Comparative Chemotaxonomic Investigations on *Physalis* angulata Linn. and *Physalis micrantha* Linn. (Solanaceae)

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ABSTRACT— The present study is set to investigate the comparative micro- and macro-morphological, anatomical, cytological and phytochemical properties of Physalis angulata Linn. and Physalis micrantha Linn., members of the family Solanaceae predominantly found in the Niger Delta Tropics, Nigeria. They are used as vegetable and medicine. Their habits are annual herbaceous plants which attain up to 50cm or more in height. The leaves are simple, ovate and dentate, acuminate at apex, cuneate to rounded at base and petiolate measuring 6 ± 2.47 cm in length and $3 \pm$ 1.24cm in width for Physalis angulata Linn. while Physalis micrantha Linn. is 2 ± 1.64 cm in length and 2 ± 0.84 cm with alternate phyllotaxy. Their glabrous stems are angular with hollow and the inflorescence has stalked solitary axilliary flowers. The petals are pale yellowish and sepals greenish which enlarges into an encapsulated 5 lobed, prominently veined, membranous structure housing a many seeded berry fruit measuring 2.5 ± 1.41 cm long for Physalis angulata Linn. and 1 ± 0.74 cm long for Physalis micrantha Linn. The epidermal studies revealed anomocytic stomata whereas the trichomes are simple uniseriate forms and the flowers are axile in placentation borne at nodes. The anatomy of mid-ribs and petioles showed bicollateral vascular systems. There are 3 vascular traces and node is unilacunar in each species, their stems have 5 to 6 vascular bundles, their petioles are associated with 2 rib traces at primary growth phases. At secondary growth phases, their mid-ribs and petioles revealed vascular arcs and the stems have rings of open vascular systems. The cytological studies showed a diploid chromosome number of 2n = 24 and n =12 for the haploids. Alkaloids, saponins, tannins, phlobatannins, flavonoids, combined anthraquinones, free anthraquinones and cardiac glycosides are present in both species.

Keywords— Morphological, Anatomical, Cytological, Phytochemical.

1. INTRODUCTION

The family Solanaceae is composed of 95 genera (1) .It is widely distributed in temperate and tropical regions, but the centre of distribution is Central and South America. In West Africa however, there are 8 genera and 53 species of Solanaceae (2).. Physalis angulata Linn. and Physalis micrantha Linn. are annual sub-shrubs (3) and (4). According to (5) defined anomocytic (irregular-celled) as stoma surrounded by a limited number of cells that are indistinguishable in size, shape, or form from those of the remainder of epidermis; anisocytic (unequaled-celled) as stoma surrounded by three cells of which one is distinctly smaller than the other two; paracytic (parallel-celled) as stoma accompanied on either side by one or more subsidiary cells parallel to the long axis of the pore and guard cells while tetracytic as four subsidiary cells being present, two lateral and two terminal, and actinocytic as stoma surrounded by a circle of radially elongated subsidiary cells. Physalis species. have simple uniseriate trichomes (6). Trichomes are termed 'simple' when unbranched. Simple trichomes could be unicellular or multicellular (7). The type of hair can be of diagnostic value at species level, sometimes also at generic level, but rarely at family level (8). The word 'uniseriate' is really an anatomical term rather than morphological and does not describe the shape. 'Multiseriate' is not unique to trichomes, it could also mean multi layers as in epidermal and hypodermal axial parenchyma (7) and (1) stated that members of Solanaceae have unilacunar node. The primary vascular tissues of Solanaceae are bicollateral (7). Most members of Solanaceae are diploids for example the genus Solanum Linn. where 2n = 24 (9), (10) and (11) .Woody plants can accumulate in their cells a great variety of phytochemicals including alkaloids, flavonoids, tannins, saponins, cyanogenic glycosides, phenolic compounds, lignin and lignans (12). The medicinal value of these plants lies in bioactive phytochemical constituents that produce definite physiological actions on the human body (13) .Some of the most important bioactive phytochemical constituents are alkaloids, essential oils, flavonoids, tannins, terpenoid, saponins, phenolic compounds and many more (14) .These natural compounds formed the foundation of modern prescription drugs as we know to day (15). Saponins are used as blood cleanser (16) .The presence of tannins aid in wound healing (12) .Cardiac glycosides have been shown to aid in treatment of congestive heart failure and cardiac arrhythmia (17).

The relevance of the study is to enhance information on the existing literature and taxonomic characteristics of *Physalis* species, this is due to the fact that they are economic plants. Thus the objectives of the study is therefore aimed at considering: the comparative morphological, anatomical, cytological and phytochemical investigations of *Physalis* angulata Linn. and *Physalis micrantha* Linn., to be more precise, the chemotaxonomy.

2. MATERIALS AND METHODS

The materials used for this study were collected from the wild and raised from seeds from the fruit. A study of the macro-morphological features of the species were made using a 30cm ruler. The plants parts measured included: leaf length, leaf width, petiole length, stamen length, style length, fruit length and width and average plant height. The presence or absence of trichomes were observed painstakingly under a light microscope, and microphotographs were taken where relevant.

Floral biology: The opening and closing time of the flowers of the species were studied.

The arrangement pattern of the petals and sepals (that is the aestivation type) were observed and the insect pollinators noted.

2.1 Epidermal Studies

Fresh materials (leaves and stem epidermal peels) were collected for this study; the fresh leaves were peeled and bleached using sodium hypochlorite for about 2 minutes following the method of (18). The clear epidermal layers obtained were then washed in several changes of distilled water and stained with Alcian blue or safranin and temporarily mounted in aqueous glycerol solution(18). Photomicrographs were taken from good preparations. Stomatal studies (Stomatal indices) were done from the cleared leaves. The length and width of the guard cells were measured using a calibrated eye piece graticule following the method of (19). The stomata observed were viewed with the light microscope

and were calculated in unit area using the stomatal index [S.I.] formula as shown below: S.I. = $\frac{S}{E+S} \times \frac{100}{1}$ where S

and *E* mean numbers of stomata and epidermal cells within the particular area under investigation. The same formula was applicable for the calculation of trichome indices (T.I.), in this case, trichomes (T) were used instead of stomata: $T.I = T_{T} = 100$

$$\frac{T}{E+T} \times \frac{100}{1}$$

2.2 Comparative Anatomical Studies

Seeds of the plant materials were plated out in petri dishes containing wetted filter paper and the germination tests were calculated using similar formula as applied to stomatal indices but based on the percentage of the number that germinated divided by total number of seeds plated. Three days to two weeks after growth had occurred, stem and root systems studied were fixed, alongside with mature leaves, flowers, fruits and petioles harvested from mature plants, in FAA in the ratio of 1:1:18 of 40% formaldehyde, acetic acid and 70% alcohol for at least 48% hours following the method of (20) with some modifications. The leaves, petioles, fruits, flowers, stems and roots.

Free hand sectioning using a systematic arrangement of 5 razor blades, with 2 sets(nacet and tiger blades) crossed and a central vertical one(nacet) lying in between the 2 sets crossed. The blades were adjusted until the holes in them synchronized. The plant part to be sectioned was placed in the hole and using the first two fingers of the left hand to hold the vertical blade sets, while pressing down the 2 crossed sets with the first two fingers of the right hand to make a transverse section of about 20 to 25µm thick. The sections made were passed through alcohol solutions in the order: 30%, 50% 70%, 95% and absolute alcohol, allowing them for 5 minutes in each solution. The dehydrated materials were cleared of their natural wax by passing them through different proportions of alcohol and chloroform in the following ratios (3:1; 1:1; 1:3) v/v for 10 minutes in each, and as the chloroform gradually replaced the alcohol, the process was repeated from the pure chloroform and down the series again within same time interval. These were rehydrated in alcohol series starting with absolute then 95%, 70%, 50%, 30% and stained with 1% Alcian Blue for 2 minutes, washed off with water before counter-staining with 1% edsSafranin for 2 minutes. The stain was washed off and placed on clean glass slide with a drop of glycerol and a clean cover slip placed on it (21). The slides so prepared are as good as those of microtomy and are near permanent ones. These slides were viewed with the light microscope and microphotographs taken from good preparations after proper examination.

2.3 Cytological Study

Healthy root tips for mitotic study were obtained from seeds of *Physalis angulata* Linn.and *Physalis micrantha* Linn. grown in a petri dish containing 110mm Whatman filter paper wetted with water for a period of three days to one week. The early germinated roots were transferred to solution of 0.002M of 8- hydroxyquinoline for 3 hours specifically

to suspend the spindle fibres or to accumulate chromosomes at metaphase between 9 and 10 a.m. to be precise. The roots were treated with Carnoy's fluid (3:1 ethanol/acetic acid v/v) for 12 to 24 hours aimed at killing the cells. The roots were then preserved in 70% alcohol and kept in the refrigerator until when needed or used immediately by hydrolyzing in 9% HCl for 8 minutes and passing them through 70% ethanol for 10 minutes. Imm of the root tip studied was excised from the apex and squashed in a drop of FLP- orcein stain(2g of orcein dissolved in 100ml of a solution of equal parts of formic acid, lactic acid, propanoic acid and water) under a coverslip, flattened out and examined under a light microscope, following the method of (22) .Photomicrographs of the chromosomes were taken from good temporary slides, using a Sony digital camera (7.2 Mega pixels). For the meiotic chromosomes, inmature flower buds were used. These were treated with carnoy's fliud for 24 hours and preserved in 70% alcohol and kept in the refrigerator or used immediately as already described for mitotic chromosomes above following the method of (22) .

2.4 Phytochemical Studies (Qualitative analyses)

The leaves of each species studied was sun dried for 72 hours (3 days) and weighed. Fifty grams (50g) of the leaves were macerated in 96% ethanol using a pestle and a mortar. The extract was thereafter filtered and evaporated to dryness using a rotary evaporator set at 45° C to constant weight and later, an exhort extraction machine. Residue yields were noted and a portion was used for the phytochemical screening. Phytochemical screening for saponin, frothing tests, was done following the method described by (23) and (24)) as shown below: The ability of saponins to produce frothing in aqueous solution and to haemolyse red blood cells was used as screening test for these compounds. 0.5g of each plant extract was shaken with water in a test tube. Frothing which persisted on warming was taken as preliminary evidence for the presence of saponins. In order to remove 'false-positive' results, the blood haemolysis test was performed on those extracts that frothed in water. 0.5g of each extract was boiled briefly with 50ml phosphate buffer, pH 7.4, and then allowed to cool and filtered; 5ml of the filtrate was passed for 3 hours through an asbestos disc (1.5mm thick and about 7mm in diameter), which had been previously soaked with two or three drops of 1 percent cholesterol in ether and dried. After filtration the disc was washed with 0.5ml of distilled water, dried and boiled in 20ml of oxylol for 2 hours to decompose the complex formed between cholesterol and any saponins in the extract. The disc was then washed in ether, dried and placed on a 7 percent blood nutrient agar. Complete haemolysis of red blood cells around the disc after 6 hours was taken as further evidence of presence of saponins.

Test for alkaloids: 0.5g of each extract was stirred with 5ml of 1 percent aqueous hydrochloric acid on (25) and (26) .A confirmatory test designed to remove non-alkaloidal compounds capable of eliciting 'false-positive' results was carried out as follows with all extracts which gave preliminary positive tests for alkaloids. A modified form of the tinlayer chromatography (TLC) method as described by (27) was used. 1g of the extract was treated with 40 percent calcium hydroxide solution until the extract was distinctly alkaline to litmus paper, and then extracted twice with 10ml portions of chloroform. The extracts were combined and concentrated in vacuo to 5ml. The chloroform extract was then spotted on thin-layer plates. Four different solvent systems (of widely varying polarity) were used to develop each plant extract. The presence of alkaloids in the developed chromatograms was detected by spraying the chromatograms with freshly prepared Dragendorff's spray reagent. A positive reaction on the chromatograms (indicated by an orange or darker coloured spot against a pale yellow background) was confirmatory evidence that the plant extract contained an alkaloid.

Test for tannins: 5g of each portion of plant extract was stirred with 10ml of distilled water, filtered, and ferric chloride reagent added to the filtrate. A blue-black, green, or blue-green precipitate was taken as evidence for the presence of tannins (26).

Test for anthraquinones: Borntrager's test was used for the detection of anthraquinones. 5g of each plant extract was shaken with 10ml benzene, filtered and 5ml of 10 per cent ammonia solution added to the filtrate. The mixture was shaken and the presence of a pink, red, or violet colour in the ammonia (lower) phase indicated the presence of free hydroxyanthraquinones.

For combined anthraquinones, 5g of each plant extract was boiled with 10ml aqueous tetraoxosulphate vi acid and filtered while hot. The filtrate was shaken with 5ml of benzene, the benzene layer separated and half its own volume of 10 per cent ammonia solution added. A pink, red, or violet coloration in the ammonia phase (lower layer) indicated the presence of anthraquinone derivatives in the extract (26).

Test for phlobatannins: Deposition of a red precipitate when an aqueous extract of the plant part was boiled with 1 per cent aqueous hydrochloric acid was taken as evidence for the presence of phlobatannins (26).

Test for cardiac glycosides: Lieberman's test was used. 0.5g of the extract was dissolved in 2ml of acetic anhydride and cooled well in ice. Tetraoxosulphate vi acid was carefully added. A colour change from violet to blue to green indicated the presence of a steroidal nucleus (i.e. aglycone portion of the cardiac glycoside) (28).

3. **RESULTS**

3.1 MORPHOLOGICAL CHARACTERISTICS

The geographic location of the parent plants studied were 040521803211N and 0060551131011E at 14m altitude for Physalis angulata Linn. and 04°5217240¹¹N and 006°5511420¹¹E at 20m altitude for Physalis micrantha Linn.. The opening and closing times of the flowers was studied. It was revealed that the flowers commenced opening at 6:30 a.m. and opened completely at 8:30 a.m. while the closing time started at 5:00 p.m. and closed completely at 7:50 p.m. for Physalis angulata Linn. while Physalis micrantha Linn. flowers started opening at 6:15 a.m., opened completely at 8:20 a.m. and commenced closing at 5:00p.m. and closed completely at 7:30p.m. This feature is of taxonomic relevance as it aids supply information patterning the breeding status of the plants. The germination test conducted was 25% for Physalis angulata Linn. and 10% for Physalis micrantha Linn. The distributional pattern of the species has been recorded by Hutchinson and Dalziel (1958). Physalis angulata Linn. Plate 1 and Physalis micrantha Linn. Plate 2. Wildcape gooseberries as commonly known are annual herbaceous plants attaining up to heights of 50cm or more. The petiolate leaves are simple, ovate and dentate, acuminate at apex cuneate to rounded at base measuring 6 ± 2.47 cm in length and 3 ± 1.24 cm in width for *Physalis angulata* Linn. while *Physalis micrantha* Linn. is 2 ± 1.64 cm in length and 2± 0.84cm with alternate phyllotaxi, not as deeply dentated as the former Their glabrous stems are angular with hollow and the inflorescence have stalked solitary axilliary and pentamerous flowers. The petals are pale yellowish and sepals greenish which enlarge into an encapsulated 5 lobed, prominently veined, membranous structure housing many seeded berry fruit measuring 2.5 ± 1.41 cm long for *Physalis angulata* Linn. and 1 ± 0.74 cm long for *Physalis micrantha* Linn. and the stamens are 0.3cm long while for *P* angulata Linn the stamens are 0.5cm long. Fruit when unripe green and vellowish-green when ripe. The seeds are 0.2cm in diameter for P. angulata Linn. and 0.1cm in diameter for P. micrantha Linn. Aestivation type for the species studied is valvate. Insect pollinators are ants, spiders, house flies, bees and caterpillars. Pollinators started appearing at 7:00 a.m. and were not seen at 2:20 p.m., and sometimes resurfaced later in the day.

3.2 EPIDERMAL STUDIES

Physalis angulata Linn. foliar epidermal study revealed the presence of anomocytic stomata and uniseriate trichomes at both the adaxial and abaxial foliar surfaces(Plates 3 and 4). It is shown that the adaxial foliar layer has 20.00% stomatal index and 13.64% for the abaxial surface. Trichome index is also studied revealing 1.19% for the adaxial and 1.32% for the abaxial surfaces, stomatal characteristics showed that adaxial stomatal length as13.8 \pm 1.14µm with 8.23% coefficient of variation (C.V.) and width of 8.4 \pm 1.17µm with 13.97% C.V. and abaxial stomatal length as 10.3 \pm 2.91µm with 28.23% C.V. and width of 7.2 \pm 1.40µm with 19.42% C.V. while *Physalis micrantha* Linn. also revealed anomocytic stomata and the stomatal characteristics showed adaxial stomatal length of 11.2 \pm 1.40µm with 12.48% C.V. and width of 5.90 \pm 0.99µm with 16.85% C.V. and that of abaxial stomatal length of 8.30 \pm 1.16µm with 13.98% C.V. and width of 5.0 \pm 0.82µm with 16.34% C.V. respectively. The trichome types are simple uniseriate forms at both adaxial and abaxial surfaces.

See plates 5 and 6. Stem epidermal study of *Physalis angulata* Linn. showed presence of anomocytic stomata, and covering uniseriate trichomes and irregularly-shaped cells (Plate 7). *Physalis micrantha* Linn. stem epidermal study revealed presence of actinocytic stomata with regularly shaped cells and glandular trichomes(Plate 8).

3.3 Anatomical Investigation

Anatomy of *Physalis angulata* Linn. mid-rib shows uniseriate trichomes in epidermis made of a layer of cells. The collenchymatous cells occupy the region of the hypodermis. Parenchymatous cells occupy the ground meristem. The primary growth phase reveals 3 vascular traces having bicollateral arrangement with no rib bundle wings in both growth phases.(Plate 9). The mid-rib of Physalis micrantha Linn. is similar to that of Physalis angulata Linn. (plate 10). The petiole of Physalis angulata Linn. is made of a layer of cells in the epidermis, 2 to 4 layers of collenchyma in the hypodermis, the general cortex is predominated by parenchymatous cells. The primary growth phase reveals 3 vascular traces having bicollateral arrangement with 2 rib bundle wings (Plate 11). The petiole anatomy for Physalis micrantha Linn, is as described for *Physalis angulata* Linn. Plate 12. The internodal anatomy of *Physalis angulata* Linn, shows a circular structure with 5 to 6 vascular bundles having a central hollow in the region of the pith. The epidermis has a layer of sclerenchymatous cells. The hypodermis is made of about 1 to 2 layers of collenchymatous cells, and the general cortex comprises 2 to 3 layers of parenchyma of thin walls. The endodermis is made of a layer of barrel-shaped cells clearly-marked. The pericycle just below the endodermis is composed of 1 to 2 cell-layers. The pith region is made of large parenchymatous cells when present. The internodal anatomy is shown in Plate 13. The nodal pattern is unilacunar for both spcies (Plate 14 and 15). Physalis micrantha Linn. intermodal anatomy revealed some irregular out growth in the structure of the roughly shaped stem having same structural arrangement from the epidermis to the pith.as in the former except that there is no visible hollow in the region of the pith(Plate16). Root anatomy of both species have exarch xylary structure. The piliferous layer is single-cell thick. The vascular bundles are radially symmetrical. Centralized parenchymatous cells occupy the pith region of the root. (Plate 17 and 18). The ovary anatomy of both species revealed the placentation as axile type. Their ovaries are bilocular and 2-celled (Plates 19 and 20).

3.4 Cytological Investigation

Cytological Studies of *Physalisangulata* Linn. and *Physalis micrantha* Linn. showed the meoitic chromosome number as n=12 at diakinesis for the former and mitotic chromosomes at late prophase for the latter. Plates 21 and 22.

3.5 Phytochemical studies

Qualitative analysis carried out revealed the presence of the following phytochemical constituents: alkaloids, saponins, tannins, phlobatannins, flavonoids, combined anthraquinones, free anthraquinones and cardiac glycosides respectively in both species, table 1.

4. DISCUSSION

Observation on vegetative and floral features of *Physalis angulata* Linn. and *Physalis micrantha* Linn. revealed the habits of the species as annual herbs as also recorded by Hutchinson and Dalziel (1958). Physalis angulata Linn. and Physalis micrantha Linn. possess simple uniseriate trichomes and anomocytic stomata. Their stem epidermal studies revealed paracytic stomata for *Physalis angulata* Linn. and actinocytic stomata for *Physalis micrantha* Linn. The structure of the stamens and carpels, and mostly their pilose nature are of taxonomic relevance in delimitations at the generic and species level. The stem anatomical investigation revealed that *Physalis angulata* Linn. is circular with a central hollow while *Physalis micrantha* Linn. has out growths in the rough shaped structure. The nodal pattern is unilacunar for both species and the roots' vascular system revealed radial symmetry. The species investigated are bisexual, hypogynous and placentation are axile which is also in accordance to the observation of Hutchinson and Dalziel (1958). Anatomically, studies on the primary growth phase revealed the mid-ribs and petioles of the species are observed with 3 vascular traces and also have bicollateral vascular system. It was observed that the departure of the rib-bundle wings are towards the position of the open vascular system. The secondary growth phase revealed vascular arc structure in the mid-ribs and petioles, while the stems and roots showed a complete ring structure of an open vascular system in all the species investigation . Cytologically, the basic chromosome number for members of Solanaceae is x = 12 Omidiji (1985), Okoli and Osuji (2008) also supported the chromosome basic number as x = 12, and diploids of 2n = 24.

The medicinal value of these plants is due to the presence of bioactive phytochemical constituents that produce definite physiological actions on the human eaters as also recorded by Akinmoladun *et al.* (2007). Some of the most important bioactive phytochemical constituents are alkaloids, essential oils, flavonoids, tannins, terpenoid, saponins, phenolic compounds and many more as also related by Edeoga *et al.* (2005). These natural compounds formed the foundation of modern prescription drugs as we know to day as observed by Goh *et al* (1995).

5. CONCLUSION

Physalis species are used as vegetables and in most African dishes and in tradomedicine. Having worked extensively on their morphological, anatomical, cytological, and phytochemical properties, other areas of interest need are DNA barcoding, Palynology, proximate analysis and quantitative aspect of phytochemistry. Interested researchers could carry on work in these areas.

Solanaceae	Alkaloi	Sapon	Tan	Phloba	Flavono	Combined	Free	Cardiac
	ds	ins	nins	tannins	ids	anthraquinon	anthraquino	glycosides
						es	nes	(Lieberman test)
Physalis	+	+	+	+	+	+	+	+
angulata Linn.I								
Physalis	+	+	+	+	+	+	+	+
micrantha Linn.								

Table 1: Phytochemical Properties of Physalis angulataa Linn. and Physalis Micrantha Linn.

+ represents presence of phytochemicals

7. ACKNOWLEDGMENT

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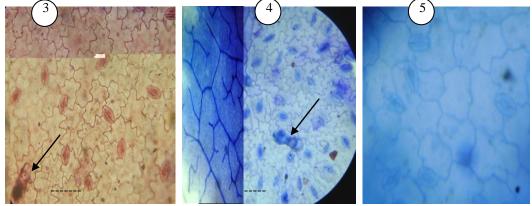
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Plate1: Physalis angulata Linn.



Plate 2 : Physali micrantha Linn.



S:15µm

T:25µm

Plate3:*P.angulata* Adaxial Foliar Surface Plate 4: *P.angulata* Abaxial Epidermis Plate5: *P.micrantha* Adaxial Foliar Surface. Plate6: *P.micrantha* Abaxial Foliar Surface. Anomocytic Stomata(S) Reveaved in 3, 4, 5, 6 and 7. Arrow Sshows Glandular Hair(T).

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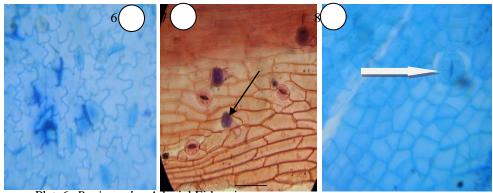
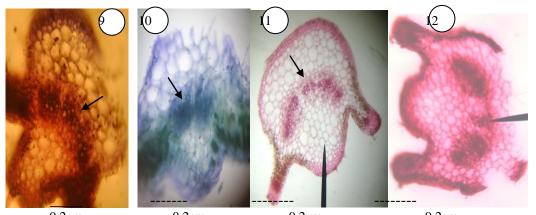


Plate6: *P.micrantha* Adaxial Eidermis. ------Plate7: *P.angulata* Stem Epidermis CT:20µm Plate 8: *P.micrantha* Stem Epidermis. Black Arrow Reveals Covering Biseriate Trichome(CT). White Arrow Shows Actinocytic Stoma in 8.



0.2cm0.2cm0.2cmPlate 9: P .angulataMid-rib Anatomy.Plate 10: P micranthaMid-rib AnatomyPlate 12 P. micranthaPpetiole AnatomyPlate 12 P. micranthaPpetiole AnatomyPlate 11: P. Physalis angulataArrow reveals vascular arc in 9 and 10 while in 11 and 12 Arrow shows 3 vascular tracesPlate 12 P. micranthaPpetiole

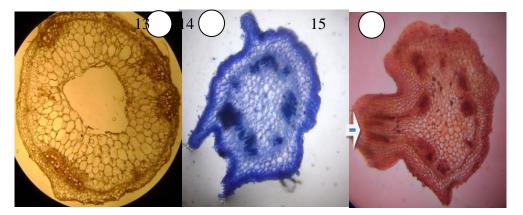


Plate13: *P.angulata* Linn Stem Anatomy. Plate 14: *P.micrantha* Linn.internodal Anatomy. Plate 15:*P. angulata* Nodal Pattern. Plate 16: *P.micrantha* Nodal Pattern with 5 to 6 Vascular Bundles and having 3 VascularTtraces from One Gap in 15 and 16. A Central Hollow in 13 and Epidermis with Protruions Prominent.

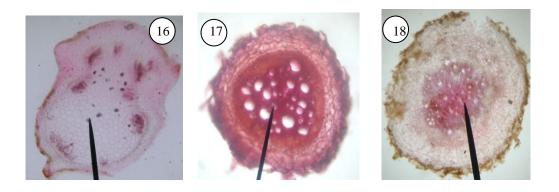
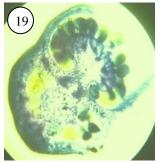


Plate17: *P. angulata* Root Anatomy. Plate 18:*P. micrantha* Root Anatomy. Exarch Xylary Rays in 17 and 18.



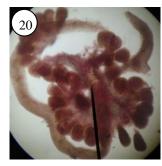
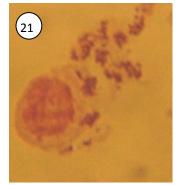


Plate 19:*P.angulata* Ovary Anatomy Plate 20: *P. micrantha* Ovary Anatomy Bilacular Ovary with Axile Placentation. Presnet in 19 and 20.



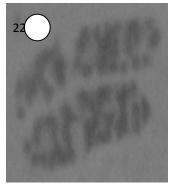


Plate 21: *P.angulata* Meiotic Chromosomes at Diakinesis. Plate 22: *P. micrantha* mitotic chromosomes at late prophase.