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EXPLORING CELLULOSE RECOVERY FROM CAULIFLOWER DISCARDS

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ABSTRACT

Agricultural and industrial wastes are among the major causes of environmental pollution. Their conversion to useful products may ameliorate the problems they cause. In the presented study, Cauliflower stem and stem leaves commonly discarded as waste materials were used for the extraction of cellulose followed by paper making as one of the applications. For the extraction of cellulose, the Acid Alkaline hydrolysis method was explored. The waste is composed of both stems and leaves, so 3 distinct batches were employed for extraction, a-only stem as source, b-only leaves, and c-entire waste including both; in order to evaluate which part of the discard contained the highest amount of cellulose. Regardless of the results, the study proceeded with batch c, as we aimed at the utilization of the entire waste. The extracted cellulose fibers were then subjected to various evaluation parameters such as microscopical analysis and chemical analysis including sulfuric acid test, vanillin test, ninhydrin test, biuret test, iodine test, Molisch test, benedict test, and Fehling test. Further, the study headed with the preparation of paper from obtained fibers, one among many applications of cellulose. Finally, the obtained paper was characterized by various tests, GSM test, pH test, thickness test, bulk test, moisture test, folding endurance and, tensile strength. There are other applications as well of extracted cellulose such as biofuel production, as film-forming agent, as thickener, as blocker, sustained release agent, blending agent, and suspending agent, ensuring discrete pharmaceutical preparations, and many more.

KEYWORDS: Cauliflower waste, Cellulose extraction, Waste utilization, Cellulose evaluation, Paper making and Cellulose applications.

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INTRODUCTION

Cauliflower

Cauliflower belongs to the species *Brassica oleracea* in the Brassicaceae family.

An annual plant reproducing by seed, the cauliflower head is composed of a white inflorescence meristem, and only the head is typically eaten, known as the "curd."

Varietal Groups

Includes white, Romanesco, brown, green, purple, and yellow cultivars

Asian

A tropical cauliflower developed in India during the 19th century. Includes old varieties Early Banaras and Early Patna.

Varieties and Colours

Hundreds of historic and current commercial varieties globally. North American varieties maintained at North Carolina State University. Colours include white, orange (beta-carotene), green (broccoflower, Romanesco), and purple (anthocyanins)¹.

Cauliflower Waste

Food waste is a pressing global issue with significant environmental consequences. The Figure ofCauliflower Show in (Figure. 1). Approximately one-third of the world's food produced for human consumption is wasted annually. This wastage contributes to greenhouse gas emissions, exacerbates resource depletion, and poses a threat to biodiversity. The environmental impact extends throughout the entire food supply chain, from production and distribution to consumption and disposal. Cauliflower waste presents a specific challenge within this broader context. As a cruciferous vegetable, cauliflower generates substantial waste during processing and consumption. Traditional disposal methods, such as landfilling, contribute to methane emissions and other environmental hazards. Moreover, the nutritional and organic value of cauliflower waste remains largely untapped. The urgent need for sustainable solutions becomes evident in addressing both the global food waste crisis and the specific challenges posed by cauliflower waste. By developing efficient methods to extract valuable components like cellulose from

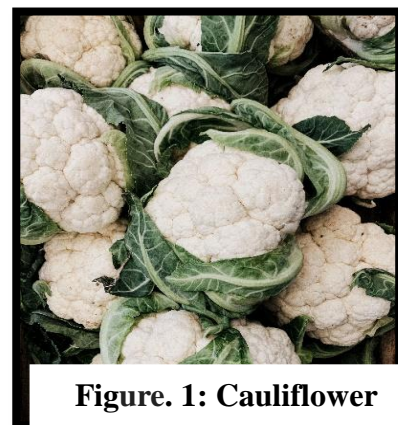


Figure. 1: Cauliflower

cauliflower waste, we not only mitigate environmental harm but also contribute to the circular economy. This initiative aligns with broader sustainability goals, offering a pathway to transform waste into valuable resources and reduce the overall ecological footprint of the food industry. Cauliflower waste is reported to be a rich source of cellulose, with a composition of approximately 16.6% cellulose. Additionally, it contains 8.4% hemicellulose, making it a substantial reservoir of complex carbohydrates². The high cellulose and hemicellulose content in cauliflower waste positions it as a promising feedstock for biofuel production, particularly for the generation of biobutanol through processes like the acetone–butanol–ethanol (ABE) fermentation. The abundance of cellulose and hemicellulose in cauliflower waste makes it an attractive substrate for enzymatic hydrolysis, where these complex carbohydrates can be broken down into fermentable sugars. This breakdown is a crucial step in biofuel production, as it provides the necessary sugars for microbial fermentation, ultimately leading to the production of biobutanol. Moreover, the utilization of cauliflower waste as a cellulose-rich source not only addresses environmental concerns associated with waste disposal but also aligns with the concept of sustainable and renewable resources in the context of biofuel production³.

MATERIAL AND METHOD

Procurement

Cauliflower discards were obtained from Chiplun local market and all other chemicals used were obtained from Chemistry Lab, Govindrao Nikam College of Pharmacy, Sawarde. Showed in Table. 1.

SR NO.	Materials	USE
1.	Cauliflower discards	Cellulose source
2.	Dil. Sulphuric Acid (64% w/v),	Acidic Agent
3.	Dil. NaOH (3% w/v)	Alkaline Agent
4.	Hydrogen Peroxide	Bleaching Agent
5.	Distilled Water	Co-solvent

Table. 1: List of Materials

Extraction

Acid hydrolysis

Cut or shred the cellulose source into small pieces and weigh about 10 g of it. Add 100 mL of dilute sulfuric acid to the beaker and heat it on the hot plate until it reaches about 80°C. Add the cellulose source to the acid solution and stir with the glass rod for about 30 minutes. Make sure the temperature does not exceed 100°C. Filter the mixture using the filter paper and the funnel. Collect the filtrate in another beaker and discard it safely. Wash the residue on the filter paper with distilled water until the pH is neutral⁴.

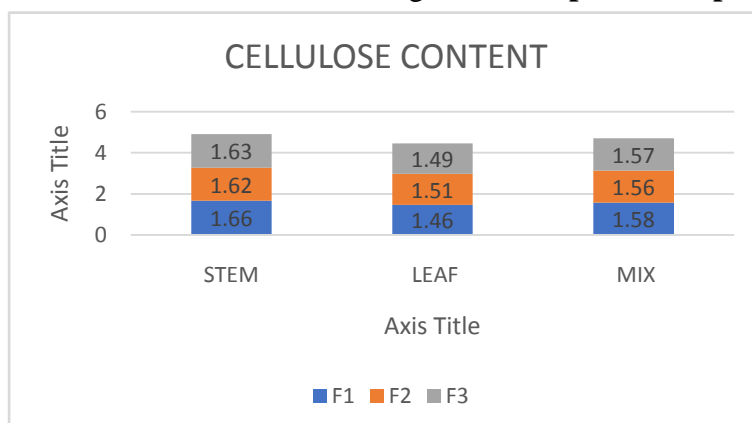
Treatment with alkali

Add 100 mL of sodium hydroxide solution to the beaker and heat it on the hot plate until it reaches about 80°C. Add the residue to the base solution and stir with the glass rod for about 30 minutes. Make sure the temperature does not exceed 100°C. Filter the mixture using the filter paper and the funnel. Collect the filtrate in another beaker and discard it safely. Wash the residue on the filter paper with distilled water until the pH is neutral. Transfer the residue to the baking tray and spread it evenly. Dry it in the oven at 105°C for about 2 hours. Scrape off the dried cellulose fibers from the baking tray using the spatula. You have obtained the cellulose extracted by the acid and alkaline hydrolyses method⁵.

Comparison Between Batches

As the cauliflower waste is composed of both stems and leaves. In order to determine which part of the waste comprised the highest cellulose content, 3 distinct batches were employed for extraction following the previously mentioned extraction process and dry weight of cellulose obtained from each batch was considered. Shown in Graph 1

- Batch a- only stem as source,
 - Batch b-only leaves as source, and
 - Batch c-entire waste including both
- Graph 1: Comparison Between Batches**



Characterization Of Cellulose Fibers

Microscopical Analysis

The cellulose fibers were observed under the compound microscope. The magnification power used was 10X. The fibers were observed for their shape and size. 300 cellulose fibers were analysed for their length and diameter, finally calculated the average of both. All Observations are showed in Table. 2 and all Figure show inFigure 4.

Chemical Analysis

Sulfuric Acid Test⁶

Chemicals: Sulfuric Acid solution

Procedure: Apply Sulfuric Acid solution with cellulose content in test tube Cellulose treated with sulfuric acid becomes hydrolysed, producing monosaccharides, which can be identified through subsequent tests. Appearance of Reddish colour in test tubes due to presence of monosaccharides.

Vanillin Test⁷

Chemicals: Vanillin-sulfuric acid reagent.

Procedure: Add vanillin reagent with cellulose content in test tube; cellulose shows no color change.

Ninhydrin Test

Chemicals: Ninhydrin solution.

Procedure: Apply Ninhydrin with cellulose content in test tube; cellulose exhibits no reaction.

Biuret Test

Chemicals: Copper sulphate and sodium hydroxide.

Procedure: Perform Biuret test with cellulose content in test tube; cellulose exhibits no violet colour.

Iodine Test

Chemicals: Iodine solution.

Procedure: Apply Iodine solution with cellulose content in test tube. Positive reaction indicates the presence of cellulose by forming a blue colour.

Molisch Test

Chemicals: Molisch reagent.

Procedure: Apply Molisch reagent with cellulose content in test tube. Detects the presence of carbohydrates, including cellulose, by forming a violet ring.

Benedict Test

Chemicals: Benedict's solution.

Procedure: Fill the test tube with Benedict's cellulose-containing solution. Benedict's solution is a specialised reagent that may be used to detect simple carbohydrates like glucose. Benedict's solution is blue, but if there are any simple carbs, it will become green or yellow when there are few, and red when there are a lot.

Fehling Test⁸

Chemicals: Fehling solution.

Procedure: Apply Fehling solution with cellulose content in test tube Similar to Benedict's test, it confirms the presence of reducing sugars derived from cellulose.

PREPARATION OF PAPER**Bleaching Process^{8,9}**

After the alkali treatment, 1.7 wt% sodium chlorite was used for the bleaching process, which lasted for 4 hours at 80°C. After allowing the mixture to cool, more distilled water was used to filter it. There were two instances of this procedure completed.

Paper Making Process^{10,11}

Soak the cellulose fibers in water for about 24 hours to soften them. Blend the soaked fibers with Mixer and Grinder more with water to form a pulp. Pour the pulp onto a fine mesh screen and press it with a sponge to remove excess water. Lift the screen and place it on a flat surface. Peel off the wet paper sheet from the screen and let it dry in Oven or sun drying accordingly. And Paper is made from cellulose Fibers. The Paper Making Process Show in Figure 2.

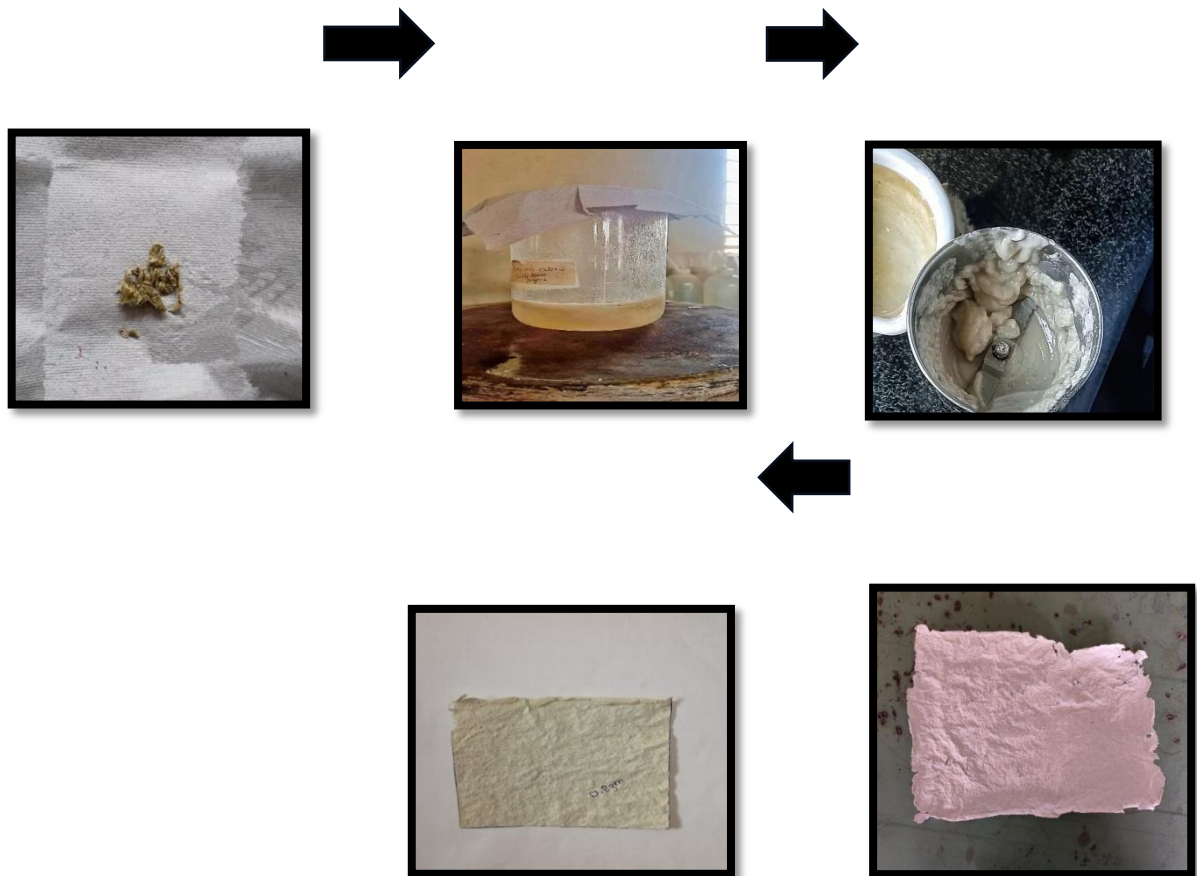


Figure 2: Paper Making Process

Evaluation Tests for Paper^{12,13}

GSM (Grams per Square Meter) Test

GSM values were calculated for paper made from cauliflower waste. Cut a square piece of paper with sides measuring 10 centimetres (1/100th of a meter). This will give you an area of 100 square centimetres. Weight the cut piece of paper using a scale. Make sure to record the weight in grams. Divide the weight (in grams) by the area (in square centimetres) to calculate the GSM. All Observations are showed in Table. 3 and all Figure show inFigure 5.

$$\text{GSM} = \text{Weight (grams)} / \text{Area (cm}^2\text{)}$$

This will give you the GSM value for the paper based on a 10x10 cm square.

pH Test

Cut a representative sample of the paper, ensuring it is free from contaminants. If using a pH meter, calibrate it according to the manufacturer's instructions using buffer solutions. Use distilled water to eliminate any interference from impurities. Moisten the paper sample with distilled water. This helps in obtaining an accurate pH reading. If using pH testing strips, dip a strip into the water-moistened paper and compare the colour change to the provided colour chart. If using a pH meter, immerse the electrode into the water-moistened paper and wait for the stable pH reading. Record the pH value obtained from either the pH testing strips or the pH meter. A neutral pH is 7. Values below 7 indicate acidity, and values above 7 indicate alkalinity. Compare the pH value with industry standards or other papers to assess the paper's acidity or alkalinity. Repeat the pH test with multiple samples to ensure accuracy and reliability. All Observations are showed in Table. 3 and all Figure show inFigure 10.

Thickness Test

Cut a representative sample of the paper, ensuring it is free from irregularities. If using a calliper or micrometre, ensure it is calibrated according to the manufacturer's instructions to maintain accuracy. Place the paper sample on a clean and flat surface. Choose multiple points on the paper for thickness measurements. It's advisable to measure at various locations to account for any unevenness. Use the calliper or micrometeor to measure the thickness of the paper at each chosen point. Apply gentle pressure to get accurate readings. Calculate the average thickness by summing up the individual measurements and dividing by the number of measurements. The thickness measurement provides insights into the density and thickness uniformity of the paper. All Observations are showed in Table. 3 and all Figure show inFigure7.

Bulk Test

Cut a representative sample of the paper, ensuring it is free from defects or irregularities, Weigh the paper sample using a precise scale, recording the weight in grams. Measure the area of the paper sample. If it's a standard size, you can use the dimensions provided. If not, use a ruler or measuring tape to determine the length and width. Use a calliper or micrometre to measure the thickness of the paper. Take measurements at multiple points for accuracy.

Calculate the bulk of the paper using the formula:

[Bulk = GSM/ Thickness]

This gives an indication of how much space the paper occupies relative to its weight.

Moisture Test

Cut a representative sample of the paper, ensuring it is free from contaminants. Weigh the paper sample accurately using a scale and record the initial weight. Place the paper sample in an oven set at a specific temperature (commonly around 105°C or 221°F). Ensure proper ventilation within the oven. Allow the paper to dry in the oven until it reaches a constant weight. This may take several hours, depending on the paper type. Allow the dried paper to cool to room temperature in a desiccator (optional) to prevent moisture. Weigh the dried paper sample once it has cooled. Record the final weight Use the formula:

[Moisture content = (Initial Weight – Final Weight / Initial Weight) X 100]

All Observations are showed in Table. 3 and all Figure show inFigure 8.

Folding Endurance

Folding endurance is a crucial property for paper, indicating its ability to withstand repeated folding without breaking or deteriorating. Place the paper sample in the tester, ensuring it's aligned with the folding edge. Start the test, and the machine will automatically fold the paper back and forth until it breaks or reaches a specified number of folds. Test piece is folded back and forth till it ruptures. All Observations are showed in Table. 3 and all Figure show inFigure 9.

Tensile Strength

Tensile strength measurement provided a practical way to determine the mechanical characteristics of the paper. An arrangement made up of a pan hung with a strong thread and the other end of the thread fastened to the paper's centre was used to measure the tensile strength of the material. Weights were kept on the pan and the entire assembly was balanced using a beam balance. Noted was the weight needed to shatter the paper. After that, tensile strength was computed using the following formula: Tensile strength is equal to Break Force / a.b(1+ ΔL/L). All Observations are showed in Table. 3 and all Figure show inFigure 6.

RESULT AND DISCUSSION

Microscopic Identification

The shape of fibers was found to be cylindrical. The average length and diameter of the cellulose fibers was found to be 3.97 micron and 1.86 micron respectively. The Microscopic Identification show in Figure 3.

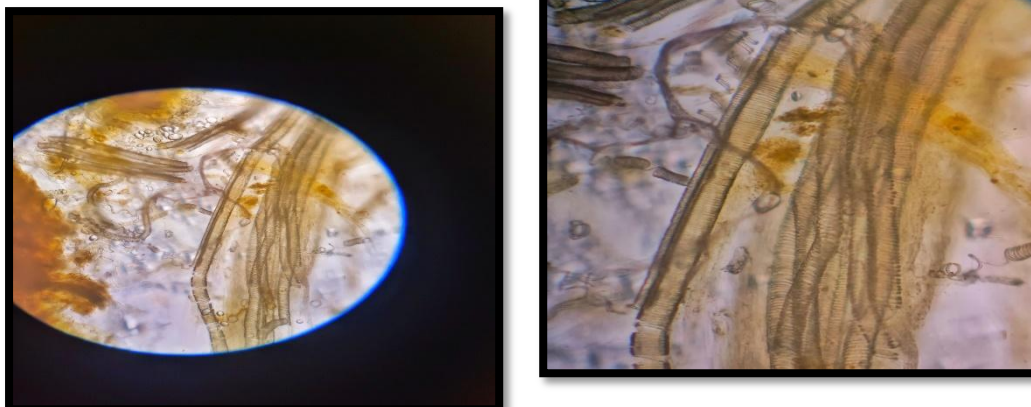


Figure3: Microscopic Identification

Chemical Analysis

Sr. No.	Tests	Observation	Inference
1.	Sulfuric Acid Test	Appearance of Reddish Colour in test tubes due to presence of monosaccharides	Cellulose present
2.	Vallin Test	No Colour Change	Cellulose present
3.	Ninhydrin Test	No Colour Change and no reaction	Cellulose present
4.	Biuret Test	Exhibits No violet Colour	Cellulose present
5.	Iodine Test	Formation of Blue Colour	Cellulose present
6.	Molisch Test	Formation of Violet Ring	Cellulose present
7.	Benedict Test	Appearance of reddish Colour precipitate in test tubes, due to presence of carbohydrates	Cellulose present

8.	Fehling Test	Appearance of deep blue Colour in test tubes due to absent of reducing sugars	Cellulose Absent
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Table. 2: Chemical Analysis



Figure4:Chemical Analysis

Evaluation Test Results for Paper

Sr. No	Tests	Paper 1	Paper 2	Paper 3
1.	GSM	148.28	148.14	148.35
2.	pH	5.56	5.60	5.58
3.	Thickness	0.3 mm	0.2 mm	0.2 mm
4.	Bulk Test	714.56	714.60	714.45
5.	Moisture %	27.45	27.55	27.58
6.	Folding Endurance	Good	Good	Good
7.	Tensile Strength	Better	Better	Better

Table. 3: Evaluation Test Results for Paper



Figure5:GSM Test

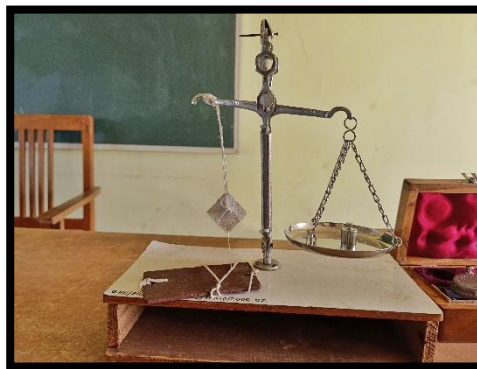


Figure 6:Tensile Strength



Figure7:Thickness Test



Figure8:Moisture Test

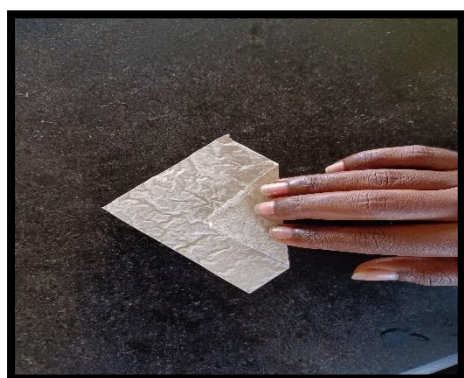


Figure 9:Folding Endurance



Figure 10:pH Test

CONCLUSION

As there is increasing attention on the exploration of waste feedstocks as economically viable substrates. So, in this study, Cauliflower stems and stem leaves commonly discarded as waste materials were put to use. Cauliflower is a vegetable with the highest waste index, contributing to 45-60% of the total weight of the vegetable. But this waste is composed of 17.32% cellulose, 9.12% hemicellulose, and 5.94% lignin. It is also an excellent source of health-promoting phytochemicals such as antioxidants like phenols, flavonoids, vitamins, and minerals. Hence, the presented work was done to encourage the use of cauliflower waste,

focusing on only one component cellulose, and its extraction. The successful extraction of cellulose was done using the Acid Alkaline hydrolysis process for systematically breakdown of the organic matter to isolate cellulose. The discards being utilized contained both stem and stem leaves as a source. So, to determine which part contained the highest amount of cellulose different batches were prepared. The results exposed that the batch extracted with the stem as source material contains more amount of cellulose as compared to leaves. The study was aimed at the use of entire waste hence further study was carried out by employing entire waste and not just stem. Further, microscopical analysis and chemical analysis confirmed the identification of the extract as cellulose. The extracted cellulose was then utilized to craft paper, showcasing the versatility and applicability of the obtained material along with the characterization of the prepared paper, which exhibited satisfactory results. Our study not only aimed at effective cellulose extraction but also emphasized the practical utilization of this renewable resource in a tangible and eco-friendly product. Through a combination of analytical techniques, hydrolysis processes, and hands-on applications like papermaking, our project contributes to sustainable waste management and underscores the potential of cauliflower waste as a valuable source of cellulose-based materials. This work aligns with the broader goal of promoting environmentally conscious practices and repurposing agricultural by-products for a greener future.

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Conflicts of Interest

The authors declare that there is no conflict of interest.

Author Contribution

Sukanya Patil suggest this topic Rushikesh Hingmire prepare manuscript and all technical work Chavan Komal, Salunkhe Prathamesh and all author perform work.

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