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Pharmacognostical evaluation on the Medicinal & Nutritive fruits of Raamaphal – *Annona reticulata* L.

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Abstract
Annona reticulata L. is one of the useful medicinal plant for various diseases, especially known for its nutritive and medicinal properties. In the present communication powder microscopy of the seed and microscopical studies on the fruit rind along with preliminary phytochemical studies have been carried out. Study revealed the presence of abundant oilgloubles, groups of stone cells, oil cavity with oil globules, epidermal cells filled with reddish tannin contents, simple starch grains in groups to mention a few characters. Fruits of *A. reticulata* have many medicinal properties like Anti-cancerous, antidysenteric, antihelmintic, and astringent, sweet, useful in blood complaints. Preliminary phytochemical studies revealed the presence of Alkaloides, proteins, Carbohydrates, terpenoides, phenols etc.

Keywords: Raamaphala, Bullock's Heart, microscopy, Annona, antidysentric.

1. Introduction

1.1 Habitat: *Annona reticulata* L. is commonly known as ***Ramaphal,*** is a low, erect tree, with a rounded or spreading crown and trunk 25-35 cm thick. Height ranges from 5-10 m. The ill-smelling leaves are deciduous, alternate, oblong or narrow lanceolate, 10-20 cm long, 2-5 cm wide, with conspicuous veins. Flowers, in drooping clusters, are fragrant, slender, with 3 outer fleshy, narrow petals 2-3 cm long; light-green externally and pale-yellow with a dark-red or purple spot on the inside at the base. The flowers never fully open. The compound fruit, 8-16 cm in diameter, may be symmetrically heart shaped, lopsided, irregular, or nearly round, or oblate, with a depression at the base. The skin, thin but tough, may be yellow or brownish when ripe, with a pink, reddish or brownish-red blush, and faintly, moderately, or distinctly reticulated. There is a thick, cream-white layer of custard-like, somewhat granular, flesh beneath the skin surrounding the concolorous moderately juicy segments, in many of which there is a single, hard, dark brown or black, glossy seed, oblong, smooth, less than 1.25 cm long. Actual seed counts have been 55, 60 and 76. A pointed, fibrous, central core, attached to the thick stem, extends more than halfway through the fruit [1].

1.2 Uses: The tree is introduced in India and is distributed all over the country up to an altitude of 1200m, and is also found in Nepal & Srilanka. The tree starts fruiting during 4-7 years, flowers open during August-December, ripened in 8 months and the annual yield of fruits is up to 70. The fruit consists of 72% of edible portion. The fruit is fair quality, but inferior to that of *A. squamosa*, *A. cherimola*. The pulp has insecticidal properties and is used to kill lice. The unripe and dry fruits yields black die. The unripe fruit is considered astringent, anthelmintic, antidysenteric and antidiarrhoeic and used in fever and enlarged spleen. The ripe fruits are alleviate biliousness, thirst and stop vomiting [2]. The flesh may be scooped from the skin and eaten as is or served with light cream and a sprinkling of sugar. Often it is pressed through a sieve and added to milk shakes, custards or ice cream. A suace can made by blending the seeded flesh with mashed banana and cream [1]. The raw fruit peel possesses antioxidant and antimicrobial properties [3]. The unripe fruit is rich in tannins, it is dried, pulverized and employed against diarrhoea and dysentery. Seeds are astringent, vermifugal and useful in diarrhea and dysentery, these are irritant to conjunctiva and abortifacient [4]. The seeds are reported to possess astringent and vermifugal properties and used in diarrhea and dysentery. The seed-meal is rich in nitrogen and oil is toxic, can be used as a contact-poison [2]. The seeds are so hard that they may be swallowed whole with no ill effects, but the kernels are very

toxic [1]. The seed oil showed anti-inflammatory activity [5]. The leaf is useful as antiphlogistic in tumours and as anthelmintic [2]. The leaves have insecticidal properties, used to kill lice. A decoction of the leaf is given as a vermifuge [1]. The root is a drastic puragative. The bark has been reported as powerful astringent and is given as tonic, bark is also yields good quality fibre used for cordage [2]. The bark is very astringent and the decoction is taken as a tonic and also as a remedy for diarrhea and dysentery Fragments of the root bark are put around the gums to relieve toothache, the root decoction is taken as a febrifuge. All parts of the plant possesses insecticidal properties [1].

1.3 The Nutritive values: The fruit is rich in nutrients, 100 g of the edible portion the fruit contains the following nutrients: Proteins-1.5 g, Fat-0.2 g, Crude fibre-2.1 g, Carbohydrates-15.8 g, Energy-71Kcal, Calcium-40 mg, Phosphorus-30 mg, Iron-0.5 g, Moisture-76.8 g, Minerals-07. g, Riboflavin-0.07 mg, Vitamin C-5.0 g, Niacin-0.6 mg, Carotene-67 µg, and Iodine-0.64 ppm, Flourine-5.6 ppm and Ascorbic acid etc. [2, 6].

1.4 Chemical constituents: From the seeds number of chemical components were isolated and identified, in which some are showed promising cytotoxic activity. A series of N-fatty acyl tryptamines [7]; γ-lactone acetogenin, *cis-/trans*-isomurisolenin, annoreticuin, annoreticuin-9-one, bullatacin, squamocin, *cis-/trans*-bullatacinone and *cis-/trans*-murisolinone [8]; reticulatain-1 and reticulatain-2, reticulatacin and uvariamicin III [9]; **()**; dieporeticanin-1 and dieporeticanin -2, dieporeticenin, trieporeticanin [10]; One hundred and eighty compounds were identified in the aroma extracts, of which alpha-pinene, beta-pinene, myrcene, limonene, terpinen-4-ol, and germacrene D were found to be the major constituents [11]. From the bark, reticulacinone, rolliniastatin-2, molvizarin [12]; reticulatacin, bullatacin, (−)-kau-16-en-19-oic acid and methyl 16β, 17-dihydro-(−)-kauran-19-oate and, liriodenine [13]; the root bark contains liriodenine [14]; anonaine, norushinsunine, retuculine [2]. Leaves contains Annonaretin A, kaurenoic acid, taraxerol, β-sitosterol, 16α-hydro-19-al-*ent*-kauran-17-oic acid, 6β- hydroxystigmast -4-en-3-one, and 17-acetoxy-16β-*ent*-kauran-19-oic acid, 16α-hydro-*ent*- kauran-17,19-dioic acid, and (2*S*)-di-*O*-methylquiritigenin[15].

1.5 Regional & other names in India: English: Bullock's Heart, corazon, West Indian custard-apple; **Indonesian:** Buah nona; **Hindi:** Anta, Louna, Ramphal, Luvuni; **Kannada**: Ramaphala; **Sanskrit:** Krishna beejam, Lavati, Lavali, Raamaaphalam, Ramphala; **Tamil:** Aninuna, Manilvatta, Raamsita; **Telugu:** Raama phalamu, Raamaseethaphalamu, Ramachita; **Malayam:** Parankichchkka, Vlathi, Manilanilam; **Marathi:** Raamaaphal; **Orissa:** Barhial, Nena; **Gujarathi:** raamaaphal; **Bengali:** Luvuni, Nona [5].

2. Materials and Methods: The ripened fruits were collected from Cheruthuruthy, Kerala, India and identified through local floras as *Annona reticulata* L. belonging to the family *Annonaceae*. Fruits were washed thoroughly, outer portion along with the pulp portions were removed, shade dried, powdered, seeds were also removed and powdered separately. Powdered material was used to carry out microscopical and chemical analysis for different parameters. Dried pulp and seeds powder used to microscopial studies and recorded in Table-3 [16, 17]. Powdered fruit rind along with pulp and seeds were extracted with different solvents with the help of soxhlet extraction apparatus, dried the extracts under reduced pressure and used for preliminary phytochemical analysis. Physico chemical and preliminary phytochemical screening of the pulp and seeds were carried out to the coarse powder according to the standard methods and recorded in Table 1 and 2 [18].

3. Results

3.1 Macroscopical characters: The fruit is variable in shape: heart-shaped or spherical. The size ranges from 7 centimetres (2.8 in) to 1 centimetre (0.39 in), depending on the cultivar. When ripe, the fruit is brown or yellowish, with red highlights and a varying degree of reticulation, depending again on the variety. The flesh varies from juicy and very aroma, compound fruit, 8-16 cm in diameter, may be symmetrically heart-shaped, lopsided, irregular, or nearly round, or oblate, with a depression at the base. The skin, thin but tough, may be yellow or brownish when ripe, with a pink, reddish or brownish-red blush, and faintly, moderately, or distinctly reticulated. There is a thick, cream-white layer of custard-like, somewhat granular, flesh beneath the skin surrounding the concolorous moderately juicy segments, in many of which there is a single, hard, dark-brown or black, glossy seed, oblong, smooth, less than 1.25 cm long. Actual seed counts have been 55, 60 and 76. A pointed, fibrous, central core, attached to the thick stem, extends more than half way through the fruit.

3.2 Microscopic characters: T.S of the fruit shows outer exocarp made up of single layered epidermis made up of rectangular cells covered with cuticle, epidermal cells are filled with abundant reddish tannin content, followed by this is many layered, compactly arranged, thin walled parenchymatous cells filled with oil globules, tannin content and sugar content cells. Beneath this is endocarp region made up of groups of stone cells with narrow lumen and highly lignified wall.

Powder microscopy of fruit rind and pulp shows different fragments of tissues like epidermal cells with abundant reddish tannin content in surface view, stone cells in groups and in single with a small lumen, abundant oil globules and elongated fiber which are highly lignified.

Powder microscopy of seed shows different fragments of tissues like Elongated fibers and groups of oil gloubles, helical to spiral xylem vessel, Groups of stone cells, Single stone cell and oilglouble, Groups of simple starch grains, oil globules and starch grains.

3.3 Diagnostic characteristics

1. Presence of abundant simple starch grains and oil globules in mesocarp region of the fruit.
2. Presence of sugar content cells in mesocarp region.
3. Presence of groups of abundant stone cells with a small lumen in mesocarp region
4. Presence of rough surface of the outer portion of the fruit.
5. Presence of elongated fibers which are highly lignified.
6. Presence of reddish tannin content in the epidermal cells of epidermis of the fruit.
7. Presence of granulated/crystalloid fruit pulp, which is sweet in taste.

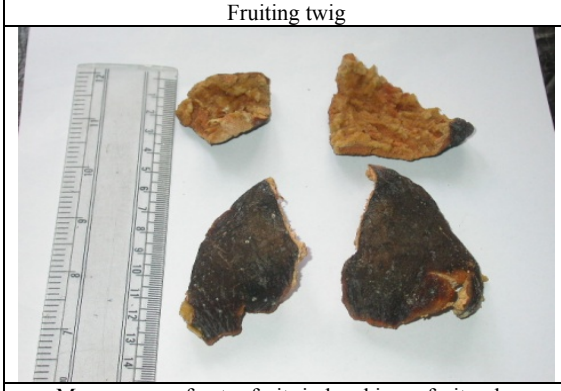
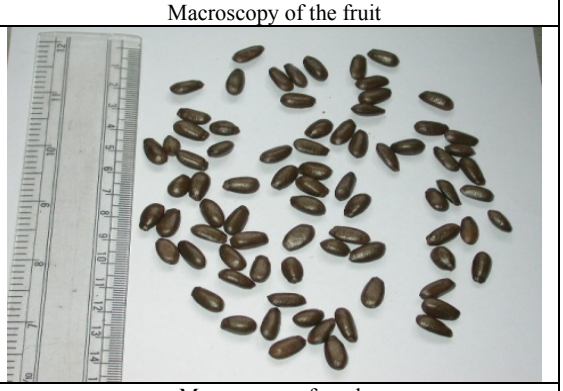
Table 1: Preliminary Phytochemical tests of *Annona reticulata* Linn. Fruit rind and Seeds.

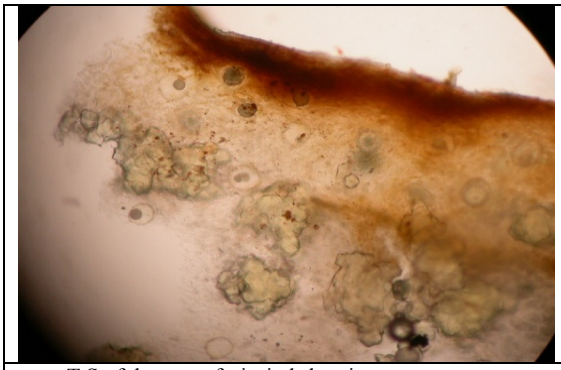
S. No.	Natural product test	Fruit rind/pericarp region	Seeds
1	Alkaloids	-ve	+ve
2	Carbohydrates	+ve	+ve
3	Flavonoids	-ve	-ve
4	Terpenoids	+ve	+ve
5	Proteins	+ve	+ve
6	Saponins	-ve	-ve
7	Steroids	+ve	+ve
8	Tannins	+ve	-ve
9	Starch	-vess	-ve
10	Phenols	+ve	+ve

Table 2: Physicochemical parameters of *Annona reticulata* Linn. Fruit rind and seeds.

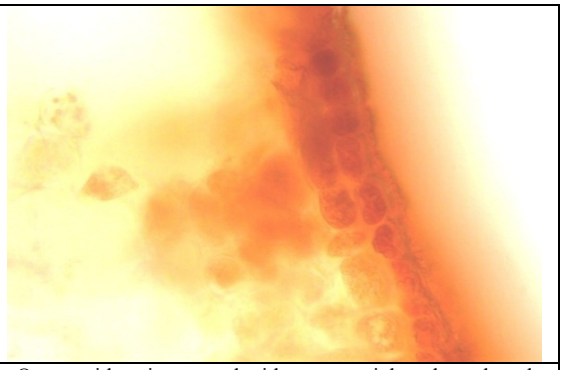
S. No.	Name of the parameter	Fruit rind/pericarp region	Seeds
1	Description	Light brown powder	White/cream clour oily powder with black particals.
2	Foreign matter	-Nil-	-Nil-
3	pH (5% w/v aq. solution)	5.90	4.55
4	Loss on drying at 105 ^{0}C	11.82%	7.36%
5	Total ash	4.05%	1.96%
6	Acid-insoluble ash	0.02%	0.05%
7	Water-soluble extractive	34.64%	6.61%
8	Alcohol-soluble extractive	17.97%	13.76%

Table 3: Macro, microscopy & powder studies of Fruit rind with pulp & seeds.

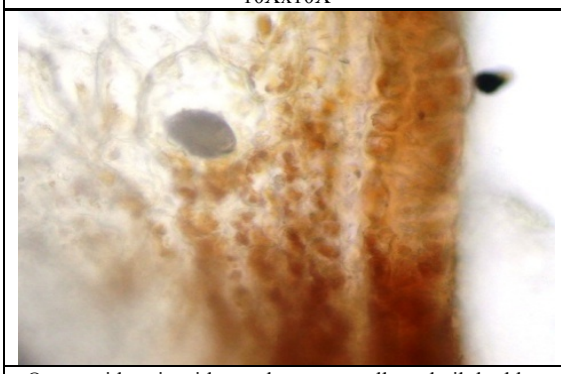
	
Fruiting twig	Macroscopy of the fruit
	
Macroscopy of outer fruit rind and inner fruit pulp.	Macroscopy of seeds



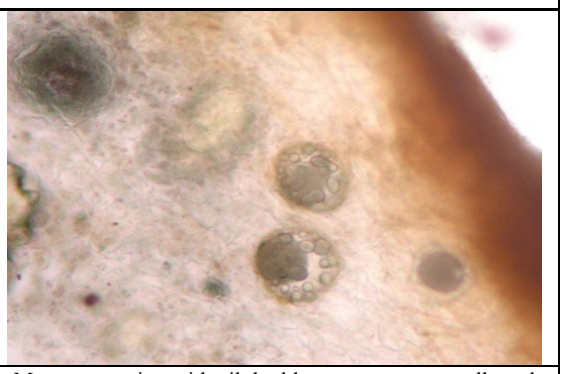
T.S of the outer fruit rind showing outer exocarp (Epidermis), mesocarp and stone cell layer in groups. 10Xx10X



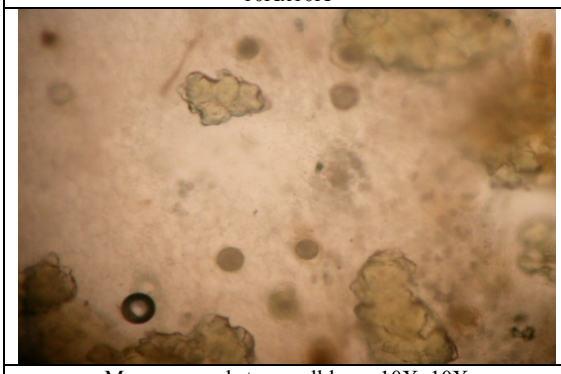
Outer epidermis covered with wavy cuticle enlarged, and with reddish tannin content. 10Xx40X



Outer epidermis with starch content cells and oilgloubles. 10Xx10X

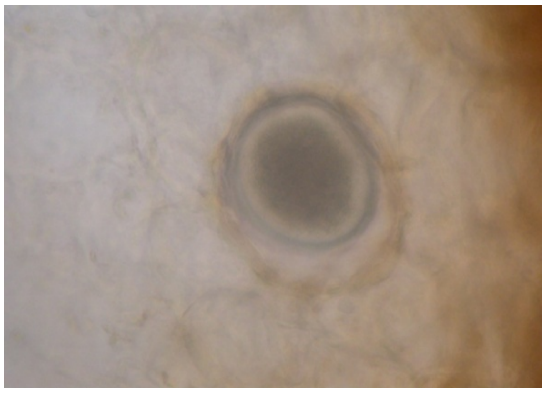


Mesocarp region with oilgloubles, sugar content cells and starch grains enlarged. 10Xx10X



Mesocarp and stone cell layer 10Xx10X

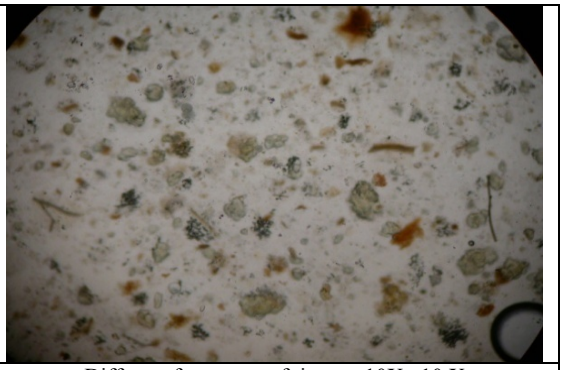
Mesocarp and stone cell layer with abundant starch grains enlarged. 10Xx40X



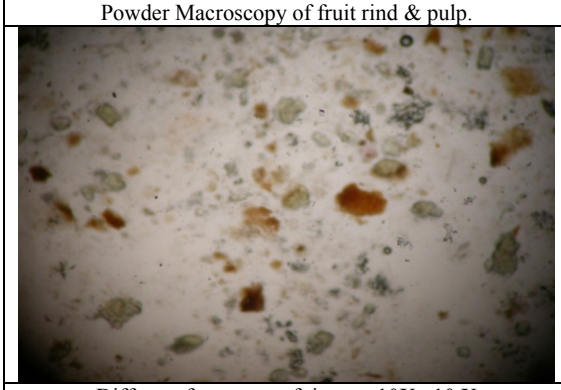
Sugar content cells enlarged. 10Xx40X

Powder Microscopy of fruit rind and pulp

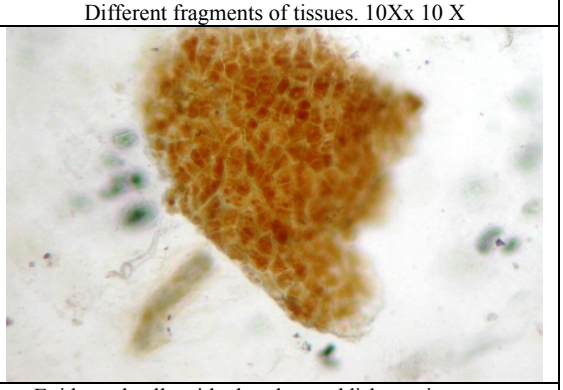
Powder Macroscopy of fruit rind & pulp.



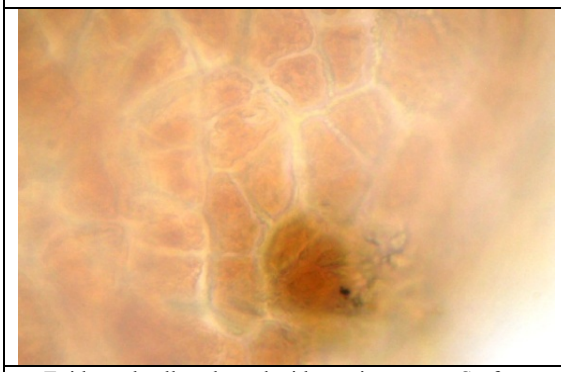
Different fragments of tissues. 10Xx 10 X



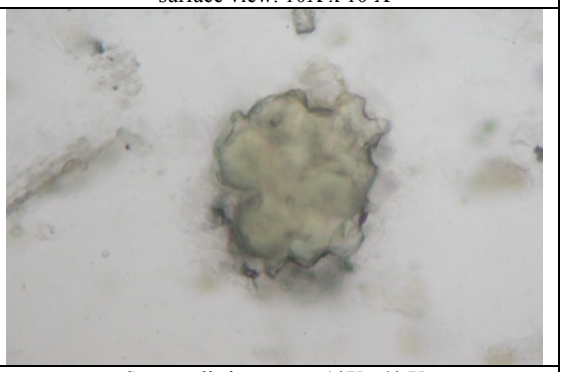
Different fragments of tissues. 10Xx 10 X



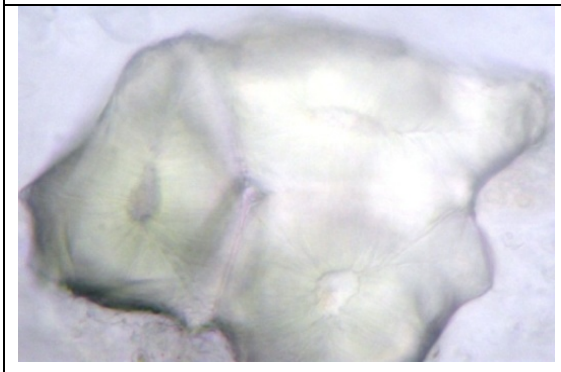
Epidermal cells with abundant reddish tannin content surface view. 10X x 10 X



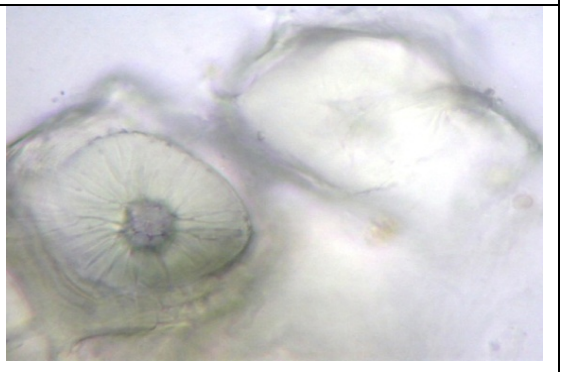
Epidermal cells enlarged with tannin content. Surface view. 10Xx 40 X



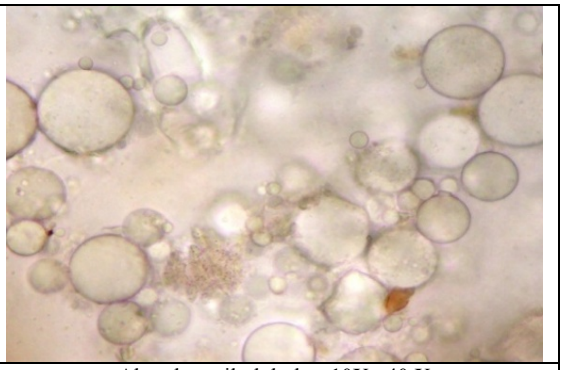
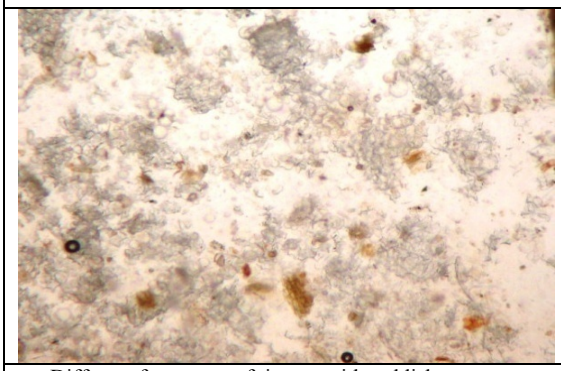
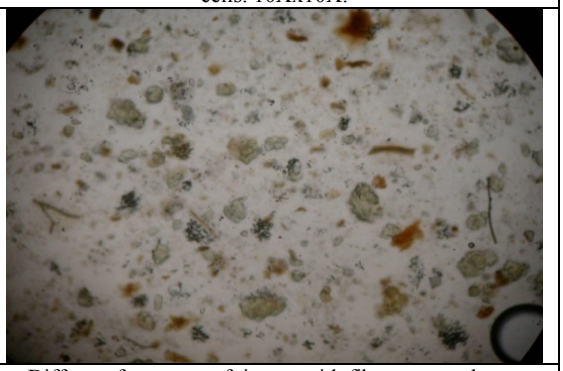
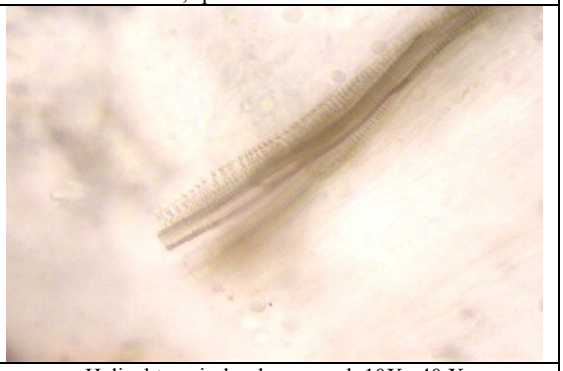
Stone cells in groups. 10Xx 40 X



Group of Stone cells enlarged.10Xx 40 X

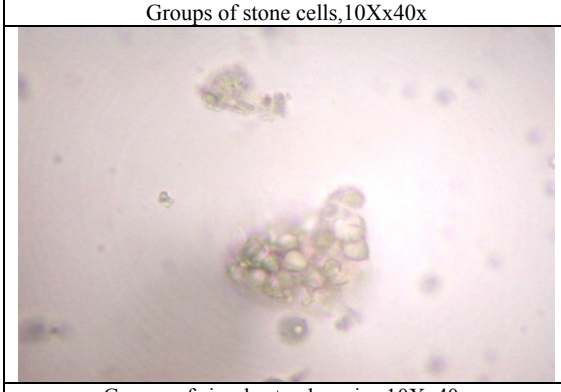


Single stone cell. 10X x 40X

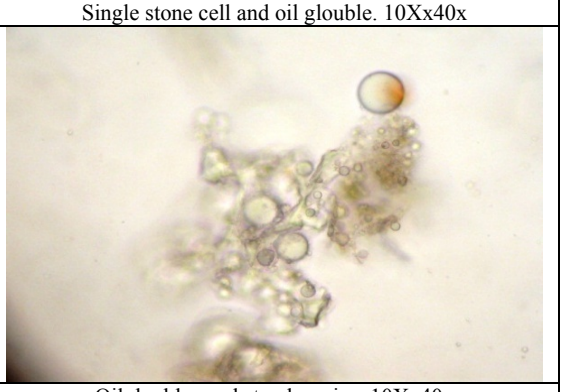
	
Elongated fibre and oil globules. 10X x 40X	Abundant oil globules. 10Xx 40 X
Powder microscopy of Seed	
	
Seed powder: Macroscopy	Different fragments of tissues With fibers, groups of stone cells. 10Xx10X.
	
Different fragments of tissues with reddish contents, parenchyma cells. 10Xx10X	Different fragments of tissues with fibers, parenchyma cells, spiral vessels.10Xx10X
	
Elongated fibers and groups of oilgloubles. 10Xx 40 X	Helical to spiral xylem vessel. 10Xx 40 X

Groups of stone cells,10Xx40x

Single stone cell and oil glouble. 10Xx40x



Groups of simple starch grains 10Xx40x



Oilgloubles and starch grains, 10Xx40x

4. Discussion/Conclusion

In the present scenario herbal/natural medicine/foods are playing a vital role in human health care system. The anti-cancer properties of custard apple appear to be mainly due to a class of compounds called acetogenins which are specific to Annonaceaous species. The different acetogenins present in *A. reticulata* possess more potent antitumor activity and cardio protective activity [19].

Pharmacognostical evaluation on the fruits of Ramaphal revealed that, the identification characteristics in fresh and in powder form. Fruits are highly nutritive, seeds possess anti cancerous and other properties. Microscopically fruit shows abundant stone cells, simple starch grains, iol globules, sugar content, granular/crystalloid pulpy region. Chemical constituents like terpenoides proteins, phenols, carbohydrates, tannins etc. Further the custard apple fruit appear to be have excellent health and medicinal benefits which deserve to be further explored for the beneficial of human beings. This study will help for the identification of Raamaphal in fresh and in powder form, it can also be useful for standardization purpose.

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