RESEARCH ARTICLE

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Xanthan Gum Production from Chicken Feathers

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Abstract: Xanthan gum is a polysaccharide produced by the bacterium *Xanthomonas campestris* which is one of the *Xanthomonas* species. Xanthan gum plays a key role in the industrial and food sectors. It shows a wide range of applications in food, cosmetics, oil, and paper industries due to its characteristics as a thickening, suspending and emulsifying agent. Due to the increased demand, there is a need for cheap and easily accessible organic nitrogen sources for production. This study produces Xanthan gum from waste chicken feathers. Poultry feather disposal annually harms our environment. Chicken feathers are renewable, easily accessible waste and economical. The waste chicken feathers are converted to CFP (Chicken Feather Peptone) through a chemical process. In this study, we discussed CFP to be an advisable substrate for Xanthan gum production.

Keywords: Chicken feather peptone, Polysaccharide, Xanthomonas campestris, Xanthan gum

1. Introduction

Many fungi and bacteria have the ability of producing polysaccharides. One of them is Xanthan gum. Xanthan gum was discovered by Allene Rosalind jeans and her research team at the United States of Agricultural department, and brought into a commercial production by Kelco, under the trade name kelzan in early 1960s [1]. Xanthan gum is one of the most universally used gums to control viscosity. It is a polysaccharide and a gelling agent. This gum is produced by the bacterium named Xanthomonas campestris which is one of the gram-negative phytopathogenic bacteria which is also seen in some vegetables and plants. These bacteria can convert Glucose, Glycerol, Sucrose, and other organic substrates into Xanthan gum [2]. Xanthan gum plays a key role in the industrial and food sectors. It shows a wide range of applications in food, cosmetics, oil, medicinal use, and paper industries due to its characteristics as a thickening, suspending and emulsifying agents. In the 1970's the use of Xanthan gum in the industrial sector increased.

Due to its industrial applications, the worldwide consumption of Xanthan has increased and hence the competition of the different sectors which needs Xanthan gum also increased. Several environmental factors get influenced by the production of Xanthan gum by microbial fermentation. Some of the factors are temperature, pH and dissolved oxygen level and production time. It was evident that there is really a need for the implementation of a low-cost substrate to make the Xanthan gum production process economical. Hence, to reduce the substrates and overall production cost, the only best solution is to use cheaper organic waste substrates. One among the low-cost substrate is chicken feathers which contain 10% chicken fat, and 90% protein composed of Keratin.

Every year tons of poultry feathers is disposed of causing adverse effects on our environment. Chicken feathers are renewable, easy access waste and economical which is suitable for the peptone production. Peptones are known as the hydrolysis products of substrates which are rich in nitrogen [3].

This research review is aimed to understand the work that is progressing in the field of production of Xanthan Gum by using chicken feather peptone which is a low- cost and eco-friendly raw material.

2. Environmental Impact of Chicken Feathers

The waste from the poultry industry creates a serious solid waste problem as it pollutes the soil and when it is burnt, it releases Sulphur dioxide which pollutes the air. In India, around 400 million chickens are processed every week (2021 data) [4]. To avoid environmental harm, we must make use of chicken feathers in diverse ways. Several types of waste are produced in the poultry industry, and it is disposed in the environment, which causes pollution. Some of such wastes are damaged eggs, spoiled feed, fecal mixtures, packing materials and feathers which is the highly produced waste which causes soil pollution. Hence it is important to degrade such waste chicken feathers to reduce the soil pollution. Chicken feathers are rich in keratin which is a reliable source of organic nitrogen. This study shows that using the peptone created from the waste chicken feathers xanthan gum can be produced economically.

2.1 Why are Chicken Feathers employed in Xanthan gum production?

Chicken feathers are renewable and easily access waste and very economical. The waste chicken feathers are converted to chicken feather peptone through a series of chemical processes. Mainly feathers contain 90% protein that composed of keratin.

The waste chicken feathers are the best example for organic nitrogen sources which are economical and eco-friendly compared to the expensive commercial nitrogen sources. The main nitrogen sources are from poultry and urban waste. The main utilization of chicken feather peptone is to produce the Xanthan gum to be economically. To produce xanthan gum, here

CFP were used as carbon and nitrogen sources. CFP is a low-cost substrate to produce xanthan gum by Xanthomonas campestris.

3. Method

Xanthan gum production consists of five methods. The methods are hydrolysis of chicken feathers, isolation of Xanthomonas campestris, media, analytical methods, and statistical analysis. Waste chicken feathers from the poultry and from the disposed feathers are collected and washed with deionized water followed by powdering using a blender. In this process chicken feather is treated using the neutralization and filtration process to get CFP. Neutralization is the method that forms a neutral solution which may react with an acid along with an alkali and in filtration process the solid matter is separated from the liquid material. The impurities are removed in this process.

3.1 Hydrolysis of chicken feathers

By using Deionized water, the feathers were washed and dried at 60°C in an oven. After drying completely, the feathers were cut into small pieces. With the help of a blender the chopped feathers were powdered. The powdered chicken feather is hydrolysated using the modified method of Kurbangolu as shown in figure 1.

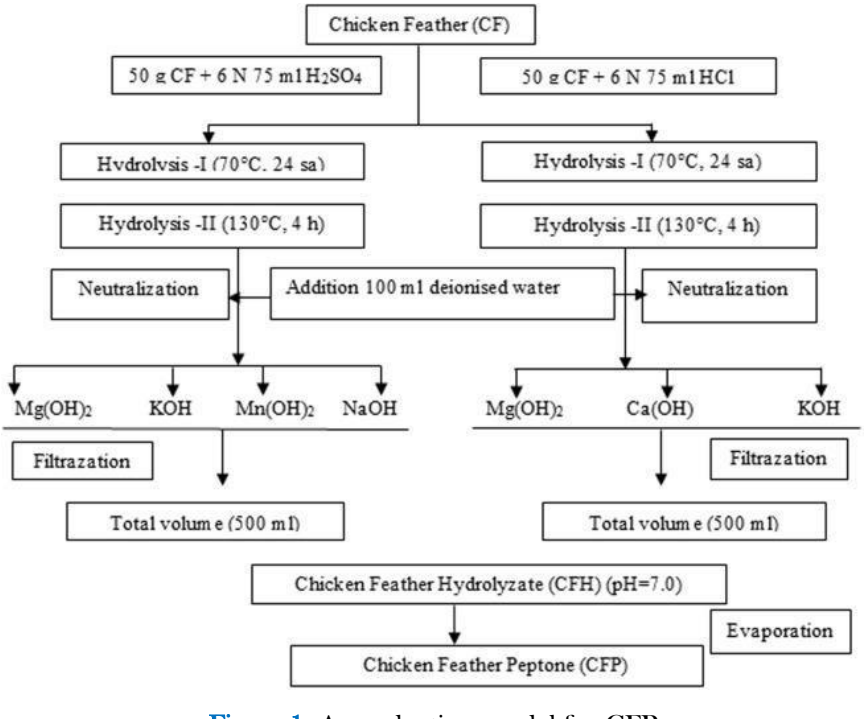

Figure 1. A production model for CFP

3.2 Isolation of Xanthan Gum Producing Bacterium

In cruciferous plants like cabbage, cauliflower and other plants infected the bacterium *Xanthomonas campestris*. By using yeast malt agar [YM] the infected plants were isolating. A bacterial colony has yellow mucus that is selecting and maintained on nutrient agar slants. The bacterial isolates were identified by using different tests like gram staining, oxidase tests and catalase test [5].

3.3 MEDIA

The cells grown on the yeast agar plates for three days are used to inoculate in a 250 ml flask containing 50 ml of the yeast malt broth. The flasks were incubated at 28°C and 200 rpm for 24 hours. The control medium compositions are concentrated HCl, Glucose, Citric acid, NH_4NO_3, $MgCl_2$.

Before autoclaving at 121°C for 15 minutes the pH was adjusted to 7.0. The culture temperature and the rate of agitation were maintained at 30°C. After which the Chicken feather peptone is compared with bacto peptone, Yeast and tryptone which are known as the commercial organic nitrogen sources at the concentration. This was found to be optimal for CFP [6].

3.4 Analytical Methods

1. ***AOAC methods*** (Amino acid Analysis) – It is used to find the total sugar and dry matter of the sample. In this study, it is used to estimate the total sugar, and dry matter contained in the chicken feather peptone. Figure 2 represents the experimental set-up of the AOAC method.

2. ***DNS (Dinitro salicylic acid method)*** – This method provides a simple and fast estimation of the reducing sugars or residual sugars in the hydrolysate process. The dinitro salicylic acid is known as a fragrant compound that reacts with reducing molecules and other reduced sugars to form 3-amino-5-nitrosalicylic acid. This method tests for free carbonyl groups in reducing sugars. Figure 3 represents the experimental set-up of DNS (Dinitro salicylic acid method)

3. ***Micro-Kjeldahl Apparatus is*** used to determine the amount of nitrogen contained in organic substances and inorganic compounds such as ammonia and ammonium. This measurement excludes inorganic compounds such as nitrate. This study aimed to determine Nitrogen content in the chicken feathers. Figure 4 represents the experimental set-up of Micro-Kjeldahl Apparatus.

Figure 2. AOAC methods (Amino acid Analysis)

4. ***Reverse-phase HPLC*** is a method to quantify, identify, and separate components by hydrophobicity and was used to analyze amino acids.

Figure 3. DNS (Dinitro salicylic acid method)

5. ***Soxhlet Apparatus*** – This apparatus is used to extract lipids from a solid material. The apparatus consists of three main parts. The first one is a percolator which is used to circulate the solvent. The second part is a thimble, which acts as a filter paper and retains the solid which is to be extracted, and the third part is a siphon mechanism which empties the thimble periodically. In this study it is used to find the crude fat content in chicken feathers. At regular intervals of fermentation, the residual sugar, microbial growth, and xanthan gum are determined. Cultures were harvested by centrifugation to achieve constant weight for estimation of biomass and ethanol is mixed with cell-free supernatant to precipitate xanthan gum. This xanthan gum is separated and dried until constant weight is gained [7].

Figure 4. The experimental set-up of Micro-Kjeldahl Apparatus

3.5 Statistical Analysis

In a randomized block design, the experiments were replicated three times. The SPSS 15.0 software program was used to perform statistical analysis. The statistical analysis was performed one way analysis of variance (ANOVA). ANOVA is a collection of statistical models, which is used to analyze the differences between the group means in a sample. In simple form, a statistical test is provided for the sample collected and which generalizes the data and provides the population means.

4. An Outline of Xanthan Gum production

First, to maintain the desired properties, the selected microbial strain is preserved for a longer period using proven methods. To obtain the inoculum for large bioreactors, a small quantity of the preserved culture is expanded by growth on solid surfaces or liquid media. The temperature, dissolved oxygen concentration and pH are the culture conditions required for the growth of microorganism and xanthan production.

Towards the end of fermentation, the broth contains bacterial cells, xanthan, and other chemicals. The cells are usually removed at the initial step to recover xanthan by centrifugation or through filtration. Using water-soluble non-solvents further purification is done and pH values are adjusted along with the addition of some required salts. Later the impurities are washed out. After a dry and evaporation method xanthan gum is formed.

5. Uses of Xanthan Gum

The unique properties of Xanthan gum have led to its widespread use as a thickener in many different industries such as the following:

- Food and beverage: In the food section, xanthan gum is used in sauces, dressings, and bakery products. The reason for the usage of xanthan gum in food products is to stabilize emulsion and help with solid particles suspension, such as spices.
- Personal care: Used in lotions and creams to prepare water gels.
- Pharmaceutical: Mainly used for oral care as a saliva substitute for dry mouth people and used as a laxative.
- Industrial: Used in paints and coatings due to its thickening ability.

6. Organic Nitrogen Sources in Xanthan Gum

Figure 5 shows the final output of the CFP growth rate in xanthan gum production. It shows the abundant nitrogen sources in chicken feather peptone. The rates confirm that the CFP is better than yeast extract, tryptone and bacto peptone when used as a supplement nitrogen sources. This positive effect is due to the high mineral content of CFP.

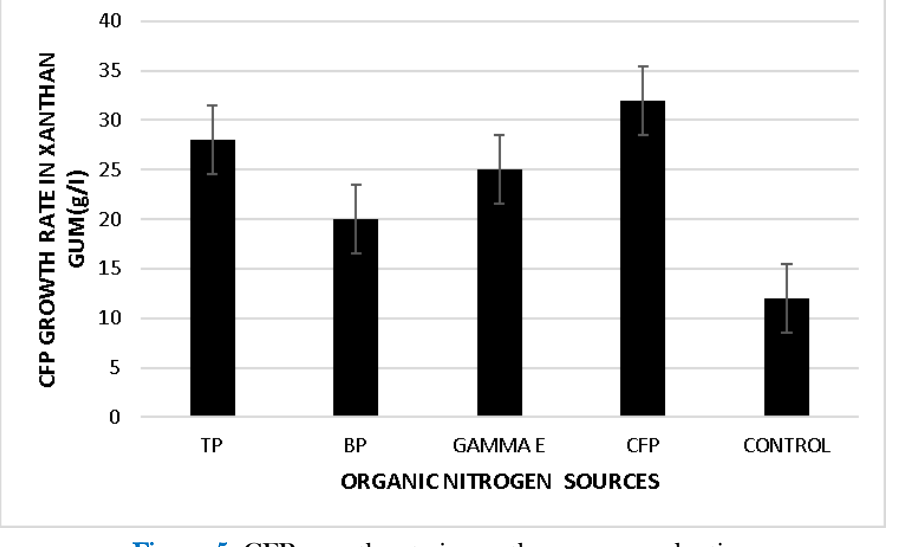

Figure 5. CFP growth rate in xanthan gum production

7. Conclusion

In conclusion, for Xanthan production, organic nitrogen sources are better than inorganic nitrogen sources. The waste chicken feathers, which are one of the best examples for organic nitrogen sources, are economical, easily available, and eco-friendly compared to expensive commercial nitrogen sources. Hence for xanthan production, waste chicken feathers

were converted to CFP through chemical processes. Chicken Feather Peptone has high protein nitrogen content. CFP contains amino acids except Methionine and Tryptophan. In the isolation and identification of Xanthan gum isolates, it is found that MO-03 strain is the best strain, which can produce maximum quantity of Xanthan gum. The quality of xanthan gum turns out to be good when CFP is used as a supplement nitrogen substrate. When compared with bacto peptone, Yeast and tryptone which are known as the commercial organic nitrogen sources, it was evident that CFP is the best one.

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