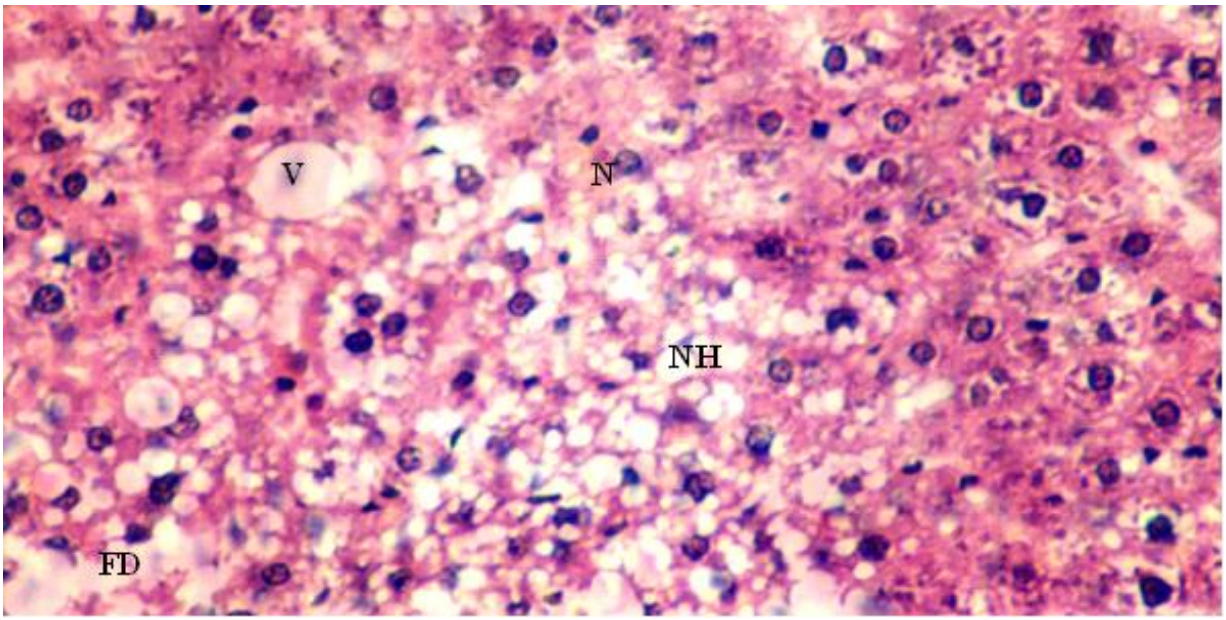

Figure 4. Photograph of the liver of a Sham control (hepatoprotective studies) rat at 40X showing necrotizing hepatocytes (NH) with degenerating nuclei (N), fatty deposition (FD), fibrosis (F) and vacuolization (V).

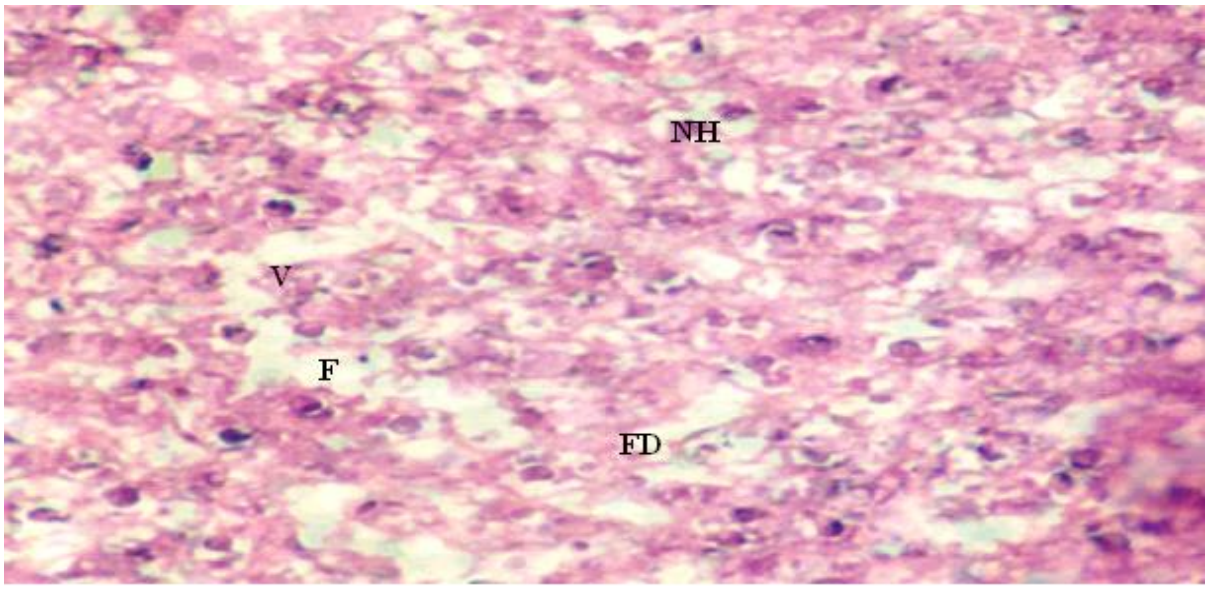

Figure 5. Photograph of the liver of a Sham control (hepatocurative studies) rat at 40X showing necrotizing hepatocytes (NH) fatty deposition (FD), vacuolization (V) and fibrosis (F).

CCl_4 induced toxicity in many species is the liver. CCl_4 when metabolized in the body is changed into a very reactive free redicals (halogenated free radical) by cytochrome P_{450} mixed function oxidase system (Meunier et al., 2004). These reactive species then induce hepatic damage. Many latest evidences show that oxidative stress caused by free radicals may induce peroxidation and damage to biomolecules (lipid protein and nucleic acids). This may further leads to aging, cancer and many other diseases in human. Further it has been observed that inflammatory state of the body (acute or chronic) is due to oxidative stress induced by the generation of free radicals (Van de straat et al., 1987; Sahi et al., 2004). CCl4 also causes severe fatty changes in liver (Afaf et al., 2008).

Hepatoprotective and hepatocurative properties of plants or plants extracts are generally attributed to the presence of chemicals which act as antioxidants or

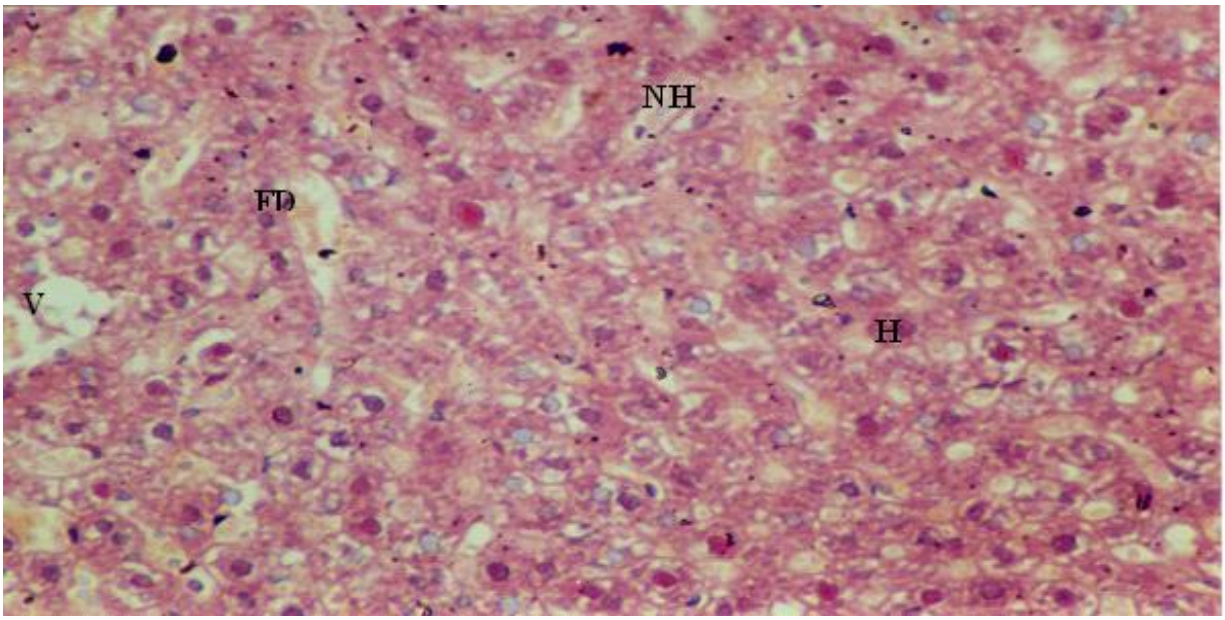

Figure 6. Photograph of the liver of alcoholic extract treated (*c. oxyacantha* seeds) rat at 40X showing normal hepatocyte (H), necrotizing hepatocytes (NH) vacuolization (V) and Fibrosis (hepatoprotective studies).

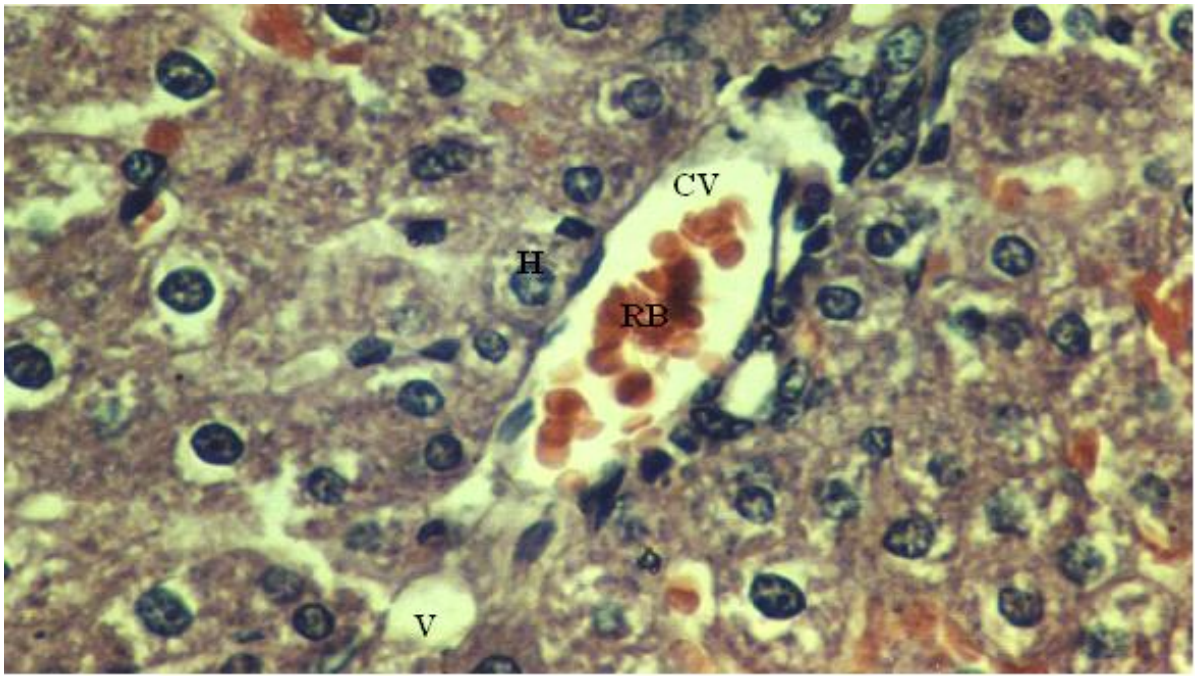

Figure 7. Photograph of the liver of alcoholic extract treated (*c. oxyacantha* seeds) rat at 40X showing normal hepatocyte (H), central vein (CV), vacuolization (V) and red blood cells (RB) (hepatocurative studies).

inhibitor of the microsomal drug metabolizing enzymes (MDME) (Shin, 1989 and Gopinathan *et al*., 2004 and Hui-Mei *et al.,* 2008). As it is widely accepted that CCl_4 is metabolically activated by hepatic microsomal cytochrome P_{450} medicated reactions to the trichloromethyl radical (Slater, 1984). Therefore, the inhibitors of cytochrome P_{450} (MDME) can impair the bioactivation of CCl_4 into its toxic species and thus provide protection against hepatocellular damage (Nelson et al., 1980). Cytochrome P_{3A2} is one of the most abundantly expressed cytochrome, which metabolizes numerous drugs including barbiturates (Desjardins and Iversen,

1995).

The hepatocurative activity in alcoholic extract of *C. oxyacantha* seeds may be due to the presence of certain antioxidants which act as scavengers and remove the free radicals formed. These antioxidants also have the ability to prevent the process of peroxidation and improve the health of hepatocytes. Moreover, it is also observed that drugs that lower triglycerides such as fibrates improve hepatic biochemical parameters and tests (Parra, 2003; Hui-Mei et al., 2008).

The future prospective includes: 1) Characterization and quantification of pharmacologically active agents in the extracts and studies of their physiological effects such as; a) antibacterial activity; b) antifungal activity; c) anti-inflammatory activity; d) anticancerous activity; 2) formulation of drugs using active agents isolated from the extracts for trials.

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Full Length Research Paper

Traditional knowledge on plant resources of Ashezai and Salarzai Valleys, District Buner, Pakistan

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An ethnobotanical study was done in the Ashezai and Salarzai Valleys, District Buner, Pakistan. A total of 163 plant species belonging to 73 families were reported with the help of standardized questionnaires for their traditional, medicinal and economic uses. Out of these, 62 families were dicots; 8 monocots and two pteridophytes. Gymnosperms were represented by one family. Asteraceae had 16 spp. which was followed by Papilionaceae and Poaceae (each with 9 spp.); Lamiaceae, Moraceae and Rosaceae (each with 6 spp.); Apiaceae, Polygonaceae and Solanaceae (each with 5 spp.); Amaranthaceae, Brassicaceae, Chenopodiaceae and Euphorbiaceae (each with 4 species); Oleaceae and Salicaceae had 3 species each. The remaining families had less number of species. They included 110 medicinal plants, 51 fodder and forage species, 37 fuel wood species, 33 vegetable/pot-herb species, 22 fruit yielding species, 20 thatching/roofing species, 13 timber species, 18 ornamental species, 8 poisonous plants, 10 fencing/ hedges plants, 4 agricultural tools making species, 5 honeybee species. Deforestation, biotic interference and overgrazing are the responsible culprits for dwindling phytodiversity in the investigated area. This study might be helpful to ethnobotanists, conservationists, ecologist, pharmacologists, taxonomists, wild life and water shed managers as baseline data.

Key words: Ethnobotanical study, plant species, medicinal plants, traditional knowledge.

INTRODUCTION

Ethnobotany can be defined as the "study of direct interrelations between humans and plants so the results from various investigations reveal important pharmacological activities of plants which may be used in developing novel therapeutic agents". Herbal medicines play an important role in health care programs worldwide, especially in developing countries, because there are no major side effects. The residents of the investigated area mostly depend on cattle rearing and their products and on forest resources. Overuse of forest resources has placed them under intense biotic pressure, which poses potential danger for wildlife habitat and medicinal plants. Mood (2008) reported some floristic and ethnobotanical aspects of 37 families, 128 genera and 160 species in Birjand area near the Afghanistan border in eastern part of Iran. Ozturk et al. (2008) published some data on the toxic and fatal behavior of some poisonous plants widely distributed in Turkey and Northern Cyprus. Studies on ethnobotany have been conducted in India (Mustafa et al., 2000; Siddiqui et al., 2000). Many of such studies have also been done on the ethnobotany of various parts of District Swat (Sher et al., 2003, 2004; Hussain et al., 2004, 2005; Ibrar et al., 2007). In Pakistan, some ethnobotanical studies have also been carried out (Dastagir, 2001; Durrani et al., 2003; Gilani et al., 2003; Sher, 2005; Hussain et al., 2007; Ahmad and Husain, 2008; Usain

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et al., 2008; Ilahi, 2008; Ali and Qaiser, 2009; Qureshi et al., 2009). Ethnobotanical studies (Sher et al., 2011) and biological spectrum (Sher and Khan, 2007) of the vegetation of Chagharzai Valley, District Buner has been worked out.

It is evident from the review of literature that no work on the ethnobotany of Ashezai and Salarzai Valleys District Buner has been done so for. The present study reports the traditional utilization of some plants of the area, which might be helpful for the future workers, ecologist, pharmacologists, taxonomists; wild life and water shed managers in their efforts to develop this area (Table 1).

MATERIALS AND METHODS

District Buner lies between latitude 34°-11 to 34°-34 and longitude 72°-13 to 72°- 45. It is bounded by Swat and Shangla districts in the north, in the west by Malakand agency, in the south by Mardan and Swabi districts and in the east by Indus River. Vegetationally and climatically, the area can be classified as tropical, sub humid temperate with alpine glimpses at certain places. Elevation varies from 366 m in the south to 2911 m in the north. The geographical setting of the area provides habitat to rich diversity of plants especially medicinal and other economic species. A survey was conducted during June, 2009 to document the traditional uses of plants. Plant specimen were collected, dried and preserved properly. They were identified through available literature (Nasir and Ali, 1971-1992; Ali and Qaisar, 1992, 2009). The plants were classified according to their economic value (medicinal, fodder, vegetables, thatching, food, fuel wood) by interviewing and filling questionnaires from drug dealers, shopkeepers, timber dealers, fuel wood seller, local hakims and farmers but priority was given to local elderly people who were the real users and had a lot of information about the plants and their traditional uses. Literature survey and general observations provided more information. The plants were submitted to Botany Department, University of Peshawar, Khyber Pukhtunkhwa, Pakistan.

RESULTS AND DISCUSSION

The people of the area depend on agriculture, fuel wood selling, timber wood selling, livestock and other natural resources of the area for a living. The ethnobotanical information revealed that 163 plants were used for various purposes in the investigated area. The reported vegetation comprised 25 trees, 33 shrubs, 100 herbs, 4 climbers and one parasite (Figure 1). There were 110 plant species used as medicine. Some of the plants were used individually, while others in mixture. A plant species had single or multiple medicinal uses. The findings agree with those of Sher et al. (2011), Iqbal et al. (2011), Hussain et al. (2004, 2005), Ahmad and Husain (2008), Husain et al. (2008), Ilahi (2008), Ali and Qaiser (2009) and Qureshi et al. (2009) with respect to medicinal uses. Among such plants, *Acacia modesta*, *Acorus calamus*, *Adiantum incisum*, *Ajuga bractiosa*, *Ammi visnaga*, *Berberis lycium*, *Calotropis procera*, *Coriandrum sativum*, *Cucimus prophetarum*, *Fumaria indica*, *Mentha longifolia, Mentha spicata*, *Morus alba*, *Morus indica*, *Oxalis corniculata, Paeonia emodi*, *Plantago lanceolata*, *Punica granatum*, *Verbascum thapsus* and *Zizyphus* spp. were usually used against various ailments. Medicinal plants used in the local community were about 64% of the total reported species. Livestock rearing is an important traditional activity of the local people. Sher et al. (2011) reported that 33% of the total plant species are used as fodder in Chaghazai valley, District Buner. Aboriginals of the present investigated area showed milk, yogurt, butter and other traditional dairy products from livestock for their daily use. These products are taken with maize bread by all members of the family. Badshah and Hussain (2011) recorded that biotic pressure in the rangelands of district Tank is beyond the regeneration capacity of plant species because of free grazing. In the present investigated area, 51 (31%) plant species were used as fodder. The most commonly used plants were *A. modesta*, *Avena sativa*, *Brassica compestris*, *Bromus japonicus*, *Cymbopogon distans*, *Cynodon dactylon*, *Dichanthium annulatum*, *Medicago minima*, *Melia azedarach*, *M. alba*, *M. indica*, *Rubus ulmifolius*, *Sorghum helepense*, *Trifolium repens* and *Zizyphus* spp. Similar findings were also reported from other parts of Pakistan (Sher, 2005; Sher et al., 2003, 2004; Hussain et al., 2004, 2005; Gilani et al., 2003; Ibrar et al., 2007).

Acacia, Ailanthus altissima, *Dodonea viscosa*, *M. azedarach*, *Mallotus philippensis*, *Morus* spp., *Pinus roxburghii*, *Populus caspica* and *Olea ferruginea* are the most common plant species used as fuel wood in the investigated area. About twenty-two percent of the total recorded plant species are used as fuel wood. There is a high pressure on fuel wood species. Fuel consumption per home is often greater because of severe winters. Khan (2000) and Awan (2000) observed that the fuel wood is collected before the commencement of winter. Most of the economically important plants are reducing due to cutting. All these species which have high fuel value, are severely damaged. Sher et al. (2011) reported that timber species like *Melia* and even *Pinus roxburghii* are used as fuel wood in Chagharzai valley. Our findings agree with them. Hussain et al. (2004, 2005), Sher et al. (2003, 2004) and Ibrar et al. (2007) also reported the same findings for fuel wood species and this support our findings. Thirty-three species were used as vegetables and potherbs comprising about 20% of the total reported plants. These vegetables are collected by women and young girls and cooked for their own need only. *Allium cepa*, *A. sativum*, *Brassica compestris* and *Luffa cylindrica* are cultivated while the remaining plant species are wild. They include *Amaranthus viridis*, *Asparagus officinalis*, *Chenopodium album*, *Malva neglecta*, *Medicago polymorpha*, *Mentha longifolia*, *Portulaca olearaceae*. Sher (2005) and Gilani et al. (2003) also reported on many wild and cultivated vegetable plants and this agree with our findings.

There were 22 plant species (13%), yielding edible fruits. Among them species like *Berberis lycium*, *Celtis*

Table 1. Ethnobotanical uses of some plants of Ashezai and Salarzai Valleys, District Buner, Pakistan.

S/N	Plants and family	Local Name	Occurrance	Habit	Part used	Ethnobotanical uses
A.	**PTERIDOPHYTES**					
1.	**Family Adiantaceae**					
	1. Adiantum incisum Forsk.	Sumbal	W	H	Fronds	Medicinal
	2. Adiantum venustum D.Done	Sumbal	W	H	Fronds	Medicinal, ornamental
2.	**Family Equisetaceae**					
	3. Equisetum arvense L.	Bandakay	W	H	Shoot	Medicinal
B.	**GYMNOSPERMS**					
3.	**Family Pinaceae**					
	4. Pinus roxburghii Sergent	Nakhtar	W	T	Wood, branches, cones, Resins, leaves	Medicinal, fuel wood, TSR,
	5. Pinus wallichiana A.B.Jackson.	Pahoch	W	T	Wood, branches, cones, Resins, leaves	Fuel wood, TSR, timber
C.	**MONOCOTYLEDONS**					
4.	**Family Aceraceae**					
	6. *Acer cappadocicum* Gled.	Chinaranga	W	T	Wood	Fuel wood, ornamental
5.	**Family Alliaceae**					
	7. Allium cepa L.	Piyaz	C	H	Bulb	Medicinal, vegetables
	8. Allium sativum L.	Ooga	C	H	Bulb, leaves	Medicinal, vegetables
6.	**Family Amaryllidaceae**					
	9. Narcissus tazzeta L.	Gul-e-nargis	W	H	Flowers	Medicinal, Ornamental, Honey bees spp.
7.	**Family Araceae**					
	10. Acorus calamus Linn.	Skha waja	W	H	Whole plant	Medicinal
8.	**Family Iridaceae**					
	11. Iris ensata Thunb.	Oogakai	W	H	Root	Medicinal
9.	**Family Liliaceae**					
	12. Asparagus officinalis L.	Tindoray	W	H	Shoot	Vegetables, ornamental
	13. Asphodalus tenuifolius Cavan	Oogakay	W	H	Leaves	Vegetables
10.	**Family Musaceae**					
	14. Musa sapientum L.	Keela	C	H	Fruit	Medicinal, fruit
11.	**Family Poaceae**					
	15. Avena sativa L.	Jamdar	W	H	Shoot	Fodder
	16. Bromus japonicus Thumb ex Murr.	Jokai	W	H	Shoot	Fodder
	17. Cymbopogon distans (Nees ex Steud.) Watson	Sargaray	W	H	Whole plant	Fodder
	18. Cynodon dactylon L.	Kabal	W	H	Whole plant	Fodder, ornamental
	19. Dichanthium annulatum Stafl.	Naram wakha	W	H	Shoot	Fodder

Table 1. Contd.

	20. Poa annua L.	Wakha	W	H	Shoot	Fodder
	21. Saccharum bengalense Ritz.	Kahay	W	H	Stem, flowering scape	TSR
	22. Saccharum spontaneum L.	Shurghashay	W	H	Stem, flowering scape	TSR
	23. Sorghum helepense (L.) Bern.	Dadum	W	H	Shoot	Fodder
D.	**DICOTYLEDONS**					
12.	**Family Acanthaceae**					
	24. Justicia adhatoda Linn.	Baikar	W	SH	Leaves, roots	Medicinal, TSR, honey bees spp.
13.	**Family Amaranthaceae**					
	25. Achyranthus aspera L.	Ghishkay	W	H	Whole plant	Medicinal
	26. Amaranthus caudatus L.	Chalwairay	W	H	Whole plant	Vegetables
	27. Amaranthus spinosa L.	Chalwairay	W	H	Whole plant	Medicinal, vegetables
	28. Amaranthus viridis Linn.	Gunhar	W	H	Shoot, Root	Medicinal, vegetables
14.	**Family Apiaceae**					
	29. Ammi visnaga (L.) Lam.	Spairkai	W	H	Dry fruit	Medicinal
	30. Coriandrum sativum L.	Dhanyal	C	H	Leaves, fruit	Medicinal, vegetables
	31. Eryngium biebersteinianum L.	Ali kanda	W	SH	Shoot	Medicinal, fodder
	32. Foeoniculum vulgare Miler.	Kaga	C	H	Leaves, seeds	Medicinal
	33. Lespedeza juncea (L.F.) Persoon	Oormaray	W	H	Shoot, leaves	Medicinal, fodder
15.	**Family Apocynaceae**					
	34. Caralluma edulis Edgew.	Pamunkay	W	H	Whole plant	Medicinal, vegetables
	35. Nerium indicum Mill.	Gundairay	W	SH	Whole plant	TSR, ornamental, poisonous
16.	**Family Araliaceae**					
	36. Hedera helix L.	Prewatai	W	Cl	Leaves	Fodder
17.	**Family Asclepiadaceae**					
	37. Calotropis procera (wild) R.Br.	Spalmay	W	SH	Whole plant, latex	Medicinal, poisonous
18.	**Family Asteraceae**					
	38. Achillea millifolium L.	Jarai	W	H	Whole plant	Medicinal, fodder
	39. Artimisia maritima L.	Juakay	W	H	Shoot	Medicinal
	40. Artimisia scoparia Walds & Kit.	Tarkha	W	H	Leaves	Medicinal
	41. Artimisia vulgaris L	Tarkha	W	H	Leaves, shoot	Medicinal, fodder, Ornamental
	42. Calendula arvensis L.	Zair Gulae	W	H	Flowers, leaves	Medicinal
	43. Calendula officinalis L.	Zair Gulae	C	H	Flowers, shoot	Medicinal
	44. Cichorium intybus L.	Kasni	W	H	Whole plant	Medicinal
	45.Lactuca serriola L.	Salad	W	H	Whole plant	Medicinal
	46. Launea procumbens Roxb.	Shodapai	W	H	Shoot, leaves	Fodder

Table 1. Contd.

	47. Onopordum acanthium L.	Wrijakai	W	H	Shoot, seeds	Fodder
	48. Sonchus arvensis L.	Shodapai	W	H	Whole plant	Fodder
	49. Sonchus asper L.	Shodapai	W	H	Whole plant	Fodder
	50. Sonchus auriculata L.	Shodapai	W	H	Whole plant	Fodder
	51. Tagetus minuta L.	Hamisha	W	H	Flowers	Ornamental
	52. Taraxacum officinale Weber.	Zair gulai	W	H	Flowering, shoots	Medicinal
	53. Xanthium strumarium L.	Ghishkay	W	SH	Leaves	Medicinal
19.	**Family Balsaminaceae**					
	54. Impatiens balsamina L.	Gul-e-mehandi	W	H	Whole plant	Medicinal
20.	**Family Berberidaceae**					
	55. Berberis lycium Royle.	Kwaray	W	SH	Leaves, fruit Bark	Medicinal, fruit, fencing and hedging
21.	**Family Betulaceae**					
	56. Alnus nitida (Spach) E.	Gairay	W	T	Wood	Fuel wood, Agri. Tools
22.	**Family Brassicaceae**					
	57. Brassica compestris L.	Sharsham	C	H	Leaves, seeds Inflorescence	Fodder, Vegetables
	58. Capsella bursa-pestoris Medic.	Bambaisa	W	H	Seeds	Medicinal
	59. Eruca sativa L.	Jamama	W	H	Leaves, seeds	Medicinal, Vegetables
	60. Nasturtium officinale R.Br.	Talmeera	W	H	Shoot	Medicinal, Vegetables
23.	**Family Buxaceae**					
	61. Buxus wallichiana Baill.	Shamshad	W	SH	Whole plant	Medicinal, Fuel wood, TSR, Poisonous
24.	**Family Cactaceae**					
	62. Opuntia dilleni Haw.	Zaqoom	W	SH	Phylloclade, fruit	Medicinal, fruit, fencing and hedging
25.	**Family Caesalpinaceae**					
	63. Bauhinia variegata L.	Kulyar	W/C	T	Wood, bark Flower buds	Medicinal, fuel wood, vegetables, TSR, timber, ornamental
26.	**Family Canabanaceae**					
	64. Cannabis sativa L.	Bhang	W	SH	Leaves Flowers	Medicinal
27.	**Family Caryophyllaceae**					
	65. Sielene conoidea L.	Mangotay	W	H	Shoot, fruit Seed	Fodder, vegetables
	66. Stellaria media (L.) Cry.	-----	W	H	Whole plant	Fodder

Table 1. Contd.

28.	**Family Celastraceae**					
	67. Gymnosporia royleana Wall ex Lawson	Soor Azghay	W	SH	Whole plant	Fodder, Fuel wood, Fencing &Hedging
29.	**Family Chenopodiaceae**					
	68. Chenopodium album L.	Sarmai	W	H	Leaves	Medicinal, Vegetables
	69. Chenopodium ambrosioides L.	Kharawa	W	H	Fruits, Leaves	Medicinal
	*70. Chenopodium botrys*L.	Skha,kharawa	W	H	Shoot	Medicinal
	71. Chenopodium murale L.	Chalwairay	W	H	Shoot	Fodder, Vegetables
30.	**Family Commelinaceae**					
	72. Commelina albescens Hassak	Pulpolakay	W	H	Whole plant	Medicinal
31.	**Family Convulvuaceae**					
	73. Convovulus arvensis L.	Prewati	W	Cl	Whole plant, Root	Medicinal, Fodder
32.	**Family Cucurbitaceae**					
	74. Cucimus prophetarum L.	Kalkunday	W	H	Fruits	Medicinal, Poisonous
	75. Luffa cylindrical (L.) Roem.	Toorai	C	Cl	Fruits	Vegetables
33.	**Family Cuscutaceae**					
	76. Cuscuta reflexa Roxb.	Maraz bootay	W	P	Whole plant	Medicinal
34.	**Family Ebenaceae**					
	77. Diospyrus kaki L.	Toor Amluk	W /C	T	Wood, Fruit	Fodder, Fuel wood, Fruit
	78. Diospyrus lotus L.	Ziar Amluk	C	T	Wood, Fruit, Leaves	Fuel wood, Fruit
35.	**Family Elaegnaceae**					
	79. Elaegnus umbellata Thumb	Ghanamranga	W	SH	Wood, Fruit	Medicinal, Fuel wood, Fruit
36.	**Family Ericaceae**					
	80. Rhododenron arborium Smith.	Gul-e-nameer	W	T	Wood, Flower	Medicinal, Fuel wood, ornamental
37.	**Family Euphorbiaceae**					
	81. Euphorbia helioscopia Mewski.	Piryano doolai	W	H	-	Poisonous
	82. Euphorbia prostrata L.	Warmagha	W	H	Whole plant	Medicinal
	83. Mallotus philippensis Muell.	Kambeela	W	SH	Wood, fruits	Medicinal, fuel wood
	84. Riccinis communis L.		W	T	Fruits	Medicinal
38.	**Family Fumariaceae**					
	85. Fumaria indica (Hsskn) H.N.	Papra	W	H	Shoot	Medicinal
39.	**Family Juglandaceae**					
	86. Juglans regia L.	Ghuz	C	T	Nuts, Bark, leaves, Wood	Medicinal, fruit, timber

Table 1. Cont.

40.	**Family Lamiaceae**					
	87. Ajuga bractiosa Wall. Benth.	Khwaga bootei	W	H	Whole plant	Medicinal
	88. Ajuga parviflora Benth.	Tarkha bootei	W	H	Whole plant	Medicinal
	89. Mentha longifolia (L.) Huds	Velanai	W	H	Leaves, inflorescence	Medicinal, vegetables
	90. Mentha spicata L.	Poodina	W	H	Leaves, inflorescence	Medicinal, vegetables
	91. Ocimum basilicum L.	Kashmalu	W	H	Flowers, seeds	Medicinal
	92. Otostegia limbata Bth.	Pishkanar	W	SH	Whole plant	Fuel wood, fencing and hedging
41.	**Family Malvaceae**					
	93. Malva neglecta Waller.	Panaruk	W	H	Whole plant, leaves	Medicinal, vegetables
	94. Malva officinalis **(L.) Schimp. & Spenn.**	Panaruk	W	H	Whole plant	Medicinal, vegetables
42.	**Family Meliacea**					
	95. Cedrella serrata Royle.	Meem	W	T	Bark, leaves	Medicinal, TSR
	96. Melia azedarach L.	Shandai	W/C	T	Wood, leaves, bark Fruit	Medicinal, fodder, fuel wood, TSR, timber
43.	**Family Menispernaceae**					
	97. Tinospora cordifolia (DC.) Meirs	Gilo	W	Cl	Stem	Medicinal, ornamental
44.	**Family Mimosaceae**					
	98. Acacia modesta Wall.	Palosa	W	T	Leaves, gum, branches flowers, wood, ashes	Medicinal, fodder, fuel wood, honey bees spp.
	99. Acacia nilotica (L.) Delile.	Kikar	W	T	Wood, leaves, gum	Medicinal, fodder, fuel wood, timber, agricultural tools
45.	**Family Moraceae**					
	100. Ficus carica L.	Baghi Inzar	W/C	T	Wood, leaves, fruit Latex	Medicinal, fodder, fuel wood, fruit
	101. Ficus palmata Forssk.	Inzar	W/C	T	Wood, leaves, fruit latex	Medicinal, fodder, fuel wood, fruit
	102. Ficus recemosa L.	Oormal	W/C	T	Wood, leaves, latex fruit	Medicinal, fuel wood, fruit
	103. Ficus religiosa Roxb.	Peepal	W	T	Wood, bark, fruit	Medicinal, fuel wood
	104. Morus alba L.	Spin Toot	W/C	T	Wood, leaves, fruit branches	Medicinal, fodder, fuel wood, fruit, TSR, timber
	105. Morus indica L.	Toor Toot	W/C	T	Wood, leaves, fruit branches	Medicinal, fodder, fuel wood, fruit, TSR, timber
46.	**Family Myrsinaceae**					
	106. Myrsine africana L.	Marorang	W	SH	Shoot	Fodder

Table 1. Cont.

47.	**Family Nyctaginaceae**					
	107. Boerhaavia diffusa L.	Ensut	W	H	Root	Medicinal
	108. Mirabilis jalapa L.	Gul-e-Nazak	C	H	Leaves	Medicinal, ornamental
48.	**Family Oleaceae**					
	109. Jasminum humile L.	Rambail chambail	W/C	SH	Flowers, root	Medicinal, ornamental
	110. Jasminum offcinale L.	Rambail chambail	W/C	SH	Flowers, root	Medicinal, ornamental
	111. Olea ferruginea Royle.	Khoona	W/C	T	Wood, leaves, bark	Medicinal, fodder, fuel wood, agricultural tools
49.	**Family Oxalidaceae**					
	112. Oxalis corniculata L.	Tarookay	W	H	Leaves	Medicinal, vegetables
50.	**Family Paeoniaceae**					
	113. Paeonia emodi Wall. Hkf.	Mamekh	W	H	Rhizome, Roots	Medicinal
51.	**Family Papaveraceae**					
	114. Argimone maxicana L.	Wild poppy	W	H	Shoot, Flowers	Fodder, ornamental
	115. Papaver nudicaule L.	Zangali kashkash	W	H	Flowers, Capsule	Medicinal
52.	**Family Papilionaceae**					
	116. Indigofera heterantha L.	Kainta	W	SH	Leaves, wood, branches	Fodder, fuel wood, TSR
	117. Lathyrus aphaca L.	Kurkamanay	W	H	Shoot	Fodder, Vegetables
	118. Lathyrus cicera L.	Wara chilo	W	H	Shoot	Fodder, Vegetables
	119. Lathyrus pratensis L.	Chilo	W	H	Shoot	Fodder, Vegetables
	120. Lathyrus sativus L.	Ghata Chilo	W	H	Shoot	Fodder, Vegetables
	121. Medicago minima (L.) Grub.	Shpaishtay	W	H	Shoot	Fodder, Vegetables
	122. Medicago polymorpha L.	Shpaishtay	W	H	Shoot	Fodder, Vegetables
	123. Vicia sativa L.	Chilo	W	H	Whole plant	Fodder, Vegetables
	124. Trifolium repens L.	Shautal	W/C	H	Shoot, seeds	Medicinal, Fodder , Vegetables
53.	**Family Plantaginaceae**					
	125. Plantago lanceclata L.	Jabai	W	H	Leaves, seeds	Medicinal
	126. Plantago major L.	Jabai	W	H	Leaves, seeds	Medicinal
54.	**Family Plantinaceae**					
	127. Platanus orientalis L.	Chinar	W/C	T	Wood, bark	Medicinal, Fuel wood, TSR, Timber
55.	**Family Polygonaceae**					
	128. Bistorta amplexicaulis (D.Don) Green	Tarva panra	W	H	Shoot	Medicinal
	129. Polygonum barbatum L.	Polpulak	W	H	Whole plant	Poisonous
	130. Polygonum serrulatum Lagasca	Polpulak	W	H	Leaves	Medicinal

Table 1. Contd.

	131. Rumex dentatus L.	Shulkhay	W	H	Leaves	Medicinal, vegetables
	132. Rumex hastatus L.	Tarookay	W	H	Leaves	Medicinal
56.	**Family Portulacaceae**					
	133. Portulaca olearaceae L.	Warkharay	W/C	H	Shoot	Medicinal, vegetables, ornamental
57.	**Family Punicaceae**					
	134. Punica granatum L.	Anar	W/C	T	Fruit bark, leaves	Medicinal, fruit
58.	**Family Ranunculaceae**					
	135. Caltha alba Jacq ex Comb.	Makhanpath	W	H	Whole plant	Medicinal,
	136. Ranunculus muricatus L.	Jaghagha	W	H	Whole plant	Medicinal, poisonous
59.	**Family Rhamnaceae**					
	137. Zizyphus jujuba Mill.	Baira	W/C	T	Wood, leaves, fruit bark	Medicinal, fuel wood, fruit, honey bees spp.
	138. Zizyphus nummularia (Burm. F.) Wight	Karkunda	W	SH	Leaves, fruit	Medicinal, fodder, fruit, fencing and hedging, honey bees spp.
60.	**Family Rosaceae**					
	139. Fragaria indica Andrew	Da zamakay toot	W	H	Fruit	Medicinal, fruit
	140. Potentilla nepalensis Hook.	Da ghar shalkhay	W	H	Root	Medicinal
	141. Pyrus pashia Ham ex. D. Done	Tangai	W/C	T	Wood, fruit	Medicinal, fuel wood, fruit
	142. Rosa webbiana Wall. Ex.Royle	Palwari	W	SH	Flowers, branches	Ornamental, fencing and hedging, honey bees spp.
	143. Rubus fruticosus Hkf none L.	Karwara	W	SH	Leaves, fruit	Medicinal, fodder, fruit, fencing and hedging
	144. Rubus ulmifolius Schott.	Goraj	W	SH	Leaves, fruit	Medicinal, fodder, fruit, fencing and hedging
61.	**Family Rubiaceae**					
	145. Gallium aparine L.	-	W	H	Whole plant	Fodder
62.	**Family Rutaceae**					
	146. Zanthoxylum aromatum D.C.	Dambara	W	SH	Wood, fruit	Medicinal, fuel wood, fruit, fencing and hedging
63.	**Family Salicaceae**					
	147. Populus caspica Bornm.	Spairdar	W/C	T	Wood, branches, leaves	Fodder, fuel wood, TSR, timber
	148. Salix babylonica L.	Wala	W/C	T	Wood	Fuel wood, TSR, timber
	149. Salix tetrasperma Roxb.	Wala	W/C	T	Wood	Fuel wood, TSR, timber

Table 1. Contd.

64.	**Family Sapindaceae**					
	150. Dodonea viscosa (L.) Jacq.	Ghwarskay	W	SH	Wood, bark	Medicinal, fuel wood, TSR, ornamental, fencing and hedging
65.	**Family Scrophulariaceae**					
	151. Verbascum thapsus L.	Khar ghaug	W	H	Leaves	Medicinal
66.	**Family Simarubaceae**					
	152. Ailanthus altissima (Mill) Swingle	Asli Bhikyanra	W/C	T	Wood, leaves, bark, gum resins	Medicinal, fodder, fuel wood, TSR, timber
67.	**Family Solanaceae**					
	153. Atropa accuminata Royle ex Mier	Barghak	W	H	Leaves	Medicinal
	154. Datura innoxia Mill.	Batora	W	H	Leaves, seeds	Medicinal, poisonous
	155. Solanum nigrum L.	Kamachoo	W	H	Shoot, leaves	Medicinal
	156. Solanum surratense Burm.f	Maraghony	W	H	Whole plant	Medicinal
	157. Withania somnifera (L.) Dunal.	Kutilal	W	SH	Leaves, fruits, roots	Medicinal
68.	**Family Thymeliaceae**					
	158. Daphne oloides Scurb.	Laighonay	W	SH	Wood, fruit	Medicinal, fuel wood, fruit
69.	**Family Tiliaceae**					
	159. Grewia optiva Drum.ex.Burret.	Pastawoonay	W	T	Leaves, bark, branches	Fodder
70.	**Family Ulmaceae**					
	160. Celtis australis L.	Tagha	W	T	Wood, leaves, fruits	Medicinal, fodder, fuel wood, fruit, agricultural tools
71.	**Family Urticaceae**					
	161. Debrrgesia salicifolia D.Done.	Ajalai	W	SH	Wood, fruit	Fuel wood
72.	**Family Verbenaceae**					
	162. Vitex negundo L.	Marwandai	W	SH	Branches, leaves, flowers	Medicinal, fuel wood, TSR
73.	**Family Zygophyllaceae**					
	163. Tribulus terrestris L.	Markundai	W	H	Fruits, roots	Medicinal

TSR = Thatching, sheltering and roofing spp.; W = Wild; C = Cultivated; H = Herb; SH = Shrub; T = Tree; Cl = Climber; P = Parasite.

australis, *Rubus ulmifolius*, *Zizyphus nummularia, Ficus cairica*, *Ficus palmata*, *Fragaria indica* grow wild. Plant species like *Diospyrus kaki*, *Diospyrus lotus*, *Juglans regia*, *M. alba*, *Punica granatum*, *Pyrus pashia* and *Zizyphus jujuba* are cultivated. Sher et al. (2011) reported the same species from their study area therefore our findings are in line with them. The locals use 20 (12%) plant species for thatching, sheltering and roofing. The leaves and branches of *Ailanthus altissima*, *Dodonea viscosa*, *Indigofera heterantha*, *Justicia adhatoda*, *M. alba*, *M. indica*, *Saccharum spontaneum* and *Saccharum bengalense* are used for this purpose, thus supporting the studies of other workers (Hussain et al., 2004, 2005; Ibrar et al., 2007;

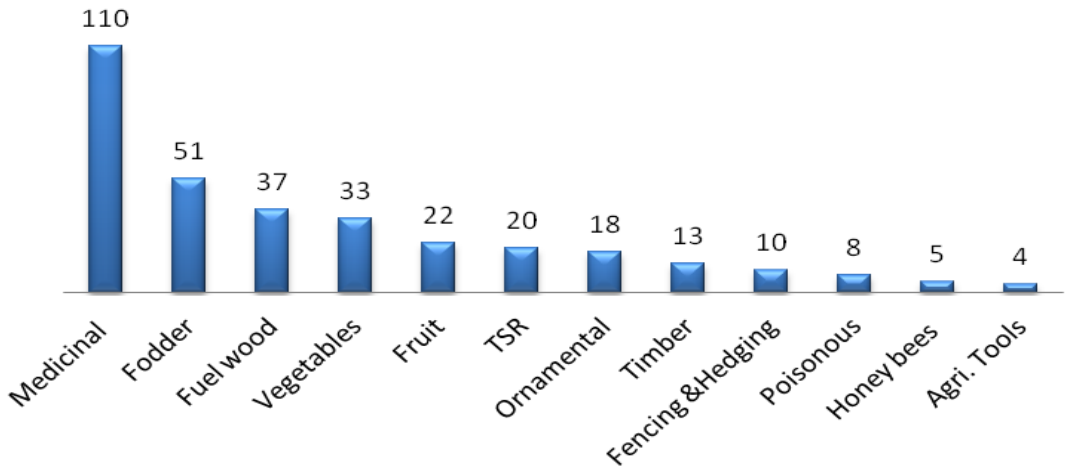

Figure 1. Percentage of plant species and their economic uses.

Sher et al., 2011). The plant species used as timber included *Ailanthus altissima*, *Juglans regia*, *Melia azedarach, Morus* spp. *Pinus roxburghii*, *Platanus orientalis* and *Salix* spp. comprising thirteen (7.9%) species of the total recorded plants. Sher et al. (2011) recorded that forests easily fulfill the requirements of the local people, but the activities of the timber maphia had greatly damaged the vegetation in Chaghazai Valley. Most of the timber species, in terms of density and frequency are decreasing due to illicit cutting. Our results also support their findings.

Some wild plant species like *Adiantum venustum*, *Artimisia vulgaris*, *Asparagus officinalis*, *Mirabilis jalapa*, *Narcissus tazzeta*, *Jasminum humile* and *Rosa webbiana* are used as ornamental plants having the potential for commercialization. These species comprised 11% of the total plants. Similar findings were reported in a number of studies in other parts of Pakistan (Sher et al., 2011; Ibrar et al., 2007; Hussain et al., 2004). Eight plant species (4.9%) including *Nerium indicum, Calotropis procera, Buxus wallichiana, Datura innoxia*, *Cucimus prophetarum, Euphorbia helioscopia* and *Polygonum barbatum* were considered poisonous to man, livestock or fish. These poisonous plants can be exploited as source of medicines. Livestock grazing is an important practice in the area therefore the people protects their crop fields by planting thorny, bushy or spiny plants around their crop fields. There were ten plants used for the purpose of fencing and hedging in the area. It comprised 6.1% of the total plants reported. Some important plants used for this purpose were: *Berberis lycium*, *Gymnosporia royleana*, *Opuntia dilleni*, *Rosa webbiana*, *Rubus* spp., *Zanthoxylum aromatum* and *Zizyphus nummularia*. The study also recorded that four species (2.4%) were used for making agricultural tools including ploughs, sticks, sickle handles, axe handles, pullies, knife handles and other agricultural appliances. *Acacia nilotica*, *Celtis australis*, *Alnus nitida*, and *Olea ferruginea* are important in this respect. Honeybees visit five species (3%). *A. modesta*, *J. adhatoda*, *Sarcococa saligna* and *Zizyphus* spp. are important. Honey obtained from *A. modesta* and *Zizyphus* is extensively used in the preparation of traditional medicines and sold at higher rates.

It is concluded that the area is under heavy biotic pressure in the form of deforestation and overgrazing, which has reduced regeneration of woody plants. Overgrazing, human population explosion, uprooting of medicinal plants by the local laypeople and other informal factors are responsible for habitat loss, soil erosion and proper functioning of ecosystems. There is dire need to conserve the biodiversity of the area in order to provide the resources and resource alternatives for the people's survival in future.

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Full Length Research Paper

Survival, growth and biomass production of *Moringa oleifera* provenances at Gairo inland plateau and Ruvu Coastal Region in Tanzania

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Survival, growth and biomass production was studied among 14 *Moringa oleifera* provenances at Gairo inland plateau-Morogoro, and Ruvu Coastal Region in Tanzania employing randomized complete block design with three replications. Growth assessment was done at six months intervals while biomass assessment was only done at 30 months. During final assessment, untransformed survival at Gairo site ranged from 65.33% for Chikwawa/Domasi (Malawi) to 98.67% for Ihumwa (Tanzania) while at Ruvu site, it ranged from 92% for PKM 2 (India) to 100% for Chikwawa/Domasi (Malawi), Mahalapye (Botswana) and Mbololo 472-029/03 (Kenya) provenances. Height ranged from 2.66 m for Makhanga (Malawi) to 5.04 m for Maun (Botswana) at Gairo site and from 4.82 m for Makhanga (Malawi) to 8.16 m for Maun (Botswana) at Ruvu site. Breast height diameter ranged from 2.80 cm for Makhanga (Malawi) to 6.07 cm for Mahalapye (Botswana) at Gairo site and from 5.18 cm for PKM 3 (India) to 8.58 cm for Maun (Botswana) at Ruvu site. Provenances Mahalapye and Maun (Botswana), Ihumwa (Tanzania), Mbololo 472-029/03 (Kenya) and PKM 1 (India) at Gairo site and Maun and Mahalapye (Botswana), Mbololo 472-029/03 (Kenya) and Ihumwa (Tanzania) at Ruvu site are recommended for planting at these and similar sites.

Key words: *Moringa,* survival, growth, biomass production, Tanzania.

INTRODUCTION

Moringa is the only genus in the family Moringaceae. This genus comprises 13 species, all of which are trees that grow in tropical and sub-tropical climates. *Moringa* is drought resistant and can be grown in a wide variety of poor soils, even barren ground, with soil pH between 4.5 and 9.0. The most popular species is *Moringa oleifera*, a multi-purpose tree originally from India and now found in most tropical countries (Africa, Asia and America). Some records indicate that the species is native to the sub-Himalayan tracts of India, Pakistan, Bangladesh and Afghanistan (Fahey, 2005). Fahey (2005) further indicated that the tree species is locally known by many names like horseradish tree, drumstick tree, benzolive tree, kelor, marango, mlonge, moonga, mulangay, nébéday, saijhan, sajna or ben oil tree. The species is easy to reproduce and its growth is very fast that have raised growing international interest due to its social, economic and environmental importance which can benefit humans and animals nutritionally, economically and as an energy source.

Moringa is an important food source in many countries. In India, *Moringa* pods are widely consumed and

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Table 1. Sources of *M. oleifera* provenances planted at Gairo and Ruvu sites, Tanzania.

Provenance	Collection site/country/state
Chikwawa/Domasi	Chikwawa/Domasi, Malawi
Makhanga	Makhanga, Malawi
Ngabu	Ngabu, Malawi
Mangochi	Mangochi, Malawi
Mtakataka/Dedza	Mtakataka/Dedza, Malawi
Mahalapye	Mahalapye, Botswana
Maun	Maun, Botswana
PKM 1	India
PKM 2	India
PKM 3	India
Jafna/Jaffana	India
Honduras	Honduras
Mbololo 472-029/03	Mbololo, Kenya
Ihumwa	Ihumwa, Tanzania

Plantations exist to produce pods for export, fresh and tinned, to overseas consumers. In West Africa, *Moringa oleifera* leaves are commonly used to make sauces. *Moringa stenopetala* leaves are the staple food of the Konso people in Ethiopia. Studies have shown *M. oleifera* leaves to be an excellent source of vitamins, minerals and protein: perhaps more than any other tropical vegetable. Many programmes use *Moringa* leaves to fight against malnutrition and its associated diseases including blindness. Fragrant/tender flowers of *Moringa* are also used as a source of vegetables and bee forage (Mbuya et al., 1994), twigs with leaves as fodder, green leaves as mulch and solid wood as energy source (FAO, 1988; Morton, 1991; D'Souza and Kulkarmi, 1993).

Moringa seeds contain a cationic polyelectrolyte that has proved efficient in water treatment, as a substitute to aluminium sulphate and other flocculents (Evans, 1991; Mayer and Stelz, 1993; Olayemi and Alabi, 1994; Folkard and Sutherland, 1996). Oil extracted from the seeds is an excellent edible vegetable oil and is also useful in the cosmetics industry. The press cake left after oil extraction from the seeds can be used as a soil conditioner or as fertilizer and forage, it has the potential use as protein supplement for livestock (ruminants) (Sá nchez et al., 2006) and poultry (Evans, 1991; Olayemi and Alabi, 1994; Folkard and Sutherland, 1996).

Although *M. Oleifera* is wide spread in many African countries its original sources is unknown (Jahn, 1988a, b, c; Folkard and Sutherland, 1996; Panga, 2002). The material therefore, is most likely of narrow genetic base. Also, the knowledge about genetic improvement and proper management of this valuable multipurpose tree species is limited. Given dangers of planting materials based on a narrow genetic base, there is a need to evaluate different provenances of this species with a view of identifying the provenances that maximize productivity in various parts of Tanzania. In addition to increasing wood biomass productivity, pod, seed and leaf quality, broadening the genetic base will serve as an insurance against pests, diseases and climatic fluctuations. This study therefore evaluated *M. oleifera* provenances in terms of survival, height, diameter growth and biomass production.

MATERIALS AND METHODS

The study sites

The study was carried out at two contrasting sites: Gairo and Ruvu sites with established replicate trials in each. Gairo site is located in Gairo District, Morogoro Region (36° 45' E; 6° 0° S; 1 300 m a. s. l.) along the Morogoro-Dodoma highway about 130 km from Morogoro town and 140 km from Dodoma. Rainfall is poorly distributed, and varies from year to year. The average annual rainfall is around 499 mm, most of which falls between November and May (Herbert et al., 2002). The geology of the site is Usagaran system. The soil is generally classified as Haplic Lixisols (Msanya and Msaky, 1994). The soil properties of the study site are as described by Mugasha et al. (2000). The soil has low inherent fertility. The soil texture is sandy clay loam with pH in the upper 50 cm soil depth varying from 6.1-6.3, total nitrogen 0.11-0.16% and Bray I available phosphorus 0.18-3.38 ug/g. The natural vegetation found around the site consists mainly of shrubs and few scattered miombo tree species.

Ruvu site is located at Kibaha District, Coast Region (6° 33' - 6° 43'; 38° 48' S - 39v 03' E) some 60 km from Dar-es-Salaam city along Dar-es-Salaam - Morogoro highway. It is within low elevations some 80 m a. s. l. The area has mean rainfall of 900 mm per annum falling in average of 81 days (Maghembe, 1979) with irregular pattern. Heavy rains fall between March and May and light rains between November - December. Temperatures are always high tending to be the highest in January and on average the temperature ranges between 23 and 27°C having occasional minima as low as 18°C and maxima of 33°C. Soils vary substantially over short distances. They are free draining, primarily sandy, sandy loam and gravel. Soil pH varies with soil depth. The top 50 cm has pH predominantly ranging from 5 - 6.5, increasing with increase in soil depth reaching extremes of pH 9.4 at 72 cm below surface (Holmes, 1988). The common natural vegetation found at Ruvu can be described as semi deciduous open to partly closed woodland to fairly open evergreen thicket and riverine forest. Main genera include, *Pteleopsis, Strichnos, Combretum, Albizia, Brachystegia, Jubernadia, Vitex, Afzelia* and *Dalbergia*.

Source of provenances

Fourteen (14) *M. oleifera* provenances were studied in each site (Table 1). Potted seedlings of the provenances were raised at Gairo and Ruvu nurseries using standard cultural techniques (Forest Division, 1982).

Experimental design

Trials were planted in a randomised complete block design (RCBD) with three replications. Each plot represented a provenance planted at 5 x 5 rows in a contiguous arrangement at an espacement of 2 x 2 m. Plots measured 8 x 8 m and contained 25 trees. Distances between blocks were 4 m. Each block had one buffer row planted at the same spacing.

Field procedures

Sites were prepared by clearing all vegetation using hand hoes followed by ploughing and pitting (pit size: 30 x 30 cm). Planting was done in January and March 2006 at Gairo and Ruvu sites respectively. Weeding was done three times during the rainy season and once during the dry season.

Data collection

Assessments were carried out at ages of 6, 12, 18, 24 and 30 months after planting. During the first assessment, survival, root collar diameter 30 cm above ground (RCD) and height (HT) were assessed while in subsequent assessments diameter at breast height (Dbh) was also measured. Biomass production was measured at final assessment. Height was measured using calibrated height measuring pole while RCD and Dbh were measured to the nearest 0.01 cm using a veneer calliper.

Data analysis

All statistical analyses were carried out using General Linear Model (GLM) of Statistical Analysis Systems (SAS) (SAS Inst. Inc., 1991). For all statistical analysis, a fixed effect model was fitted (equation 1) and a type III SS analysis was carried out. All data were subjected to analysis of variance (ANOVA) using plot means. Data for percentage survival was arcsine transformed prior to analysis to remove bias (Sokal and Rohlf, 1969). For significantly different provenance means, the Duncan's multiple range test (DMRT) was used for separating means (Gomez and Gomez, 1983) at 5% probability level.

The general linear model was denoted as:

Y = replication + provenance + error (1)

Where, Y = the measurement

To identify the best and the worst overall performing provenance at final assessment, ordinal ranking was performed. This was done as follows; for each significant variable evaluated, provenances were assigned ranks from the best (assigned 1 point) to the worst (assigned 14 points) performing provenance. Thereafter, ranks were added, averaged, and the overall score was taken as a basis of the overall provenance ranking.

RESULTS

Survival

The results for tree survival for different *M. oleifera* provenances planted at Gairo and Ruvu sites are presented in Tables 2 and 3, respectively. The differences in survival were significant ($P<0.05$) throughout the assessment period at Gairo site while at Ruvu site provenances did not differ significantly ($P>0.05$).

During the last assessment occasion (30 months after planting), untransformed survival at Gairo site ranged from 61.33% for Chikwawa/Domasi (Malawi) to 98.67% for Ihumwa (Tanzania) while at Ruvu site untransformed survival ranged from 92% for PKM 2 (India) to 100% for Chikwawa/Domasi (Malawi), Mahalapye (Botswana) and Mbololo 472-029/03 (Kenya).

Root collar diameter

The results of tree RCD for different *M. oleifera* provenances planted at Gairo and Ruvu sites are presented in Table 4. Provenances differed significantly ($P<0.05$) in RCD at both sites in all months of assessment. RCD ranged between 5.36 cm for Mangochi (Malawi) and 8.91 cm for Mahalapye (Botswana) at Gairo site and 7.43 cm for PKM 3 (India) and 10.67cm for Maun (Botswana) at Ruvu site during the final assessment occasion.

Diameter at breast height

The results of tree Dbh for both sites are presented in Table 5. Provenances differed significantly ($P<0.05$) in Dbh at both sites in all months of assessment. Dbh ranged between 2.86 cm for Makhanga (Malawi) and 6.07 cm for Mahalapye (Botswana) at Gairo site and 5.28 cm for PKM 3 (India) and 8.58 cm for Maun (Botswana) at Ruvu site during the final assessment occasion.

Height growth

Trends for height growth of *M. oleifera* provenances for Gairo and Ruvu sites are as shown in Table 6. Significant variation ($P<0.05$) in height development was observed at all assessment occasions. The mean height ranged between 2.66 m for Makhanga (Malawi) and 5.04 m for Maun (Botswana) at Gairo site and 4.82 m for Makhanga (Malawi) and 8.16 m for Maun (Botswana) at Ruvu site during the final assessment occasion (30 months after planting).

It is interesting to note that at many assessment occasions in both sites, Maun (Botswana) showed superiority in height growth even though no one single provenance maintained one rank throughout the period. Makhanga (Malawi) provenance showed poor height growth at all assessment occasions in both sites.

Biomass production

The results for stem and foliar biomass for *M. oleifera* provenances at Gairo and Ruvu sites are presented in Table 7. Provenances differed significantly ($P<0.05$) in biomass production at Gairo site during the last assessment occasion while at Ruvu site provenances did not differ significantly ($P>0.05$) in biomass production. Stem and foliar biomass at Gairo site ranged from 5 901.08 kg ha^{-1} for Mangochi (Malawi) to 24 733.60 kg ha^{-1} for PKM 2 (India) and 491.02 kg ha^{-1} for Ngabu (Malawi) to 4 407.18 kg ha^{-1} for Maun (Botswana), respectively. At

Table 2. Survival and transformed survival of 6, 12, 18, 24 and 30 months old *M. oleifera* provenances planted at Gairo, Morogoro, Tanzania.

Species/provenances	Age (months)									
	6		12		18		24		30	
	UTSv	TSv	UTSv	TSv	UTSv	TSv	UTSv	TSv	UTSv	TSv
Chikwawa/Domasi - Malawi	*61.33 (10.41)	51.92^{d} (6.35)	61.33 (10.41)	51.92^{d} (6.35)	61.33 (10.41)	51.92^{d} (6.35)	61.33 (10.41)	51.92^{d} (6.35)	61.33 (10.41)	51.92^{e} (6.35)
Makhanga - Malawi	85.33 (2.67)	67.63^{c} (2.10)	85.33 (2.67)	67.63^{cd} (2.10)	84.00 (2.31)	66.53^{cd} (1.82)	82.67 (1.33)	65.43^{cd} (0.99)	78.67 (2.67)	62.59^{de} (1.92)
Ngabu - Malawi	89.33 (4.81)	71.82^{bc} (4.42)	88.00 (4.00)	70.19^{bc} (3.38)	88.00 (4.00)	70.19^{bcd} (3.38)	88.00 (4.00)	70.19^{cd} (3.38)	85.33 (4.81)	67.99^{bcde} (3.82)
Mangochi - Malawi	86.67 (11.39)	73.86^{abc} (10.89)	85.33 (10.91)	72.23^{bc} (10.66)	84.00 (12.22)	71.45^{bcd} (11.37)	80.00 (12.22)	65.55^{cd} (8.91)	80.00 (12.22)	65.55^{cde} (8.91)
Mtakataka/Dedza - Malawi	97.33 (1.33)	82.31^{abc} (3.85)	97.33 (1.33)	82.31^{abc} (3.85)	97.33 (1.33)	82.31^{abc} (3.85)	97.33 (1.33)	82.31^{abc} (3.85)	96.00 (0.00)	78.46^{abcd} (0.00)
Mahalapye - Botswana	96.00 (0.00)	78.46^{abc} (0.00)	96.00 (0.00)	78.46^{abc} (0.00)	96.00 (0.00)	78.46^{abc} (0.00)	96.00 (0.00)	78.46^{abc} (0.00)	96.00 (0.00)	78.46^{abcd} (0.00)
Maun - Botswana	98.67 (1.33)	86.15^{ab} (3.85)	98.67 (1.33)	86.15^{ab} (3.85)	98.67 (1.33)	86.15^{ab} (3.85)	98.67 (1.33)	86.15^{ab} (3.85)	97.33 (2.67)	84.52^{ab} (5.48)
PKM 1 - India	90.67 (5.81)	75.67^{abc} (7.74)	90.67 (5.81)	75.67^{abc} (7.74)	90.67 (5.81)	75.67^{abc} (7.74)	90.67 (5.81)	75.67^{abc} (7.74)	90.67 (5.81)	75.67^{abcd} (7.74)
PKM 2 - India	98.67 (1.33)	86.15^{ab} (3.85)	98.67 (1.33)	86.15^{ab} (3.85)	98.67 (1.33)	86.15^{ab} (3.85)	98.67 (1.33)	86.15^{ab} (3.85)	97.33 (2.67)	84.52^{ab} (5.48)
PKM 3 - India	93.33 (4.81)	78.29^{abc} (6.81)	93.33 (4.81)	78.29^{abc} (6.81)	93.33 (4.81)	78.29^{abc} (6.81)	93.33 (4.81)	78.29^{abc} (6.81)	93.33 (4.81)	78.29^{abcd} (6.81)
Jafna/Jaffana - India	98.67 (1.33)	86.15^{ab} (3.85)	98.67 (1.33)	86.15^{ab} (3.85)	98.67 (1.33)	86.15^{ab} (3.85)	98.67 (1.33)	86.15^{ab} (3.85)	97.33 (2.67)	84.52^{ab} (5.48)
Honduras - Honduras	92.00 (2.31)	73.92^{abc} (2.53)	90.67 (1.33)	72.29^{bc} (1.28)	90.67 (1.33)	72.29^{abc} (1.28)	89.33 (1.33)	71.01^{bcd} (1.28)	88.00 (0.00)	69.73^{abcde} (0.00)
Mbololo 472-029/03 - Kenya	96.00 (4.00)	83.24^{abc} (6.76)	96.00 (4.00)	83.24^{ab} (6.76)	96.00 (4.00)	83.24^{ab} (6.76)	94.67 (3.53)	79.40^{abc} (5.87)	94.67 (3.53)	79.40^{abcd} (5.87)
Ihumwa - Tanzania	100.00 (0.00)	90.00^{a} (0.00)	100.00 (0.00)	90.00^{a} (0.00)	98.67 (1.33)	86.15^{ab} (3.85)	98.67 (1.33)	86.15^{ab} (3.85)	98.67 (1.33)	86.15^{a} (3.85)
P>F Ratio	0.0011	0.0038	0.0004	0.0013	0.0013	0.0032	0.0016	0.0008	0.0006	0.0032
RMSE	8.4784	9.2076	7.26	8.59	7.70	8.90	7.48	7.95	7.96	9.08
CV	9.24	11.87	7.90	11.02	8.33	11.31	8.19	10.41	8.84	12.07

*Means of individual species/provenance with standard error in parenthesis. Means of the same letter within the same column are not significantly different. UTSv, untransformed survival; TSv, transformed survival.

Table 3. Survival and transformed survival of 6, 12, 18, 24 and 30 months old *M. oleifera* provenances planted at Ruvu, Coast Region, Tanzania.

Species/provenances	Age (months)									
	6		12		18		24		30	
	UTSv	TSv	UTSv	TSv	UTSv	TSv	UTSv	TSv	UTSv	TSv
Chikwawa/Domasi - Malawi	* 100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)
Makhanga - Malawi	98.67 (1.33)	86.15[a] (3.85)	98.67 (1.33)	86.15[a] (3.85)	98.67 (1.33)	86.15[a] (3.85)	97.33 (2.67)	84.52[a] (5.48)	96.00 (2.31)	80.00[a] (4.87)
Ngabu - Malawi	98.67 (1.33)	86.15[a] (3.85)	98.67 (1.33)	86.15[a] (3.85)	98.67 (1.33)	86.15[a] (3.85)	98.67 (1.33)	86.15[a] (3.85)	98.67 (1.33)	86.15[a] (3.85)
Mangochi - Malawi	96.00 (2.31)	80.68[a] (4.87)	96.00 (2.31)	80.68[a] (4.87)	96.00 (2.31)	80.68[a] (4.87)	96.00 (2.31)	80.68[a] (4.87)	96.00 (2.31)	80.68[a] (4.87)
Mtakataka/Dedza - Malawi	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	98.67 (1.33)	86.15[a] (3.85)
Mahalapye - Botswana	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)
Maun - Botswana	98.67 (1.33)	86.15[a] (3.85)	98.67 (1.33)	86.15[a] (3.85)	98.67 (1.33)	86.15[a] (3.85)	96.00 (2.31)	80.68[a] (4.87)	93.33 (3.53)	77.77[a] (6.22)
PKM 1 - India	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	97.33 (2.67)	84.52[a] (5.48)	96.00 (2.31)	80.68[a] (4.87)
PKM 2 - India	96.00 (2.31)	80.68[a] (4.87)	96.00 (2.31)	80.68[a] (4.87)	96.00 (2.31)	80.68[a] (4.87)	94.67 (3.53)	79.40[a] (5.87)	92.00 (2.31)	79.40[a] (5.87)
PKM 3 - India	97.33 (1.33)	82.31[a] (3.85)	97.33 (1.33)	82.31[a] (3.85)	97.33 (1.33)	82.31[a] (3.85)	97.33 (1.33)	82.31[a] (3.85)	97.33 (1.33)	82.31[a] (3.85)
Jafna/Jaffana - India	100.00 (0.00)	90.00[a] (0.00)	98.67 (1.33)	86.15[a] (3.85)	98.67 (1.33)	86.15[a] (3.85)	96.00 (0.00)	78.46[a] (0.00)	96.00 (0.00)	78.46[a] (0.00)
Honduras - Honduras	96.00 (2.31)	80.68[a] (4.87)	96.00 (2.31)	80.68[a] (4.87)	96.00 (2.31)	80.68[a] (4.87)	96.00 (2.31)	80.68[a] (4.87)	94.67 (3.53)	79.40[a] (5.87)
Mbololo 472-029/03 - Kenya	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)	100.00 (0.00)	90.00[a] (0.00)
Ihumwa - Tanzania	98.67 (1.33)	86.15[a] (3.85)	98.67 (1.33)	86.15[a] (3.85)	98.67 (1.33)	86.15[a] (3.85)	98.67 (1.33)	86.15[a] (3.85)	98.67 (1.33)	86.15[a] (3.85)
P>F Ratio	0.1593	0.188	0.69	0.75	0.32	0.38	0.28	0.20	0.0251	0.1963
RMSE	2.23	5.48	3.01	5.92	1.91	5.09	2.72	6.01	2.79	6.38
CV	2.26	6.34	3.05	6.83	1.93	5.82	2.78	7.10	2.87	7.64

*Means of individual species/provenance with standard error in parenthesis. Means of the same letter within the same column are not significantly different. UTSv, untransformed survival; TSv, transformed survival.

Ruvu site stem and foliar biomass ranged from 15 796.17 kg ha^{-1} for Makhanga (Malawi) to 41 403.26 kg ha^{-1} for Mahalapye (Botswana) and 697.10 kg ha^{-1} for Makhanga (Malawi) to 1 917.21 kg ha^{-1} for Maun (Botswana) respectively.

Ordinal ranking

When the ranking of provenances in six tree variables (survival, mean height, RCD, Dbh, stem biomass and foliage biomass production) was computed, provenances were ranked as shown in Tables 8 and 9 for Gairo and Ruvu sites, respectively.

The most outstanding provenances were Mahalapye (Botswana), Maun (Botswana), Ihumwa (Tanzania), Mbololo 472-029/03 (Kenya) and PKM 2 (India) for Gairo site and Maun (Botswana), Mahalapye (Botswana), Mbololo 472-029/03 (Kenya) and Ihumwa (Tanzania) for Ruvu site.

DISCUSSION

Survival

General assessment at all occasions, showed good performance among provenances. All provenances in both sites except two at Gairo site had survival assessments above 80%. The significant differences in survival at Gairo site could be attributable to provenances difference in tolerance to arid/semi arid conditions of the study site (Edward et al., 2006). In this case provenances from Ihumwa (Tanzania), Mtakataka/Dedza (Malawi), Maun (Botswana), PKM 2 (India) and Jafna/Jaffana (India) appear to tolerate dry conditions than Chikwawa/-Domasi (Malawi). The excellent performance of these provenances is an indication of better adaptation to the site condition while the insignificant differences among provenances at Ruvu site implies that all provenances have adapted well in the sub-humid conditions of this

Table 4. Root collar diameter of 6, 12, 18, 24 and 30 months old *Moringa oleifera* provenances planted at Gairo site, Morogoro and Ruvu site, Coast Region, Tanzania.

Species/provenances	Gairo site					Ruvu site				
	Age (months)									
	6	12	18	24	30	6	12	18	24	30
Chikwawa/Domasi - Malawi	3.12^{c}	4.25^{cde}	5.67^{de}	6.12^{de}	6.63^{defg}	2.69^{def}	6.45^{cde}	8.40^{cde}	8.54^{cde}	8.81^{bcd}
	(0.07)	(0.12)	(0.20)	(0.13)	(0.53)	(0.21)	(0.33)	(0.34)	(0.12)	(0.48)
Makhanga - Malawi	2.14^{c}	3.16f	4.88^{ef}	5.28^{e}	5.73^{fg}	2.30f	5.88^{e}	7.77^{de}f	8.17^{de}	8.38^{cd}
	(0.16)	(0.18)	(0.47)	(0.45)	(0.56)	(0.24)	(0.52)	(0.89)	(0.74)	(1.15)
Ngabu - Malawi	2.41^{de}	3.51^{ef}	4.75^{ef}	5.46^{e}	6.19^{efg}	2.79^{cdef}	6.73^{bcde}	8.59^{bcd}	8.84^{bcd}	8.98^{bcd}
	(0.21)	(0.34)	(0.20)	(0.29)	(0.22)	(0.16)	(0.08)	(0.12)	(0.04)	(0.11)
Mangochi - Malawi	1.89^{e}	3.09^{f}	4.59f	5.15^{e}	5.36g	2.44f	6.58^{cde}	8.65^{bcd}	8.84^{bcd}	8.93^{bcd}
	(0.04)	(0.05)	(0.08)	(0.10)	(0.11)	(0.52)	(0.38)	(0.50)	(0.63)	(0.65)
Mtakataka/Dedza - Malawi	2.82^{cd}	4.10^{de}	6.35^{cd}	6.82^{bcd}	7.48^{bcde}	2.51^{e}f	7.07^{bcd}	9.36^{abc}	9.57^{abc}	10.12^{ab}
	(0.20)	(0.03)	(0.27)	(0.11)	(0.34)	(0.22)	(0.40)	(0.46)	(0.47)	(0.44)
Mahalapye - Botswana	4.01^{a}	6.11^{a}	7.94^{a}	8.31^{a}	8.91^{a}	3.40^{abcd}	7.29^{abc}	9.19^{bc}	9.34^{abc}	9.38^{abc}
	(0.08)	(0.12)	(0.21)	(0.15)	(0.28)	(0.28)	(0.49)	(0.52)	(0.43)	(0.17)
Maun - Botswana	3.71^{ab}	5.44^{ab}	7.56^{ab}	8.07^{a}	8.46^{ab}	3.82^{ab}	8.11^{a}	10.24^{a}	10.54^{a}	10.67^{a}
	(0.28)	(0.31)	(0.43)	(0.50)	(0.33)	(0.23)	(0.32)	(0.27)	(0.41)	(0.24)
PKM 1 - India	4.11^{a}	5.33^{ab}	7.16^{abc}	7.72^{ab}	7.79^{abcd}	3.93^{a}	7.04^{bcd}	8.58^{bcd}	8.70^{bcd}	8.70^{bcd}
	(0.37)	(0.52)	(0.63)	(0.50)	(0.84)	(0.19)	(0.32)	(0.39)	(0.36)	(0.45)
PKM 2 - India	3.80^{ab}	5.24^{b}	6.94^{abc}	7.46^{abc}	7.90^{abcd}	3.18^{bcde}	6.06^{de}	7.74^{def}	7.77^{de}	8.13^{cd}
	(0.18)	(0.11)	(0.33)	(0.30)	(0.18)	(0.04)	(0.26)	(0.30)	(0.28)	(0.35)
PKM 3 - India	3.98^{a}	5.26^{b}	6.86^{abc}	7.37^{abc}	7.54^{bcde}	3.24^{abcd}	6.10^{de}	7.15^{e}	7.35^{d}	7.43^{e}f
	(0.14)	(0.25)	(0.17)	(0.12)	(0.14)	(0.31)	(0.32)	(0.23)	(0.25)	(0.35)
Jafna/Jaffana - India	3.86^{ab}	4.68^{bcd}	6.54^{bcd}	6.75^{bcd}	6.88^{def}	3.47^{abc}	6.32^{cde}	7.82^{def}	7.96^{de}	8.03^{cd}
	(0.06)	(0.16)	(0.13)	(0.34)	(0.27)	(0.06)	(0.25)	(0.27)	(0.41)	(0.35)
Honduras - Honduras	2.85^{cd}	4.07^{de}	6.15^{cd}	6.59^{cd}	7.03^{cdef}	2.77^{cdef}	5.68^{e}	7.35^{f}	7.43^{de}	7.77^{cd}
	(0.21)	(0.38)	(0.55)	(0.45)	(0.63)	(0.57)	(0.37)	(0.19)	(0.20)	(0.13)
Mbololo 472-029/03 - Kenya	3.07^{c}	4.69^{bcd}	7.14^{abc}	7.59^{abc}	8.32^{abc}	3.78^{ab}	8.14^{a}	9.79^{ab}	9.80^{ab}	10.08^{ab}
	(0.16)	(0.35)	(0.37)	(0.44)	(0.29)	(0.11)	(0.04)	(0.12)	(0.09)	(0.39)
Ihumwa - Tanzania	3.31^{bc}	5.01^{bc}	6.86^{abc}	7.46^{abc}	8.00^{abcd}	3.40^{abcd}	7.67^{ab}	9.66^{abc}	9.67^{abc}	10.13^{ab}
	(0.14)	(0.11)	(0.25)	(0.22)	(0.22)	(0.07)	(0.25)	(0.24)	(0.16)	(0.26)
P>F Ratio	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.0008
RMSE	0.3327	0.46	0.58	0.55	0.72	0.38	0.55	0.62	0.72	0.84
CV	10.33	10.09	9.15	8.02	9.83	12.18	8.05	7.13	8.35	9.36

*Means of individual species/provenance with standard error in parenthesis. -Means of the same letter within the same column are not significantly different.

site.

Root collar diameter

Significant variation (P<0.05) in RCD development was observed at all sites during the whole assessment period. At Gairo site, the study indicates the superior RCD development of Mahalapye (Botswana) and Maun (Botswana), Mbololo 472-029/03 (Kenya) and Ihumwa (Tanzania) and poor RCD development of Mangochi (Malawi) and Makhanga (Malawi). Similar results have been observed in Zimbabwe (Gadzirayi et al., 2013; Goss, 2012). At Ruvu site, the study indicates the superior RCD development of Maun (Botswana), Ihumwa (Tanzania), Mtakataka/Dedza (Malawi) and Mbololo 472-029/03 (Kenya) and poor RCD development of PKM 3 (India) and Honduras (Honduras). The differences in RCD development within a site could be attributed to variations in adaptability among provenances, but generally the provenances at Ruvu site showed good performance in RCD development, which is expected due to the sub-humid conditions of the site.

Diameter at breast height

Significant variation (P<0.05) in Dbh was observed on all

Table 5. Diameter at breast height of 12, 18, 24 and 30 months old *M. oleifera* provenances planted at Gairo site, Morogoro and Ruvu site, Coast Region, Tanzania.

Species/provenances	Gairo site				Ruvu site			
	Age (months)							
	12	18	24	30	12	18	24	30
Chikwawa/Domasi - Malawi	2.27^{de}	3.38^{ef}	3.52^{def}	3.87^{efg}	3.93^{ef}	5.66^{cde}	5.84^{def}	6.30^{cde}
	(0.09)	(0.23)	(0.16)	(0.34)	(0.33)	(0.35)	(0.25)	(0.38)
Makhanga - Malawi	1.89^{f}	2.80^{f}	2.84^{f}	2.86^{h}	3.69^{f}	4.85^{e}	5.48^{ef}	6.05^{cde}
	(0.09)	(0.39)	(0.28)	(0.23)	(0.34)	(0.43)	(0.71)	(0.91)
Ngabu - Malawi	1.95^{ef}	2.70^{f}	2.86^{f}	3.11^{gh}	4.74^{cde}	5.74^{cde}	6.23^{cdef}	6.35^{cde}
	(0.06)	(0.16)	(0.11)	(0.11)	(0.06)	(0.17)	(0.25)	(0.28)
Mangochi - Malawi	1.83^{f}	3.02^{f}	3.11^{ef}	3.22^{gh}	5.06^{bcd}	6.31^{bc}	6.69^{bcd}	6.98^{bcd}
	(0.04)	(0.07)	(0.07)	(0.16)	(0.41)	(0.64)	(0.47)	(0.65)
Mtakataka/Dedza - Malawi	1.96^{ef}	3.37^{ef}	3.61^{de}	3.74^{fgh}	4.87^{bcde}	6.38^{bc}	7.19^{abc}	7.81^{ab}
	(0.21)	(0.07)	(0.09)	(0.04)	(0.43)	(0.32)	(0.30)	(0.27)
Mahalapye - Botswana	3.90^{a}	5.61^{a}	5.67^{a}	6.07^{a}	5.71^{abc}	7.23^{ab}	7.26^{abc}	7.32^{abc}
	(0.22)	(0.25)	(0.22)	(0.10)	(0.36)	(0.28)	(0.40)	(0.31)
Maun - Botswana	3.49^{ab}	5.09^{ab}	5.32^{ab}	5.72^{ab}	6.21^{a}	7.73^{a}	8.25^{a}	8.58^{a}
	(0.18)	(0.31)	(0.37)	(0.28)	(0.51)	(0.27)	(0.27)	(0.29)
PKM 1 - India	3.30^{b}	4.49^{bcd}	4.79^{bc}	5.01^{bcd}	5.48^{abc}	6.21^{bcd}	6.49^{bcde}	6.77^{bcd}
	(0.26)	(0.41)	(0.36)	(0.64)	(0.20)	(0.36)	(0.27)	(0.45)
PKM 2 - India	3.29^{b}	4.52^{bcd}	4.64^{bc}	4.93^{bcd}	4.37^{def}	5.03^{de}	5.65^{def}	6.07^{cde}
	(0.06)	(0.12)	(0.15)	(0.03)	(0.23)	(0.06)	(0.27)	(0.18)
PKM 3 - India	3.30^{b}	4.53^{bcd}	4.67^{bc}	4.84^{bcde}	4.11^{def}	5.18^{cde}	5.21^{f}	5.28^{e}
	(0.19)	(0.25)	(0.24)	(0.26)	(0.33)	(0.34)	(0.29)	(0.74)
Jafna/Jaffana - India	2.66^{cd}	3.99^{cde}	4.01^{cd}	4.17^{defg}	4.76^{cde}	5.61^{cde}	6.07^{def}	6.27^{cde}
	(0.22)	(0.06)	(0.31)	(0.16)	(0.05)	(0.24)	(0.18)	(0.25)
Honduras - Honduras	2.53^{cde}	3.82^{de}	4.08^{cd}	4.21^{cdef}	4.18^{def}	5.21^{cde}	5.61^{def}	5.67^{de}
	(0.15)	(0.28)	(0.25)	(0.41)	(0.49)	(0.31)	(0.24)	(0.25)
Mbololo 472-029/03 - Kenya	3.03^{bc}	4.59^{bc}	4.74^{bc}	5.14^{abc}	6.27^{a}	7.28^{ab}	7.81^{a}	7.82^{ab}
	(0.36)	(0.36)	(0.46)	(0.44)	(0.13)	(0.11)	(0.06)	(0.22)
Ihumwa - Tanzania	3.00^{bc}	4.59^{bcd}	4.71^{bc}	5.24^{abc}	5.81^{ab}	6.97^{ab}	7.48^{ab}	7.87^{ab}
	(0.14)	(0.33)	(0.27)	(0.34)	(0.27)	(0.40)	(0.31)	(0.25)
P>F Ratio	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	0.0001
RMSE	0.33	0.42	0.39	0.55	0.52	0.64	0.60	0.73
CV	11.86	10.27	9.31	12.51	10.43	10.43	9.16	10.69

*Means of individual species/provenance with standard error in parenthesis. -Means of the same letter within the same column are not significantly different.

sites during the whole assessment period. At Gairo site, the study indicates the superior Dbh development of Mahalapye (Botswana) and Maun (Botswana) and poor Dbh development of Makhanga (Malawi) and Ngabu (Malawi). These results are in agreement with those reported in Zambabwe where Mutoko provenance from Zimbabwe performed very well as compared to Malawi provenance (Gadzirayi et al., 2013). For Ruvu site, the study indicates the superior Dbh development of Maun (Botswana) and Ihumwa (Tanzania) and poor Dbh development of PKM 3 (India) and Honduras - Honduras. The differences in Dbh development could be attributed to variations in adaptability among provenances, but generally the provenances at Ruvu site showed good performance in Dbh development. Good climatic condition, that is, availability of rainfall and genetic superiority could have contributed to the good performance of these provenances at Ruvu site and those from Botswana at Gairo site.

Height growth

This study has shown superior height growth for Maun (Botswana) and Mahalapye (Botswana) provenances at Gairo site and poor height growth of Makhanga (Malawi), Ngabu (Malawi) and Mtakataka/Dedza (Malawi) provenances. At Ruvu site, only Maun (Botswana)

Table 6. Height growth of 6, 12, 18, 24 and 30 months old *M. oleifera* provenances planted at Gairo site, Morogoro and Ruvu site, Coast Region, Tanzania.

Species/provenances	Gairo site					Ruvu site				
	Age (months)									
	6	12	18	24	30	6	12	18	24	30
Chikwawa/Domasi - Malawi	1.28^{d} (0.03)	1.74^{c} (0.05)	2.89^{e} (0.09)	3.05^{d} (0.04)	3.12^{e} (0.23)	2.11^{d} (0.17)	4.10^{de} (0.27)	4.74^{de} (0.26)	4.93^{e} (0.37)	5.02^{ef} (0.33)
Makhanga - Malawi	1.11^{de} (0.10)	1.48^{c} (0.10)	2.57^{e} (0.18)	2.65^{d} (0.20)	2.66^{e} (0.22)	2.08^{d} (0.10)	3.92^{e} (0.26)	4.43^{e} (0.39)	4.48^{f} (0.37)	4.82^{f} (0.40)
Ngabu - Malawi	1.22^{d} (0.09)	1.64^{c} (0.10)	2.76^{e} (0.08)	2.82^{d} (0.19)	3.02^{e} (0.13)	2.63^{c} (0.03)	4.59^{cde} (0.12)	5.03^{cde} (0.25)	5.09^{cdef} (0.17)	5.25^{cdef} (0.29)
Mangochi - Malawi	0.89^{e} (0.01)	1.53^{c} (0.03)	2.56^{e} (0.05)	2.96^{d} (0.19)	3.15^{e} (0.15)	2.00^{d} (0.22)	4.82^{bcd} (0.31)	5.77^{bc} (0.29)	5.85^{bc} (0.27)	6.16^{bcd} (0.35)
Mtakataka/Dedza - Malawi	1.23^{d} (0.07)	1.76^{c} (0.04)	2.88^{e} (0.16)	2.99^{d} (0.09)	3.09 (0.11)	2.07^{d} (0.13)	4.38^{cde} (0.37)	5.23^{cde} (0.37)	5.32^{bcde} (0.34)	5.81^{bcdef} (0.28)
Mahalapye - Botswana	2.28^{ab} (0.10)	3.30^{a} (0.06)	4.58^{ab} (0.07)	4.72^{ab} (0.08)	4.83^{ab} (0.06)	3.26^{ab} (0.12)	5.35^{b} (0.22)	5.98^{b} (0.10)	6.20^{b} (0.19)	6.22^{bc} (0.62)
Maun - Botswana	2.15^{b} (0.12)	3.22^{a} (0.13)	4.84^{a} (0.09)	4.93^{a} (0.14)	5.04^{a} (0.17)	3.38^{ab} (0.20)	6.38^{a} (0.39)	7.46^{a} (0.52)	7.73^{a} (0.50)	8.16^{a} (0.57)
PKM 1 - India	2.53^{a} (0.06)	3.07^{a} (0.14)	4.14 ^{c} (0.27)	4.24^{bc} (0.20)	4.28^{cd} (0.26)	3.73^{a} (0.18)	5.08^{bc} (0.12)	5.63^{bcd} (0.09)	5.72^{bcd} (0.02)	6.06^{bcde} (0.22)
PKM 2 - India	2.40^{ab} (0.04)	3.04^{a} (0.05)	4.11^{c} (0.12)	4.22^{bc} (0.07)	4.28^{cd} (0.10)	3.14^{b} (0.04)	4.43^{cde} (0.19)	4.93^{de} (0.25)	5.03^{cdef} (0.24)	5.26^{cdef} (0.26)
PKM 3 - India	2.49^{a} (0.14)	2.98^{a} (0.03)	3.97^{c} (0.13)	3.99^{c} (0.13)	4.06^{cd} (0.07)	3.36^{ab} (0.19)	4.46^{cde} (0.12)	5.01^{cde} (0.03)	5.08^{cdef} (0.02)	5.12^{cde} (0.04)
Jafna/Jaffana - India	2.48^{a} (0.10)	2.98^{a} (0.06)	3.78^{c} (0.17)	3.79^{c} (0.23)	3.87^{d} (0.14)	3.68^{a} (0.01)	4.48^{cde} (0.09)	4.88^{de} (0.13)	5.12^{cdef} (0.21)	5.16^{def} (0.29)
Honduras - Honduras	1.64^{c} (0.12)	2.44^{b} (0.24)	3.44^{d} (0.16)	3.76^{c} (0.23)	3.81^{d} (0.20)	2.64^{c} (0.44)	4.37^{cde} (0.25)	4.99^{de} (0.16)	5.08^{cdef} (0.12)	5.33^{bcdef} (0.18)
Mbololo 472-029/03 - Kenya	1.87^{c} (0.22)	2.60^{b} (0.24)	4.07^{c} (0.07)	4.22^{bc} (0.14)	4.47^{bc} (0.18)	3.50^{ab} (0.21)	5.42^{b} (0.16)	6.04^{b} (0.13)	6.11^{b} (0.12)	6.30^{b} (0.17)
Ihumwa - Tanzania	1.79^{c} (0.05)	2.55^{b} (0.07)	4.10^{c} (0.19)	4.20^{bc} (0.19)	4.22^{cd} (0.17)	3.08^{bc} (0.11)	5.09^{bc} (0.12)	5.72^{bcd} (0.25)	5.81^{bcd} (0.27)	6.16^{bcd} (0.29)
P>F Ratio	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
RMSE	0.1656	0.20	0.22	0.30	0.28	0.28	0.38	0.46	0.45	0.53
CV	9.14	8.11	6.13	8.10	7.21	9.56	7.85	8.41	8.24	9.24

*Means of individual species/provenance with standard error in parenthesis. -Means of the same letter within the same column are not significantly different.

provenance showed superior height growth while Makhanga (Malawi) and Chikwawa/Domasi (Malawi) provenances showed poor growth in height. The differences in height growth within a site could be attributed to variations in adaptability among provenances while the between site differences in growth relate with rainfall differences between the two sites with Ruvu having higher rainfall as compared to Gairo. Generally, the provenances showed good performance in height development at Ruvu site. The growth performance of outstanding provenances in the present study compare favourably with results reported in Song Hau State farm, Cantho City, Vietnam (Manh et al., 2005) and in Zimbabwe (Gadzirayi et al., 2013; Goss, 2012). The Vietnam site has higher annual rainfall of 1800 - 2000 mm as compared to that of the study sites.

Biomass

Most provenances grown at the Ruvu site showed high production of foliar and stem biomass than those grown at Gairo site. This could be due to sufficient availability of growth resources (rainfall) at Ruvu site unlike the Gairo site. The lowest yield in stem biomass at Gairo site were recorded from Ngabu (Malawi) and Mangochi (Malawi) while at Ruvu site were recorded for Mangochi (Malawi) and Makhanga (Malawi) provenances. The lowest yield in

Table 7. Biomass production of 30 months old *M. oleifera* provenances planted at Gairo site, Morogoro and Ruvu site, Coast Region, Tanzania.

Species/Provenances	Age (months)			
	Gairo site		Ruvu site	
	Stems(Kg/ha)	Foliage(Kg/ha)	Stems(Kg/ha)	Foliage (Kg/ha)
Chikwawa/Domasi - Malawi	8235.60^{cde} (2173.46)	1324.10^{cde} (378.86)	21036.74^{ab} (4163.94)	738.19^{ab} (107.89)
Makhanga - Malawi	8481.34^{cde} (803.95)	982.04^{de} (461.58)	15796.17^{b} (5022.39)	697.10^{b} (187.76)
Ngabu - Malawi	6200.41^{de} (1781.16)	491.02^{e} (98.81)	20858.93^{ab} (6500.60)	1423.19^{ab} (542.02)
Mangochi - Malawi	5901.08^{e} (1027.08)	1035.61^{de} (104.09)	15816.98^{b} (2015.65)	972.51^{ab} (31.67)
Mtakataka/Dedza - Malawi	11386.00^{bcde} (1053.55)	1723.31^{bcde} (598.34)	17292.03^{b} (4111.31)	1383.46^{ab} (243.10)
Mahalapye - Botswana	21212.24^{ab} (4618.29)	3108.98^{ab} (590.92)	41403.26^{a} (12290.79)	1717.77^{ab} (587.61)
Maun - Botswana	20285.88^{ab} (2910.27)	4407.18^{a} (1227.52)	34888.16^{ab} (4362.86)	1917.21^{a} (205.66)
PKM 1 - India	21012.81^{ab} (7635.45)	2213.56^{bcd} (129.57)	31943.36^{ab} (11629.09)	1865.19^{ab} (550.69)
PKM 2 - India	24733.60^{a} (1063.24)	2562.68^{bcd} (628.82)	19061.91^{b} (4741.96)	1506.82^{ab} (361.19)
PKM 3 - India	16183.97^{abcde} (3687.52)	2159.61^{bcd} (152.48)	22859.98^{ab} (3360.61)	1484.22^{ab} (164.91)
Jafna/Jaffana - India	19981.74^{ab} (2759.02)	2740.89^{bc} (552.82)	24946.83^{ab} (1697.49)	1437.73^{ab} (130.37)
Honduras - Honduras	12280.77^{bcde} (1835.96)	1574.84^{bcde} (685.94)	19679.77^{b} (2399.60)	1450.30^{ab} (250.07)
Mbololo 472-029/03 - Kenya	17542.76^{abcd} (4795.64)	2280.33^{bcd} (599.18)	28120.39^{ab} (8479.69)	1398.37^{ab} (553.10)
Ihumwa - Tanzania	18859.69^{abc} (3138.39)	2987.07^{ab} (971.92)	17610.75^{b} (3767.78)	1104.62^{ab} (225.14)
P>F RATIO	0.0049	0.007	0.1820	0.3987
RMSE	6016.97	853.42	10969.71	612.24
CV	39.68	40.38	46.35	44.88

*Means of individual species/provenance with standard error in parenthesis. Means of the same letter within the same column are not significantly different

foliar biomass at Gairo site were recorded for Ngabu (Malawi) and Makhanga (Malawi) provenances while at Ruvu site were recorded at Makhanga (Malawi) and Chikwawa/Domasi (Malawi) provenances. This poor performance could probably be due to poor genetic adaptations influenced by climatic conditions in these areas.

Conclusions and recommendations

The present study has shown that provenances differ significantly in survival and biomass production (Gairo site), and diameter and height growth in both sites. The best performing provenances at Gairo site were Mahalapye (Botswana), Maun (Botswana), Ihumwa (Tanzania), Mbololo 472-029/03 (Kenya) and PKM 1 (India) while at Ruvu site were Maun (Botswana), Mahalapye (Botswana), Mbololo 472-029/03 (Kenya) and Ihumwa (Tanzania). These provenances have shown promising growth throughout the study period indicating their suitability to the locality and other areas with similar soil/climatic conditions. On the other hand Chikwawa/-Domasi (Malawi), Makhanga (Malawi), Ngabu (Malawi) and Mangochi (Malawi) at Gairo and Mangochi (Malawi), Honduras (Honduras), Chikwawa/Domasi (Malawi) and Makhanga (Malawi) at Ruvu site failed to put on promising growth on these sites. The best performing provenances are recommended for planting at these and similar sites.

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Table 8. Ordinal ranking of 30 months old *M. oleifera* provenances planted at Gairo site, Morogoro, Tanzania.

Species/Provenances	Surv1	ht	rcd	dbh	fbio	sbio	Mean	Overall rank
Chikwawa/Domasi - Malawi	11	10	11	10	11	12	11	7
Makhanga - Malawi	10	13	13	14	13	11	12	8
Ngabu - Malawi	8	12	12	13	14	13	12	8
Mangochi - Malawi	9	9	14	12	12	14	12	8
Mtakataka/Dedza - Malawi	4	11	8	11	9	10	9	6
Mahalapye - Botswana	4	2	1	1	2	2	2	1
Maun - Botswana	2	1	2	2	1	4	2	1
PKM 1 - India	6	4	6	5	7	3	5	3
PKM 2 - India	2	4	5	6	5	1	4	2
PKM 3 - India	5	6	7	7	8	8	7	5
Jafna/Jaffana - India	2	7	10	9	4	5	6	4
Honduras - Honduras	7	8	9	8	10	9	9	6
Mbololo 472-029/03 - Kenya	3	3	3	4	6	7	4	2
Ihumwa - Tanzania	1	5	4	3	3	6	4	2

[1]Surv., Survival; ht, height; rcd, root collar diameter; dbh, diameter at breast height; fbio, foliar biomass; sbio, stem biomass.

Table 9. Ordinal ranking of 30 months old *M. oleifera* provenances planted at Ruvu site, Coast Region, Tanzania.

Species/Provenances	Surv[1]	ht	rcd	dbh	fbio	sbio	Mean	Overall rank
Chikwawa/Domasi - Malawi	1	12	8	9	13	7	8	7
Makhanga - Malawi	5	13	10	12	14	14	11	9
Ngabu - Malawi	2	9	6	8	8	8	7	6
Mangochi - Malawi	4	4	7	6	12	13	8	7
Mtakataka/Dedza - Malawi	2	6	3	4	10	12	6	5
Mahalapye - Botswana	1	3	5	5	3	1	3	2
Maun - Botswana	8	1	1	1	1	2	2	1
PKM 1 - India	4	5	9	7	2	3	5	4
PKM 2 - India	6	8	11	11	4	10	8	7
PKM 3 - India	3	11	14	14	5	6	9	8
Jafna/Jaffana - India	7	10	12	10	7	5	9	8
Honduras - Honduras	6	7	13	13	6	9	9	8
Mbololo 472-029/03 - Kenya	1	2	4	3	9	4	4	3
Ihumwa - Tanzania	2	4	2	2	11	11	5	4

[1]Surv.-Survival; ht-height; rcd-root collar diameter; dbh-diameter at breast height; fbio-foliar biomass; sbio-stem biomass.

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Full Length Research Paper

Nutrient composition of *Moringa oleifera* leaves from two agro ecological zones in Ghana

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***Moringa oleifera* Lam. (*MO*) is a small size multipurpose tree of approximately 5 to 10 m in height which is cultivated all over the world. The study was conducted in the Guinea Savanna and the semi-deciduous forest zones of Ghana in December 2011 to compare the nutrient levels of *MO* leaves from the two agro-ecological zones. Leaf samples were collected from three randomly selected districts in each ecological zone for proximate and chemical analysis of some macro and micro nutrient. The results show no significant difference in crude protein and carbohydrate levels of *MO* leaves from the two ecological zones, however *MO* leaves from the semi-deciduous forest zone recorded a slightly higher mean crude protein value of 26.54% than the Guinea savanna value of 25.65%. *MO* leaves from the semi-deciduous forest and the Guinea Savanna recorded mean calcium values of 1880 and 1474.33 mg per 100 g of leaf powder, respectively. The comparison of mean values of potassium and iron of *MO* leaves showed no significant difference between *MO* leaves from the two ecological zones. The research concluded that agro ecological zonation had no significant effect on the levels of most nutrients in *MO* leaves.**

Key words: *Moringa oleifera*, ecological zone, Guinea Savanna, semi-deciduous forest, nutrients.

INTRODUCTION

Moringa oleifera Lam *(MO)* is the most widely cultivated species of a monogeneric family, the *Moringaceae* (Fahey, 2005). The tree is an aboriginal of Indian subcontinent and has become naturalized in the tropical and subtropical areas around the world (Farooq et al., 2012). While it grows best in dry sandy or loamy soil that is slightly alkaline (Abdul, 2007; Anjorin et al., 2010), it is adaptable to various soil conditions from 4.5 to 8.0 pH, but does not tolerate water logging, freezing or frosts conditions (Radovich, 2011). India is rated as the largest producer of Moringa, with an annual production of between 1.1 to 1.3 million tonnes of tender fruits from an area of 380 km² (Rajangam et al*.,* 2011). In the past 5000 years, *MO* has been used as a regular component of conventional eatables in the Indian sub-continent (Anwar et al., 2005, Anwar and Bhanger, 2003). Its trunk is soft, white corky and branches bearing a gummy bark. Each tripinnately compound leaves bear several small leaflets. The flowers are white and the three wing seeds are scattered by the winds (Farooq et al., 2012).

According to Fuglie (2005), *MO* plant forms the basis for several nutritional programmes in many poor countries by charitable organizations, given that the leaves of *MO* tree are rich in essential nutrients.

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The leaves of *MO* are considered to give immense possibilities for those who are nutritionally challenged and may be regarded as a protein and calcium supplement (Rajangam et al., 2001). Bamishaiye et al. (2011) reported MO leaves of all stages having varying percentages of nutritional composition. Fuglie (1999) was quick to say the "so good to be called syndrome was the biggest challenge for Moringa", it offers so much to us so that it's so difficult to believe if such simple plant can be so useful. This little-plant has the potential to improve nutrition, boost food security and support sustainable landuse practices (Price, 2007). It is estimated that 82% of the population in developing countries depend on herbal treatment (Abbiw, 1990).

Researchers at the Asian Vegetable Research and Development Center (AVRDC, 2006) reported that leaves from four different *Moringa* species (*Moringa oleifera, Moringa peregrina, Moringa stenopetala* and *Moringa drouhardii*) all contained high levels of nutrients and antioxidants. Vitamin A was found to be at its peak during the hot-wet season, where as iron and vitamin C was highest during the cool-dry season (Price, 2007). Bureau of plant industry reported *MO* as an outstanding source of nutritional components. Its leaves (weight per weight) have the calcium equivalent of four times that of milk, the vitamin C content is seven times that of oranges, while its potassium is three times that of bananas, three times the iron of spinach, four times the amount of vitamin A in carrots, and two times the protein in milk (Kamal, 2008). In addition, the leaves can serve as a rich source of beta-carotene (Nambiar and Seshadri, 2001), vitamin C and E, and polyphenolics (Ross, 1999). Also, *Moringa* is suggested as a viable supplement of dietary minerals. The pods and leaves of *Moringa* contains high amount of Ca, Mg, K, Mn, P, Zn, Na, Cu and Fe (Aslam et al., 2005).

MO leaf extracts are rich in pterygospermin and other related compounds such as isothiocyanates which is used in the treatment of many skin infections because of its antibiotic and fungicidal properties (Price, 2007).

It is generally known in the developing world as a vegetable, a medicinal plant and a source of vegetable oil (Bennet et al., 2003). Six tablespoons full of *MO* leaf powder will provide nearly the woman's daily iron and calcium during pregnancy and breastfeeding hence *MO* has been used to combat malnutrition among infants and nursing mothers (TFL, 2011). *M. oleifera* can survive in harsh climatic condition including impoverished soils without being much affected by drought (Morton, 1991). It can tolerate wide range of rainfall requirements estimated at 250 mm and maximum at over 3000 mm and a pH of 5.0 to 9.0 (Palada and Chang, 2003). Easy cultivation of *Moringa* within adverse environmental condition and wide availability attract attention for economic and health related potential in resource limited developing countries (Farooq et al., 2012).

The mineral contents in *M. oleifera* and their bioavailability continue to be a subject of tremendous interest. There are however limited reports on the influence of variation in geographical locations or agro-ecology of *M. oleifera* on the mineral composition in various organs of the plant. Aslam et al. (2005) suggested the contents of different minerals in leaves and pods of *M. oleifera* to significantly differ from region to region in Pakistan. Anjorin et al. (2010) confirmed that there were variations in macro and trace minerals in *M. oleifera* leaves, pods and seeds from different locations in Nigeria. The increasing conviction and confidence in the consumption of this plant calls for a need to document well the nutritional comparison of *Moringa* leaves from different agro ecological zones in Ghana. The objective of this study was to investigate and compare the nutritional components of *MO* leaves in two agro-ecological zones of Ghana.

MATERIALS AND METHODS

Study area

The study was conducted in the Guinea Savanna (GS) and the semi-deciduous forest (SDF) zones of Ghana in December 2011. Leaf samples were taken from three selected districts in the Guinea Savanna zone of Ghana: Tolon-Kumbungu District (latitude 09° 25″N and longitude 0° 58″W in the Northern region), Kassena-Nankana East District (Latitude 10°53′5″N and longitude 01°5′25″W in the Upper East region) and the Sissala West District (Latitude 10° 11″ N and Longitude 02°13′36″ in the Upper West region). The Guinea Savanna records a unimodal rainfall pattern (Figure 1), which starts in April and ends in September-October. The mean annual rainfall ranges between 900 -1100 mm (NAES, 1991). The vegetation of the area is generally grassland with few trees interspersed (NAES, 1993). The temperature of the zone ranges between a minimum of 15°C in January and a maximum of 42°C in March.

The semi-deciduous forest zone is considered the most important for timber production in Ghana (Owusu et al., 1989). Leaf samples were taken from three randomly selected districts of the semi-deciduous forest zone of Ghana: Techiman municipality (Latitude 7°35′13″N Longitude 1°56′06″W), Wenchi municipality (latitudes 7° 30′ and 8°05′N and longitudes 2°15′ W), and Nkoranza South District (Latitudes 7°20′55″N and Longitudes 1°10′55″W) all in the Brong Ahafo Region. The zone records a bimodal rainfall pattern (Figure 2). The major raining season starts in early March and reaches its peak in June, and tapers off gradually through July. The minor season starts in late August and reaches its peak in September/November. The mean annual rainfall figures range from 1150 to 1250 mm. The mean monthly temperature is between 24°C in August and 30°C in March. The relative humidity is also high ranging between 90 and 95% in the rainy season.

Field work

Moringa leaves and soil samples were taken from both the savannah (Tolon-kumbugu,Kassena-Nankana and Sissala West) and transitional zones (Techiman, Wenchi and Nkoranza). Leaf samples were plugged from tree growing points across the lower, mid and upper parts of tree canopy. Leaves were defoliated from rachis and samples from aforementioned districts within each zone were bulked and thoroughly mixed. Final samples were kept in to

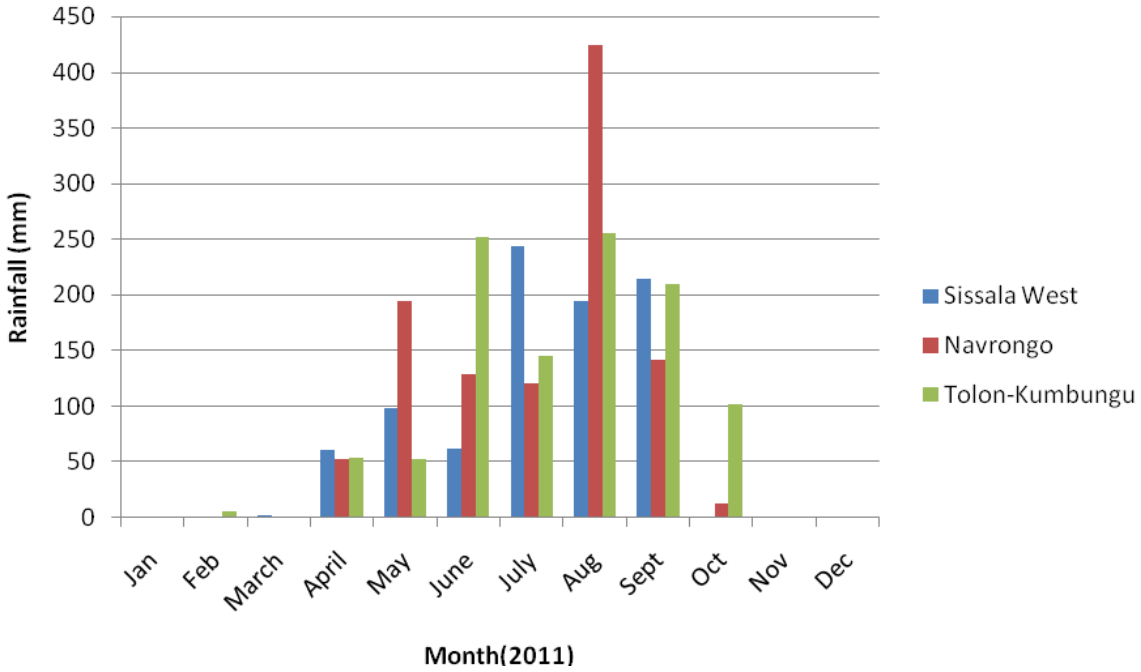

Figure 1. Monthly figures showing unimodal rainfall for areas sampled in the Savanna zone

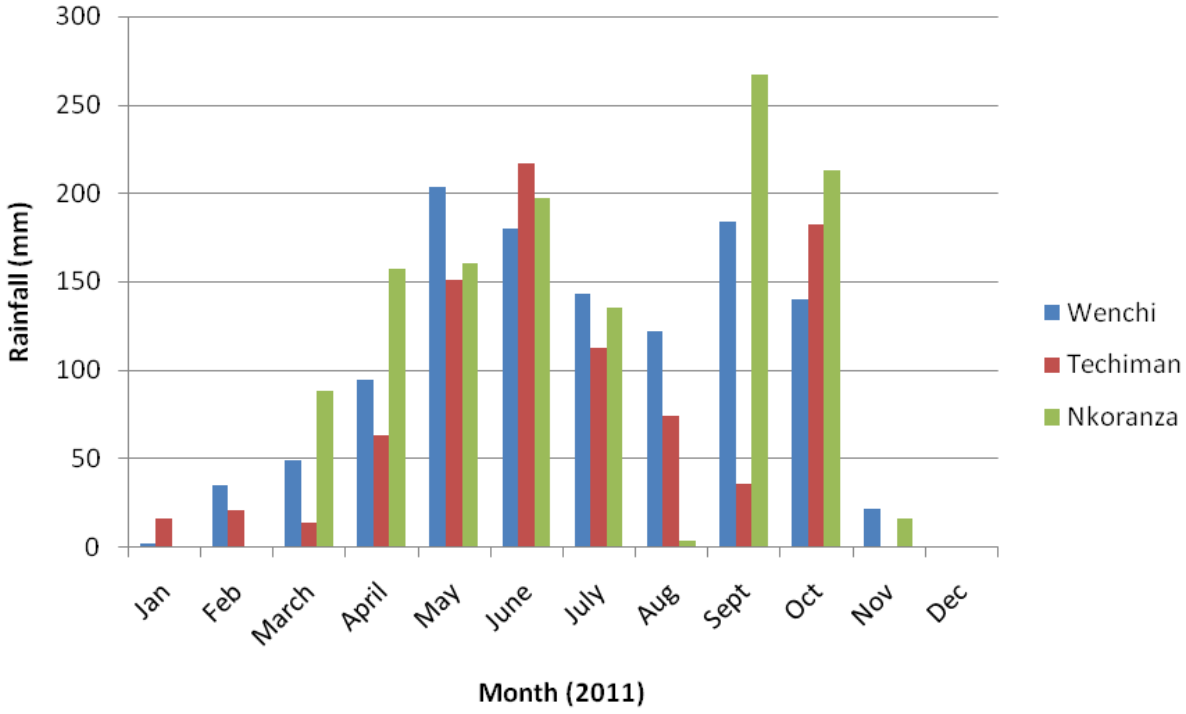

Figure 2. Monthly figures showing bimodal rainfall for areas sampled in the semi-deciduous forest zone.

ice-chest and transported to the laboratory. The harvested *Moringa* leaves were air dried in shade to ensure they retain their greenish coloration and to ensure that their nutritional values were maintained. The dry leaves were milled and sieved. The powder was then packaged into small containers and labeled based on ecological zones to ensure that right samples were picked.

Soil samples were taken at 0-15 m from the spots where the *MO* leaves were sampled. This was done using the soil auger. The soils of various zones were sieved and packaged in their labeled containers based on their ecological zones.

Table 1. Comparison of mean levels of nutrients per 100 g of *MO* soils from the Guinea Savanna and semi-deciduous forest of Ghana.

Ecological zone	N (%)	P (mg/kg)	K (mg/kg)	Ca (mg/kg)	Mg (mg/kg)
Semi-deciduous forest	0.11	4.61	202.67	813.33	202.50
Guinea savanna	0.05	9.36	207.00	505.33	136.18

Analysis of soil samples

Three samples of soil were assayed from each agro ecological zone and analyzed individually in triplicate. The amount of phosphorus present in the soil sample was determined using bray 1 extraction with the help of the spectrophotometer. Ammonium acetate was used to extract potassium, calcium and magnesium, however, in determining the rate of nitrogen present in the soil, a sample weight of soil of 0.2 g was added to 10 ml concentrated sulphuric acid. One Kjeldahl was added and placed on the block digester. The Kjeldahl distillation process was used to determine the rate of soil nitrogen.

Determination of minerals in *MO* leaves

Three samples of leaves were assayed from each agro ecological zone and analyzed individually in triplicate. Moisture content was determined by air drying, and protein by the Kjedahl method (AOAC, 1990). The pH of *Moringa* leaves was taken with an electronic pH metre (Bates, 1954). The amount of carbohydrate was determined by the calometric method (AOAC, 1990). Calcium was measured directly on the atomic absorption spectrophotometer using 1 g of the sample solution which was topped to 100 ml. The potassium was measured using the Barnes et al. (1954) method while iodine and iron were analyzed calorimetrically using the ultra violet visible spectrophotometer (Hald, 1946).

Data was subjected to Analysis of Variance (ANOVA) and differences among the mineral components and pH of the leaves were determined with Duncan Multiple Range Test, using SAS 9.0 Statistical Package. *P*-values ≤ 0.05 was considered statistically significant. Comparison was made of minerals content and pH in leaf of *MO* from savanna and semi-deciduous forest.

RESULTS

Nutrient content of soils where *MO* samples were taken from in the two ecological zones

The soils under the sample plants of *MO* in both ecological zones were fairly fertile. Soils under *MO* plants from the semi-deciduous forest and the Guinea savanna zones recorded 4.61 and 9.36 mg/kg of phosphorous (P), respectively (Table 1), but there was no significant difference. Levels of K in soils similarly showed no significant difference recording 202.67 and 207 mg/kg for the semi-deciduous forest and the Guinea Savanna zones, respectively. Levels of Ca were higher in the semi-deciduous forest zone (813.33 mg/kg) than in the Guinea savanna (505.33 mg/kg) however, there was no statistical difference between the two zones. Magnesium (Mg) in soils of the semi-deciduous forest were similarly higher (202.5 mg/kg) than soils from the Guinea savanna (136.18 mg/kg) but not significantly different. Percentage nitrogen was low but not significant in soils from semi-deciduous forest (0.11%) and the Guinea Savanna soils (0.05%), respectively.

Proximate results and pH of *Moringa* leaves from the two ecological zones

A comparison of the mean values of moisture, crude protein, carbohydrate and pH of *Moringa* leaves from the Guinea Savanna and the semi-deciduous forest zone are shown in Figure 3. *Moringa* leaves from the semi-deciduous forest recorded a higher average moisture content of 71.34%. Analysis of variance at P>0.05 showed no significant difference in moisture content of *Moringa* leaves from the two ecological zones.

Crude protein and carbohydrate in *MO* leaves from the two ecological zones recorded similar mean values. Analysis of variance at P>0.05 showed no significant difference in crude protein and carbohydrate of *MO* leaves from the two ecological zones. There was however a significant difference in pH (Table 2) of *MO* leaves from the two ecological zones (P>0.05).

Levels of macro and micro nutrients in *MO* leaves from the two ecological zones

MO leaves from the semi-deciduous forest and the Guinea savanna zones recorded 1209.67 mg/100 g and 997.3 mg/100 g of potassium (K), respectively as shown in Table 2. There was a significant difference (P>0.05) in calcium (Ca) levels between *MO* leaves from the two ecological zones. The mean levels of iron (Fe) in *MO* leaves from the semi-deciduous forest and the Guinea savanna zones recorded close mean values of 26.83 mg/100 g and 25.043 mg/100 g, respectively (Table 2).

DISCUSSION

Effect of agro ecological zone on crude protein, carbohydrate and pH levels of *MO* leaves

Crude protein levels of *MO* leaves from the two ecological zones ranges between 25.34 and 26.98 g in every 100 g of *MO* leaf powder (Figure 3). This is similar to the findings of Fuglie (2005) that suggested *MO* as

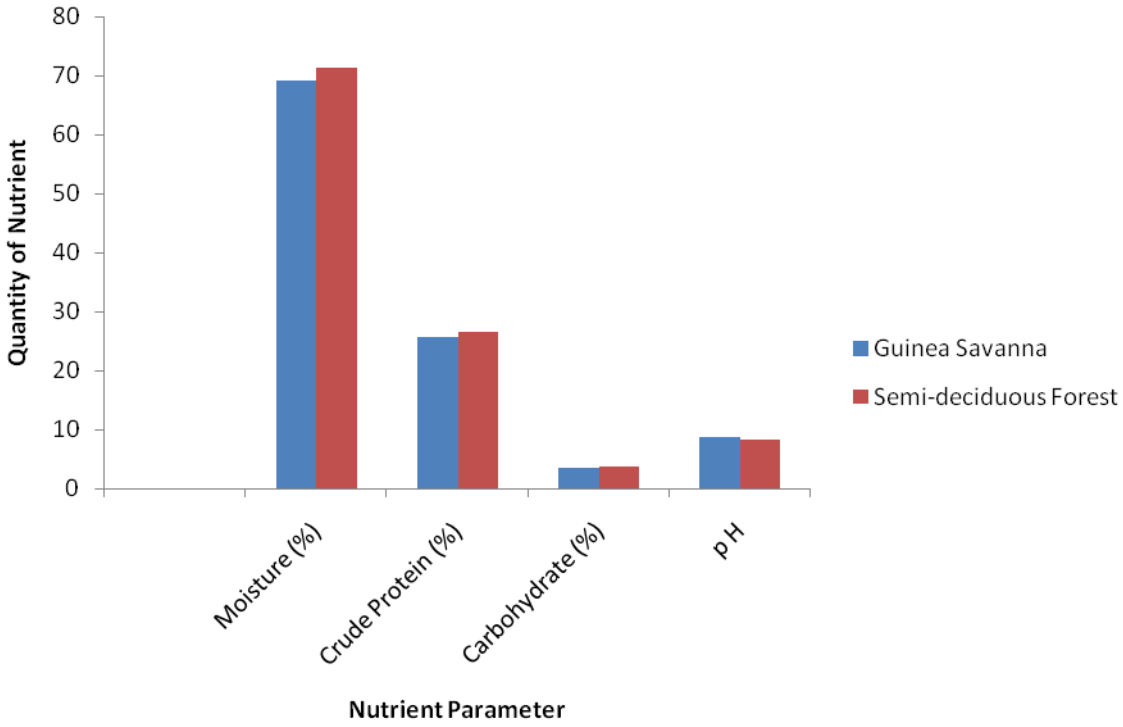

Figure 3. Comparison of mean proximate values and pH of *MO* leaf powder from the Guinea Savanna and the semi-deciduous forest of Ghana.

Table 2. Comparison of mean levels of macro and micro nutrients per 100 g of *MO* leaf powder from the Guinea Savanna and semi-deciduous forest of Ghana.

Ecological zone	Macro nutrient		Micro nutrient	
	K (mg/kg)	Ca (mg/kg)	Fe (mg/kg)	I (mg/kg)
Semi-deciduous forest	1209.67	1880.00	26.83	0.674
Guinea savanna	997.33	1474.33	25.043	0.517

containing high amount of proteins. The crude protein of *MO* leaves from the semi-deciduous forest of Ghana was higher than that of Guinea Savanna though there was no significant differences (P>0.05). These variations may be due to the periodic higher temperatures in the Guinea Savanna at certain times of the year. Temperatures in Guinea Savanna rise beyond the actual climatic requirements of the *MO* plant (25 - 35°C) to a maximum of about 42°C in March.

Although increasing tempe-ratures activates enzymatic activities, higher tempera-tures beyond the plant requirement causes many cell proteins that function as enzymes or structural com-ponents to become unfolded or misfolded, there by leading to loss of proper structure and activity (Taiz and Zeiger, 2002). This agrees with the findings of Modi (2007) that reported that cool environmental conditions are associated with high total protein in leafy vegetables while hot temperatures had a significant decrease in leaf protein content. Apart from the higher temperatures in the savanna, the long periods of drought during the dry season could be a contributing factor.

The higher levels of carbohydrates in the samples from the semi-deciduous forest zone than that of the Guinea Savanna could be due to higher temperatures. Although photosynthesis and respiration are inhibited at higher temperatures, photosynthetic rates drop before the respiration (Modi, 2007). At any temperature above the plant temperature compensation point, photosynthesis cannot replace the carbon used as a substrate for respiration. As a result, carbohydrate reserves decline, and also lead to loss of sweetness as well. Taiz and Zeiger (2002) reported imbalances between photosyn-thesis and respiration and associated it to the deleterious effects of high temperatures on plant development.

The mean pH values of *MO* leaves from the semi-deciduous forest zone and the Guinea Savanna were 8.35 and 8.71, respectively (Figure 3). The pH reading of

Table 3. ANOVA of pH values, Ca and I nutrients per 100 g of *MO* leaf powder from the Guinea Savanna (GS) and semi-deciduous forest (SDF) of Ghana.

Source of variation	d.f	s.s	m.s	v.r	F.pr.
ANOVA for pH of *MO* leaves from the GS and the SDF zone.					
Treatment	1	0.98067	0.98067	383.26	0.001
Residual	4	0.00207	0.00052		
Total	5	0.20008			
ANOVA for Ca per 100 g *MO* leaf powder from the GS and the SDF zone					
Treatment	1	246848	246848	47.54	0.002
Residual	4	20771	5193		
Total	5	267619			
ANOVA for I per 100 g of *MO* leaf powder from the GS and the SDF zone					
Treatment	1	0.03373	0.03373	119.39	0.001
Residual	4	0.00125	0.00031		
Total	5	0.03854			

MO leaves from both ecological zones falls within the slightly alkaline range of the pH scale. The analysis of variance showed a significant difference in pH (Table 2). Research by Diet.myfit (2012) indicates that there exist no recommended dietary pH for food substances, but the human body operates best within a pH range of 7.35 - 7.45. Therefore, the consumption of any food with a higher or lower pH value is an indication of it being alkaline or acidic to the human body.

Effect of agro ecological zone on moisture content of *MO* leaves

Samples from the two ecological zones recorded high moisture content ranging from 67 to 75% of the weight of fresh leaves (Figure 3). The leaf samples from the semi-deciduous forest zone recorded higher average moisture content than leaves from the Guinea Savanna. The low moisture values recorded in *MO* leaves from the Guinea Savanna zone may be associated with hot and dry winds of the harmattan during sample collection (December) which comes with high rates of evapotranspiration. Modi (2007) reported that transpiration rates are at their peak in hot dry/windy environment. The effects of annual harmattans are more severe in the Guinea savanna than the semi-deciduous forest. During this period, atmospheric humidity becomes very low in the Guinea savanna. However, with plant leaves relative humidity between cells approach 100%, therefore when stomata opens, water vapour inside the leaf moves out forming a bubble of higher humidity around the plant. The difference in relative humidity around the stomata and adjacent air regulates transpiration rates and pulls water up through the xylem tissues (Taiz and Zeiger, 2002).

Moringa leaves from the Guinea Savanna zone recorded higher mean dry matter content of 30.92% than that of the semi-deciduous forest zone of 28.66%. The high dry matter content of savanna leaves makes them more favourable for animal fodder. Bell (2006) indicated that the higher the dry matter of fodder, the higher its digestibility (the proportion of a feed an animal can use to satisfy its nutritional requirements). He further reported that digestibility is positively related to the energy content and protein of animal fodder. Nuhu (2010) also indicated that *MO* leaf meal (MOLM) could be used to improve daily weight gain, dry matter (DM) and crude protein (CP) digestibility of rabbits. In the current situations where natural rangelands are getting extinct due to rapid encroachment, *MO* can be incorporated into agroforestry practices to provide fodder to increase animal production in the savanna regions of Ghana.

Effect of agro ecological zones on macro and micro nutrients of *MO*

There was no significant differences in K and Fe levels of *MO* leaf samples (P>0.05) from the two ecological zones. Although savanna soils are known to be low in soil organic matter as compared to that of the semi-deciduous forest, high levels of soil nitrogen in the semi-deciduous forest zone did not influence the accumulation of some nutrient elements in *MO* leaves. This agrees with Ore-Oluwa et al. (2003) where nitrogen application did not record any significant effect on the accumulation of Ca, K, Na, Cu and Zn in tropical leafy vegetables however, decreasing levels of soil nitrogen reduces the Fe content of tropical vegetables.

Also, significant difference in calcium and iodine (Table 3) may be due to the long droughts in the Guinea Savanna which makes soils dry for longer periods of time

inhibiting the absorption of soil nutrients for leaf development. Potassium, calcium and phosphorus move primarily by diffusion therefore the drier the soil, the lesser the flow of these nutrients to the plant root. Kessel (2005) reported that calcium and potassium levels in leaves decreased with decreasing soil moisture. However, the low levels of Ca and iron recorded in the Guinea Savanna zone as compared to the semi-deciduous forest contradicts the findings of Modi (2007) were mineral elements, calcium and iron increased in the leaves in response to increasing temperatures. Aslam et al. (2005) reported that mineral content in the leaves and seeds of *MO* varied in Pakistan with location while Anjorin et al. (2010) reported significant variation in macro and micro elements in *MO* leaves, pods and seeds from different regions in Nigeria.

MO leaves generally contain significant quantities of some macro and micro food nutrients needed by the human body. The research revealed that agro ecological zones have no significant effect on the levels of most nutrients of *MO* leaves in Ghana. However, *MO* leaf samples from the semi-deciduous forest zone recorded slightly higher nutrient values than that of the Guinea savanna.

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Full Length Research Paper

Phytochemical studies on the extract and essential oils of *Artemisia dracunculus* L. (Tarragon)

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***Artemisia dracunculus* L. (Tarragon) is a species of flowering plant within the family Asteraceae, commonly used as a dietary seasoning. During the present study, the plant was collected from Indian Institute of Integrative Medicines (IIIM Srinagar). Air dried shoots (room temperature 25-35°C) were used to extract essential oil using Clevenger type apparatus for 3 h and analyzed. Thirty-four (34) compounds were identified using gas chromatography- flame ionization detector (GC-FID) and Gas chromatography-mass spectrometry (GC-MS) analysis. Major constituents of the essential oil were trans-Anethole (28.06%), Z-β-ocimene (15.79%), α- Terpenolene (10.12%), Elemecin (10.08%), 1, 8 cineole (7.71%) and α-copaene (2.78%), etc. Comparing our results with those of other *Artemisia* species already published in the literature revealed considerable qualitative and quantitative similarity of the major constituents of the essential oils. As trans-Anethole is the major constituent, this chemo type may be useful for industrial exploitation as well as chemotaxonomic characterization.**

Key words: *Artemisia dracunculus*, Terpenolene, Z-β-ocimene, trans-Anethole.

INTRODUCTION

Artemisia is a genus of small herbs or shrubs widely distributed throughout the world but found mostly in Northern temperate regions. It belongs to the important family compositae (Asteraceae), which comprises about 1000 genera and over 20,000 species. Within this family, *Artemisia* is included into the tribe Anthemideae and comprises over 500 species. The 500 species of *Artemisia* are mainly found in Asia, Europe and North America. This genus is industrially important due to its insecticidal, antifungal, antibacterial, allelopathic and other properties. The genus is useful in Ayurveda, Homeopathy, Unani, Siddha and Western medicinal system (Ved and Goraya, 2008).

Chemical composition and biological activities of *Artemisia* spp. essential oils has been reported recently (Lopes-Lutz et al., 2008). *Artemisia dracunculus* L. (commonly known as Tarragon) finds an important place in the genus *Artemisia* and remains a subject of interest due to great variability in traditional medicinal use, plant morphology, reproductive behaviour, essential oil con-tent, composition, etc. Tarragon is a perennial, erect, herb or small shrub, widely distributed in India, China, Japan, North America, European countries, etc., between altitudes of 3000-4000 msl (Hooker, 1882). The species is under cultivation for long time in France, Germany, Holland, Russia, Georgia, Hungary, California, Cuba, etc. for its aromatic value in seasoning salads, edibles, medicinal and in the preparation of Tarragon vinegar.

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Tarragon is safe to use as dietary supplements or in functional foods (Poulev et al., 2004). Biological characteristics and useful properties of tarragon are reported in a review recently (Aglarova et al., 2008). The dried aerial parts of *A. dracunculus* are used orally to treat epilepsy in Iranian traditional medicine (Khorasani, 1992).The species is also useful as sleep aid to mild sedative properties (Chevallier, 1996). It is reported that monoterpenes present in essential oil of *A. dracunculus* are responsible for anticonvulsant and sedative effect (Sayyah et al., 2004). Antidiabetic property is also supported recently by ethanolic extract (Ribnicky et al., 2004). Reports are available on chemical composition of *A. dracunculus* from different parts of the world (Pino, 1996; Irena and Krystyna, 1996; Pappas and Sturtz, 2001; Sayyah et al., 2004). The aim of the present study was to determine the chemical profile of essential oil of *A. dracunculus* L. by using different spectroscopic procedures like GC and GC-MS.

MATERIALS AND METHODS

Plant material

The Himalaya is a well known source of variety of Medicinal and aromatic plants. The plant material of *A. dracunculus* was collected from various geographical regions like Sonmarg, Gulmarg, Gurez and was then transfered to IIIM germplasm and Field station Bonera, Pulwama. The plant material was then taken from IIIM germplasm for the extraction and isolation purposes.

Extraction of essential oil

The shade dried leaves and stem of the plant were finely chopped and then subjected to hydro distillation separately in a Clevenger like apparatus at 60°C for 3 h. The oil obtained was dried over anhydrous sodium sulphate and stored at 4°C in a sealed vial until analysis.

Essential oil analysis

GC/FID was carried out on Perkin Elmer auto system XL Gas Chromatograph 8500 series equipped with flame ionization detector (FID) and head space analyzer using a fused silica capillary RTX-1 Column (30 m x 0.25 mm, film thickness 0.25 mm) coated with dimethyl polysiloxane (RT × 1). Oven temperature was programmed from 60 to 290°C with injector temperature of 230°C and detector temperature of 250°C. Injection volume was 1 µl, and nitrogen was used as a carrier gas (1.0 ml/min). GC- MS analysis was carried on a varian gas chromatograph series 3800 fitted with a VF 5 ms fused silica capillary column (30 m x 0.25 mm, film thickness 0.25 mm) coupled with a 4000 series mass detector under the following conditions: injection volume of 0.5 ml with split ratio 1:60, helium as carrier gas at 1.0 ml/min constant flow mode, injector temperature of 230°C, oven temperature of 60 to 280°C at 3°C /min. Mass spectra was electron impact (EI+) mode,70 eV and ion source temperature was 250°C. Mass spectra were recorded over 50-500 a.m.u range. Identification of the essential oil constituents was done on the basis of Retention Index [RI, determined with respect to homologous series of n-alkanes (C9-C24), Polyscience Corp., Niles IL] under the same experimental conditions), and co-injection with standards (Sigma Aldrich and standard isolates), MS Library search (NIST 98 and WILEY), by comparing with the MS literature data (Jennings and Shibamoto, 1980; Adams, 2007).

RESULTS AND DISCUSSION

The essential oil obtained using Clevenger-type apparatus of the dried aerial part was analyzed and oil percentage was found to be 0.05%. The total oil percentage present in aerial parts was found to be 88.46%. GC and GC-MS analyses of essential oil resulted in the identification of 34 components. The essential oil was found to be rich in trans-Anethole (28.06%), Z-β-ocimene (15.79%), α- Terpenolene (10.12%), Elemecin (10.08%), 1,8 cineole (7.71%) and α-copaene (2.78%) (Table 1). The essential oil was found to be dominated by Monoterpene hydrocarbons (35.22%), Sesquterpene hydrocarbons (13.01%) and Oxygenated sesquiterpenes (2.09%) (Table 2). Besides this, other components belonging to different classes were found to be 30.02%. In the present study, the concentration of trans-Anethole was highest (29%) which was found to be almost in higher concentration as in case of *A. dracunculus* from Italy (up to 53%). In Russian Tarragon, the concentration of β- ocimene (12%) was found to be almost in higher concentration (15%). The essential oils of most *Artemisia* species are characterized by high content of oxygenated monoterpenes with 1,8-cineole and camphor being the most represented. In some other species, such as *A. annua, A. vulgaris, A. diffusa, A. santonicum, A. spicigera, A. afra, A. abiatica, A. austriaca* and *A. pedemontana*, bornane derivatives (camphor, borneol and bornyl acetate) and 1,8-cineole are the major characteristic components (Perez-Alonso et al., 2003; Kordali et al., 2005). Existing literature reveals that *Artemisia absinthium* oil was characterized by high amounts of myrcene (10.8%), trans-thujone (10.1%) and trans-sabinyl acetate (26.4%).

Approximately 71.0% of *A. absinthium* oil composition was identified. The remaining unidentified components were monoterpene esters and sesquiterpenes. Phenyl propanoid compounds comprised 52.2% of *A. dracunculus* oil, with methyl chavicol and methyl eugenol being the most representative constituents. *A. biennis* yielded an oil rich in (Z)-beta-ocimene (34.7%), (E)-beta-farnesene (40.0%) and the acetylenes (11.0%) (Z)- and (E)-en-yndicycloethers. Previous research showed that bornane derivatives (camphor, borneol and bornyl acetate) and 1,8-cineole are major characteristic components of many species of *Artemisia* genus, such as: *A. Annua, A. vulgares, A. diffusa, A. santonicum, A. spicigera, A. afra, A. asiatica, A. austriaca* and *A. pedemontana* (Perez-Alonso et al., 2003; Kordali et al., 2005). In the young leaf of *Artemisia Scoparia* oil, b-myrcene (24.13%) was the major constituent monoterpene, whilst p-cymene (27.06%) was the major component in mature leaf oil. The other major

Table 1. Essential oil composition of aerial parts of *Artemisia dracunculus* L.

Compound Name	RI	Area %
α-thujene	923.7	0.0731
α-pinene	932.4	1.0572
Camphene	947	0.8372
Sabinene	970.2	0.1095
β-pinene	973.9	0.3629
Myrecene	984.8	1.9397
Hexanoic acid	994.2	0.1323
α-phellendrene	999.4	0.5691
Delta-3 carene	1007.7	0.1359
P-cymene	1015.7	0.1198
1,8-cineole	1022.8	7.7162
(Z)-β-ocimene	1030.5	15.7927
(E)-β-ocimene	1041.1	3.7592
Gama terpene	1050.7	0.3335
α- Terpenolene	1080.6	10.123
Linalool	1088.2	0.0741
α-thujone	1092.2	0.033
Hexyl isobutanoate	1132.4	0.726
P-cymene-8-ol	1168.7	0.984
α-terpeneol	1187.2	0.829
Cis-piperitol	1184.8	0.1162
Nerol	1217.5	0.0111
Carrone	1221.1	0.0307
trans-Anethole	1258	28.064
Bornyl acetate	1271.9	0.7026
α-terpinylacetate	1331.7	0.0236
Noryl acetate	1340.9	0.236
Gernyl acetate	1361.9	0.1626
Hoxyl hexonate	1365.4	0.0475
α-copaene	1377.1	2.7876
β-caryophyllene	1419.7	0.747
Alpha humiene	1452.5	0.0204
Elemicin	1473.8	10.0837
Carophyllene oxide	1569.6	0.4038
Total	88.46%	

Table 2. Composition of the essential oil of *A. dracunculus* L residues by class.

Chemical class	% in essential oil
Monoterpene hydrocarbons	35.22
Oxygenated sesquiterpenes	2.09
Sesquterpene hydrocarbons	13.01
Other Constituents	30.02
Total	80.34

monoterpene constituents in young leaf oil included caryophyllene oxide (a sesquiterpene; 7.86%) and monoterpenes such as p-cymene (16.47%), (+)-limonene (8.03%) and oxygenated compounds such as capillin (a polyacetylene ketone; 7.13%) (Singh et al., 2010). Lutz et al. (2008) and Chauhan et al. (2010) also studied 24 compounds using GC-FID and GC-MS analysis. Major constituent of the essential oil was capillene (58.38%), whereas other constituents were Z-β- ocimene (8.63%), β-phellandrene (7.03%), terpenolene (5.87%), camphene (4.16%), spathulenol (2.02%), β-pinene (1.02%), etc. Chemical composition of essential oil was described and compared with earlier studies. The population was categorized as chemotype of *A. dracunculus*. As capillene is the major constituents, this chemotype may be useful for industrial exploitation as well as chemotaxonomic characterization. The composition of the volatile oils obtained from the aerial parts of *Artemisia deserti* and *Artemisia oliveriana* was analyzed by GC and GC/MS. While the oil of *A. deserti* contained camphor (45.5%), 1,8-cineole (16.7%), piperitone (8.6%), b-pinene (5.7%) and isoborneole (3.2%), the oil of *A. oliveriana* contained a-thujone (65.4%), camphor (11.5%), 1,8-cineole (9.2%) and pinocarvone (8.8%) (Rustaiyan et al., 2000). Comparing our results with those of other *Artemisia* species already published in the literature revealed considerable qualitative and quantitative similarity of the major constituents of the essential oils.

Conclusion

The essential oil found to be rich in trans-Anethole (28.06%), Z-β-ocemene (15.79%), α- Terpenolene (10.12%), Elemecin (10.08%), 1,8 cineole (7.71%) and α-copaene (2.78%) is due to its chemotypic variability which makes it an alternate source of industrial exploitation as well as chemotaxonomic characterization.

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Full Length Research Paper

A preliminary approach to halo sensitivity of sorghum cultivars

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Effect of five salt concentrations viz, 0, 130, 160, 190, 220 and 250 mM on germination and seedling growth of sorghum cultivars (*Asian, Indian, Mycho* and *Summer graze*) was evaluated under laboratory condition. Analysis of variance (ANOVA) revealed significant differences for germination percentage, plumule and radicle growth, fresh and dry weight for different salt concentrations except moisture contents. Inter cultivars genetic variation and concentration x cultivars interaction showed significant differences for all the parameters studied. Data for germination percentage, plumule and radicle growth, fresh and dry weight and moisture contents clearly demonstrated varietal differences. Though results clearly showed that salinity stress decreased all the parameters studied for sorghum cultivars significantly, tolerance of sorghum cultivars to such high levels of salt stress is worth mentioning and it can be suggested that sorghum can be tried for cultivation on moderately saline areas.

Key words: germination percentage, plumule and radicle growth, fresh and dry weight, moisture contents.

INTRODUCTION

Salinity is one of the fore most important abiotic factors that retard the activity of embryo in arid and semi arid regions. Most part of the land is under the influence of salinity and water logging. Crops growing under these saline regimes are stunted and weak resulting in low productivity (Ahmed, 2009; Ahmad et al., 2012). Although Pakistan is an agrarian economy yet we are facing scarcity of cereals and forages because of the afore-mentioned problems. Salt stress adversely affects plants at all stages of their life cycle.

Salinity affects seed germination by creating an external osmotic potential that prevents water uptake due to the toxic effects of sodium and chloride ions on the germinating seed (Khajeh-Hosseni, 2003; Kandil et al., 2012). Plants differ in their ability, to develop under saline conditions (Greenway and Munns, 1980; Kandil et al., 2012). Salinity compacts root length and plant height as the level of salt increases (Bashir et al., 2011; Saberi et al., 2011; Kandil et al., 2012). Plant scientists are confronting a challenge of salinity and water logging and they are trying to find out salinity resistant valuable plants. The aim of the present study was to search out salinity stress tolerant sorghum cultivars for vast barren salt effected land of Pakistan. Various researchers studied the effect of salinity on sorghum (Asfaw, 2011; Bashir et al., 2011; Kafi et al., 2011; Kandil et al., 2012; El-Naim et al., 2012; Rani et al., 2012). Though the study is preliminary, yet it will provide important clues and guide line for researchers to study further in this regard.

MATERIALS AND METHODS

Seeds of sorghum cultivars (*Asian, Indian, Mycho* and *Summer graze*) obtained from open market were subjected to 4 salt concentrations (130, 190, 220 and 250 mM). Tap water was used

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Table 1. Mean squares analysis of the varience table for plumule and radicle growth, germination percentage (%), fresh and dry weight and moisture contents of sorghum cultivars.

Source	Degree of freedom	Plumule growth	Radicle growth	Germination (%)	Fresh weight	Dry weight	Moisture contents
Concentration	4	0.0000^{S}	0.0000^{S}	0.0000^{S}	0.0000^{S}	0.0000^{S}	******
Cultivars	3	0.0001^{S}	0.0000^{S}	0.0000^{S}	0.0000^{S}	0.0000^{S}	0.0053^{S}
Concentration x cultivars	12	0.0000^{S}	0.0048^{S}	0.0325^{S}	0.0000^{S}	0.0000^{S}	0.0011^{S}
Error	76						
Total	99						

S = Significant NS = non significant.

Table 2. Effect of salinity on plumule growth (cm) of sorghum cultivars.

Concentration (milli moles)	*Asian*	*Indian*	*Mycho*	*Summer graze*	Concentration means
Control	7.51^{ab}	7.68^{a}	6.80^{b}	5.84^{c}	6.96^{a}
160	5.00^{c}	1.05^{fgh}	3.71^{d}	1.99^{e}	2.94^{b}
190	0.64^{gh}	0.71^{gh}	1.81^{ef}	1.49^{efg}	1.16^{c}
220	0.60^{h}	0.48^{h}	0.49^{h}	0.58^{h}	0.54^{d}
250	0.53^{h}	0.48^{h}	0.32^{h}	0.38^{h}	0.43^{d}
Cultivars means	2.86^{a}	2.08^{b}	2.63^{a}	2.06^{b}	

Lsd value at 5% level of significance for concentration = 0.4286, cultivars = 0.3833 and concentration x cultivars interaction = 0.8571. Values bearing similar letters in rows and columns are statistically non significant at 5% level of significance.

as control. Seeds viability was checked prior to the start of the experiment. Seeds were placed equidistantly on two folded Whatman # 1 filter paper as seed bed in Petri dishes of equal size (5 cm). Each treatment was replicated 5 times with 10 seeds in each replicate. Equal volume of solutions was added to the Petri dishes from respective salt concentrations. The glass wares were properly washed and sterilized at 65°C for 24 h in oven prior to use. The dishes were incubated at 25°C for 72 h. After 72 h, data for germination percentage (%), plumule and radicle growth (cm) and fresh weight (gm) were collected. Dry weight (g) and moisture contents (%) were determined after the seedlings were dried in oven at 65°C for 72 h following Hussain (1989). Fisher analysis of variance technique (1985) and LSD test at 5% probability was applied on the data to compare the differences among treatment means (Steel and Torie, 1980).

RESULTS AND DISCUSSION

Effect on plumule growth (cm)

ANOVA for plumule growth revealed significant differences for concentrations, cultivars and concentrations x cultivars interaction (Table 1). Maximum mean value (6.96 cm) for plumule growth was recorded in control, followed by 160 (2.94 cm), 190 (1.16 cm), 220 (0.45 cm) and 250 (0.43 cm) mM salt concentration, respectively. Among cultivars, *Asian* showed maximum mean value (2.86 cm) for plumule growth, while cultivar *Summer graze* showed minimum mean value (2.06 cm) for plumule growth. Differences between cultivar *Asian* and cultivar *Mycho* was not significant for plumule growth. Similarly, cultivar *Indian* and cultivar *Summer graze* showed non-significant differences. In concentrations x cultivars interaction, maximum mean value (7.68 cm) was recorded in control for cultivar *Indian,* while minimum (0.32 cm) for cultivar *Mycho* in 250 mM salt concentration (Table 2). Concentration dependent decrease was observed in plumule growth (cm) of sorghum cultivars (Figure 1). Jamil et al. (2006) reported that root and shoot lengths are important traits in salt stress sensitivity evaluation. Arrest of plumule growth of sorghum cultivars may be seen due to the toxic or injurious effects of different ions on embryo or may be due to osmotic disturbance uptake of ions was effected resulting in abnormal metabolism (Farooq and Azam, 2006; Maghsoudi-Moudi and Maghsoudi, 2008; Kazemi and Eskandari, 2011; Ahmad et al., 2012).

One may also conclude that ions of different charges bind with the domain of DNA or enzymes responsible for normal metabolism resulting in retarded plumule growth. Mechanisms of plant adaptation under salt stress are complex. The initial sites of the cell injury by any environmental stress firstly appears in cell membranes (Ashraf and Ali, 2008; Kafi et al., 2011). The imbibition of water was reduced due to the increased osmolality of salt

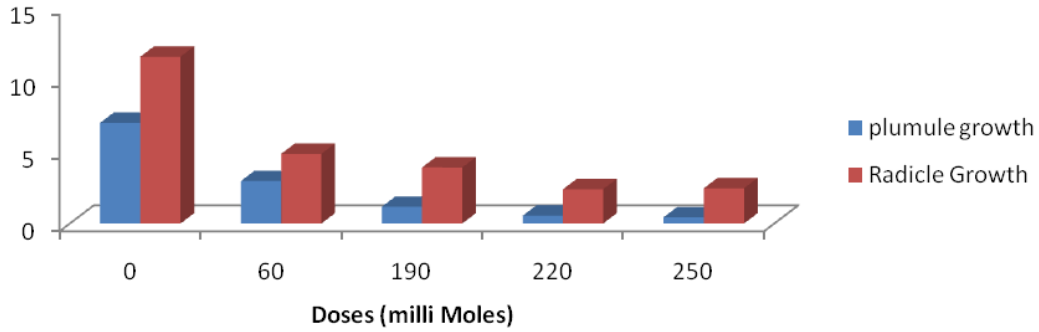

Figure 1. Effect of salinity on plumule and radicle growth (cm) of sorghum.

Table 3. Effect of salinity on radicle growth (cm) of sorghum cultivars.

Concentration (milli moles)	*Asian*	*Indian*	*Mycho*	*Summer graze*	Concentration means
Control	10.98^{b}	10.97^{b}	13.63^{a}	10.57^{b}	11.54^{a}
160	5.54^{cd}	2.57^{fhg}	7.09^{c}	4.08^{def}	4.82^{b}
190	2.23^{fgh}	2.78^{fgh}	5.45^{cd}	5.07^{de}	3.88^{b}
220	2.61^{fgh}	1.09^{h}	3.49^{efg}	2.39^{fgh}	2.37^{c}
250	2.02^{gh}	2.70^{fgh}	1.98^{gh}	3.05^{fg}	2.44^{c}
Cultivars means	4.67^{bc}	4.02^{c}	6.31^{a}	5.03^{b}	

Lsd value at 5% level of significance for concentration = 0.9550, cultivars = 0.8541 and concentration x cultivars interaction = 1.910. Values bearing similar letters in rows and columns are statistically non significant at 5% level of significance.

solutions (Wahid et al., 1998). Among cultivars, *Asian* was more tolerable to salinity and showed maximum plumule growth (Figure 4). It may be concluded that cellular membrane permeability of cultivar *Asian* is less as compared to other cultivars (Ismail, 2003). Naseer (2001) reported that the reduction in shoot length is due to excessive accumulation of salt in the cell wall which modifies the metabolic activities and limits the cell wall elasticity. Further, secondary cell appears sooner and cell wall becomes rigid, as a consequence the turgor pressure efficiency in cell enlargement decreases. These processes may cause the shoot to remain small. Our findings are parallel to those of Jamil et al. (2006) on vegetables species, Rahman et al. (2008) on wheat, Baybordi and Tabatabaei (2009), Keshavarzi (2012) on *Artemesia* and Moradi and Zavareh (2013) on chickpea, thus confirmed our findings for plumule growth.

Effect on radicle growth (cm)

ANOVA for radicle growth exhibited significant differences for concentrations, cultivars and concentrations x cultivars interaction (Table 1). Maximum mean value (11.54 cm) for radicle growth was recorded in control which was significantly different from the rest of all the salt concentrations. Differences between 160 and 190 mM salt concentrations for radicle growth were non-significant. Similarly, differences between 220 and 250 mM salt concentrations were also non significant for radicle growth. Cultivar *Mycho* showed maximum mean value (6.31 cm) for radicle growth, followed by *Summer graze* (5.03 cm), *Asian* (4.67 cm) and *Indian* (4.02 cm), respectively. In concentrations x cultivars interaction, maximum mean value (13.36 cm) for radicle growth was recorded in control for cultivar *Mycho* while minimum mean value (1.09 cm) was observed in 220 mM salt concentration for cultivar *Indian* (Table 3). Water stress condition caused marked reduction in radicle growth (Ahmad et al., 2012) (Figure 1). Salinity affects the seedling growth of plant by slow or less mobilization of reserved food, suspending the cell division, enlargement and injuring hypocotyls (Tezara et al., 2003).

The data on the average length of radicle and plumule shows that decrease in length of plumule was more pronounced as compared to radicle in all NaCl salt treatment. This indicated that plumule is more sensitive than radicle. It may be due to ability of the radicle system to control entry of ions as compared to plumule (Moradi and Zavareh, 2013). Stress tolerance is dependent upon the genetic and biochemical characteristics of the cultivars (Barakat, 2003).

Our study shows that cultivar *Mycho* is genetically well adapted to salt stress than other sorghum cultivars (Figure 4). Our findings are in complete accordance with the Keshavarzi (2012), Saberi et al. (2012) and Moradi

Table 4. Effect of salinity on germination percentage (%) of sorghum cultivars.

Concentration (milli moles)	*Asian*	*Indian*	*Mycho*	*Summer graze*	Concentration means
Control	86.00^{ab}	82.00^{abc}	96.00^{a}	76.00^{bcd}	85.00^{a}
160	68.00^{cde}	44.00^{h-k}	68.00^{cde}	40.00^{i-l}	55.00^{b}
190	64.00^{d-g}	54.00^{e-i}	58.00^{e-h}	44.00^{h-k}	55.00^{b}
220	66.00^{c-f}	36.00^{j-m}	48.00^{g-j}	20.00^{mn}	42.50^{c}
250	28.00^{k-n}	12.00^{n}	50.00^{f-j}	26.00l^{mn}	29.00^{d}
Cultivars means	62.40^{a}	45.60^{b}	64.00^{a}	41.20^{b}	

Lsd value at 5% level of significance for concentration = 8.858, cultivars = 7.923 and concentration x cultivars interaction = 17.72. Values bearing similar letters in rows and columns are statistically non significant at 5% level of significance.

and Zavareh (2013) whom studied effect of salinity on *Trifolium*, *Artemesia* and chick pea, respectively, thus confirming our results regarding radicle growth.

Effect on germination (%)

ANOVA for germination percentage (%) exhibited significant differences for concentrations, cultivars and concentrations x cultivars interaction (Table 1). Maximum mean value (85.00%) for germination percentage (%) was observed in control while minimum in 250 mM salt concentration. Control showed significant differences for all the salt concentrations. Cultivar *Mycho* showed maximum mean value (64.00%) for germination percent-tage (%) while cultivar *Summer graze* showed minimum mean value (41.20%).

Differences between cultivar *Mycho* and cultivar *Asian* was non-significant for germination percentage (%). Similarly cultivar *Indian* and cultivar *Summer graze* was non-significantly different. In interaction of concentrations x cultivars, maximum mean value (96.00%) for germination percentage (%) was recorded in control for cultivar *Mycho* while minimum mean value (12.00%) was observed for cultivar *Indian* in 250 mM salt concentration (Table 4). Salinity concentrations are inhibitory for germination percentage of sorghum cultivars (Figure 3). Salinity exerts its undesirable effects through osmotic inhibition and ionic toxicity (Munns et al., 2006). The differences in germination percentage of forage sorghum cultivars may be due the genetic factors and heredity variation among the four sorghum cultivars (Figure 6). Our results regarding germination percentage of sorghum cultivars are confirmed by Kausar et al. (2012), Keshawarzi (2012), Saberi et al. (2012) and Moradi and Zavareh (2013) who studied ill effects of salinity on germination of sorghum, *Artemesia*, *Trifolium* and chick pea, respectively. Furthermore Anwar et al. (2001), Zia and Khan (2002) and Muhammad and Hussain (2010) also reported reduced germination under saline conditions in some medicinal plants that also strengthen our findings.

Effect on fresh weight (g)

ANOVA for fresh weight was significantly different for concentrations, cultivars and concentration x cultivars interaction (Table 1). Maximum mean value (1.29 g) for fresh weight was observed in control which was significantly different from all the salt concentrations. Differences among the salt concentrations were significant for fresh weight. Cultivar *Indian* showed maximum mean value (0.75 gm) for fresh weight. While cultivar *Summer graze* showed minimum mean value (0.32 g). In interaction of concentrations x cultivars, maximum mean value (1.77 g) for fresh weight was observed for cultivar *Indian* in control which was higher than control of salt concentrations while minimum mean value (0.11 g) was recorded for cultivar *Indian* in 250 mM salt concentrations (Table 5). Increase in salinity level caused simultaneous reduction of seedling fresh weights in all sorghum cultivars (Figure 2).

This is in line with previous reports in wheat (Afzal et al., 2005), sugar beet, cabbage, amaranth and pak-choi (Jamil et al., 2006) and in sorghum (Asfaw, 2011). Thus our findings regarding fresh biomass are confirmed. Reduction in fresh biomass at higher concentration might be due to poor absorption of water from the growth medium due to physiological drought (Muhammad and Hussain, 2010). Salt stress disturbed the biochemical machinery resulting in reduced biomass. Cultivar *Indian* has got good genetic variations for salinity tolerance thus showed increase in fresh biomass (Figure 4).

Effect on dry weight (g)

ANOVA for dry weight exhibited significant differences for concentrations, cultivars and concentrations x cultivars interaction (Table 1). Maximum mean value (0.53 g) for dry weight was recorded in control while minimum in 250 mM salt concentration. Among cultivars, *Asian* showed maximum mean value (0.31 g) for dry weight while minimum mean value (0.16 g) was recorded for *Summer graze*. In concentration x cultivars interaction, control of

Table 5. Effect of salinity on fresh weight/seedling (gram) of sorghum cultivars.

Concentration (milli moles)	*Asian*	*Indian*	*Mycho*	*Summer graze*	Concentration means
Control	1.44^{b}	1.77^{a}	1.37^{b}	0.56^{ef}	1.29^{a}
160	0.84^{c}	0.81^{cd}	0.59^{de}	0.27^{ghi}	0.63^{b}
190	0.57^{ef}	0.56^{ef}	0.52^{ef}	0.39^{e-h}	0.51^{c}
220	0.35^{fgh}	0.50^{efg}	0.27^{ghi}	0.19^{hi}	0.33^{d}
250	0.26^{hi}	0.11^{i}	0.22^{hi}	0.18^{hi}	0.19^{e}
Cultivars means	0.69^{ab}	0.75^{a}	0.60^{b}	0.32^{c}	

Lsd value at 5% level of significance for concentration = 0.1161, cultivars = 0.1039 and concentration x cultivars interaction = 0.2323. Values bearing similar letters in rows and columns are statistically non significant at 5% level of significance.

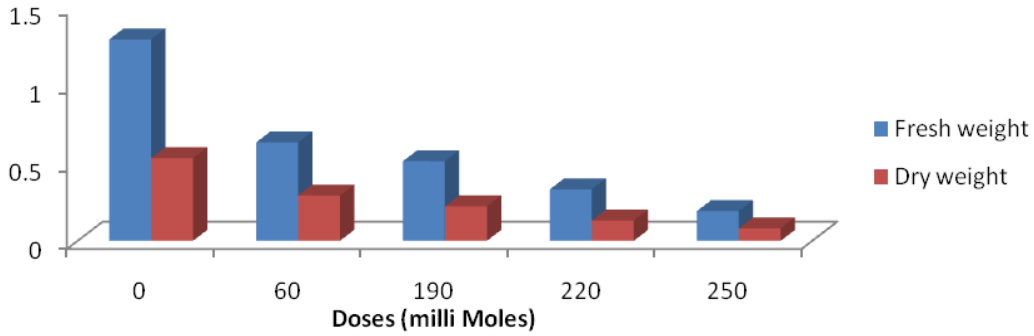

Figure 2. Effect of salinity on fresh and dry weight/seedling (gram) of sorghum.

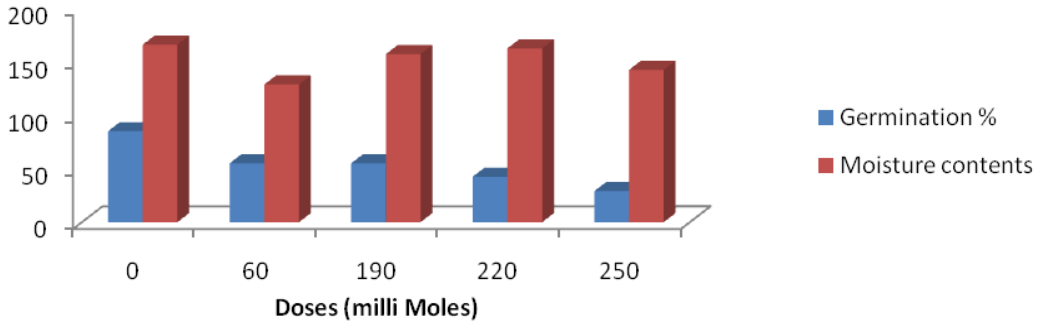

Figure 3. Effect of salinity on germination and moisture contents (%) of sorghum.

cultivar *Mycho* represented maximum mean value (0.71 g) which was higher than control of salt concentrations while minimum mean value (0.03 gm) was observed for cultivar *Indian* in 250 mM salt concentration (Table 6). Increased salinity level caused simultaneous reduction of seedling dry weights in all the cultivars (Figure 2). Salt stress disturbed the biochemical machinery resulting in reduced dry biomass. Increase dry biomass of cultivar *Asian* indicated good genetic variations for salinity (Figure 4). This is in line with previous reports in wheat (Afzal et al., 2005), sugar beet, cabbage, amaranth and pak-choi (Jamil et al., 2006) and in sorghum (Asfaw, 2011) and we reported the same. The decrease in seedling dry weight with increasing salinity level is in contrast with the findings of Muhammad and Hussain (2010) who reported increase in seedling dry weight of *Lepidium sativum*, *Linum usitatissimum*, *Plantago ovata* and *Trigonella foenum-graecum* (Figure 5).

Effect on moisture contents (%)

ANOVA for moisture contents revealed non-significant differences for concentrations while differences were

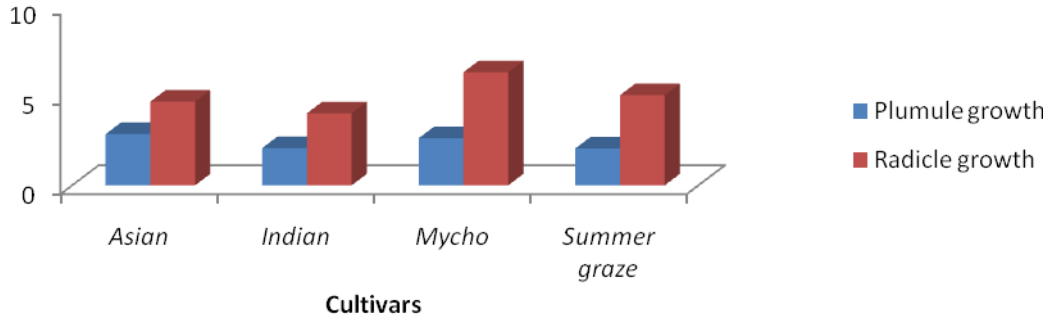

Figure 4. Effect of salinity on plumule and radicle growth (cm) of sorghum cultivars.

Table 6. Effect of salinity on dry weight/seedling (gram) of sorghum cultivars.

Concentration (milli moles)	*Asian*	*Indian*	*Mycho*	*Summer graze*	Concentration means
Control	0.60^{b}	0.43^{c}	0.71^{a}	0.36^{cd}	0.53^{a}
160	0.42^{c}	0.32^{de}	0.29^{def}	0.12^{h-k}	0.29^{b}
190	0.23^{fg}	0.25^{efg}	0.23^{fg}	0.16^{ghi}	0.22^{c}
220	0.18^{gh}	0.11^{h-k}	0.14^{hij}	0.07^{jk}	0.13^{d}
250	0.11^{h-k}	0.03^{k}	0.09^{h-k}	0.08^{ijk}	0.08^{d}
Cultivars Means	0.31^{a}	0.23^{b}	0.29^{a}	0.16^{c}	

Lsd value at 5% level of significance for concentration = 0.04454, cultivars = 0.03983 and concentration x cultivars interaction = 0.08907. Values bearing similar letters in rows and columns are statistically non significant at 5% level of significance.

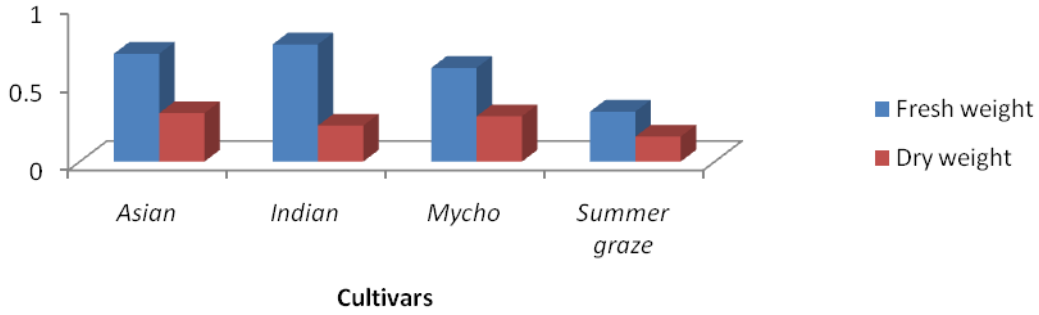

Figure 5. Effect of salinity on fresh and dry weight/seedling (gram) of sorghum cultivars.

significant for cultivars and between concentrations and cultivars interaction (Table 1). Maximum mean value (165.9 g) for moisture contents (%) was recorded in control. Cultivar *Indian* showed maximum mean value (195.4%) for moisture contents. While minimum mean value (128.9 g) was observed for cultivar *Asian*. In concentrations x cultivars interaction, maximum mean value (432.1%) for moisture contents was observed for cultivar *Indian* in control while minimum (98.8%) in 220 salt concentration for cultivar *Mycho* (Table 7). Moisture contents of seedling play an important role in various physiological processes including growth (Muhammad and Hussain, 2010; Ahmad et al., 2012). In the present study, decrease in moisture contents was recorded with salinity treatments (Figures 3 and 6). Shadded and Zaidan (1989), Naqvi and Mahmood (1994) and Ahmad et al. (2012) recorded reduction in moisture contents with increase salt stress in *Trigonella*, *Sesbania sesban* and canola, respectively, thus confirmed our findings. The decrease in moisture contents with increasing salinity level is in contrast with the findings of Ibrar and Hussain (2003), Akhtar and Hussain (2008) and Muhammad and Hussain (2010) who reported increase in moisture contents with increase in salinity concentrations while working on *Medicago*, grasses and medicinal plants, respectively.

Table 7. Effect of salinity on moisture contents (%) of sorghum cultivars.

Concentration (milli moles)	*Asian*	*Indian*	*Mycho*	*Summer graze*	Concentration Means
Control	146.1^{cd}	324.1^{a}	101.1^{d}	92.2^{d}	165.9
160	93.0^{d}	155.0^{bcd}	119.0^{cd}	147.9^{cd}	128.7
190	150.7^{cd}	163.3^{bcd}	178.0^{bcd}	136.2^{cd}	157.1
220	155.0^{cd}	195.0^{bc}	98.8^{d}	241.3^{ab}	162.5
250	140.0^{cd}	139.5^{cd}	164.0^{bcd}	126.0^{cd}	142.4
Cultivars Means	128.9^{b}	195.4^{a}	132.2^{b}	148.7^{b}	

Lsd value at 5% level of significance for Cultivars = 40.31 and Concentration x Cultivars interaction = 90.15. Values bearing similar letters in rows and columns are statistically non significant at 5% level of significance.

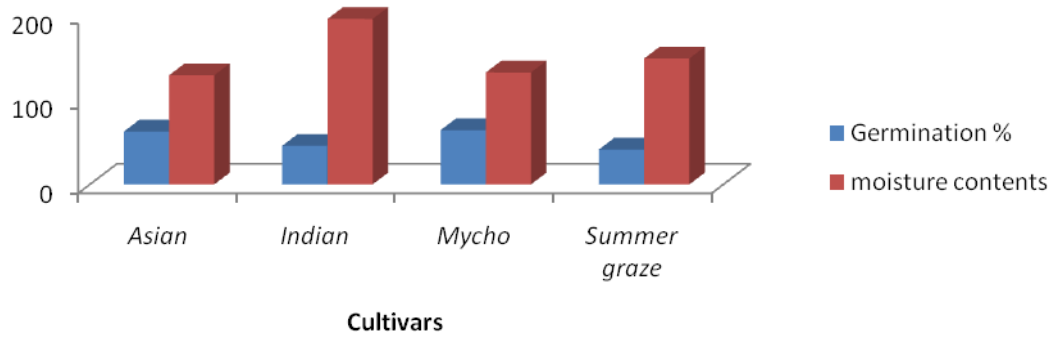

Figure 6. Effect of salinity on germination and moisture contents (%) of sorghum cultivars.

This preliminary laboratory study suggests that the tested sorghum cultivars could be grown on marginally saline habitats due to their tolerance to moderate salinity at germination and seedling stage. However, further study is needed to test their salt tolerance under field conditions to assess the possibility of cultivation of sorghum cultivars on otherwise unproductive lands.

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