

TAXONOMIC CHARACTERISTICS OF *SPINACIA OLERACEA* L.

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ABSTRACT

The research focused on the taxonomic study and proximate analysis of *Spinacia oleracea* L. The samples were fixed in formaldehyde acetic acid alcohol, dehydrated in alcohol solutions of 50%, 70% and 90%, sectioned, stained in 2% of Safranin O, counter stained in Alcian blue for 5 minutes, mounted in glycerine and photo-micrographed using Leica WILD MPS 52 camera on Leitz Draplan microscope. The proximate analysis was done using the Association of Official Analytical Chemists' methods. The result revealed that spinach is glabrous and reach up to 4 to 8ft in length. The foliar organs are simple, ovate to ovate-triangular and alternate to rosette phyllotaxy. Margins are entire to slightly serrated, reticulate veined, up to 5 ± 4 cm in length and 3 ± 2 cm in width. The basal leaves are far larger than the ones towards the apical meristem. The inflorescence has spikelet of actinomorphic flowers pollinated by wind. The fruit is a berry/ drupe. The foliar epidermal study revealed presence of simple elongated, linear and glandular trichomes, and amphistomatic diacytic stomata. Anatomical study showcased a layer of epidermis, hypodermis of 2 to 3 rolls of collenchyma, general cortex of 7 to 10 rolls of parenchyma and the pith of parenchyma. The node is unilacunar with 2 lateral leaf traces and 1 leaf trace median. The proximate compositions of: 0.19 ± 0.00 Carbohydrate (%); 3.75 ± 0.00 Protein (%); 0.02 ± 0.00 Lipid (%); 94.74 ± 0.36 Moisture (%) content; 0.62 ± 0.32 Fiber (%) and 1.18 ± 0.03 Ash (%). These information would assist for further delimitation of the species.

Keywords: Taxonomic, Spinach, morphology, anatomy, proximate.

INTRODUCTION

The history of spinach was known as far back as 2000 years ago and spread separately to the East and West in accordance to Dong and Liu (2008). It was first introduced into China around the 17th century and is now widely cultivated all over the country. Spinach spread from China to Japan and other South Asian countries and during this time, the cultivars with pointed leaves were discovered (Wang and Li, 2008). In the west, spinach spread to the northern Africa and then to

Spain in the 11th century and was widely planted in Germany in the 13th century, followed by expansion into other European countries. Spinach cultivars developed in these areas had round leaves (Bark *et al.*, 1974). Spinach was introduced into England in 1568 and then into America starting in 1806, Ryder (1979).

Spinacia oleracea L. is of the Order Caryophyllales and a member of Chenopodioideae (subfamily of Amaranthaceae/Chenopodiaceae alliance)

Kadereit *et al.* (2003). It remains one of the most nutritious leafy vegetables grown worldwide (Food and Agricultural Organization of the United Nations, 1998).

It is commonly considered a dioecious species with wind pollination, although certain cultivars, lines and crosses can produce individual plants with both male and female flowers, that is, monoecious plants (Janick and Stevenson, 1955a; Onodrea *et al.*, 2008). Spinach has a large nutritional value especially when fresh, steamed and quickly boiled. It is a rich source of vitamin A, such as carotene, vitamin C, folate and fiber (Bender and Bender, 2005). Spinach is now widely planted all over the world. There are only two confirmed taxonomic varieties of spinach, one with spiny fruits [*S. oleracea* var. *spinosa* (Moench)], and the other with spineless fruits [*S. oleracea* var. *inermis* (Moench) Peterm]; there are yet to be known additional recognized varieties or subspecies. The genus *Spinacia* is known to contain two wild relatives (*S. turkestanica* Iljin. and *S. tetrandra* Steven ex M.) of cultivated spinach, *S. oleracea* L. (Sneep, 1983; Hammer, 2001). It is noted that *S. tetrandra* Roxb. is now regarded as synonymous with *S. oleracea* L. (Lorz, 1937; Sneep, 1983). The foliar organs are within 2 to 30 cm in length and 1 to 15 cm wide; the larger ones are arranged alternately at basal part of the plant, while the smaller foliar organs are closer to the apical meristem. The flowers are inconspicuous, yellow-green, 3 to 4 mm in diameter, maturing into a small hard dry lumpy fruit cluster 5 to 10 mm in diameter containing seed (Williams, 1993). The proximate composition of spinach showed that moisture content ranged from (1.00-1.45 %), ash content (8.63-8.74 %), crude protein (14.13-14.44%), crude fiber (2.52-2.63%), crude fat (23.02-23.11%) and carbohydrate (50.10-50.59%) as reported by Amos *et al.* (2023). Apart from the descriptions given as regards spinach, there are much potential uses of the plant that are yet to be discovered. Hence, it is needful to add more information to the existing knowledge of the plant. The

objectives of the research, therefore, focuses on the taxonomic study and proximate analysis of *Spinacia oleracea* L.

MATERIALS AND METHODS

Geographic Location

The plants were collected fresh in a garden located in Young Chief Lane, Umuchiolu Aluu in Ikwerre Local Government Area of Rivers State of Nigeria.

Morphological Studies

The meter rule was used to ascertain morphological measurements of plant parts such as: plant height from the root-collar to the terminal bud, the leaf length from the leaf tip to the petiole base and the leaf width across the leaf lamina, from one margin to another at the widest region.

Epidermal Studies

Fresh leaves collected were peeled chemically with nitric acid, and made to pass through alcohol solutions in the ratios of 50%, 75% and absolute alcohol for 5 minutes in each and thereafter stained with Safranin O, rinsed with distilled water and counter stained with Alcian blue for 5 minutes in each, rinsed again and mounted in aqueous glycerol solution placed on glass slide with coverslip following the method of Cutler (1977). Slides with good sections were placed on the stage, viewed and photo-micro graphed using Leica WILD MPS 52 microscope camera on Leitz Dra plan microscope.

Anatomical Studies

Spinacia oleracea L. stems, leaves, petioles, flowers, fruits and roots harvested for the study, were fixed in FAA prepared in the ratio of 1:1:18 of 40% formaldehyde, glacial acetic acid and 70% alcohol for 2 to 48 hours following the methods of Johansen (1940) modified; Free hand sections were done as described by Wahua (2020). Slides with good sections were placed on the stage, viewed and photo-micro graphed using Leica WILD MPS 52 microscope camera on Leitz Dra plan microscope.

Proximate Properties

The AOAC (1990) method was employed for the analysis of the proximate composition done as shown below:

Carbohydrate (Cleg Anthrone Method)

The formula used for carbohydrate analysis is as showcased below:

Proteins (Kjeldahl method)

The protein analysis was done using the formula shown below:

$$\begin{aligned} \% \text{CHO as glucose} &= \frac{25 \text{ ml x absorbance of}}{\text{Absorbance of standard glucose x}} \\ &= \frac{\text{Titre value x } 1.4 \times 100 \times 100}{1000 \times 20 \times 0.1} \end{aligned}$$

Where 1.4 = Nitrogen equivalent to the normality of the HCl used in the titration 0.1N

100 = the total volume of digest dilution

100 = percentage factor

0.1g of the sample

1000 = conversion from gram to milligram

20 = integral volume of digits analyzed or distilled

0.1g = the weight of sample in gram digested

Lipid (Soxhlet Extraction Method)

The % lipid composition was calculated using the formula:

$$\% \text{ Lipid} = \frac{\text{Weight of flask and extract-weight of empty}}{\text{Weight of sample extracted}} \times \frac{100}{1}$$

Moisture (Air Oven Method)

The % moisture content was analyzed using the formula as shown below:

$$\% \text{ Moisture} = \frac{\text{Weight of fresh sample} - \text{Weight of dried sample}}{\text{Weight of sample used}} \times \frac{100}{1}$$

Crude Fiber

The % crude fiber composition was done using the formula below:

Where W_1 = weight of sample

W_2 = weight of oven dried residue

W_3 = weight of dried residue

$$\% \text{ Crude Fiber} = \frac{W_2 - W_3}{W_1} \times \frac{100}{1}$$

Ash (Furnace Method)

The % ash was calculated using the formula as shown below:

$$\% \text{ Ash} = \frac{\text{Weight of crucible + Ash sample - weight of}}{\text{Weight of sample}} \times \frac{100}{1}$$

RESULTS**Morphological Study**

It is glabrous and grows up to 4 to 8 ft. or more in length. The petiolate simple foliar organs are ovate triangular and alternate to rosette phyllotaxy, having margins that are entire to slightly serrated, palmate veined, measuring up to 5 ± 4 cm in length and 3 ± 2 cm in width. The inflorescence has spikelet of actinomorphic flowers. The fruit has a strong nut or seed. The results for the morphological studies are presented in plate 1 and table 1.

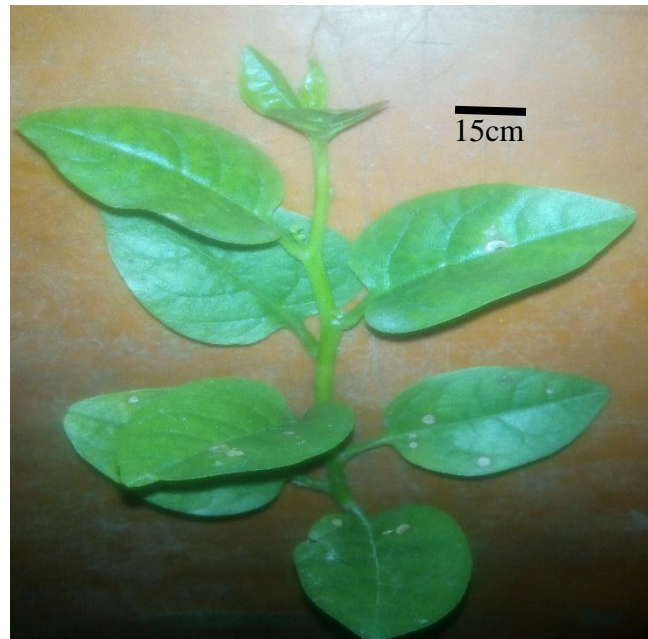


Plate 1: *Spinacia oleracea* L.

Table 1: summary of morphological characteristics of *Spinacia oleracea* L.

Characters	<i>Spinacia oleracea</i> L.
Habit	Herbaceous
Duration	Annual or biennial
Root	Produced from the nodes and shallow rooted.
Stem Description	Climber and trailing habit, glabrous and grows up to 4 to 8 ft. in length
Leaf type	Petiolate and simple, cuneate at base
Leaf venation type	Reticulate
Phyllotaxy	Alternate to rosette
Leaf outline or shape	Ovate to ovate-triangular
Leaf margin	Entire to slightly serrated
Length of leaf (cm)	7cm
Range	2.0 -9.0cm
Mean	5 ± 4 cm
Breadth of leaf (cm)	4cm
Range	1.0 -5.0cmm
Mean	3 ± 2 cm
Nodal anatomy	Unilacunar with 2 lateral leaf traces

Flower description	Numerous, actinomorphic, yellowish green.
Fruit description	A berry (drupe).

Epidermal Study

The foliar epidermal study revealed presence of simple elongated, linear and glandular trichomes as showcased with the red arrows in plates 2a and 2c; diacytic stomata which are amphistomatic in nature, more numerous in the abaxial foliar epidermis, see plate 2b.

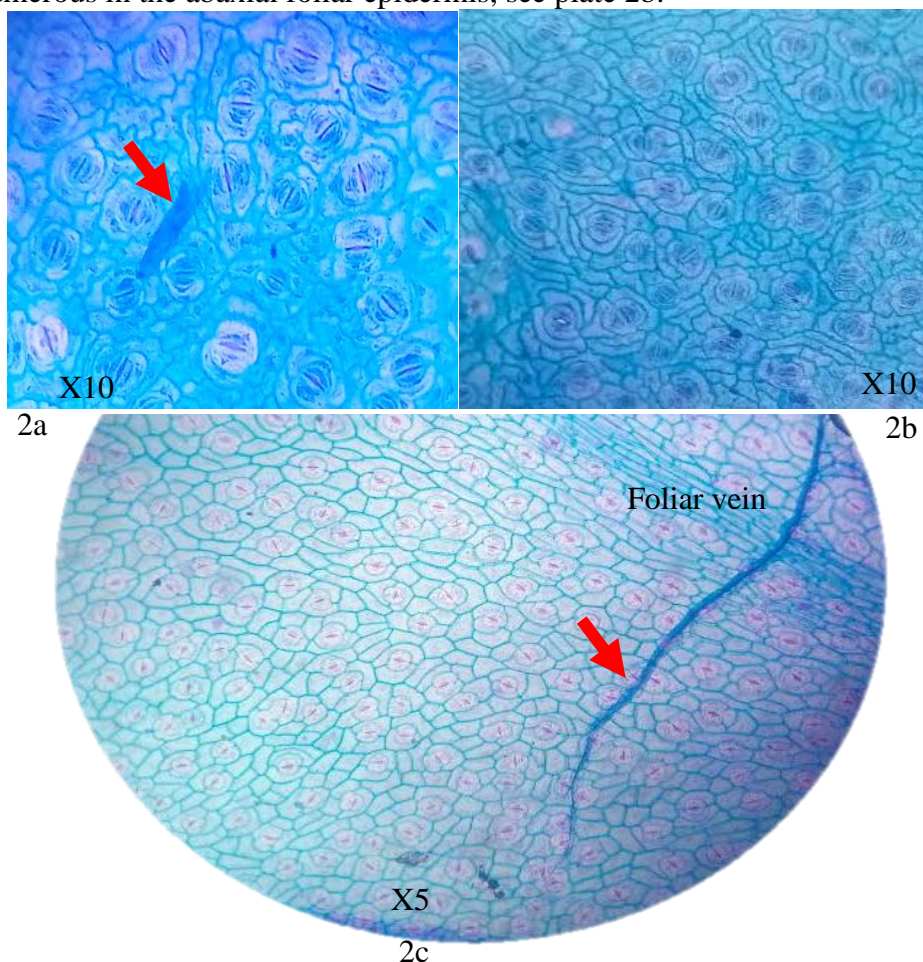


Plate 2a: *Spinacia oleracea* L. Adaxial foliar Epidermis. Arrow revealed linear shaped trichome,

Plate 2b: Abaxial epidermis showing numerous diacytic stomata; Plate 3: *Spinacia oleracea* L. Abaxial epidermis arrow showcased a linear and lengthy trichome transversing leaf vein.

Anatomical study

Anatomical study showed a single roll of cells in the epidermis. The hypodermis is made of 2 to 3 layers of collenchyma, the general cortex and pith is composed 7 to 10 rolls of parenchyma and this cuts across the mid-ribs, petioles, stems, nodes and roots, occurring in similar locations except that the number of rolls of cells are not the same and vascular system is an open type. The central star-shaped vessels is the xylem tissues (X) shown in plate 3d, hence no pith, while the phloem (P) alternates with the xylem. See plates 3a to 3e below:

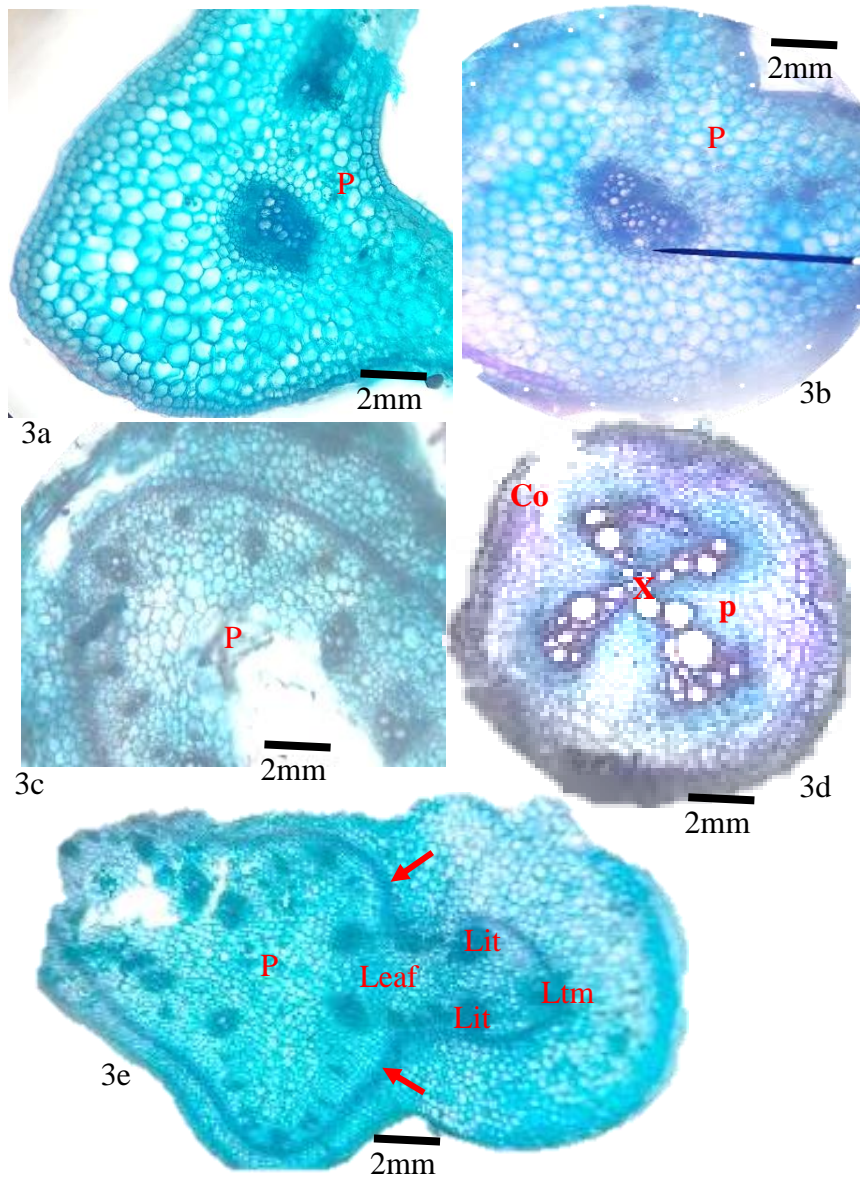


Plate 3: *Spinacia oleracea* T.S. anatomy; 3a: Mid-rib anatomy; 3b: Petiole anatomy with a pair of vascular wings; 3c: Stem anatomy; 3d: Root anatomy revealing xylem vessels (X) occupying the central position alternating with the phloem tissues (p). Below the root piliferous layer is the collenchyma, (Co.); 3e: Nodal anatomy with unilacunar node with 1 leaf gap, 2 lateral leaf traces (Lit) and 1 leaf trace median (Ltm). Arrows revealed stem vascular ring.

Proximate Analysis

Table 2: Proximate composition of *Spinacia oleracea* L

Carbohydrate %	0.19±0.00
Proteins %	3.75±0.00
Lipids %	0.02±0.00
Moisture content %	94.74±0.36
Fiber %	0.62±0.32
Ash content %	1.18±0.03

DISCUSSION

The morphological description of *Spinacia oleracea* L. used for this work is in consonance with the one given by FAOSTAT (1998). It is a rich source of fiber content and other nutrients as also supported by Bender and Bender (2005). Spinach is a prominent climber and the flowers are inconspicuous, yellowish green and the inflorescences are spikelet of actinomorphic flowers which produce seeds, early formed ones are (berries) and mature into small hard dry lumpy fruits (drupe), this is also in accordance to the work of Williams (1993). The stomatal complex is diacytic and amphistomatic in nature, more numerous on the abaxial foliar surface than as in the adaxial region and consists of linear and glandular trichomes. The nodal anatomy revealed unilacunar state with two lateral leaf traces and one leaf trace, median. The percentages of proximate compositions were far more of lower contents than those of Amos *et al.* (2023), except that the moisture content was on the contrary, being highest with (94.74 ± 0.36 %) as opposed to (1.00-1.45 %).

CONCLUSION

Spinacia oleracea L. has scanty work in anatomy especially as it has to do with nodal anatomy. A very clear background of types of trichomes and stomatal complexes are contributions made. More research findings in the DNA barcode is recommended.

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