Journal of Biology and Today's World
ISSN 2322-3308
http://www.journalbio.com

Received: 02 November 2012 • Accepted: 29 November 2012

Research

doi:10.15412/J.JBTW. 01020101

Evaluation of Locally Sourced Raw Materials for use as A Microbiological Media

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ABSTRACT

Local prepared nutrient media made from guinea corn and maize were used to culture bacteria such *Staphylococcus aureus*, *Klebsiella*sp, *Salmonella typhi*, *Escherichia coli*, and *Pseudomonas aeruginosa* and compared with the growth on commercial nutrient agar and broth. Bacteria growth was higher in the commercial nutrient agar and broth but with little significant difference. Locally prepared potato dextrose agar was also used to culture fungal isolates such as *Aspergillusniger*, *Aspergillusfumigatus*, *Aspergillusflavus*, *Cephalosporium*sp, and *Mucor*sp and compared with commercial made potato dextrose agar using mycelia extension technique. Fungal growth was higher in commercially prepared potato dextrose agar with little difference. The results of these studies suggest that these locally sourced materials could be used for media preparation in place of commercially produced media.

Key words: Microbiological Media, Locally prepared Nutrient Media, Bacteria, Fungi

1. INTRODUCTION

A medium is solid or a liquid preparation containing materials for the culture (growth) of microorganisms, animal cells or plant tissue cultures (1). To culture organisms in laboratory, it requires the preparation of substances which can be used as food. Such nutrient preparations which microorganisms can use as food are called culture media. Different microorganisms require different nutrient materials. Thus, culture media vary in form and in composition depending on the species to be cultivated (2). A medium may be formulated as either permissive with the intent of allowing the growth of whatever organisms are present, or restricted or selective with the intent of only selecting for growth only a particular subset of those organism (2). This may take the form of a nutritional requirement, for instance providing a particular component such as lactose as the only source of carbon for energy including a particular antibiotics or other substances in order to select only organisms which are resistant to that substances. This correlates to some degree with defined and undefined media are made from natural product and containing an unknown combination of very many organic molecules, while defined media can be precisely tailored to select organisms with very properties (3). Such media are very useful as a simple complex medium may be sufficiently rich to completely meet the nutritional requirement of many different microorganisms. In addition, complex media often are needed because the nutritional requirements of a particular microorganism are unknown and thus, a defined medium cannot be constructed. This is the situation with many fastidious bacteria that have complex nutritional or cultural requirement; they may even require a medium containing blood serum (4). Complex media contain undefined component like peptone, meat extract and yeast extract. Peptones are protein hydrolyzate prepared by partial proteolytic digestion of meat, casein, soy malt, gelatin and other protein sources. They serve as source of carbon, energy and nitrogen. In composition, peptones are mixtures containing only partly known concentration and identify, a variety of peptides and polypeptides, protease, amino acids and carbohydrate including inorganic and many organic micronutrients. Peptone provides insoluble and assimilable form of all the phosphorous and sulfur and essential mineral content of living materials as well as the organic carbon and nitrogen source [4]. Beef extract contains

amino acid, peptides, nucleotides, organic acids, vitamins and minerals. Among meat extract and infusions beef extract and aqueous meat infusion made by infusing (soaking) fresh ground meat in water are common ingredient of media useful for many species. Beef infusions are rich in mineral, organic micronutrient, proteins, protein derivatives and carbohydrates. They are often supplemented with 1% peptone. Culture media fluids made from beef extract as extract broth. All such nutritional media including simple solutions of peptone are sometimes loosely included in the general terms nutrient broth (5).

2. MATERIALS AND METHODS

2.1. Collection of Samples

Maize, guinea corn and Irish potato was obtained from a retail outlet in Bosso area of Minna, Nigeria and was transported to the Microbiology laboratory of federal university of Technology, Minna, Nigeria. Meat (beef) was obtained from Minna Abattoir along western bypass and was equally transported to the laboratory.

2.2. Processing of Materials

Two hundred grams of maize and guinea corn was washed with distilled water and manually grinded using a clean grinder. It was then sieved using sterile filter cloth into 500ml beaker and covered with aluminum foil. The meat (beef) was boiled for 30 minutes and the extract was transferred into a sterile beaker and covered with aluminum foil. Two hundred ml of guinea corn and maize extract each was mixed with 100ml of meat extract, 20g of agar-agar, 5g of sodium chloride and the medium was sterilized at 121^{o}C for 15 minutes.Two hundred grams of Irish potato was boiled with water for 30 minutes, and the extract is allowed to cool and transferred to a 500ml beaker and was covered with aluminum foil. 20g of agar-agar, 10g of glucose was dissolved in a 500ml beaker containing Irish potato extract and 0.2g of chloramphenicol powder was added to inhibit the growth of bacteria. Each of the mixture was sterilized using a validated autoclave at a temperature of 121^{o}C for 15 minute and it was dispensed into petri-dish and labeled accordingly.

2.3. Inoculation of Organisms

Pure isolates of each of *Staphylococcus aureus, Klebsiellasp, Salmonellatyphi, Escherichia coli* and *Pseudomonas aeruginosa* was sub-cultured into commercially prepared nutrient media and locally prepared medium using a pour plate technique. The plate was incubated at 37^{o}C for 24 hours and the growth rate in the commercial nutrient agar was compared to the locally prepared media using a colony counter to count the colonies that was formed. Also, 5mm of pure fungal isolates of each of *Aspergillusniger, Aspergillus fumigates, Aspergillusflavus, Cephalosporim*sp, and *mucor*sp was also sub cultured into locally prepared potato dextrose agar and commercially prepared potato dextrose agar. The plates were incubated at room temperature, and the growth was examined using radial extension method at 48 and 72 hours.

3. RESULTS AND DISCUSSION

3.1. Bacterial count on locally prepared and commercially prepared nutrient agar

Table 1 show that the counts of some selected bacteria i.e *Staphylococcusaureus, Klebsiella*sp, *Escharichia coli, Salmonella typhi* and *Pseudomonasaeruginosa* on locally prepared nutrient agar using guinea corn and meat extract. *Escherichia coli* had the highest microbial count on the guinea corn agar with a mean growth of 7.1 + 0.1 when compared with the maize agar, followed by *Klebsiella*sp with a mean growth of 4.2 + 0.3 on guinea corn agar. *Staphylococcus aureus* had the lowest count with the mean growth of 2.05 + 0.01. Commercial prepared nutrient agar had the highest microbial count when compared with the guinea corn and maize agar.

Table 1. Count (cfu/ml) of some selected bacteria on locally prepared nutrient agar using guinea corn and meat extract

Organism	*GCA (x105cfu/ml)*	*MA (x105cfu/ml)*	*CPN (x105cfu/ml)*
Staphylococcus aureus	1.005 + 0.01	1.05 + 0.05	2.15 + 0.05
*Klebsiella*sp	4.20 + 0.30	4.15 + 0.15	7.20 + 0.10
Escherichia coli	7.10 + 0.10	4.95 + 0.05	2.30 + 0.16
Salmonella typhi	3.55 + 0.55	2.60 + 0.10	1.35 + 0.05
Pseudomonas aeruginosa	1.06 + 0.04	1.10 + 0.10	1.05 + 0.05

Key:GCA –Guinea corn agar; MA – Maize agar**;** CPNA – Commercial prepared nutrient agar

3.2. Bacterial count on locally prepared and commercially prepared nutrient broth

Table 2 shows the count of some selected bacteria *Staphylococcus aureus, Klebsiella*sp, *Escharichia coli, Salmonella typhi* and *Pseudomonasaeruginosa* on locally prepared nutrient broth using maize extract and meat extract *Pseudomonas aeruginosa* had the highest microbial count on the guinea corn broth with a mean growth of 8.10 + 0.10 when compared with the maize broth followed by *Klebsiella*sp with a mean growth of 5.05 + 0.05 on guinea corn broth. *Escharichiacoli* had the lowest count with the mean growth of 3.10 + 0.10. Commercially prepared nutrient broth had the highest microbial count when compared with the guinea corn and Mize agar.

Table 2. Count (cfu/ml) of some selected bacteria on locally prepared nutrient broth using guinea corn and maize extract

Organism	*GCA (x10^{5}cfu/ml)*	*MA (x10^{5}cfu/ml)*	*CPN(x10^{5}cfu/ml)*
Staphylococcus aureus	4.05 + 0.05	4.10 + 0.10	2.15 + 0.05
*Klebsiella*sp	5.05 + 0.05	1.95 + 0.05	1.15 + 0.05
Escherichia coli	3.10 + 0.10	3.10 + 0.10	8.30 + 0.10
Salmonella typhi	4.35 + 0.15	2.10 + 0.10	1.90 + 0.10
Pseudomonas aeruginosa	8.10 + 0.04	6.05 + 0.05	2.10 + 0.70

Key: GCA –Guinea corn agar;MA – Maize agar**;** CPNA – Commercial prepared nutrient agar

Table 3. Fungal growth (mycelia extension) on locally prepared potato dextrose agar and commercially prepared potato dextrose agar after 48 and 72 hours of incubation

Organism	48 hours (mm)		72 hours (mm)	
	LPDA	CPDA	LPDA	CPDA
Aspergillusniger	19.70 + 0.70	30.30 + 0.20	23.50 + 1.50	44.00 + 1.00
Aspergillusfumigatus	20.50 + 0.50	20.50 + 0.50	25.00 + 0.70	27.00 + 1.00
Aspergillusflavus	19.00 + 1.00	22.00 +1.00	25.50 + 0.5	34.00 + 1.00
*Cephalosporium*sp	42.50 + 2.50	73.50 + 1.50	63.00 +2.00	90.50 + 0.60
*Mucor*sp	51.00 + 1.50	77.00 + 1.00	81.50 + 1.50	85.00 + 2.20

Key: LPDA –Locally prepared potato dextrose agar; CPDA – Commercially prepared potato dextrose agar

Microbiological media provide an artificial means for the growth of microorganisms because they contain some essential nutrients that could enhance their growth and metabolism (6). When locally prepared nutrient agar using guinea corn and maize was compared with commercial prepared nutrient agar, there was a higher microbial count in the commercially prepared nutrient agar, there was a higher microbial count in the commercially prepared nutrient agar than the locally prepared nutrient agar but with a little significant difference (Table 1). The high microbial count in commercially prepared nutrient agar and broth is as a result of presence of peptone. This is in line with Nagel and Kulz, 1973 who stated that the "simplest way of ensuring adequate supply of nitrogen source is by addition of peptone which is a hydrolyzed product of protein and consists of a mixture of protease, polypeptide and amino acid". However, there was a higher microbial count in guinea corn agar when compared with maize agar. This might be as a result of higher nutritional content when compared with maize extract. When the organism were grown in the locally prepared broth using guinea corn and maize as energy source, there was a higher growth in guinea corn broth than in the maize broth (Table 2). There is a higher microbial growth of the test organism in the guinea corn broth compared with guinea corn agar with *Pseudomonas aeruginosa* having the highest count (8.0 x 10^5 cfu/ml) in the guinea corn broth and *E. coli* having highest count (7.0 x 10^5 cfu/ml) in the guinea corn agar (Table 1 and Table 2). Also, there is a higher count of the test organism in the maize broth than in the maize agar with *P. aeroginosa* having a count of (5.0 x 10^5 cfu/ml) in the maize broth and *E. coli* having a count of (5.0 x 10^5 cfu/ml) in the maize agar. This suggests that the organism grows mush in a liquid media compared to solid media. The result in Table 3 shows the growth of various fungi in the commercially prepared potato dextrose agar and locally prepared potato dextrose agar after some period of incubation. There is a higher radial growth (mycelia extension) in the commercial prepared potato dextrose agar when compared to locally prepared potato dextrose agar but with a little significant difference. The result of this research work suggest that locally prepared media from local subtract such as guinea corn, maize and potato could be used in the absence of commercial prepared nutrient media and pota dextrose agar.

4. CONCLUSION

The growth of various bacterial cultivated on commercially prepared nutrient agar and locally prepared nutrient agar and also broth were compared and there were no much significant difference. In addition, when locally prepared potato dextrose agar (PDA), were compared with commercial one, locally prepared PDA also gave a better growth (mycelia extension method or redia growth method) at various incubation temperature examined.

5. RECOMMENDATION

Based on the above result of findings, the following recommendations are made;

*The preparation of media from natural substrate should be encouraged.

*School authority and government should also encourage the use of locally prepared media for microbiological use.

6. ATTACHMENT

Appendices: Enumeration of microorganisms using the various media prepared from locally sourced Raw Materials.

Appendix I Counts (cfu/ml) of some selected bacteria on locally prepared nutrient agar using guinea corn and meat extract

Organism	*Counts (cfu/ml)*
Staphylococcus aureus	1.01 x 10^5
*Klebsiella*sp	4.50 x 10^4
Escherichia coli	7.00 x 10^4
Salmonella typhi	3.00 x 10^4
Pseudomonas aeruginosa	3.00 x 10^4

Appendix II Counts (cfu/ml) of some selected bacteria on locally prepared nutrient agar using maize and meat extract

Organism	*Counts (cfu/ml)*
Staphylococcus aureus	1.10 x 10^5
*Klebsiella*sp	4.00 x 10^4
Escherichia coli	5.00 x 10^4
Salmonella typhi	4.50 x 10^4
Pseudomonas aeruginosa	9.40 x 10^4

Appendix III Counts (cfu/ml) of some selected bacteria on commercially prepared nutrient agar

Organism	*Counts (cfu/ml)*
Staphylococcus aureus	2.10 x 10^5
*Klebsiella*sp	7.10 x 10^4
Escherichia coli	2.50 x 10^5
Salmonella typhi	1.30 x 10^5
Pseudomonas aeruginosa	1.00 x 10^5

Appendix IV Counts (cfu/ml) of some selected bacteria on locally prepared nutrient broth using guinea corn and meat extract

Organism	*Counts (cfu/ml)*
Staphylococcus aureus	1.00x 10^5
*Klebsiella*sp	5.10 x 10^4
Escherichia coli	3.00 x 10^4
Salmonella typhi	4.50 x 10^4
Pseudomonas aeruginosa	8.00 x 10^4

Appendix V Counts (cfu/ml) of some selected bacteria on locally prepared nutrient agar using maize and meat extract

Organism	*Counts (cfu/ml)*
Staphylococcus aureus	8.00 x 10^4
*Klebsiella*sp	2.00 x 10^4
Escherichia coli	3.00 x 10^4
Salmonella typhi	2.00 x 10^4
Pseudomonas aeruginosa	5.00 x 10^4

Appendix VI Counts (cfu/ml) of some selected bacteria on commercially prepared nutrient broth

Organism	*Counts (cfu/ml)*
Staphylococcus aureus	2.00 x 10^5
*Klebsiella*sp	1.10 x 10^5
Escherichia coli	8.20 x 10^4
Salmonella typhi	1.80 x 10^5
Pseudomonas aeruginosa	1.50 x 10^5

ACKNOWLEDGMENT

No mentioned any acknowledgment by authors.

AUTHORS CONTRIBUTION

This work was carried out in collaboration between all authors.

CONFLICT OF INTEREST

The authors declared no potential conflicts of interests with respect to the authorship and/or publication of this article.

REFERENCES

1. Talaro K. Foundations in Microbiology 5th editions. Boston Mc GrawHill. 2005:510-1.
2. Ryan KJ, Ray CG. Medical microbiology. Appleton & Lange, Norwalk, Connecticut. 1994.
3. Madigan MT, Martinko JM, Parker J, Brock TD. Biology of microorganisms: prentice hall Upper Saddle River, NJ; 1997.
4. Prescott LM, Harley J, Klein D. Microbiology, 6th. McGrawJHill Higher Education. 2005.
5. Omer E-FE-O. Review of medical bacteriology1990.
6. ASPINALL LJ, Kilsby D. A microbiological quality control procedure based on tube counts. Journal of Applied Microbiology. 1979;46(2):325-9.