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Evaluating the Potential of Wild Cocoyam "Caladium Bicolor" for Ethanol Production using Indigenous Fungal Isolates

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Abstract

Ethanol is a renewable bio-fuel which impacts the environment minimally. Different plant materials have been assessed as carbon source for bio-ethanol production in previous studies. In the present study, flour from peeled tuber of Caladium bicolor (wild cocoyam) was used as the carbon source and microorganisms isolated from rotten C. bicolor tubers and palm wine were utilized for the fermentation. This research was aimed at using indigenous microbial isolates and wild cocoyam (non-edible) as substrates for bio-ethanol production. Eighteen fungal strains were isolated from rotten tubers of C. bicolor and fresh palm wine and thereafter screened for amylase production and fermentation capability. The best isolate was used to optimize reducing sugar production by varying the fermentation parameters (time, pH, substrate concentration, and nitrogen source and inoculum size). The results obtained revealed nine isolates with high starch degrading ability. Aspergillus spp. were found to be the predominant microorganisms and Aspergillus sp. (Org 2) gave the highest amylolytic activity with a concentration of 6.019 mg/ml reducing sugar during submerged fermentation. Under optimal conditions (pH 5, 5% substrate concentration, 0.5% soya bean and 2% inoculum size (1.0 x 10^8 spores/ml), on day 5) results from hydrolysis showed that the highest amount of reducing sugar produced was 6.989 mg/ml. The hydrolysate was further fermented using Saccharomyces sp. (4.0 x 10^7cells/ml) at pH 5.2 for144 hours. The fermented liquid was then distilled to yield a maximum ethanol concentration of 0.485%. This study demonstrates that indigenous microbial isolates and substrates when properly propagated and processed can be utilized for amylase and ethanol production.

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Keywords:Bioethanol, Caladium bicolor, Aspergillus sp., Amylase, Hydrolysis, Saccharomyces sp., Fermentation;

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1.0 Introduction

Energy in all its forms is essential to humanity and is central to the improvement in people's quality of life. The continuous increase in energy demand, the inevitable decline in the availability of fossil fuels, and the growing concerns about climate change have sparked a number of initiatives from governments around the world to increase production of energy from renewable sources (Quintero *et al*., 2008). Bio-fuels, and in particular bio-ethanol, *i.e.* ethanol obtained from crops or lignocellulosic biomass, are getting a lot of attention as possible options for renewable fuel. At the global level, Brazil is world leading producer of biofuel and recently, Nigeria joined the league of biofuel users with the aim of generating wealth (Aisien*et al*., 2010).

Ethanol is one of the preferable liquid fuels due to its combustion properties and its use as an additive with gasoline (Galbe and Zacchi, 2002). Most importantly is the mix of ethanol and gasoline which reduces green house gas emissions at certain levels but also minimizes dependence on fossil fuel. The design had generally been production from sugarcane and cassava. Klass (1998) categorized cassava alongside with sweet potato and yam as main starches that serve as staple foods for people through the world's hot and humid regions. These plants are so proficient at supplying essential calories that they are considered the quintessential subsistent crops. However, the success of these starch crops as staple foods limits their potential development and general economic growth, for instance, cassava which has become an important bio-fuel crop is a crop crucial for food security especially in Nigeria. The implication of this is that threats to food security exist in the face of growing fuel ethanol demand. Perhaps, the diversion of food resource to bio-fuel production may to a large extent have fuelled the current food crises worldwide (Srinorakutara*et al*., 2008). It therefore becomes imperative that the searchlight be turned at present to the use of non-food starchy items for the production of fuel ethanol.

Caladium bicolor 'Florida Clown' is a wild cocoyam commonly known as '*Ede Umuagbara'*, in the eastern part of Nigeria. They look like our normal edible cocoyam (*Colocasiaesculenta*) but can be differentiated by the red and white patches found on the leaves. They are tuberous, heart-shaped leaves may vary in size from 15 cm to 60 cm in length and features leaves with randomly white and red blotches. *C. bicolor* is a non-human edible plant that grows along river banks, lakes, streams, brooks, oases and shady areas in Nigeria. Also here in Nigeria, they grow indiscriminately, without being cultivated. Unlike the edible cocoyam, it has a prominent and non-hardy cocoyam corm. It is self sustaining as it has broader leaves, which enable it to suppress other weeds under and around it. All parts of the plant are inedible. Eating of the corm produces an intense irritation in the throat. All parts should not be ingested and may irritate sensitive skin. The gainful use of *C. bicolor* will not only bring about the practical exploitation of this inedible abundant natural resource, which is wild and available; but also will encourage local farmers and boost their economy. In addition, the use of this biomass (*C. bicolor*) for bio-ethanol production will help solve the problem of the diminishing fossil fuel reserve and at the same time not lead to food insecurity that can come about while using staple food like cassava, yam, sweet potato, and cocoyam.

2.0 Materials and methods\

2.1 Sample Source

The wild cocoyam used in this study was harvested from a farm in Oboloafor in Nsukka, Enugu state and transported to the Microbiology Laboratory, University of Nigeria Nsukka for analysis. Sample was identified in the laboratory of Botany Department, University of Nigeria Nsukka,. The wild cocoyam plant (sample) was identified as *Caladium bicolor* or "Florida Clown".

2.2 Processing of Samples

The fresh and healthy unpeeled tubers were sort, then washed and cleaned of soil contamination with sterile distilled water. They were air-dried at room temperature. Using a pre-sterilized knife, the tubers were peeled and sliced into smaller pieces. They fleshy part of the tuber was air dried to a constant weight and ground separately to fine powders using manual grinder to obtain stock sample. The ground sample was screened through a fine mesh sieve to remove any large particle and to obtain the same size range.