Exploration of Fungi Growth on Media Formulated from Agro-allied Wastes

Umedum, C.U.*, Enejekwute, N.P.

Department of Microbiology, Chukwuemeka Odumegwu Ojukwu University, Anambra State, Nigeria

*Corresponding author’s email: chyemmy2000@yahoo.com

Abstract
This study was carried out to assess the suitability of some agro-allied wastes in media formulation for fungi cultivation. Sweet potato peel glucose broth (SPPGB), Soybean hull glucose broth (SBHGB) and sweet potato peel and soybean hull glucose broth (SPPSBHGB) were prepared and used in comparison to a well-known commercial fungi growth medium (Sabouraud Dextrose Broth (SDB)). Four fungi species obtained from a previous study on isolation of fungi from throats of boiler chicken were grown on the formulated media at room temperature. These included: Aspergillus niger, Aspergillus fumigatus, Aspergillus flavus and Trichoderma viride. The extent of their growth was determined by measuring the absorbance of the formulated media containing the growing fungi using a spectrophotometer with a wavelength of 540nm and was compared with their growth on Sabouraud Dextrose Broth (SDB) as positive control and water as the negative control. The proximate compositions of the potato peels and soybean hulls were determined using the standard methods. Aspergillus niger, Aspergillus fumigatus, Aspergillus flavus and Trichoderma viride grew on the formulated media within four days at room temperature. The availability and low cost of these agro-allied wastes makes them a good substitute for already made media which are not readily available and very expensive. The result of this research work will go a long way in solving this problem.

Keywords: Agro-wastes; Media; Proximate analysis; Fungi growth.

Introduction

Large amount of wastes are generated every year from the industrial processing of agricultural raw materials and individual homes (Chuang et al., 1996). Most of these wastes are used as animal feed or burned as alternative for elimination. However, such wastes usually have a composition rich in sugars, minerals and proteins, and therefore, making them useful for other processes directly or indirectly. The presence of carbon sources, nutrients and moisture in these wastes provides conditions suitable for the development of microorganisms and this open up great possibilities for their reuse. The economical aspect is based on the fact that such wastes may be used as low-cost raw materials for the production of other value-added compounds, with the expectancy of reducing production costs (Chuang et al., 1996).

The environmental concern is because most of the agro-industrial wastes contains phenolic compounds and/or other compounds of toxic potential; which may cause deterioration of the environment when the waste is discharged to the nature (Chuang et al., 1996). Wastes are materials that have not yet been fully utilized. They are leftovers from production and consumption. However, waste is an expensive and sometimes unavoidable result of human activity. It includes plant materials, agricultural, household, industrial and municipal wastes and residues (Okonkwo et al., 2006). The agricultural-based industries generate significant quantities of organic wastes including plants from cassava, plantain, banana, oranges and straw from cereals. Rather than allow these wastes to become solid municipal wastes, it is necessary to convert them to useful end products. It is now realized that these waste could be utilized as cheap raw materials for some industries or as cheap substrates for microbiological processes (Nwabueze and Otowa, 2006).

Fungi constitute one of the largest groups of plants with richest arrays of species. They are a group of eukaryotic spore bearing, achlorophyllous organisms that generally reproduce asexually and sexually (Pelczar et al., 1993). Fungi grow on diverse habitat in nature and are cosmopolitan, requiring...
several specific elements for growth and reproduction. In the laboratory, fungi are isolated on specific culture media for cultivation, preservation, macroscopic examination and biochemical and physiological characterization. A wide range of media are used for isolation of different groups of fungi. These media influence vegetative growth, and colony, morphology, pigmentation and sporulation depending on their composition, pH, temperature, light, water availability and surrounding atmospheric gas mixture (Northolt and Bullerman, 1982; Kuhn and Ghonnomou, 2003).

The need to develop alternative media for various culture media has become imperative as the conventional media used are either not readily available or relatively expensive in most developing countries like Nigeria (Amadi and Moneke, 2012) and other developing countries of the world. The aim of this research was basically to formulate culture media from sweet potato peels and soybean hulls for fungi cultivation.

MATERIALS AND METHODS

Study Area

The study was carried out in Uli, Ihiala Local Government Area, Anambra State between March and July, 2017.

Sample Collections

Sweet potato peels and soybean hulls were collected from two (2) different area of agricultural wastes dumping sites in Uli community (Ezeama and Umuoma), Ihiala L.G.A, Anambra state. The samples were aseptically collected using hand glove and a sterile sealed polythene bags, all the sample collected were labeled and taken to the laboratory where they were sliced into small pieces, rinsed severally in clean water and spread on clean sacks and were sundried. The dried samples were blended into smooth powder, packed in thick nylon and stored in a cool dry place before use (Akharaiyi and Abiola, 2016).

Proximate Analysis of Agro-allied Wastes

The proximate compositions of the samples were determined using the standard methods of analysis of Association of Official Analytical Chemists, AOAC (1995).

Composition of Culture Medium from Agro-allied Wastes

This was carried out using modified method of Akharaiyi and Abiola (2016). Exactly 25 g, 50 g and 75 g of each powdery samples of sweet potato peels and soybean hulls were weighed and mixed with 150 ml, 200 ml and 350 ml of 100°C boiled sterile water respectively and were stirred with a glass rod and allowed to cool. Each mixture was filtered using muslin cloth. To a one hundred milliliter filtrate, 20 g of glucose was added. Exactly 15 ml of each medium were dispensed into bijou bottles using 4 bottles for each medium. Exactly 75 g of each of the powdery sample were mixed together and prepared in the same way. The mixtures were sterilized at 121 °C for 15 minutes in an autoclave. After autoclaving, each of the medium were allowed to cool to about 45°C before adding 1 ml of 0.05 g of chloramphenicol (this was added to inhibit bacterial growth).

Determination of pH of the Composed media

The pH of each of the composed media and the water used in formulating the media were determined using a Hanna pH meter (Clarkson Laboratory, USA) calibrated to 7.0 with pH buffer.

Fungal Isolates

The fungi isolates used were; Aspergillus niger, Aspergillus fumigatus, Aspergillus flavus and Trichoderma viride. Aspergillus niger, Aspergillus fumigatus. They were obtained from a previous work on isolation of fungi from throats of broiler chicken. The purity and identity of each fungi species were confirmed and the cultures were maintained at room temperature on potato dextrose agar until they were needed. Prior to the cultivation the isolates were subcultured into peptone water and incubated for 72 hours at room temperature.

Cultivation of Molds

The prepared culture bottles of sweet potato peel glucose broth (SPPGB); soya bean hull glucose broth (SBHGB), sweet potato peel and soya bean hull glucose broth (SPPSBHGB) and Sabouraud Dextrose Broth (SDB) were seeded with a loopful of each of the fungi isolates. Uninoculated sterile water was used as negative control while the inoculated SDB bottles served as positive control

Absorbance Measurement

The absorbance of each prepared media and control were measured using a spectrophotometer at wavelength of 540nm. The measurements were done in triplicates.

Statistical analysis

The absorbance was replicated three times. The results were expressed as mean±standard deviation (SD) and Student t-test at p≤0.05 (95% confidence level) were applied to access the difference between the means.

RESULTS

Proximate analysis

Table 1 showed the result of proximate analysis, The sweet potato peel had the highest percentage of starch and moisture content (74.97% and 5.4% respectively) among the employed agro-allied wastes product, while the soya bean hull had (64.02% and 4.1% respectively) Soya bean hull had the highest percentage of protein, crude fat, crude fiber and ash of 8.4%, 8.3%, 8.1% and 6.9% respectively while the sweet potato peel had 4.2%, 1.85%, 6.75% and 6.85% respectively).
Table 1: Proximate composition of Agro-allied Waste

<table>
<thead>
<tr>
<th>Source</th>
<th>Starch (%)</th>
<th>Protein (%)</th>
<th>Crude fat (%)</th>
<th>Ash (%)</th>
<th>Crude fibre (%)</th>
<th>Moisture content (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sweet potato peel</td>
<td>74.97</td>
<td>4.20</td>
<td>1.85</td>
<td>6.85</td>
<td>6.75</td>
<td>5.40</td>
</tr>
<tr>
<td>Soybean hull</td>
<td>64.20</td>
<td>8.40</td>
<td>8.30</td>
<td>6.90</td>
<td>8.10</td>
<td>4.10</td>
</tr>
</tbody>
</table>

pH of the Composed Media

The pH of different concentrations of the soyabean hull glucose medium and sweet potato peels glucose medium taken at ambient temperature is shown in Table 2. The result of pH determination showed that sweet potato peel glucose broth was more acidic than soybean hull glucose broth.

Table 2: pH of Composed Media at Different Concentrations

<table>
<thead>
<tr>
<th>Composed media</th>
<th>Concentrations 25 mg/ml</th>
<th>21 mg/ml</th>
<th>17 mg/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>SPPGB</td>
<td>4.5</td>
<td>4.6</td>
<td>4.7</td>
</tr>
<tr>
<td>SBHGB</td>
<td>4.7</td>
<td>5.0</td>
<td>5.3</td>
</tr>
<tr>
<td>SPPSBHBG</td>
<td>5.2</td>
<td>5.4</td>
<td>5.0</td>
</tr>
</tbody>
</table>

Keys: SPPGB= Sweet Potato Peel Glucose Broth; SBHGB= Soya Bean Hull Glucose Broth

Absorbance of Formulated Media Cultivated with known fungal Isolates

The result of the absorbance of the formulated media is shown in Tables 3, 4, and 5. Soyabean hull glucose medium and sweet potato peels glucose medium had more absorbance than the standard medium SDA. The statistical analysis of mean absorbance showed that there were significant difference between the mean of the test (formulated media) and the mean of control (SDA). The highest absorbance of 1.495 was observed in soyabean hull glucose broth inoculated with Trichoderma viride. Soyabean hull glucose broth yielded higher absorbance than potato peel glucose broth.

Table 3. Absorbance (OD\textsubscript{540nm}) of Formulated Media Cultivated with known Fungal Isolates at Concentration of 25 mg/ml

<table>
<thead>
<tr>
<th>Cultivated organism</th>
<th>SBHGB</th>
<th>SPPGB</th>
<th>SPPSBHBG</th>
<th>SDB</th>
<th>Water</th>
</tr>
</thead>
<tbody>
<tr>
<td>T. viride</td>
<td>1.495±0.000</td>
<td>1.328±0.000</td>
<td>1.483±0.000</td>
<td>0.960±0.000</td>
<td>0.165</td>
</tr>
<tr>
<td>A. flavus</td>
<td>1.311±0.000</td>
<td>1.300±0.000</td>
<td>1.432±0.000</td>
<td>0.929±0.000</td>
<td>0.157</td>
</tr>
<tr>
<td>A. niger</td>
<td>1.399±0.000</td>
<td>1.392±0.000</td>
<td>1.472±0.000</td>
<td>0.949±0.000</td>
<td>0.163</td>
</tr>
<tr>
<td>A. fumigatus</td>
<td>1.486±0.000</td>
<td>1.341±0.000</td>
<td>1.481±0.000</td>
<td>0.936±0.000</td>
<td>0.165</td>
</tr>
</tbody>
</table>

Mean absorbance ± standard deviation (p≤0.05). Keys: SPPGB= Sweet Potato Peel Glucose Broth; SBHGB= Soya Bean Hull Glucose Broth; SPPSBHBG= Sweet Potato Peel and Soya Bean Hull Glucose Broth; SDB= Sabouraud Dextrose Broth.

Table 4: Absorbance (OD\textsubscript{540nm}) of Formulated Media Cultivated with known Fungal Isolates at Concentration of 21 mg/ml

<table>
<thead>
<tr>
<th>Cultivated organism</th>
<th>SBHGB</th>
<th>SPPGB</th>
<th>SPPSBHBG</th>
<th>SDB</th>
<th>Water</th>
</tr>
</thead>
<tbody>
<tr>
<td>T. viride</td>
<td>1.522±0.000</td>
<td>1.517±0.000</td>
<td>1.513±0.000</td>
<td>0.960±0.000</td>
<td>0.165</td>
</tr>
<tr>
<td>A. flavus</td>
<td>1.497±0.000</td>
<td>1.512±0.000</td>
<td>1.509±0.000</td>
<td>0.929±0.000</td>
<td>0.157</td>
</tr>
<tr>
<td>A. niger</td>
<td>1.513±0.000</td>
<td>1.510±0.000</td>
<td>1.520±0.000</td>
<td>0.949±0.000</td>
<td>0.163</td>
</tr>
<tr>
<td>A. fumigatus</td>
<td>1.486±0.000</td>
<td>1.510±0.000</td>
<td>1.517±0.000</td>
<td>0.936±0.000</td>
<td>0.165</td>
</tr>
</tbody>
</table>

Mean absorbance ± standard deviation (p≤0.05). Keys: SPPGB= Sweet Potato Peel Glucose Broth; SBHGB= Soya Bean Hull Glucose Broth; SPPSBHBG= Sweet Potato Peel and Soya Bean Hull Glucose Broth; SDB= Sabouraud Dextrose Broth.

Table 5: Absorbance (OD\textsubscript{540nm}) of Formulated Medium Cultivated with known Fungal Isolates I at Concentration of 17 mg/ml

<table>
<thead>
<tr>
<th>Cultivated organism</th>
<th>SSPGB</th>
<th>SBHGB</th>
<th>SPPSBHBG</th>
<th>SDB</th>
<th>Water</th>
</tr>
</thead>
<tbody>
<tr>
<td>T. viride</td>
<td>1.493</td>
<td>1.493</td>
<td>1.477</td>
<td>0.960</td>
<td>0.165</td>
</tr>
<tr>
<td>A. flavus</td>
<td>1.490</td>
<td>1.493</td>
<td>1.498</td>
<td>0.929</td>
<td>0.157</td>
</tr>
<tr>
<td>A. niger</td>
<td>1.479</td>
<td>1.481</td>
<td>1.498</td>
<td>0.949</td>
<td>0.163</td>
</tr>
<tr>
<td>A. fumigatus</td>
<td>1.439</td>
<td>1.457</td>
<td>1.470</td>
<td>0.936</td>
<td>0.165</td>
</tr>
</tbody>
</table>

Mean absorbance ± standard deviation (p≤0.05). Keys: SPPGB= Sweet Potato Peel Glucose Broth; SBHGB= Soya Bean Hull Glucose Broth; SPPSBHBG= Sweet Potato Peel and Soya Bean Hull Glucose Broth; SDB= Sabouraud Dextrose Broth.
DISCUSSION

The result of this study showed that all the formulated media supported the growth of fungi. The growth of the fungi on the formulated media implies that the agro wastes which were used in formulating the media contained the required nutrients for fungal growth in the right proportion. Similar observations were made by Amadi and Moneke (2012), Itelima et al. (2014) and, Akharaiyi and Abiola (2016) who reported the use of alternative culture media for growing fungi.

The proximate analysis done showed that the wastes contained the following nutrients: protein, carbohydrate and minerals. The details are as follows: sweet potato peel and soya bean hull showed the following; Starch (74.97%, 64.2%), Protein (4.2%, 8.4%), Crude fat (1.85%, 8.3%), Crude fibre (6.75%, 8.1%), moisture content (5.4%, 4.1%), and Ash (6.85%, 6.9%) respectively which led to variation in the concentration of media components and also played a role in the overall outcome of the formulated media in good support of fungal growth. This also in line with the findings of Akinyele and Adetuyi (2005). Protein constitutes a significant portion of microbial cells and thus is necessary for the growth of microorganisms. The protein content of the formulated media must have served as source of nitrogen while the carbohydrate content served as carbon source both of which are essential for good fungal growth. The concentration of 21 mg/ ml of each of the agro wastes was observed to favor fungi growth more than other concentrations.

Of all the fungi cultivated, the formulated media highly favored mostly the growth of Trichoderma viride than the Aspergillus species, it may be probable be that the nutritional contents of these media are optimal for its growth. This is in conformity the findings of Akharaiyi and Abiola (2016) who reported a maximum yield of T. virde, A. flavus and A. fumigatus upon cultivation on yam glucose agar and plantain glucose agar. The pH concentrations of the formulated media ranged from 4.6-5.4 which is within the optimal range of pH for fungal growth. According to Andualem and Gessesse (2013) media containing high carbohydrate source, nitrogen source are required for the growth of fungi at pH range of 4 to 6, and a temperature range from 15 to 37 °C, this is in accordance with the present study.

The formulated media competed favorably well with the positive control (SDB) Similar result was obtained by Akharaiyi and Abiola (2016) using yam glucose agar, plantain glucose agar and pineapple broth seeded with eleven species of fungi. Microbiological studies depend on the ability to grow and maintain microorganisms under laboratory conditions by providing suitable culture media that offer favorable conditions (Akharaiyi and Abiola, 2016). Laley (2002) reported a modification of Potato medium supplemented with cow dung, Soy milk and other growth factor for cultivation of fungi. Also, Adesemoye and Adefere (2005) studied the feasibility of developing alternative media to Potato Dextrose Agar (PDA) using local cereal species as a basal media. They observed that all the fungal species grew to some extent better on the formulated media in relation to the standard set up.

The results obtained with sweet potato peel glucose broth (SPPGB) and soya bean hull glucose broth (SBHGB) showed optimal fungi growth which may be as a result of the enriching nutritional constituents of the agro-allied wastes. The proximate analysis report showed that sweet potato peel had higher carbohydrate content than the soybean hull, this could be used as a raw material to formulate media for cultivation of fungi, which can break down the sugars to serve as source of carbon and energy. The ideal medium for cultivation of microorganisms is mandated to be totally defined, reproducible, well buffered to maintain pH, and should be readily available. The ability of the agro-allied wastes to support good growth of the fungi showed that they not only contained the right nutrients but also probably contained them in the right proportions. The fact that the fungi grown on the formulated media thrived better than when they were grown on the conventional SDB revealed that the formulated media could serve as a good and possibly cheaper alternative medium for the cultivation of some fungi.

CONCLUSION

This study revealed that the sweet potato peels and soybeans hull contains minerals and nutrients that can meet the nutritional requirements of fungi; consequently they can be utilized as substitutes in the formulation of culture media for the in vitro cultivation of fungi. More so, the agro-allied wastes used in this study are readily available and cheap if at all they are to be purchased.

REFERENCES


Laley, S.A., Tedela, P.O., Adesua, B. and Famurewa, O. (2007). Growth of some microorganisms on media formulated from...


How to cite this article

Licensed under a Creative Commons Attribution 4.0 International License