

## Short Communication

# Production of peptone from soya beans (*Glycine max* L merr) and African locust beans (*Parkia biglobosa*)

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Peptone was produced from soya beans and African locust beans. The produced peptones were evaluated as component of microbiological media for the growth of some bacteria and compared with some commercial peptones. Some of the tested bacteria are *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Bacillus subtilis*. The soya beans and African locust beans peptones supported the growth of all the tested bacteria favourably well when compared to the commercial peptones. From the peptone broth cultures at absorbance 540 nm, the best growth was obtained from *Escherichia coli* in laboratory-produced soya beans peptone and *Staphylococcus aureus* in Bacto peptone (a commercial peptone). This may reduce the cost of peptone in Nigeria.

**Key words:** African locust beans, peptone, soya beans.

## INTRODUCTION

The nitrogen requirements of bacteria grown by artificial means, were first met by the addition to media of such naturally occurring substances as blood, urine, and other body fluids. Naegeli (1880) was probably the first to use egg albumin, which he called "peptone". However, it was later discovered that peptones, obtained by partial digestion of proteins, furnished organic nitrogen in a more readily available forms. Fish peptones have been developed from red hake (*Urophycis cluss*) and from fishery by-product for microbial media (Green et al., 1997). The use of legumes as sources of protein hydrolysates in microbial culture media is limited, and thus presents a challenge to explore this area for the production of comparatively standard culture media. These legume peptones could serve as inexpensive peptones for microbial culture and might prove to be advantageous in microbial growth or product formation by some species of microorganism.

The objectives of this research therefore are to produce peptone from soya beans and African locust beans, and to evaluate the suitability of the produced peptone as component of microbiological media.

## MATERIALS AND METHODS

### Proximate analysis of soyabeans and African locust beans

Samples of soyabeans and African locust beans were analyzed for protein, fat and moisture. The Microkjedhal estimation of nitrogen using a conversion factor of % N x 6.25 (AOAC, 1980) was used to determine total proteins. Fat was determined by using standard method (Bligh and Dyer, 1959). Moisture was measured after drying the samples at 100°C for 4-6 h.

### Peptone production

The soyabeans and African locust beans were sorted to remove stones, oven-dried at 50°C, dehulled, and milled to powder form. This was defatted with soxhlet extractor using hexane as the extracting solvent. For the hydrolysis, 40 g of defatted soyabeans was weighed and suspended in 200 ml of distilled water. This was

**Table 1.** Proximate composition of soyabeans and African locust beans.

| Parameters    | Soyabeans (%) | African Locust beans (%) |
|---------------|---------------|--------------------------|
| Protein       | 47.73         | 30.69                    |
| Moisture      | 4.83          | 3.4                      |
| Ether extract | 17.72         | 23.03                    |

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**Table 2.** Comparison of growth of some bacteria in different peptone broths (absorbance at 540 nm).

| Test bacteria                 | Laboratory produced peptones |                              | Commercial peptones |                         |               |
|-------------------------------|------------------------------|------------------------------|---------------------|-------------------------|---------------|
|                               | Soyabeans peptone            | African locust beans peptone | Soy peptone         | Bacteriological peptone | Bacto peptone |
| <i>Escherichia coli</i>       | 0.92                         | 0.63                         | 0.69                | 0.63                    | 0.66          |
| <i>Bacillus subtilis</i>      | 0.11                         | 0.79                         | 0.81                | 0.72                    | 0.81          |
| <i>Staphylococcus aureus</i>  | 0.09                         | 0.91                         | 0.24                | 0.84                    | 0.92          |
| <i>Streptococcus pyogenes</i> | 0.81                         | 0.68                         | 0.72                | 0.53                    | 0.62          |
| <i>Pseudomonas aeruginosa</i> | 0.11                         | 0.64                         | 0.56                | 0.42                    | 0.59          |
| <i>Salmonella sp</i>          | 0.16                         | 0.58                         | 0.57                | 0.46                    | 0.72          |
| <i>Micrococcus luteus</i>     | 0.18                         | 0.27                         | 0.54                | 0.67                    | 0.30          |
| <i>Flavobacterium rigense</i> | 0.12                         | 0.40                         | 0.63                | 0.38                    | 0.43          |
| <i>Streptococcus faecalis</i> | 0.12                         | 0.47                         | 0.81                | 0.53                    | 0.45          |
| <i>Klebsiella aerogenes</i>   | 0.18                         | 0.40                         | 0.72                | 0.69                    | 0.59          |
| <i>Enterobacter aerogenes</i> | 0.1                          | 0.53                         | 0.63                | 0.73                    | 0.70          |
| <i>Alcaligenes sp</i>         | 0.07                         | 0.28                         | 0.56                | 0.49                    | 0.36          |
| <i>Acinetobacter sp</i>       | 0.09                         | 0.39                         | 0.71                | 0.51                    | 0.48          |

**Table 3.** Comparison of growth of some bacteria on different peptone agar.

| Test bacteria                 | Laboratory      |                   |                              | Commercial peptones |             |
|-------------------------------|-----------------|-------------------|------------------------------|---------------------|-------------|
|                               | Dilution Factor | Produced peptones |                              | Bacto peptone       | Soy peptone |
|                               |                 | Soyabeans peptone | African Locust beans peptone |                     |             |
| <i>Escherichia coli</i>       | 10 <sup>5</sup> | 237*              | 271                          | 226                 | 241         |
| <i>Pseudomonas aeruginosa</i> | 10 <sup>5</sup> | 256               | 260                          | 277                 | 253         |
| <i>Bacillus subtilis</i>      | 10 <sup>5</sup> | 178               | 187                          | 261                 | 141         |
| <i>Streptococcus faecalis</i> | 10 <sup>5</sup> | 49                | 231                          | 80                  | 115         |
| <i>Staphylococcus aureus</i>  | 10 <sup>5</sup> | 264               | 279                          | 239                 | 231         |

\*Bacterial counts.

stirred to form slurry of the protein material. The pH of the protein slurry was adjusted to pH 6.5 with 1 N NaOH. At optimum temperature of the digest (60°C) 0.25% of commercial papain was added. Digestion was allowed to take place for 3 h and the pH was controlled using 1 N NaOH. The temperature was then raised to 80°C for 5 min to inactivate the enzyme. Liquor from the hydrolysed soyabeans was separated from the protein/enzyme slurry by repeated centrifugation. The peptone solution was then freeze-dried to obtain soya peptone. This process was also carried out in the case of African locust beans to obtain African locust beans peptone.

#### Evaluation of laboratory produced peptones as culture medium for bacterial growth

The laboratory produced soya and African locust beans peptones were prepared and used for bacterial growth. Peptone broth media was prepared by the addition of 0.5% peptone, 0.1% dextrose to distilled water, and the pH adjusted to 7.0 with 1 N NaOH. Each medium was dispensed in 10 ml aliquots into MacCartney bottles and autoclaved at 121°C for 15min. Peptone agar media were prepared as above and supplemented with 1% agar.

*Bacillus subtilis*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Streptococcus faecalis* and *Staphylococcus aureus* were obtained from our laboratory collection. Each was cultured in nutrient broth for 18 h and then 1 ml was serially diluted in sterile distilled water. From the 10<sup>-4</sup> dilution, 0.1 ml was used to inoculate peptone broth

and peptone agar each in duplicate. This was followed by incubation at 37°C for 24 h. Growth turbidity was measured from the broth culture with spectrophotometer at 540 nm. Uninoculated peptone broth was used as control. While from the peptone agar, developed colonies were counted.

## RESULTS AND DISCUSSION

The proximate composition of soya beans and African locust beans is shown in Table 1 with protein contents of 47.73 and 30.69%, respectively. With the level of moisture, there was need for drying before milling. Values obtained for their ether extract level made it necessary to defat the substrates prior to peptone production, because many microorganisms do not have the ability to breakdown fat in their culture medium. Soya beans and African locust beans are very much available in Nigeria and their protein content makes them suitable substrates for the production of peptone in Nigeria.

When the laboratory produced soya beans and African locust beans peptones were evaluated as culture media for bacterial growth, they supported the growth of the test organisms and this compared favourably well with growth

obtained from the commercial peptones (Tables 2 and 3). This means that the produced peptones contain the necessary nutrients required for the bacterial growth, especially in the area of nitrogen requirement. Green et al. (1997) also obtained growth of their test organisms when they produced peptones from soluble fish extract and soluble hake autolysate. Peptones have been known to serve as either culture media on their own or as nitrogen component in other culture media. In the peptone broth cultures at absorbance 540 nm, the best growth was obtained from *E. coli* in soya beans peptone and *S aureus* in Bacto peptone with each having turbidity value of 0.92.

The test bacteria however grew better on the African locust beans peptone than the soya beans peptone. We recommend the commercialization of both peptones because of the impressive way they supported the growth

of the test bacteria. Production of these peptones in Nigeria may help to reduce cost of peptones which at the moment are being imported into the country and sold at a very high rate.

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