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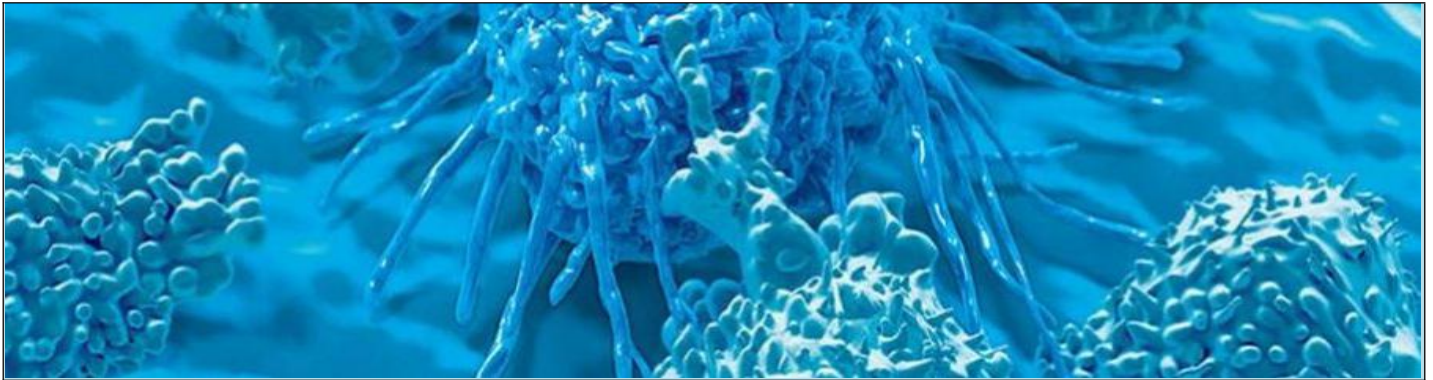
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## An indigenous substitute for peptone as the basic ingredient of culture media

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### NAME OF THE AUTHOR'S

<sup>1</sup>Raghavendra Rao M.V\*,  
<sup>2</sup>Raja Rajeswari M,  
<sup>3</sup>Vijay Kumar Chennamchetty,  
<sup>4</sup>Dilip Mathai,  
<sup>5</sup>Abrar A Khan,  
<sup>6</sup>Surekha Bhat,  
<sup>7</sup>Frank Michael Navarrete,  
<sup>8</sup>Tiara Calvo Leon,  
<sup>9</sup>Abigail Apacible,  
<sup>10</sup>Mahendra Kumar Verma

<sup>1</sup>Scientist-Emeritus and Director, Central Research Laboratory, Apollo Institute of Medical Sciences and Research, Jubilee Hills, Hyderabad, Telangana, India

<sup>2</sup>Former Professor and Head , Department of Microbiology, Guntur Medical college, Guntur, AP, India

<sup>3</sup>Assistant Professor, Department of Pulmonology, Apollo Institute of Medical Sciences and Research, Jubilee Hills, Hyderabad, Telangana, India

<sup>4</sup>Professor, Department of Medicine, Dean, Apollo Institute of Medical Sciences and Research, Jubilee Hills, Hyderabad, Telangana, India

<sup>5</sup>Professor, Dean, American University School of Medicine Aruba, Central America

<sup>6</sup>Professor, Department of Biochemistry and Genetics, American University School of Medicine Aruba, Central America

<sup>7</sup>Associate professor, American University School of Medicine Aruba, Central America

<sup>8</sup>Associate Professor, Department of Pharmacology, American University School of Medicine Aruba, Central America

<sup>9</sup>Associate Professor of Behavioral Science and Histology, American University School of Medicine Aruba, Central America

<sup>10</sup>Scientist, Centre for Molecular Biology Research, Bhopal, Madhya Pradesh, India.

## ABSTRACT

From times immemorial, peptone, a semi digested protein is being used as a basic ingredient in microbiological culture media throughout the globe. Peptone hold up and broadening the growth of bacteria from small inoculate, free from ferment -able carbohydrates, has a very low content of contaminating bacteria and a very low content of copper. Keeping in view the above criteria, every effort was made to find a suitable and cost effective substitute for peptone. The use of solid media, dates back 1884, on the advice of Frau Hesse to Robert Koch who used agar as coagulate and consolidate liaison in culture media. This made him the pioneer in isolation of pure cultures. Since a long time peptones are the basic ingredients of culture media. They are the breakdown products of proteins of animal origin. The cost of 500 gm of Peptone is Rs 1999/- Because their high nutritive value they proved to be good as cattle feeds. At the same time they are very cheap. The vegetable proteins obtained from these cakes of oil seeds are comparable to peptone in their composition and the cost is only 1/100 the cost of peptone or even less. Keeping this in view an attempt is made to substitute peptone with the powders obtained from the cakes of oil seeds like coconut, groundnut and gingelly and a preliminary report on this work is presented here.

**KEYWORDS:** Sesame oil meal (SME), Coconut oil meal (CME), Sidirophores, Escherichia coli, Salmonella, Shigella and Klebsiella.

## CITATION OF THE ARTICLE



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\* Corresponding Author

## I. INTRODUCTION

Deoiled cakes of oil seeds are known for their nutritive value. Oil seeds like groundnut, Coconut and gingly have been analyzed and their constituents are reported. These cakes are quit rich in proteins, essential amino acids, vitamins and minerals as reported by the above workers. [1] In de oiled cakes protein is present. These cakes are used as feed for poultry. [2] Groundnut flour enhances or enriches the nutritive value of wheat and other flour and used in bakery [3] Protein in the majority of formulated fish diets (worldwide) depends greatly on fishmeal which is more costly than high quality plant-based protein sources, such as soybeans. [4] The shelf life of vanilla cake is improved by the phenolic extracts of sesame oil meal (SME) and coconut oil meal (CME) [5] Lysine and methionine amino acids are absent in Ground not cake, but it is an alternate protein source in aquaculture [6] The fingerlings of fish were fed with de oiled ground nut cake which has improved growth. [7] The purpose of a general laboratory medium should support the growth of microorganisms and should contain all microbial nutrients [8] Microorganisms observed microscopically to be growing in a natural environment may prove exceedingly difficult to grow in pure culture in an artificial medium [9] Iron is required for humans, as well as the growth of bacteria. To obtain free iron, some pathogens secrete proteins called sidirophores which take the iron away from iron transport proteins by binding to the iron even more tightly. Once the iron

sidirophore complex is formed, it is taken up by receptors on the bacterial surphace and finally brought into the bacterium [10] A growth medium or culture medium is a solid or liquid containing nutrients.

It is designed to support the growth of microorganisms or cells or small plants like moss. There are different types of media for growing different types of cells. [11] Enrichment culture is the use of certain growth media to favor the growth of a particular microorganism over others [12]. An ideal culture should contain required ingredients for growth of bacteria. [13] Fat soluble antioxidants are present in sesame seeds. They are sesamin, sesamol, sesamolol and tocopherols. They are helpful against oxidative processes in cell [14]. The groundnut cake composition mixture (10%) comprised of protein 45%, fibre 15%, fat and oil 7% and Sand and silica 2%. Coconut cake composition as dry matter 91.6%, crude protein 20.5%, crude fibre 12.9%, Crude fat 9.2%. The gingly cake composition; Hull Moisture 5.4%, protein content 19.80%, crude fibre 3.20, mineral matter 4.8%, calcium 1.06% and phosphorus 0.47%.

In India, the cheaper protein source like groundnut cake and fish meal are used as major protein sources in poultry feed formulation. [15] compared with that of other nuts it is found that every 100 grams of ground nut contain more plant protein than any other legumes or nuts [16] Because of its high



nutritive content, peanut cake in Nigeria is prone to contamination by a wide variety of microorganisms [17] Ground nut cake is contaminated by enterotoxigenic pathogens such as *Escherichia coli*, *Salmonella*, *Shigella* and *Klebsiella*, and multidrug resistant (MDR) strains of these organisms [18] Among the plant protein sources, oil cakes are considered as good protein sources for the diet and are available in large quantity as by-products of the edible oil industry. [19] *Klebsiella oxytoca*, *Staphylococcus aureus*, *Bacillus cereus*, *E. coli*, *P. aeruginosa* and *Streptococcus faecalis* bacteriological isolates were found in groundnut cake powder [20]

## II. MATERIAL AND METHODS

Coconut cake, Gingelly cake and ground nut cake procured from the market dried and finely ground. The fine powders are used as a substitute for peptone. The medium was prepared in a manner exactly similar to that of the routinely used basal medium like Nutrient agar with 1% peptone, substituting peptone powder with an equal amount of powders of the three cakes separately. With the same base 1. Enriched medium blood agar with 5% human blood. 11. Differential medium ie MacConkey agar and 111. Selective media, Deoxy citrate agar for *Salmonella* and *Shigella* Sp and Thiosulphate citrate, Bile sucrose Agar for *Vibrio cholerae* were also prepared. The basal medium was tested for isolation of common pathogens viz *Staphylococcus aureus* among gram positive cocci, *Klebsiella pneumoniae*, *Proteus mirabilis* and *Pseudomonas aeruginosa* among gram negative bacilli. Growth of specific pathogens like *Salmonella* spp *Shigella* spp and *Vibrio cholerae* was also tested on basal medium. The enriched medium, differential medium and selective medium were also inoculated with respective organisms. Parameters like growth, morphology, motility and colony count were studied. Single colonies from the from the growth were inoculated in to respective broths and incubated and are studied for growth and mobility. Single colonies from all plates were inoculated separately in to peptone water broth and incubated and biochemical profile of the organisms was studied. the basal medium is tested for its efficacy as a base for antibiotic sensitivity testing.

## III. RESULTS

The results were promising and we report here following including the growth. Growth of the organisms on all the media is good. For all the common isolates preserving their morphological and biochemical characters. 111. *Pseudomonas* gave good pigmentation 1V. *proteus* showed good swarming V. Blood agar gave good growth and hemolysis is seen well. V1. Lactose and non-Lactose fermentors are well differentiated on MacConkey agar. V11. *Salmonella typhi* and *Shigella* have grown well on DCA and also

*Vibrio cholera* on TCBS giving yellow colonies V111. Growth was good in all broths. 1X. The media were as effective for Antibiotic sensitivity testing as was on Nutrient agar. Standardization test revealed that the media prepared with substituent were on par with Nutrient agar at any given concentration. While Nutrient agar failed to give growth at 15% and 20% concentration, these media yielded a growth. X. When quantitative cultures were done, colony count revealed that the the count is more in other media even at as a high dilution as 1:10,000 compared to Nutrient agar X1. Chromatography study of all these powders along with peptone revealed that the amino acids present in all of them are almost similar.

## IV. DISCUSSION

From times immemorial peptone, a semi digested protein is being used as basal ingredient in microbiological culture media all over the globe. Peptone which is known to support the growth of moderately exacting bacteria from small inocula free from ferment able carbohydrates, has a very low content of contaminating bacteria and very low content of copper. [21] The report on analysis of the cake powders used in present study proved themselves as good as peptone. The growth of all organisms on the media prepared using the cake powders is on par with that on nutrient agar. Standardization test showed that the media prepared with the substitutes are as efficacious as nutrient agar in all concentrations ranging from 0.2 to 10 %.Nutrient agar at a concentration of 15% and 20% failed to give growth where as the other media are effective in these concentrations. Colony count is also more even in higher dilution when compared to Nutrient agar. This shows that these media are better sources of available nutrients for the growth of organisms. The greatest advantage of these media is their cost effectiveness which is incomparable as shown in the table given below:

Sl. No.	Ingredients	Quantity (gm)	Cost (INR)
1.	Peptone	500	1999.025
2.	Coconut cake	500	990.03
3.	Ground nut cake	500	990.04
4.	Gingelly cake	500	99.00

As such the present expenditure on the use of peptone is reduced 100 fold or even more there by drastically reducing the cost of media preparation at institution level, state level, and national level resulting in net savings of cores of rupees of national exchequer.

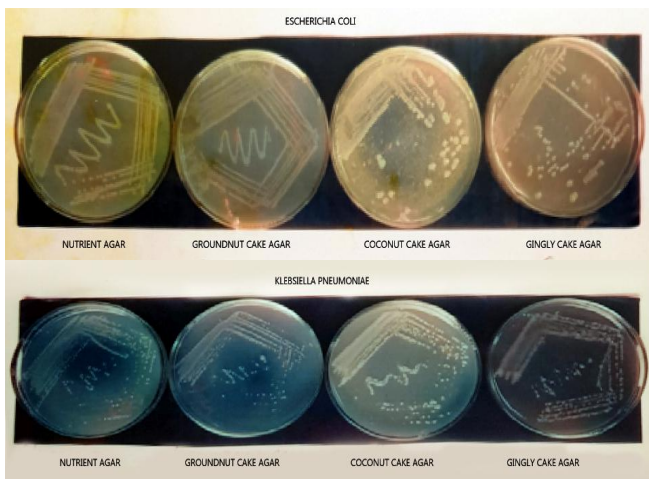


Fig.1



Fig.2

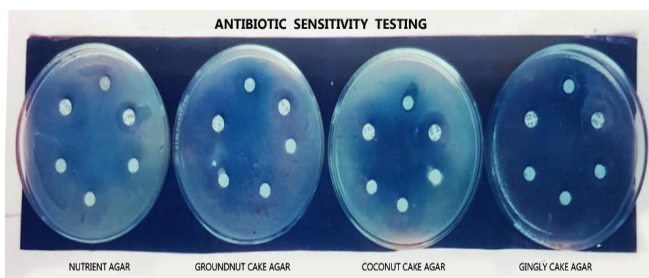


Fig.3



Fig.4

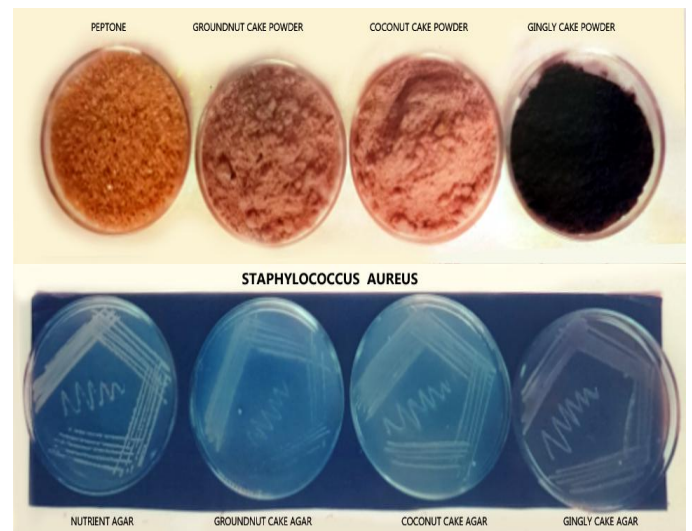


Fig.5

## V. CONCLUSION

From this we can conclude that because of their

- 1) Easy availability,
- 2) Very low cost and
- 3) Equal efficacy,

The use of these indigenous substances in culture media in place of peptone will save us a lot of expenditure.

## VI. SUMMARY

Failure of groundnut crop and susceptibility of groundnut cake to aflatoxin are the limiting factors whereas wide variation in quality due to adulteration in fish meal has initiated search for other potential protein sources. Amongst the other protein sources Soybean meal has emerged as the most promising one because of its better protein quality and fairly consistent nutrient content.

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