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Identification & Extraction of Fresh Water Snail (*Filopaludina bengalensis*) Polysaccharide

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Abstract:- Northeastern India's great biological diversity has earned it a reputation. For this study, *Filopaludina bengalensis* were selected. The aim of the present study was to investigate the presence of phytochemicals of the selected Mollusca. Morphoanatomical study on *Filopaludina bengalensis* was carried & different images was formed. Mollusca shells were digested with HCl. Centrifugation was done to take out the insoluble matter. Along with that snail enzyme was collected by starving it for days. Isolation of N-Acetyl D-glucosamine was done with extracted chitin and snail enzyme. FTIR analysis of Chitin & N-Acetyl D glucosamine was determined. With the help of graph peak was studied, N-acetyl D-glucosamine & chitin was compared with standard component. Our findings provided evidence that crude aqueous and organic solvent extracts of these tested snail contain medicinally important bioactive compounds and it justifies their use in the traditional medicines for the treatment of different diseases.

Keywords:- *Filopaludina bengalensis, Chitin, Mollusca, Marine Organism, D-glucosamine*

I. INTRODUCTION

Molluscs are the second most abundant group of invertebrates after insects. The number of valid recent species is currently estimated around 50,000 to 55,000 marine, 25,000 to 30,000 terrestrial and 6,000 to 7,000 freshwater (Mollusca Base eds. 2023).[7] Freshwater Mollusca is found in all aquatic habitats such as rivers, lakes, streams, swamps, springs, temporary ponds, drainage ditches and other water bodies.[6] Groombridge (1992) non-marine molluscs have experienced the highest number of recorded extinctions, with a total of 284 species becoming extinct within the past 300 years.[16]

There are about 80 species of freshwater molluscs including 30 species of bivalves and 50 species of gastropods are known from Nepal (Budha 2010). Freshwater gastropods are widely distributed except Antarctica.[10] The Vivipary snails are found in various regions around the world including North and South America, Europe, Asia, and Africa. Some species have very limited distributions, while others are widespread. The family Viviparidae has a temperate to tropical global distribution but is not native to Antarctica or South America. [12]

Vivipary snails or individuals of the family Viviparid are freshwater snails that display the regenerative procedure of vivipary.[13] Vivipary snails are a intriguing bunch of freshwater snails that have advanced a interesting regenerative technique to increment the survival of their sibling. This regenerative procedure in which embryos create interior the parent's body and are born as live descendant is uncommon among spineless creatures but has advanced in a few species of snails as a implies of expanding the survival of descendant (Lydeard et al. 2004).[20] Viviparidae is right now separated into three subfamilies (Bouchet&Rocroi 2005): Viviparinae (Gray 1847), Bellamyinae (Rohrbach 1837) and Lioplacinae (Gill 1863). The North American genera have agents of all three subfamilies, whereas the African, Asian, and Australian genera are all individuals of the Bellamyinae subfamily and all European genera are individuals of the Viviparinae subfamily.18 As of now, there are around 31 genera and 150 species of Viviparidae known. (Franke et al. 2007).[22]

Inside the subfamily Bellamyniae, the shell of Bellamya is exceptionally variable and due to covering shell characters the species are troublesome to separate (Mandahl-Barth 1954)[4] There are hundreds of species of gastropod molluscs which display polymorphism for shell colour and design, one of them is Filopaludina bengalensis. Shell bearing species of molluscs is the exceedingly different and broadly dispersed freshwater gastropod from family Viviparidae (Sengupta et al. 2009; Hirano et al. 2015; Hirano et al. 2019).[9] Viviparid snails are eminent for the parallel advancement of phenotypes, such as shell morphology and regenerative methodology which may be the result of rehashed nearby adjustment to particular environments (Hirano et al. 2015 and Hirano et al. 2019).[30] Viviparidae has a wealthy fossil record that dates back to the Center Jurassic which has been utilized by a few past thinks about to calibrate broadening ages of viviparid snails based on fossil records (Stelbrink et al. 2020).[15]

- *Taxonomy and Distribution of Filopaludina bengalensis*

Filopaludina bengalensis (Lamarck, 1882) was originally described as Paludina bengalensis by Lamarck in 1822. This species has been used as Bellamya bengalensis

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for several years. It is treated as Filopaludina (Filopaludina) bengalensis by Brandt (1974). Nesemann (2009) treated this species as Bellamya (Filopaludina) bengalensis (Lamarck, 1822). Currently accepted name of the Bellamya bengalensis is *Filopaludina bengalensis* (Lamarck, 1822) (MolluscaBase eds. 2023).[28] There exist multiple Filopaludina species that exhibit distinguishing characteristics such as variations in shell structures, apex shape, and color patterns whereas identification can prove challenging because of ambiguous and inconsistent depictions in the literature. They are distributed in India, Bangladesh, Myanmar, and Sri Lanka.[24]

➢ *Food value of F. bengalensis*

Filopaludina bengalensis is edible freshwater snails and has been consumed by humans in Asia and Africa. The flesh of snails is commonly utilized in both traditional medicine and cuisine within various ethnic communities.[17] Baby et al. (2010) reported that these organisms are the main source of food for 80.81% of households from more than 30 cases of general schedule and tribal peoples. F. bengalensis contains saturated fatty acids (48–60%), monogenic (18–30%), and polyunsaturated fatty acids (21-33%) in its flesh, which has low cholesterol (Misra et al. 2002).[26]

Shell characteristics including shape, size, and texture can also vary within F. bengalensis populations. Some individuals may have shells that are more elongated, while others may have rounder or flatter shells. Shell polymorphism can be influenced by genetic factors and environmental conditions, such as resource availability and predation pressure.[21] Size polymorphism could occur in F. bengalensis, with some individuals growing larger or smaller than the average size. Genetic factors, environmental conditions, and individual growth rates can contribute to the observed size variations.[27] It is important to note that detailed research specific to F. bengalensis may be necessary to provide a more comprehensive understanding of the extent and mechanisms of polymorphism in this particular snail species.[16]

There are about 22 forms of this species recognized based on the shell morphology and variations including B. bengalensis *F. balteata*, B. *bengalensis* F. *typica*, B. bengalensis F. *annandalei*, B. bengalensis F. *mandiensis*, B. *bengalensis* f. *colairensis*, B. bengalensis f. doliaris, B. bengalensis f. *nepalensis* and B. *bengalensisfeburnea* (Annandale 1921). The goal of the current study is to present morphological shell variations on the edible freshwater viviparous snail species F. *benglanensis* from various habitats and geographic locations of Nepal including variation in operculum and anatomical parts of this species.[22]

➢ *Habitat and Ecological Importance:*

F. bengalensisis a snail that lives in freshwater which can be found in practically any sort of lowland water body, mostly stagnant water, and low-saline water resources, including rivers, streams, lakes, ponds, wetlands, contaminated roadside marshes and ditches, and paddy fields (Ramakrishna &Dey 2007).[25] F. bengalensis have great significance role in aquatic ecology because they form the food of fishes and their productivity play an important link in the food chain. F. bengalensis are mainly filter feeder and detritivore that is why they are able to form an important link in the food chain. Because, as the significance of detritus in the food chain becomes clear, they can transform low-quality, low-energy waste into higher-quality food for higher creatures in the food web. These snail populations in particular are excellent markers of local conditions that indicate the quality of the water.(Roy et al, 2022)[28]

➢ *Distribution of F. bengalensis in India*

Freshwater snails are found in various regions across India, particularly in freshwater bodies such as rivers, lakes, ponds, streams, and reservoirs. They play important roles in freshwater ecosystems and are often indicators of water quality. Here are some key regions where freshwater snails are distributed in India:

- Ganges River Basin: The Ganges River and its tributaries support diverse freshwater snail species. These rivers flow through several states in northern India, including Uttarakhand, Uttar Pradesh, Bihar, and West Bengal.[17]
- Brahmaputra River Basin: The Brahmaputra River, along with its tributaries, provides habitat for freshwater snails in the northeastern states of Assam, Arunachal Pradesh, and Meghalaya.[17]
- Western Ghats: The Western Ghats Mountain range is a biodiversity hotspot and is home to numerous freshwater ecosystems, including rivers, streams, and freshwater marshes. States such as Kerala, Karnataka, and Maharashtra have diverse freshwater snail populations.[17]
- Eastern Ghats: The Eastern Ghats also support freshwater snails in states like Odisha, Andhra Pradesh, and Tamil Nadu.[17]
- Central India: Several rivers and lakes in central India, including those in Madhya Pradesh, Chhattisgarh, and Jharkhand, harbor freshwater snail species.
- Northeastern States: Apart from the Brahmaputra River Basin, other northeastern states like Manipur, Mizoram, Tripura, and Nagaland have freshwater snail populations in their rivers and wetlands.(stelbrink et al, 2023)[17]

➢ *Chitin:*

Chitin is a long-chain polymer of N-acetylglucosamine, a subordinate of glucose. It is one of the most inexhaustible biopolymers in nature, essentially found in the exoskeletons of arthropods (such as shellfish and creepy crawlies) and the cell dividers of parasites. Filopaludina bengalensis, like other molluscs, moreover contains chitin in its shell.[19]

➢ *Structure:*

Chitin comprises of β-(1→4)-linked N-acetylglucosamine units, comparative to cellulose's structure. The essential distinction lies in the substitution of the hydroxyl bunch at the C-2 position with an acetyl amine gather:

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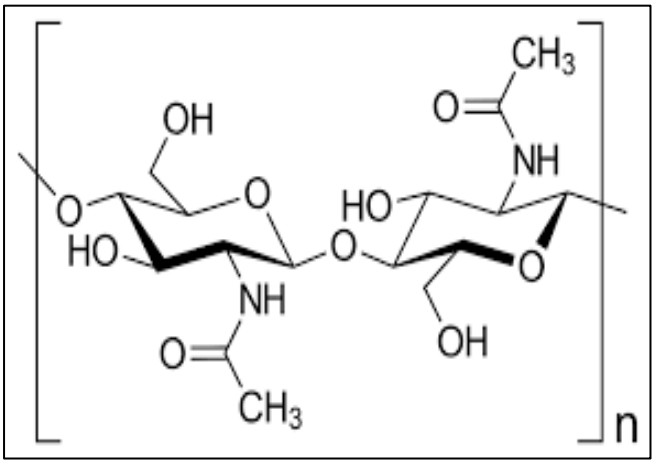

Fig 1 Structure of Chitin

- *Properties:*

Chitin is insoluble in water and most organic solvents but can be dissolved in acidic solutions, concentrated alkalis, and certain ionic liquids. It exhibits excellent biocompatibility, biodegradability, and antimicrobial properties, making it valuable in various industrial and biomedical applications (Rinaudo et al. 2006).[6]

- *Chitosan:*

Chitosan is a derivative of chitin obtained by deacetylation, a process that removes a portion of the acetyl groups from the chitin polymer. This results in a polymer with both primary amino groups from (deacetylated N-acetylglucosamine units) and remaining acetyl groups.

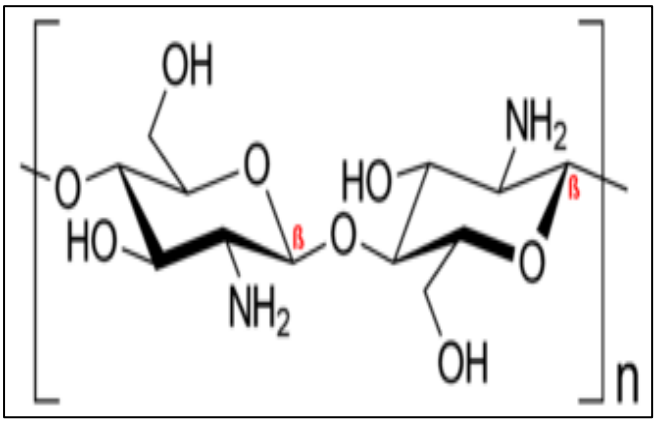

Fig 2 Structure of Chitosan

- *Properties:*

Chitosan is dissolvable in acidic arrangements, shaping polycationic chains due to protonation of amino bunches. This dissolvability makes it flexible for different applications. Chitosan shows bio cement, mucoadhesive, and wound-healing properties (Rinaudo et al. 2006)[10]

- *N-Acetyl-D-Glucosamine:*

N-Acetyl-D-Glucosamine (GlcNAc) is a monosaccharide and an amino sugar derivative of glucose. It is a key component of chitin and chitosan, constituting the repeating unit of their polymer chains.

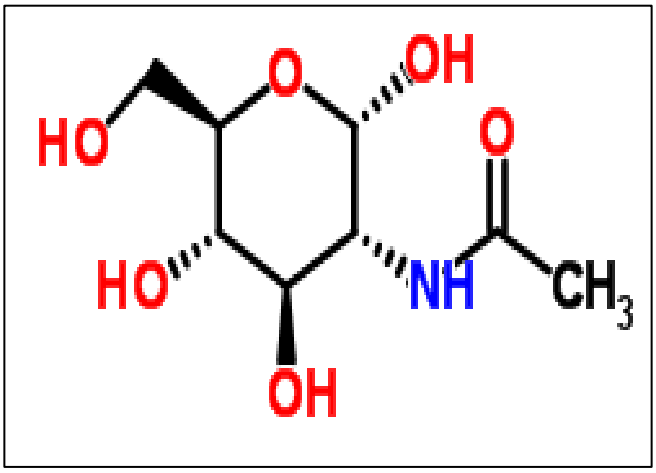

Fig 3 Structure of N-Acetyl-D Glucosamine

➢ *Function:*

- GlcNAc plays essential roles in various biological processes. In addition to being a structural component of chitin, it serves as a substrate for glycosylation reactions in protein modification. GlcNAc is also a precursor for the synthesis of other important molecules, such as glycosaminoglycans found in connective tissues and the glycan portion of glycoproteins.[12]
- GlcNAc has attracted attention for its potential therapeutic applications, particularly in promoting joint health and as a dietary supplement for conditions like osteoarthritis.
- In *Filopaludina bengalensis,* GlcNAc contributes to the structural integrity of the shell, and its extraction could provide a source of this compound for further research and applications.
- The estimated half-life for glucosamine is 15 hours after an oral dose.
- About 90% of glucosamine administered orally as a glucosamine salt get absorbed from the small intestine, and from there it is transported via the portal circulation to the liver. It appears that a significant fraction of the ingested glucosamine is catabolized by first-pass metabolism in the liver (Jain et al. 2013).[17]

➢ *Mechanism of Action of N-Acetyl-D-glucosamine*

The component of activity of glucosamine in joint wellbeing is vague, in any case there are a few conceivable instruments that contribute to its restorative impacts. in joint wellbeing is vague, in any case there are a few conceivable instruments that contribute to its restorative impacts. Since glucosamine is a antecedent for glycosaminoglycans, and glycosaminoglycans are a major component of joint cartilage, glucosamine supplements may offer assistance to revamp cartilage and treat the indications of joint pain. A few in vitro considers appear prove that glucosamine decreases aggravation by means of hindrance of intergalactic gamma.(van et al, 2016).[23]

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When taken up by living cells, glucosamine responds with ATP to shape glucosamine-6-phosphate, the normal antecedent of glycosaminoglycans (Chokes) that contain N-acetylglucosamine (keratan sulfate and Hyaluronan) and those that have N-acetyl galactosamine (heparan sulfate and chondroitin sulfate). These Chokes are polysaccharides composed of hexosamines and monosaccharides organized as a straight chain of rehashing disaccharide units. With the exemption of hyaluronan, Chokes do not exist alone in nature but are connected to particular "core" proteins, and the composite structures are called proteoglycans. Both hyaluronan and numerous diverse sorts of proteoglycans (such as aggrecan, versican, and syndecan) are copious all through the body where they perform different functions.(Van et al, 2016).[11]

II. MATERIALS & METHODS

A. Collection

➢ *Exterior Shell Structure:*

- **Apex:** This is the oldest portion of the shell and its pointed end. Because fresh whorls are added to the shell from the base of the apex, the apex also contributes to the shell's growth.
- **Whorls:** The shell is composed of these swirling layers. The snail's shell develops new whorls as it becomes bigger.
- **Suture**: The whorls converge at this line. The suture helps to reinforce the shell and comes in a variety of forms.
- **Aperture:** The snail inserts its body through this gap in the shell. The operculum, a thin, horny plate, can seal it in certain species.
- **Peristome:** This is the aperture's edge, and depending on the species, it may have a variety of forms and structures. The snail's ability to hold onto surfaces can be aided by the peristome.
- **Columella:** The shell is supported by this central pillar. Additionally, it can aid in controlling the snail's buoyancy and motion.
- **Radial ribs:** These ridges extend from the aperture to the apex. They give the shell structure and strength.
- **Growth lines:** The snail's growth rate is indicated by these lines, which run parallel to the suture.
- **Color bands:** The shells of certain snail species exhibit recognizable bands of color. These can give predators warning signals or concealment. Depending on the species, the shell's size and shape might vary significantly.

➢ *Interior Shell Structure:*

The shell of a snail is composed of three main layers:

- **Periostracum:** Conchiolin is a thin, organic substance that makes up the shell's outermost layer. Typically brown or black, the periostracum might aid in shielding the shell from erosion and harm.
- **Prismatic layer:** This layer, which is composed of prism-like calcium carbonate layers, is situated under the periostracum. The prismatic layer is in charge of giving the shell its stiffness and strength.
- **Nacreous layer:** The deepest layer of the shell, also referred to as the mother-of-pearl layer, is composed of thin, flat layers of calcium carbonate. In addition to giving the shell its smooth, iridescent appearance, the nacreous layer gives it extra strength.
- **Shell Gland:** Near the mantle, it secretes the substance that makes up the snail's shell. These are the primary internal body parts that *Filopaludina bengalensis* usually has. Age, sex, and environmental factors can all have an impact on the precise anatomy, which may differ slightly from person to person. Additional information about the composition and operation of these organs can be obtained by examination using microscopy and dissection.

➢ *Preparation of Chitin*

- **Cleaning the snail Shells:** The periwinkle shells are cleaned by washing them under water at 100°C to remove any impurities or contaminants.
- **Digestion with Aqueous HCl:** The cleaned shells are digested with 2.0N aqueous hydrochloric acid (HCl) for 5 hours at room temperature. This step helps in breaking down the organic material present in the shells.
- **Washing and Drying:** After digestion, the shells are washed to remove excess acid and dried at 100°C to evaporate any remaining moisture.
- **Grinding into Powder:** The dried shells are then ground into a fine powder using a hammer mill. This increases the surface area and facilitates further extraction.
- **Extraction with Aqueous HCl:** The powder is extracted for 48 hours with 2.0N aqueous HCl. The content of the flask is vigorously agitated at frequent intervals to ensure thorough extraction.
- **Centrifugation:** After extraction, the mixture is centrifuged(3000rpm) to separate the insoluble material from the liquid.
- **Washing:** The insoluble material is washed with water to remove any residual acid.
- **Extraction with Aqueous NaOH:** The washed material is then extracted with 1N aqueous sodium hydroxide (NaOH) for 12 hours at 100°C. The flask is shaken frequently during this process.
- **Collection of Insoluble Material:** The insoluble material is collected by centrifugation after extraction with aqueous NaOH.
- **Repeat Extraction:** The extraction with aqueous alkali is repeated three times to ensure thorough removal of impurities.

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- **Washing and Drying:** The collected insoluble material is washed with water until neutral and then with ether and ethanol to remove any remaining impurities. Finally, it is dried in a desiccator over phosphorous pentoxide to ensure complete dryness.

➢ *Preparation Snail Enzyme*

- **Collection of Periwinkles:** Periwinkles are collected from their natural habitat.
- **Starvation:** The collected periwinkles are starved for several days to empty their gut of food contents.
- **Immersion in Water:** The periwinkles are completely submerged in water in a closed dish, causing them to be killed by lack of air.
- **Removal of Intestinal Tracts:** The intestinal tracts of the periwinkles are carefully removed without losing any intestinal contents.
- **Grinding:** The collected intestinal tracts are ground in a mortar with toluene and sand. This mixture is then washed with a small amount of water and filtered through asbestos pulp.
- **Filtration:** The brown filtrate obtained from the grinding process is cleansed against running water for three days. It is then diluted to a volume such that each volume represents the enzyme obtained from one periwinkle.
- **Preservation:** A few drops of toluene are added to the preparation as a preservative to maintain its stability.
- **Enzyme Concentration:** The preparation contains an enzyme that can filter through a cellophane membrane, indicating the presence of the desired enzyme in the final product.

➢ *Isolation of N-acetyl-D-glucosamine from the Enzymic Concentration of Chitin*

- *Preparation of Reaction Mixture:*

Finely powdered chitin, snail enzyme preparation, toluene, and 0.1N aqueous hydrochloric acid are mixed thoroughly. The pH of the solution is adjusted to the desired level (usually acidic) using the hydrochloric acid solution. The mixture is then kept at a constant temperature of 37 ± 0.25°C for 30 days to allow enzymatic degradation of chitin.

- *Agitation of Reaction Mixture:*

The reaction mixture is shaken vigorously twice daily to ensure thorough mixing and promote enzymatic action on chitin.

- *Removal of Protein and Unchanged Chitin:*

Ethanol is added to the reaction mixture to precipitate proteins and any unchanged chitin. The mixture is then subjected to centrifugation to separate the precipitated solids from the clear solution.

- *Evaporation of Solution:*

The clear solution, free from protein and unchanged chitin, is evaporated to dryness under vacuum at 50°C in an inert atmosphere.

- *Extraction of N-acetyl-D-glucosamine:*

The dried residue is dissolved in absolute methanol and boiled under reflux for 90 minutes to dissolve N-acetyl-D-glucosamine.

- *Filtration and Concentration:*

The hot methanol solution containing N-acetyl-D-glucosamine is filtered to remove any insoluble impurities. The filtrate is then concentrated to a small volume at 40°C under vacuum in an inert atmosphere.

- *Crystallization of N-acetyl-D-glucosamine:*

The concentrated solution is left in the refrigerator to allow N-acetyl-D-glucosamine to crystallize.

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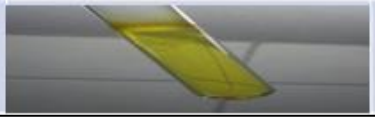
III. RESULTS

Table 1 Identification Test

SL NO	CONSTITUENTS	TEST	OBSERVATION	RESULT
1	Carbohydrate	Molisch test		Positive
2	Carbohydrate	Fehling test		Positive
3	Carbohydrate	Benedict test		Positive
4	Flavonoids	Sulfuric acid test		Positive
5	Fe3+	Nitric acid + △ = brown ppt Add HCl + potassium ferrocyanide		Positive
6	Amino Acid	Ninhydrin test		Positive
7	Amino Acid	1% Sodium Nitrite		Positive
8	Iodine	Iodine test		Positive
9	Protein	Biuret test		Negative
10	Protein	Millon's test		Negative
11	Fats and fixed oil	1%$CuSO_4$+NaOH test		Positive
12	Ca^+Ion	Ammonium oxalate + Ammonium hydroxide		Positive
13	Phosphate	Nitric acid + ammonium molybdate		Positive

➢ *Microscopical Evaluation of Filopaludina bengalensis: -*

- *Shell Morphology*

Juvenile shells have an elevated embryonic shell with fine spiral threads, transitioning subtly to the teleoconch. Early teleoconch features include a bluntly-angular basal carina and prominent cords with varying hairs. Adults lack these features, with deeper sutures, rounded shoulders, and inflated whorls. The adult aperture is large and ovate with a well-rounded base and an open umbilicus.

Fig 4 Shell of Filopaludina benghalensis

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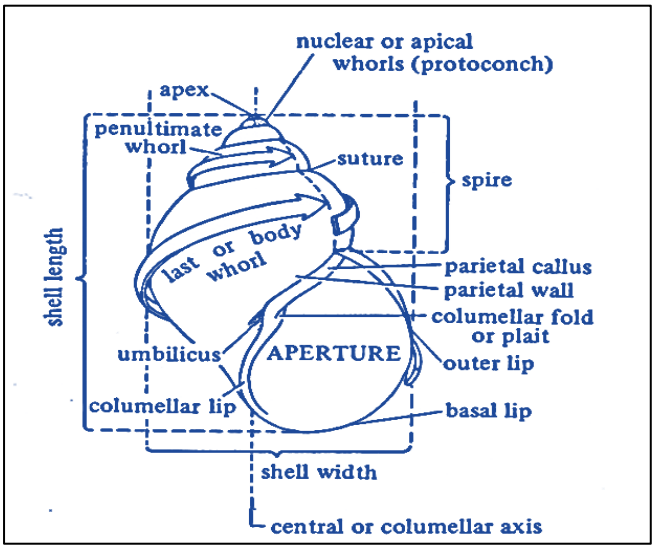

Fig 5 Structure of Shell

➢ *External Anatomy*

The operculum is thick and brown, thicker at the parietal and columellar edges. Delicate tissues of grown-ups incorporate a wide, strong foot with a pedal organ, and cephalic limbs. In guys, the right limb is adjusted into a penis. Eyes are terminal on brief peduncles. The neck is altered into a tall nuchal flap at the level of the urinary pore, butt, and female gonopore.

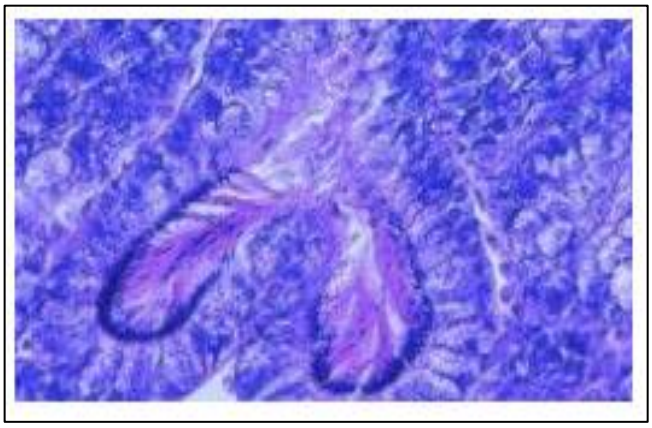
Fig 6 External Anatomy of Fresh Water Snail

➢ *Alimentary System*

The mouth forms a transverse slit at the ventral end of the oval snout. The buccal mass is long and slender, with the odontophore occupying most of its length. A small paired jaw flanks the mouth dorsolateral. Shallow buccal pouches are behind the buccal ganglia. Salivary gland ducts pass through the nerve ring to large glands covering it dorsally and laterally. Buccal retractors extend from the buccal mass to the cephalic hemocoel. The radular sac is short, projecting slightly past the buccal mass. The radula is small and slender, with about 80 rows of developed teeth.

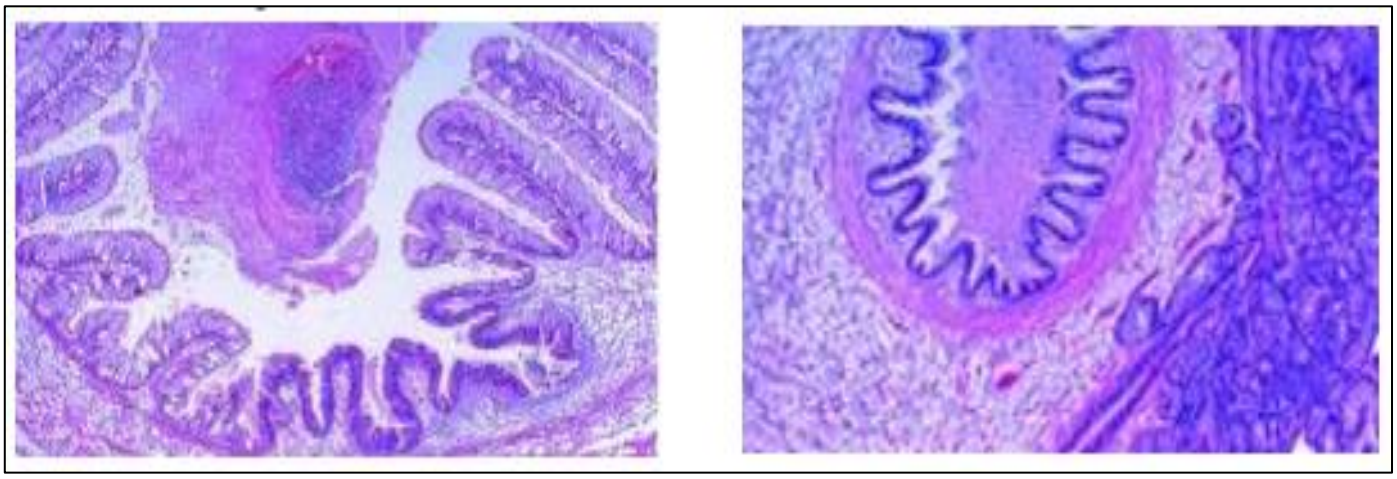
Fig 7 Alimentary System of Fresh Water Snail

➢ *Digestive Gland: -*

The stomach related organ, found in the visceral bump, involves roughly 2.0-2.5 apical whorls, with its front parcel supporting the right angle of the gastric chamber. It's not completely separated into littler front and bigger back flaps. Tubules deplete into stomach related organ channels along the columellar angle of the whorls, opening into the gastric chamber. The fashion sac, approximately 1.5 times the length of the gastric chamber, needs a crystalline fashion and bears two ventral typhlosoles. These typhlosole's turn strongly to the cleared out some time recently proceeding anteriorly, with shallow pyloric caeca between them. The digestive tract rises from the fashion sac's front conclusion, bending posteriorly overlying the pericardium and the fashion sac's right side. The typhlosole's of the fashion sac proceed along the side into the digestive tract, with the major typhlosole show for half its length.

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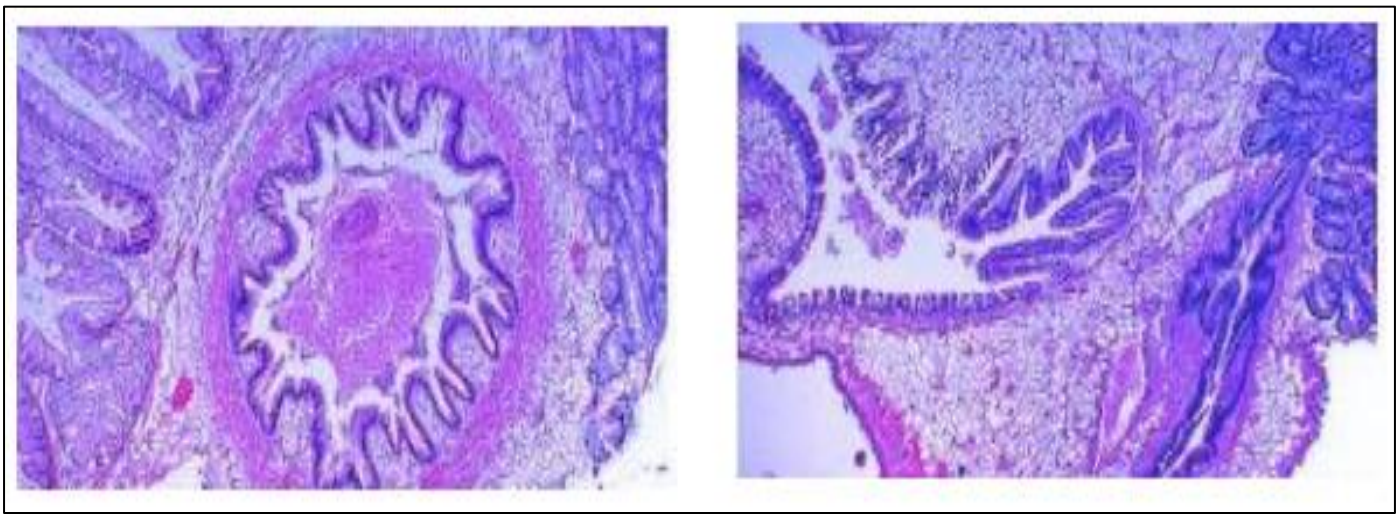

Fig 8 Digestive System of Fresh Water Snail

➢ *Characterization by FTIR Analysis*

FTIR range was utilized to distinguish the useful bunches of the dynamic constituents that were isolated from test. The FTIR spectra given by Bruker optics alpha FTIR spectrometer, were utilized to recognize the item and to decide the degree of closeness. The comes about are spoken to in taking after fig 9,10,11,12. FTIR crests of commercial Chitin and chitin gotten from test were compared.

The FTIR spectra of commercial chitin shown a characteristic wide band at 3447 cm−1 ascribing to O–H extending. The retention groups at 1660 cm−1 and 1559 cm−1 compared to Amide I C=O extending and N–H twisting and C-N extending of Amide II .The top at 1073 cm−1 was due to the C–O–C vibration interior the chitin ring structure .

Analysis of utilitarian bunches to recognize utilitarian bunches and calculates the degree of deacetylation was performed by utilizing FTIR (Fourier change infra-red). FTIR Spectra of N-Acetyl-D-glucosamine appears the assimilation at wave number of 3345.53cm-1 as a result of vibration of –OH gather and taken after by assimilation at wave number 3293.47 cm-1 determined from extending vibration of NH amines. Test is about comparative to standard N-Acetyl -D-Glucosamine.

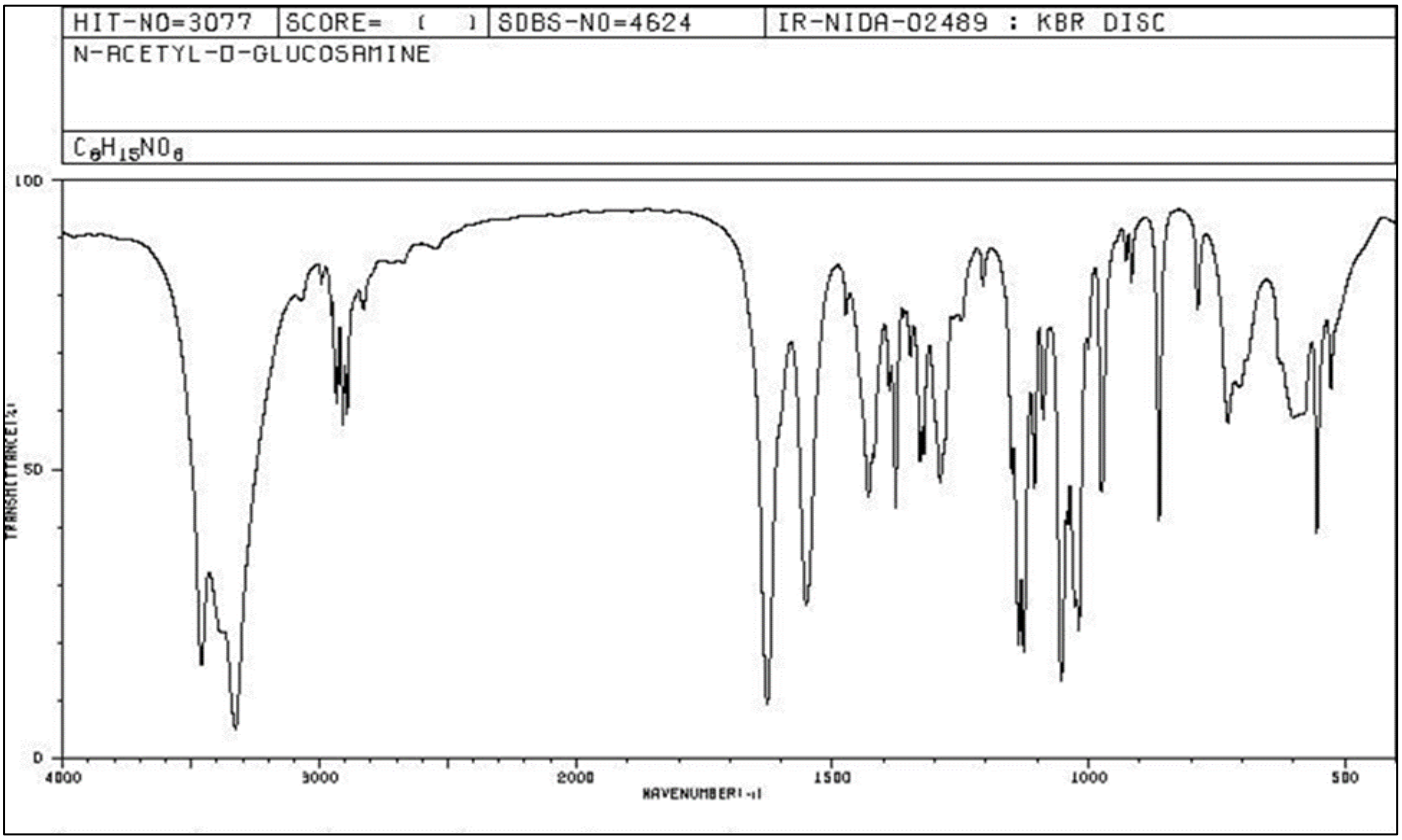

Fig 9 FTIR Analysis of N-Acetyl-D-glucosamine (Standard)

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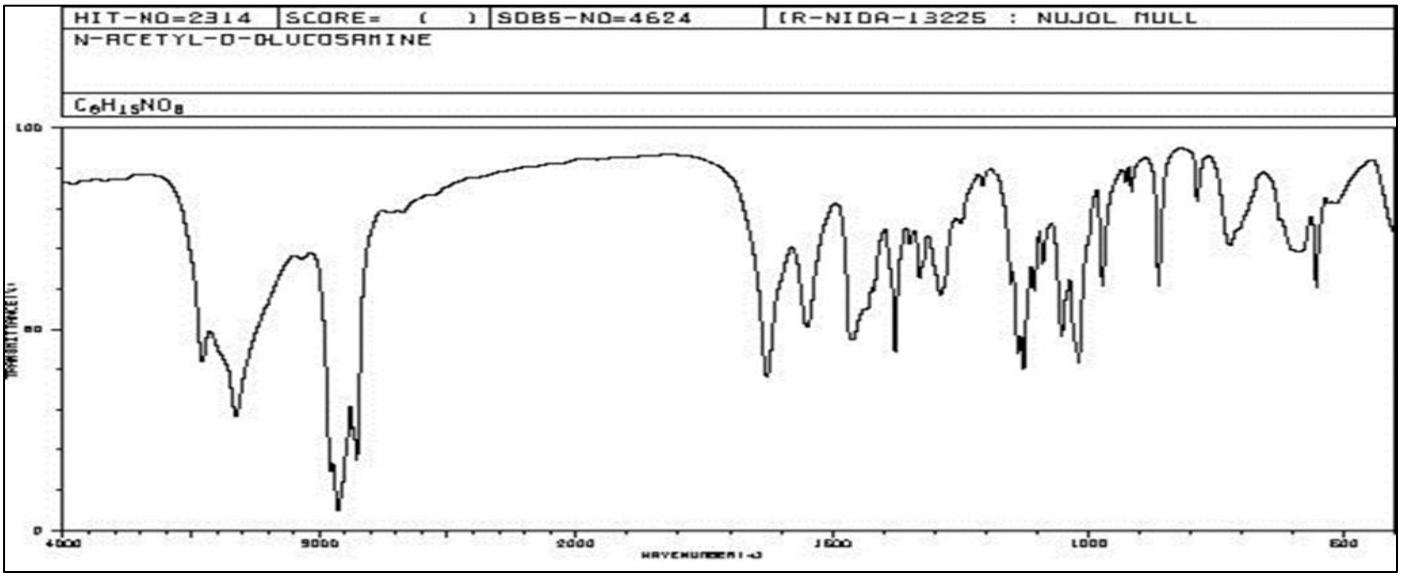

Fig 10 FTIR Analysis of N-acetyl-D-glucosamine (*Filopaludina bengalensis*)

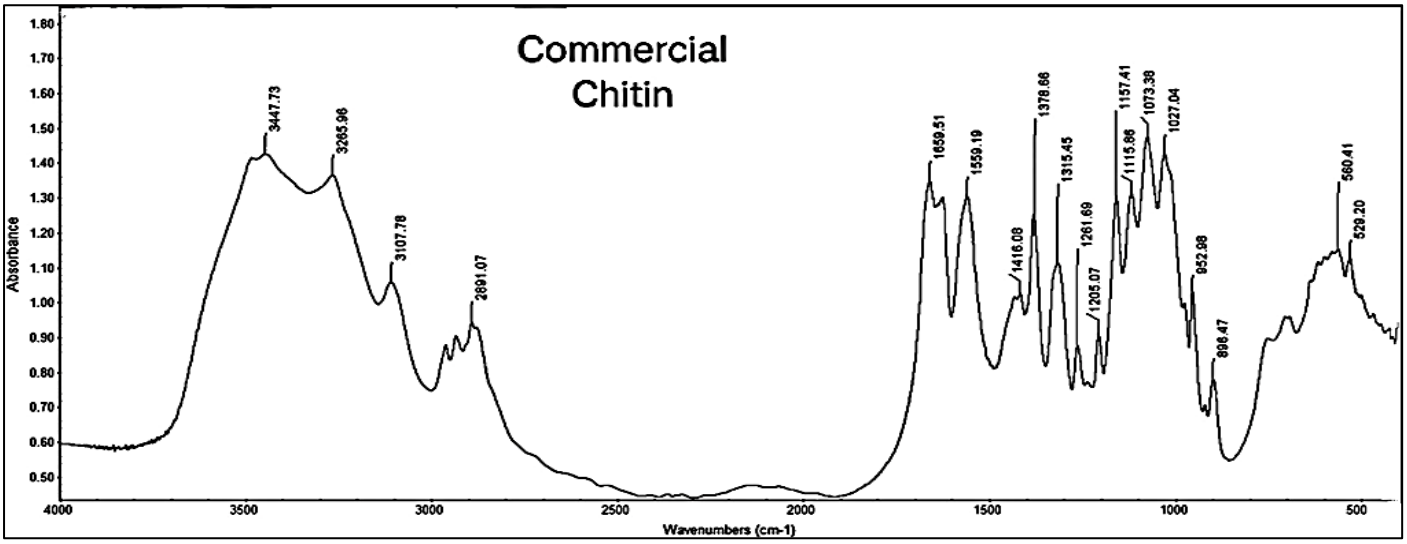

Fig 11 FTIR Analysis of Commercial Chitin (Standard)

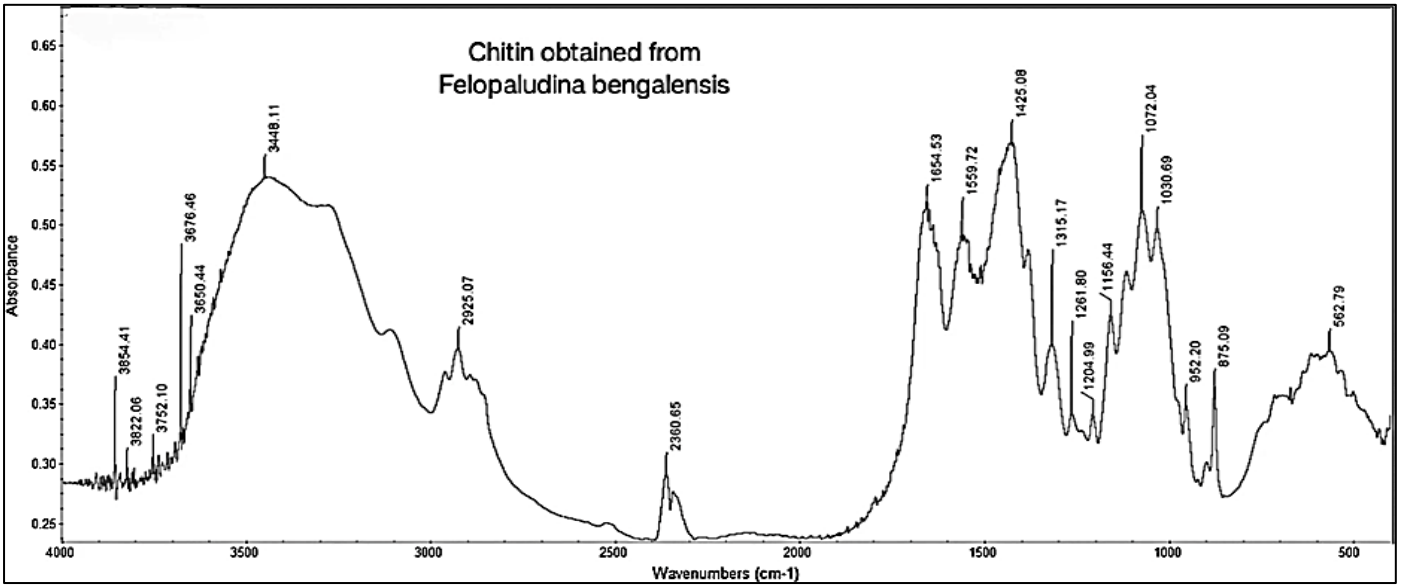

Fig 12 FTIR analysis of Chitin Obtained from *Filopaludina bengalensis*

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IV. DISCUSSION

The text discusses the biodiversity, taxonomy, food value, and polymorphism *of Filopaludina bengalensis*, a freshwater snail species. It highlights its ecological importance, nutritional value, and potential medicinal uses, focusing on its distribution in India and the properties of chitin and chitosan extracted from its shell.

The freshwater snail *Filopaludina bengalensis* is an aquatic gastropod found in various freshwater habitats worldwide. It possesses a radula for feeding, tentacles for sensory perception, and a muscular foot for movement. Its shell comprises distinct layers, and internally, it has systems for digestion, respiration, circulation, excretion, and reproduction.

Freshwater snail shells were cleaned, digested with HCl, ground, and extracted with HCl and NaOH to obtain chitin. Periwinkles were collected, starved, and their enzymes extracted and concentrated. Chitin was enzymatically degraded, and N-acetyl-D-glucosamine was isolated through a series of steps including filtration and crystallization.

Microscopic evaluation of *Filopaludina bengalensis* reveals juvenile shells with fine spiral threads, transitioning to adult shells with deeper sutures and a large ovate aperture. Soft tissues include a thick brown operculum, a broad muscular foot, cephalic tentacles, and a modified male penis. The alimentary system comprises a long buccal mass, shallow buccal pouches, and a digestive gland with anterior and posterior lobes, typhlosoles, and pyloric caecae.

- *Conflict of Interest*

There is no conflict of interest

V. ACKNOWLEDGEMENT

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VI. CONCLUSION

The present study entitled **"Identification & extraction of Fresh Water Snail Polysaccharide"** support the following conclusion :-

- *Systematic Extraction Approach:*

The study outlines a methodical process for extracting polysaccharides from freshwater snails, specifically targeting chitin and N-acetyl-D-glucosamine. It involves multiple steps, including rigorous cleaning, digestion, and extraction, aimed at ensuring the purity and quality of the final product.

- *Focus on N-acetyl-D-glucosamine:*

The extracted N-acetyl-D-glucosamine shows promise for biomedical applications due to its diverse functionalities. This compound holds potential for various therapeutic uses in the medical field.

- *Optimization of Extraction Parameters:*

Further research could focus on optimizing extraction parameters to enhance yields, purity, or both. Fine-tuning the extraction process may lead to improved efficiency and cost-effectiveness.

- *Evaluation of Bioactivity:*

Assessing the extracted polysaccharides bioactivity is crucial.- This evaluation could involve studying their effects on biological systems, such as wound healing, inflammation, or immune response. Understanding the bioactivity could unveil potential therapeutic applications and mechanisms of action.

- *Potential Therapeutic use:*

Investigating the therapeutic potential of the extracted polysaccharides is crucial. This research could explore their efficacy in treatinsg various health conditions, potentially leading to the development of new drugs or therapies.

- *Consideration of Sustainability:*

Sustainable sourcing practices should be considered, taking into account the ecological implications of extracting materials from freshwater snails. Environmental impact assessments and sustainable harvesting methods could ensure long-term viability without harming ecosystems.

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