

Anatomical and Phytochemical Studies on *Benincasa hispida* (Thunb.) Cogn. (Cucurbitaceae)

Chimezie EKEKE*, Chinedum A. OGAZIE, Ikechukwu O. AGBAGWA

University of Port Harcourt, Faculty of Science, Department of Plant Science and Biotechnology, Nigeria; ekeke.uche@uniport.edu.ng;
ogazie.chinedum@uniport.edu.ng; ikechukwu.agbagwa@uniport.edu.ng (*corresponding author); ekeke.uche@uniport.edu.ng

Abstract

The morphological and anatomical studies of *Benincasa hispida* (Thunb.) Cogn. from Nigeria was carried out with the view to reporting morphological and anatomical characteristics for the first time. Physical and microscopic (microtomy) observations were used. *B. hispida* is a monoecious climbing or trailing herb, stem hairy, 5-angled, with suborbicular stipuliform bract at the petiole-base; leaves simple, very hairy on both surface, alternate, blade palmately or ovate in young plant, base cordate. Flowering occurs between April and May. Female flowers solitary, male flowers solitary or in a slender-pedunculate racemes, petals-5, cream, yellow or pale yellow, ovary ellipsoid, ovules many, stigma 3-lobed and stamen 3. Fruits are large, weighs 8.5 - 9.0 kg, succulent, densely hairy when young, with a thick waxy deposit when mature, cylindrical to oblong with hairy stalk. Seeds are ovate-obovate, cream. Leaves and petals of male flower are amphistomatic with anomocytic, tetracytic and anisocytic stomata while petals of the female flower are hypostomatic with anomocytic stomata only on the abaxial surface. It has glandular and non-glandular trichomes with uniseriate stalk, clavate and multicellular gland heads. The midrib, petiole, stem, tendril, male and female flower stalks and tendril have hollow pith with 3, 9, 6-7, 16, 14 and 10 bicollateral vascular bundles respectively. The percentage crude protein, ash, carbohydrate, lipid, crude fibre, alkaloid, flavonoid, tannin and phytate could account for the numerous medicinal properties.

Keywords: amphistomatic; anatomy; *Benincasa hispida*; glandular trichome; hypostomatic; morphology; phytochemical

Introduction

The Cucurbitaceae is most diverse in tropical and subtropical regions with hotspots in Southeast Asia, West Africa, Madagascar, and Mexico (Schaefer and Renner, 2011) and is divided into two subfamilies Nandiroboideae also called Zanonioideae (Jeffrey, 1990) with 19 genera and 60 species and Cucurbitoidae with 111 genera and 740 species. The subfamily Cucurbitoidae comprises some of the most important tribes and genera within cucurbits. These include Benincaseae (*Benincasa*, *Citrullus*, *Coccinia*, *Lagenaria*, *Cucumeropsis*, *Cucumis* and *Zehneria*), Luffeae (*Luffa*), Cucurbitae (*Cucurbita*), Sicyeae (*Cyclanthera*, *Sechium*), Joliffieae (*Momordica*, *Telfairia*), Schizopeponeae (*Schizopepon*), Trichosantheae (*Hodgsonia*, *Trichosanthes*), Bryonieae (*Echallium*), Herpetospermae and Coniandreae (Jeffrey, 1990; Rubatzky and Yamaguchi, 1997; Jeffrey, 2005).

Benincasa (Cucurbitaceae) is a monotypic genus with a single species. It is indigenous to both arid and temperate regions of the earth and requires long periods of warm, dry weather for their optimal growth (Whitaker and Davis,

1962). It is under cultivation in large areas in Indian states (Cheran *et al.*, 2010). *B. hispida* has a chromosome number of $2n = 24$ (Mini-Raj *et al.*, 1993; Robinson and Decker-Walters, 1993) and four cultivars based on the size, shape, fuzziness, waxiness, and presence or absence of a dusty or ashy layer (Rubatzky and Yamaguchi, 1997). The fruits of this species could be medium or large and weigh from 7.5 to 45 kg depending on genotype and the production system (Rubatzky and Yamaguchi, 1997; Stephens, 1988) with varying shapes (oblong to cylindrical or elongated) (Stephens, 1988; Rubatzky and Yamaguchi, 1997).

B. hispida is widely used as medicine and has nutritional, pharmacological and medicinal properties some of which include antioxidant activity (Mingyu *et al.*, 1995; Yoshizumi *et al.*, 1998; Huang *et al.*, 2004; Du *et al.*, 2005), anti-compulsive effect (Girdhar *et al.*, 2010), angiotension-converting enzyme (ACE) inhibitor activity *in vitro* (Huang *et al.*, 2004; Bhalodia *et al.*, 2009), Alzheimer disease treatment (Cantuti *et al.*, 2000; Maciuk *et al.*, 2002; Roy *et al.*, 2008), anti-ulcer (Grover *et al.*, 2001; Shetty *et al.*, 2008; Rachchh and Jain, 2009), anti-inflammatory (Cuzzocrea *et al.*, 2001; Chandrababu and Umamaheshwari, 2002; Shetty

et al., 2008; Gill et al., 2010), anti-obesity (Zhang, 1996; Kumar and Vimalavathini, 2004; Nadhiya et al., 2016) and anti-diarrheal agent (Ammon et al., 1974; Gaginella et al., 1975; Mathad et al., 2005). The fruits contain volatile oils, flavonoids, glycosides, saccharides, proteins, carotenes, vitamins, minerals, β -sitosterin and uronic acid (Nurul et al., 2011).

Among Nigerian cucurbits, there are many works on their anatomy and morphology (Okoli 1984; Ndukwu and Okoli, 1992; Agbagwa and Ndukwu, 2004; Aguoru and Okoli, 2012; Ajuru and Okoli, 2013; Agogbua et al., 2015a; Ekeke et al., 2015), cytology (Okoli, 1984, 1987; Agbagwa, et al., 2007), ethno-botanical values (Chike et al., 2006; Omara-Achong et al., 2012) and polyploidy induction (Agogbua et al., 2015b; Agogbua and Ekeke, 2017). However, there is no record on the occurrence of *B. hispida* in Nigerian. It is yet to be described and is scarcely studied. There is relatively no report on the morphology, anatomy, phytochemistry and cytology of this species from Nigeria. This work presents the first report on the morphology, anatomy and phytochemistry *B. hispida* from Nigeria.

Materials and Methods

Source of plant material

The plant material studied in this work was collected from the University of Port Harcourt Biodiversity Center (a forest over 50 year old), South-South, Nigeria. The plant was identified, processed and deposited in the University of Port Harcourt Herbarium (UPH/V/1264). The morphological attributes of the species were described and recorded.

Epidermal studies

Foliar materials for epidermal studies were collected fresh from plants growing in the wild. The adaxial and abaxial epidermal surfaces were peeled, stained with 1% safranin or alcian blue, rinsed with distilled water to remove excess stain, mounted in a drop of pure glycerine on clean glass slides; coverslips placed over the peels and sealed with nail varnish to prevent dehydration (Ndukwu and Okoli, 1992). The slides were observed using a microscope and the epidermal features described by Metcalfe and Chalk (1979) and stomatal types described according to Malvey (2004).

Anatomical studies

Cut sections of petiole, midrib from matured leaves and young stems were fixed in FAA (formaldehyde:glacial acetic acid:ethanol in the ratio of 1:1:18 parts of 70% ethanol v/v) for at least 48 hours. These materials were washed in several changes of distilled water, dehydrated through alcohol series (30%, 50%, 70% and 100%) solution 2 hours in each and embedded in wax. Sections were cut on a Leitz 1512 rotary microtome at thickness between 15 - 20 μ m. The thin sections selected were de-waxed and stained with 1% Safranin O and counterstained with Alcian blue, mounted on slides and photomicrographs of the anatomical sections taken with Leitz Diaplan photomicroscope fitted with Leica WILD MPS 52 camera.

Proximate and phytochemical studies

The leaves of these plant species were detached washed with distilled water, air-dried, ground into powder using an electronic blender, sieved with 80 micron mesh and 200g of each sample stored in glass bottles. The samples were analysed for crude protein, ash and crude fibre according to the AOAC Official Method (AOAC, 2000), 920.152, 930.05, 948.22 and 935.53 respectively. The method of determination of phytochemicals were oxalates (Sanchez-Alonso and Lachica, 1987), alkaloids and flavonoid (Harborne, 1973), saponins (Obadoni and Ochuko, 2001), lipid (Yaniv et al., 1999) and tannin (AOAC, 2006).

Results

Habit, habitat and morphology of *B. hispida*

B. hispida grows on moist soils, sandy or seasonal flood plain. It is a monoecious climbing or trailing herb; stem hairy, 5-angled, with suborbicular stipuliform bract at the petiole-base; internode 10.2 - 13.1 cm; leaves simple, 9.8 - 14.1 cm long, 1.0 - 1.7 cm wide, very hairy on both surface, alternate, blade palmately 5 - 7-lobed or ovate in young plant, lobes ovate-triangular or triangular, apex acute - acuminate, base cordate; petiole hairy, 5.8 - 11.0 cm long; 6 - 7-angled; tendril bifid at 1.8 - 2.7 cm (Figs. 1a, 1b, 1c and 1d). Flowering occurs between April and May. Female flowers solitary, petals-5, cream, yellow or pale yellow, 4.0 x 5.0 cm; ovary ellipsoid, ovules many, receptacle-tube short, stigma 3-lobed, pedicle very hairy, 1.0 - 1.5 cm long (Fig. 1e). Male flowers solitary or in a slender-pedunculate racemes; fairly coxillary with female flowers, petals-5, yellow or cream, entire, hairy, campanulate, receptacle-tube relatively short and broad, tube 0.6 - 1.0 cm; sepals hairy, ovate-obovate, 3.8 x 4.6 cm, apex acute, pedicles slender, 4.8 - 8.1 cm long; stamen 3, filaments free, inserted on the tube (Fig. 1f).

Fruits are large, succulent, densely hairy when young (Figs. 2a and 2b), with a thick waxy deposit when mature and 58.6 - 59.4 x 14.0 - 15.5 cm (Figs. 2c and 2d), cylindrical to oblong with hairy stalk (Fig. 2e). Matured fruits weigh 8.5 - 9.0 kg. Seeds are ovate-obovate, cream, 7.8 - 12.9 mm long, 5.7 - 7.0 mm wide, 1.5 - 2.4 mm thick, 100 seeds weigh 4.12 - 5.10 g (Fig. 2h).

Epidermal characteristics

Three stomata types namely anomocytic, tetracytic and anisocytic occurred in this species (Fig. 3 and Table 1).

Leaves

The leaf is amphistomatic. The abaxial (upper) and adaxial (lower) epidermises have anomocytic, tetracytic and anisocytic stomata with the anomocytic and tetracytic stomata predominating. The shape of the adaxial epidermal cells are irregular (Fig. 3A) while the abaxial epidermal cells are polygonal (Fig. 3B). The adaxial anticlinal cell walls are slightly straight or curved while the abaxial ones are undulating. The stomatal indexes are 80.86 ± 2.59 and 32.39 ± 5.11 on the lower and upper epidermises respectively (Table 1).

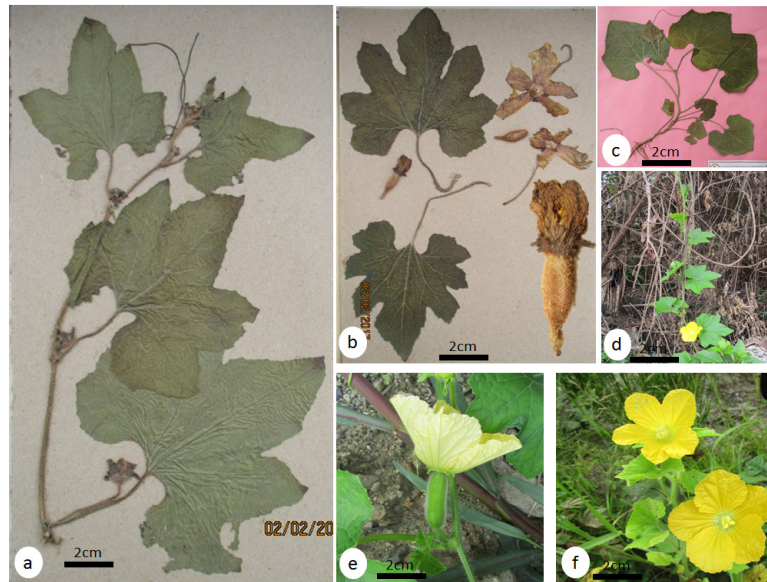


Fig. 1. Vegetative and reproductive parts of *B. hispida*: a – c. voucher specimens-UPH/V/1264, a & b. mature plants, c. seedling, d. plant habit, e. female flower and f. male flower

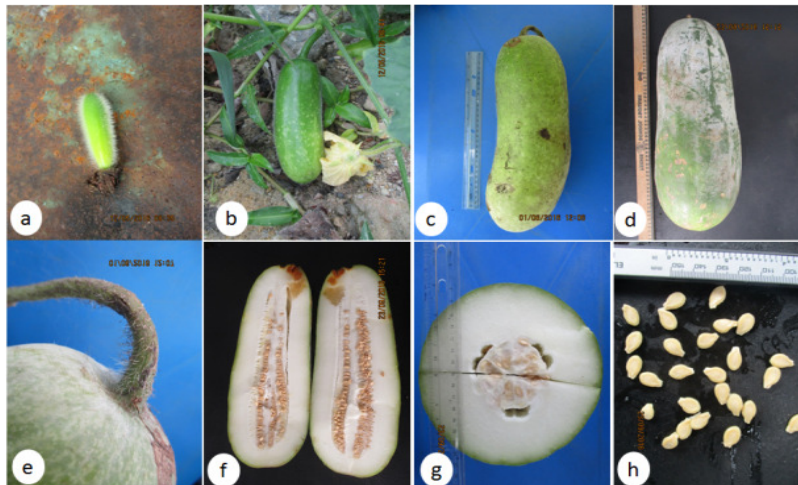


Fig. 2. Fruits and seeds of *B. hispida*: a – c. young/immature fruits, d. mature fruit, e. hairy fruit-stalk, f. longitudinal view of the fruit, g. transvers view of the fruit, and h. seeds

Table 1. Epidermal characteristics of leaf, male and female flowers of *B. hispida*

| Epidermal character | Leaf | | Male flower | | Female flower | |
|------------------------------|---------------------------------------|---------------------------------------|-----------------------------|----------------------------|------------------------------|------------|
| | Adaxial | Abaxial | Adaxial | Abaxial | Adaxial | Abaxial |
| Shape of epidermal cell | Polygonal | Irregular | Polygonal | Irregular | Regular and partly Irregular | Irregular |
| Anticlinal cell wall pattern | Slightly straight or curved | Undulating | Slightly straight or curved | Undulating | Undulating | Undulating |
| Stomata type | Anomocytic, tetracytic and anisocytic | Anomocytic, tetracytic and anisocytic | Anomocytic | Anomocytic and tetracytic | - | Amomocytic |
| Stomata index (S.I) | 29.03 - 40.0 (32.39±5.11) | 77.78- 83.87 (80.86±2.59) | 0.0-1.05 (0.77±0.51) | 7.14 - 7.69 (7.45±0.24) | 4.76 - 10.53 (7.99±2.95) | 0.00 |
| Trichome | + | + | + | + | | |
| Glandular | + | + | - | + | + | |

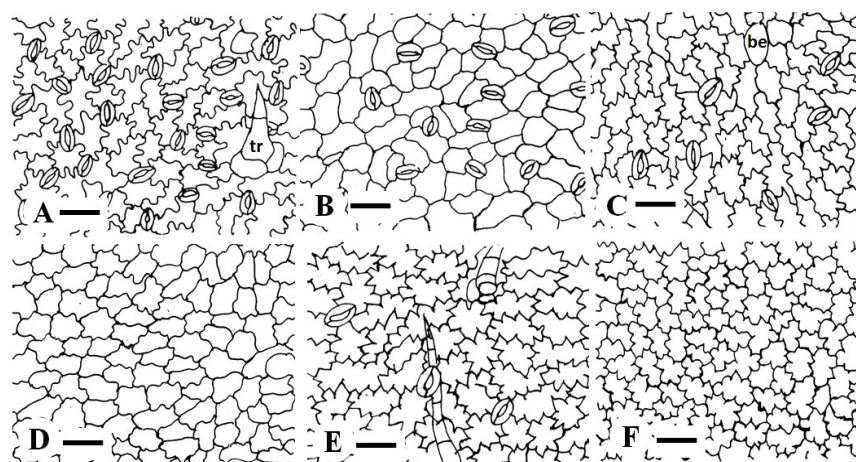


Fig. 3. Epidermal characteristic of *B. hispida*: Leaf (A and B); female flower (C and D) and male Flower (E and F) bar = 45μm

Male flower

The petals of the male flower are amphistomatic having anomocytic and tetracytic stomata. Anomocytic and tetracytic stomata were recorded on the abaxial surface while adaxial surface had only anomocytic stomata (Figs. 3C and 3D). The adaxial anticlinal cell walls are slightly straight or curved while the abaxial ones are undulating. The stomatal indexes are 7.45 ± 0.24 and 0.77 ± 0.51 on the abaxial and adaxial surfaces respectively.

Female flower

The petals of the female flower are hypostomatic with anomocytic stomata only on the abaxial surface (Figs. 3E and 3F). The shape both abaxial and adaxial epidermal cells irregular and the anticlinal cell walls are undulating.

Trichome types and distribution

The trichomes present in this species are glandular and non-glandular with uniseriate stalk, clavate and multicellular gland heads (Fig. 4). The non-glandular trichome types (Figs. 4a, 4b, 4c and 4d) are found on the adaxial and abaxial surfaces of the male flower petals while glandular trichome types (Figs. 4g, 4h, 4i, 4j and 4k) occurred on the adaxial epidermal surface of the male flower petals. The petals of the female flowers have glandular (multicellular gland head) trichome (Fig. 4i) and all the forms of non-glandular trichome identified on the lower epidermal surface of the male flower. Also the non-glandular trichome (Fig. 4l) and glandular trichome (Fig. 4i) were recorded on the adaxial surface of the leaf with the non-glandular types occurring mainly on the veins while the abaxial surface had trichome types (Figs. 4f and 4g).

Anatomical characteristics

The result of the anatomical characteristics of the midrib, petiole, stem, tendrils, male and female flower stalks and tendrils is presented in Table 2, Figs. 5, 6 and 7.

Midrib/lamina

The midrib has three bicollateral vascular bundles arranged parallel to each other (Figs. 5A, 5B). The lamina contains 1-layer of palisade mesophyll and loosely packed spongy mesophyll with intercellular spaces. The upper and lower epidermises comprised 1-layer of periclinally elongated cells (Fig. 5C).

Petiole

The petiole has hollow pith, 320 - 360 μm wide, cortex 20 - 24 μm thick, collenchyma 2 - 4 μm thick with 9 vascular bundles (Table 2, Figs. 6A and B).

Tendrils

The tendril has hollow pith, 180 - 220 μm wide, cortex 64 - 76 μm thick and sclerenchyma 2 - 3 μm thick with 6 - 7 vascular bundles (Table 2, Figs. 6E and D).

Male flower stalk

The male flower stalk has hollow pith, 180 - 220 μm wide, and cortex 53 - 64 μm thick, and sclerenchyma 2 - 3 μm thick and 16-vascular bundles (Table 2, Figs. 7A and B).

Female flower stalk

The young female flower stalk pith is not hollow, cortex 112 - 116 μm thick, sclerenchyma 2 - 3 μm thick with 14 vascular bundles (Table 2, Figs. 7C and D).

Table 2. Anatomical Characteristics of *B. hispida*

| Plant part | Nature of pith | TOC (μm) | DP (μm) | NVB | TS (μm) |
|---------------------|----------------|-----------|-----------|-------|---------|
| Female flower stalk | Not hollow | 112 - 116 | 340 - 400 | 14 | 6 - 7 |
| Stem | Hollow | 83 - 96 | 250 - 370 | 10 | 3 - 4 |
| Midrib | Not hollow | NA | NA | 3 | NA |
| Tendrils | Hollow | 64 - 76 | 180 - 220 | 6 - 7 | 2 - 3 |
| Petiole | Hollow | 20 - 24 | 320 - 360 | 9 | NA |
| Male flower stalk | Hollow | 53 - 64 | 500 - 540 | 16 | 2 - 3 |

Note: DP = Diameter of pith, TOC = Thickness of cortex, TS = Thickness of Sclerenchyma, NA = Not applicable and NVB = Number of vascular bundle

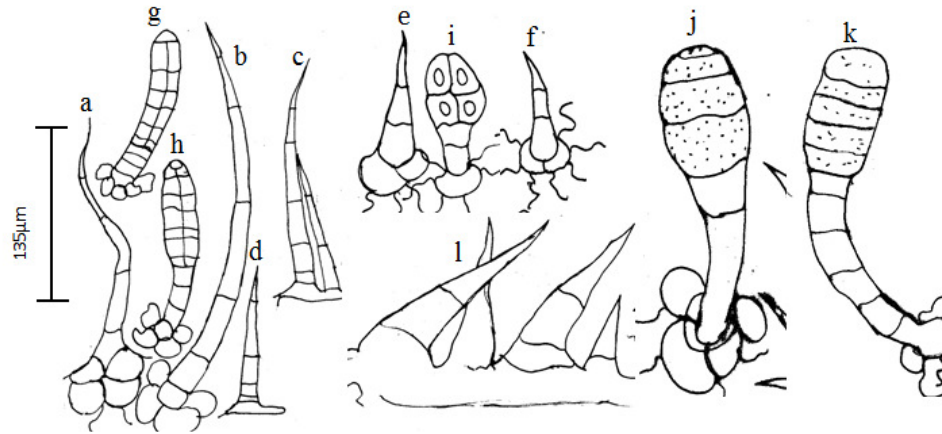


Fig. 4. Trichome types in *B. hispida* (a, b, c, d, e, f and l) non-glandular trichomes and (g, h, i, j and k) glandular trichomes, (g, h and i) multicellular gland head and (j and k) clavate gland head

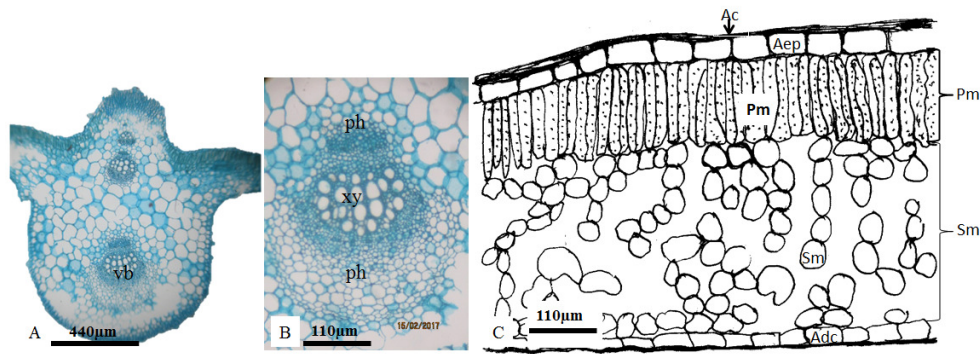


Fig. 5. Anatomy of *B. hispida* (A and B) midrib and (C) leaf lamina (vb = vascular bundle, xy = xylem, ph = phloem, Ac = adaxial cuticle, Aep = adaxial epidermis, Pm = palisade mesophyll, Sm = spongy mesophyll, Adc = abaxial epidermis)

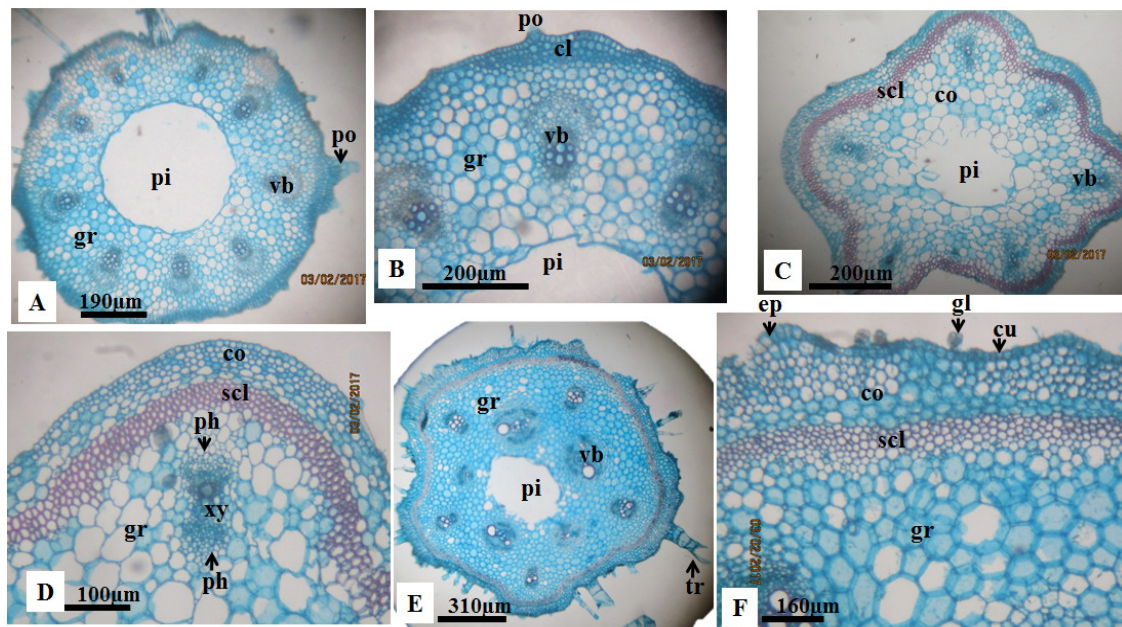


Fig. 6. Anatomy of stem of *B. hispida* (A and B), petiole (C and D) and Tendril (E and F) Abbreviations: vb = vascular bundle, pi = pith, cu = cuticle, ep = epidermis, po = papillous, co = cortex, gr = ground tissue, ph = phloem, xy = xylem, tr = trichome, scl = sclerenchyma

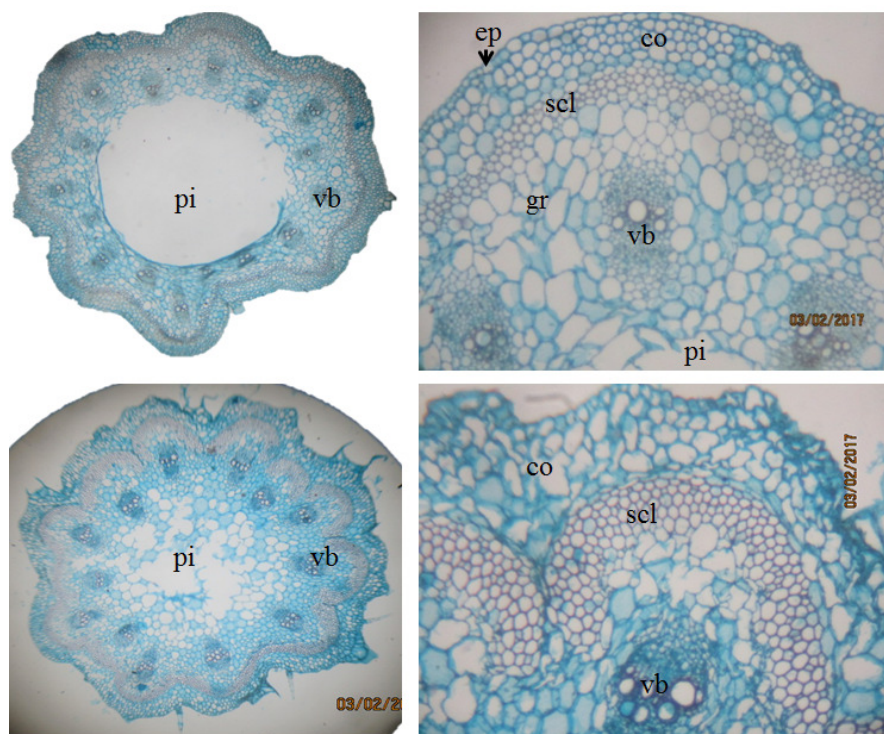


Fig. 7. Anatomy of flower stalks of *B. hispida* (A and B) = male flower and (C and D) = female flower (pi = pith, vb = vascular bundle, scl = sclerenchymatous cells, co = cortex, ep = epidermal cell, gr = ground tissue)

Stem

The stem has hollow pith, 250 - 370 μm wide, cortex 83 - 96 μm thick, sclerenchymatous cells 3 - 4 μm thick with 10 vascular bundles in two concentric rings (Table 2, Figs. 6E and F).

Proximate and phytochemical composition

The percentage crude protein, ash, carbohydrate, lipid, moisture, crude fibre, alkaloid, flavonoid, tannin and phytate concentrations are 0.37 ± 0.03 , 0.52 ± 0.08 , $3.23 \pm$

0.11 , 0.42 ± 0.07 , 94.20 ± 0.10 , 1.25 ± 0.06 , 4.20 ± 0.10 , 1.13 ± 0.15 , 0.09 ± 0.01 and 0.06 ± 0.00 and 0.49 ± 0.02 , 0.21 ± 0.01 , 3.36 ± 0.05 , 0.60 ± 0.10 , 95.40 ± 0.53 , 0.32 ± 0.02 , 4.35 ± 0.05 , 0.95 ± 0.05 , 0.07 ± 0.02 and 0.06 ± 0.01 for skin and pulp respectively (Table 3). Also, in the seeds the percentage concentrations of crude protein, ash, carbohydrate, lipid, moisture, crude fibre, tannin and phytate are 0.58 ± 0.02 , 9.33 ± 0.58 , 9.43 ± 0.07 , 0.77 ± 0.15 , 37.60 ± 0.53 , 42.49 ± 0.50 , 0.09 ± 0.01 and 0.06 ± 0.00 (Table 3).

Table 3. Proximate and phytochemical composition of mature *B. hispida* (g/100g)

| Parameter | Present work | | | Wills <i>et al.</i> (1984) | Morton, 1971 | Tee <i>et al.</i> (1997) | Mingyu <i>et al.</i> (1995) | FAO, 1972 |
|---------------|-----------------|------------------|------------------|-------------------------------|-----------------|-----------------------------|--------------------------------|-----------|
| | Pulp | Skin | Seed | | | | | |
| Moisture | 95.4 \pm 0.53 | 94.20 \pm 0.10 | 37.60 \pm 0.53 | 96.80 | 96.2 | 94.5 | 96.7 | 96.20 |
| Crude Protein | 0.49 \pm 0.02 | 0.37 \pm 0.03 | 0.58 \pm 0.02 | 0.30 | 0.40 | 0.50 | 0.40 | 0.50 |
| Carbohydrate | 3.36 \pm 0.05 | 3.23 \pm 0.11 | 9.43 \pm 0.07 | 1.10 | 2.24 | 4.00 | 2.56 | 2.30 |
| Crude fibre | 0.32 \pm 0.02 | 0.42 \pm 0.07 | 42.49 \pm 0.50 | 1.50 | 0.68 | 0.5 | 0.58 | 0.60 |
| Lipid | 0.60 \pm 0.10 | 1.25 \pm 0.06 | 0.77 \pm 0.15 | 0.00 | 0.03 | 0.20 | 0.00 | 0.10 |
| Ash | 0.21 \pm 0.01 | 0.52 \pm 0.08 | 9.33 \pm 0.58 | 0.30 | 0.45 | 0.30 | 0.27 | 0.30 |
| Alkaloid | 4.35 \pm 0.05 | 4.20 \pm 0.10 | ND | NA | NA | NA | NA | NA |
| Flavonoid | 0.95 \pm 0.05 | 1.13 \pm 0.15 | ND | NA | NA | NA | NA | NA |
| Tannin | 0.07 \pm 0.02 | 0.09 \pm 0.01 | 0.09 \pm 0.01 | NA | NA | NA | NA | NA |
| Phytate | 0.06 \pm 0.01 | 0.06 \pm 0.00 | 0.06 \pm 0.00 | NA | NA | NA | NA | NA |
| Country | Nigeria | Nigeria | Nigeria | Australia | Florida | Malaysia | China | FAO |

Discussion

Plant morphology is an important character used in plant classification. It is always used in combination with floral and anatomical features. In Cucurbitaceae, morphological characteristics have been employed in their classification (Hutchinson and Dalziel, 1954; Jeffrey, 1980, 2005; Okoli, 2013) while the application of anatomical features have been emphasized by Agbagwa and Ndukwu (2004), Ajuru and Okoli (2013), Ekeke *et al.* (2015), Agobua *et al.* (2015a). In this study, morphology and anatomy of fruit, flower stalks, tendril, leaf and stem anatomy were investigated. Also, the epidermal characteristics of the leaf, petals male and female flowers were studied.

Morphological attributes

The morphological attributes of the *B. hispida* studied conform to the existing information on the species. For instance, Alejandro (1998) reported average fruit size and weight of two accessions of *B. hispida* 55.5 cm x 24.7 cm and 11.7 kg for green winter melon and 48.9 cm x 22.1 cm and 6.1 kg for fuzzy white gourd. Also Sudhakar *et al.* (2008) recorded an average fruit weight of 3.06 kg to 13.67 kg and equatorial diameter of 52.99 cm to 80.11 cm in 34 accessions of *B. hispida* in India and in our study we recorded fruit size of 58.6 - 59.4 cm long, 14.0 - 15.5 cm wide and 8.5 - 9.0 kg (Table 3). These morphological characteristics are in line with the works of Jeffrey (1990), Alejandro (1998), Ahmed (2011) and suggest that this is a cultivar or an accession *B. hispida* (Alejandro, 1998; Sudhakar *et al.*, 2008).

B. hispida is a monoecious climber with yellow flowers, hairy stem, petiole, leaf, petals and young fruits. The male flowers are solitary or in a slender-pedunculate racemes, fairly coxillary with female flowers (Jeffery, 1980; Okoli, 1982; Ndukwu, 1988; Agobua *et al.*, 2015). However, majority of cucurbits such as *Telfiaria esculenta*, *Citrullus lanatus* Thunb and *Cucurbita pepo* L. are dioecious (Agbagwa and Ndukwu, 2004) while some other species are hermaphroditic (Hutchinson and Dalziel, 1954; Okoli, 1984; Ndukwu and Okoli, 1992). *B. hispida* exhibits similar characteristics with these species. This however supports the placement of these species in the Cucurbitaceae (Jeffrey, 1990).

Epidermal characteristics

The epidermal cells in the leaves and the petals are polygonal to irregular with anomocytic, tetracytic and anisocytic stomata. The anticlinal cell wall patterns are slightly straight, curved or undulating. Also, the leaf is amphistomatic. Okoli (1989), Ekeke *et al.* (2015), Agbagwa and Ndukwu (2004), Ajuru and Okoli (2013), Agobua *et al.* (2015a) opined the fact that most members of this family are amphistomatic with anomocytic, tetracytic and isotricytic stomata types. Also Adebooye *et al.* (2012) reported more stomata on the abaxial than the adaxial surface of *Trichosanthes cucumerina*. In the same way, the stomatal indices in this study were more on the abaxial surfaces of the leaf and petals. This finding supports the

existing data on the members of the family and thereby confirms the interspecific relationship between this species and other cucurbits.

Trichome types

Trichomes are of certain systematic significance and sometimes common types are even used for diagnostic purposes in association with other characters (Cutler 1984). The range of trichome variation in the Cucurbitaceae is enormous. The important aspects of variation of trichomes include the degree of modification of the adjacent epidermal cells and the scars left by lost trichomes, in addition to its distribution and anatomy. Generally, trichomes are categorised into glandular and the eglandular types. The significance of trichomes in relation to taxonomy has been shown in angiosperm families earlier by Gupta and Murty (1977) and Rao (1991).

Metcalf and Chalk (1950) had made a survey of various types of trichomes in different genera of the Cucurbitaceae. According to them glandular hairs with multiserial stalks is one of the major characteristics of this family, but simple unicellular or uniseriate types as well as wart-like or spiny trichomes are also present in the Cucurbitaceae. Both glandular and non-glandular trichomes were encountered during. Furthermore, other glandular trichomes identified among cucurbits include: club-shaped or globose with a multicellular head and a single stalk cell, club-shaped or elongate consisting of a multicellular head and two-celled stalk and multicellular clavate head with multicellular stalk. Also non-glandular trichomes such as uniseriate, 1-2 celled with a single basal cell, uniseriate and multicellular with a single basal cell, uniseriate and multicellular with biserial basal cells and uniseriate and multicellular with a multicelled base trichomes are characteristics of Cucurbitaceae. Both glandular and eglandular trichomes were present in *Benincasa hispida* and noted that this wide range of trichome characters observed can be used for diagnostic purposes (Mohammad and Fahad, 2011). This is also consistent with the different types of glandular and eglandular trichomes that have been studied and described in cucurbits (Okoli, 1989; Agbagwa and Ndukwu, 2001; Kolb and Muller, 2004).

Anatomy of stem, tendril, petiole and flower stalks

The arrangement of the vascular bundles and hollow piths in the stem, tendril, petiole and the male flower stalk have been reported (Metcalf and Chalk, 1950; Ekeke *et al.*, 2015) and the concentric nature of the vascular bundles (Ekeke *et al.*, 2017) have made similar observation in other members of cucurbits. The vascular bundles in cucurbits are bicollateral with varying sizes and mostly forming concentric rings in the stem, petiole and tendril (Metcalf and Chalk, 1950; Agbagwa and Ndukwu, 2004; Ekeke *et al.*, 2015; Agobua *et al.*, 2015a). Also, the tendril, female flower/fruit stalk and the stem showed remarkable layers of sclerenchymatous cells. The sclerenchymatous cells in the tendril, stem of *B. hispida* and male flower stalk are continuous while the ones in the female flower/fruit stalk are partly continuous or discontinuous. This is evident because they are used for anchoring the plant on other plants or to support the weight of the fruits (Ekeke *et al.*, 2015). This report supports previous works by Metcalf

and Chalk (1950) and Ekeke *et al.* (2015) and a strong phylogenetic bond between *B. hispida* and other members of Cucurbitaceae.

Chemical composition and uses

Regarding the chemical composition of *B. hispida*, Wills *et al.* (1984) noted that the pulp of *B. hispida* contains glucose and fructose and that their concentrations are 0.5 - 0.9% and 0.5 - 0.8 % respectively in matured fruits. Also, other authors from different parts of the world have analysed and reported the concentrations of moisture, carbohydrate, fibre, fat and ash in the pulp of mature fruit of *B. hispida* (Morton, 1971; FAO, 1972; Wills *et al.*, 1984; Mingyu *et al.*, 1995; Tee *et al.*, 1997) in Australia, Florida, Malaysia and China (Table 3). Our result on the concentrations of these nutrients in the pulp of the specimen collected from the University of Port Harcourt Biodiversity Centre is similar to the works of these authors. However we further reported the concentration in the skin and seeds. The seeds had the highest concentrations of crude protein, ash, carbohydrate, lipid and fibre. This however suggests that the combination of the skin, pulp and seeds will be for medicinal and other purposes.

The fruits of *Benincasa* have been reported to contain different amino acids and several antioxidant properties (Mingyu *et al.*, 1995; 2010; Yoshizumi *et al.*, 1998) and the oil for *B. hispida* seeds as the best cucurbit seed oil from the hot and humid tropics (Martin, 1984; Lee *et al.*, 2005; Sew *et al.*, 2010). The presence of high crude protein and lipid in the seeds compared to other parts of the fruit studied in this work upholds the works of these previous authors and could account for the health benefits, therapeutic uses, pharmacological, medicinal and food properties of *B. hispida* (Uchikoba *et al.*, 1998; Shih *et al.*, 2001; Lee *et al.*, 2005; Akinmoladun *et al.*, 2007; Roy *et al.*, 2008; Bhalodia *et al.*, 2009; Qadrie *et al.*, 2009; Monson, 2010; Girdhar *et al.*, 2010).

Conclusions

The morphological, anatomical and phytochemical characteristics of *B. hispida* from Nigerian are described for the first time. This species share the same ancestral characteristics in common with other members of Cucurbitaceae and supports the placement of this species in the family and the phytochemical constituents supports the medicinal, anti-compulsive effect, angiotensin-converting enzyme (ACE) inhibitor activity *in vitro*, Alzheimer disease treatment, anti-ulcer, anti-inflammatory, anti-obesity and anti-diarrheal agent of this species.

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