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Integrated bioethanol and biomanure production from potato waste

Anjani Devi Chintagunta [a], Samuel Jacob [b], Rintu Banerjee [b,*]

[a] Advanced Technology Development Centre, Indian Institute of Technology, Kharagpur 721302, West Bengal, India
[b] Agricultural & Food Engineering Department, Indian Institute of Technology, Kharagpur 721302, West Bengal, India

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ABSTRACT

Disposal of potato processing waste and the problem of pollution associated with it is a vital issue that is being faced by the potato processing plants. The conventional peeling methods presently followed in the processing plants for removing the potato peel, also result in the loss of some portion of the mash which is rich in starch. Indiscriminate discharge of the waste causes detrimental effects in the environment, so this problem can be resolved by successful utilization of the waste for the generation of value added products. Hence, the present work focuses on integrated production of bioethanol and biomanure to utilize the waste completely leading to zero waste generation. The first part of the work describes a comparative study of ethanol production from potato peel and mash wastes by employing co-culture of *Aspergillus niger* and *Saccharomyces cerevisiae* at various incubation time (24–120 h) instead of application of enzymes. The solid state fermentation of potato peel and mash inoculated with co-culture, resulted in bioethanol production of 6.18% (v/v) and 9.30% (v/v) respectively. In the second part of the work, the residue obtained after ethanol production was inoculated with seven different microorganisms (*Nostoc muscorum*, *Fischerella muscicola*, *Anabaena variabilis*, *Aulosira fertilissima*, *Cylindrospermum muscicola*, *Azospirillium lipoferum*, *Azotobacter chroococcum*) and mixture of all the organisms in equal ratio for nitrogen (N), phosphorous (P) and potassium (K) enrichment. Among them, *A. variabilis* was found to enrich N, P and K content of the residue by nearly 7.66, 21.66 and 15 fold than that of the initial content, ultimately leading to improved N:P:K ratio of approximately 2:1:1. The application of simultaneous saccharification and fermentation (SSF) for the conversion of potato waste to ethanol and enrichment of residue obtained after ethanol production with microorganisms to be used as manure envisages environmental sustainability.

1. Introduction

Changing life style and craving for processed foods are factors responsible for the significant growth of global food processing industries. Among several varieties of processed foods, the products from potatoes are in great demand. Potato is a starchy tuber with carbohydrates constituting as much as of 13–30% with a little protein (0.7–4.6%) (Puttongsiri et al., 2012). The production of potato in India for the year 2012–13 was reported to be 45.3 million tonnes from 1.99 million hectares of land with an yield of 22,763.1 kg/ha (Agricultural statistics at a glance, 2013). The potato processing industry has multiplied in India owing massive potato production and a booming demand for its products in the market. Potato peeling is the initial step in any potato processing industry and losses caused during this stage may range from 15% to 40% according to the method followed for peeling (Arapoglou et al., 2009). The processing loss during production must be reduced by employing efficient peeling methods to obtain maximum economic returns and at the same time the disposal problem needs to be addressed by converting the waste into value added products.

Production of consumable alcohol from the food waste can be an impressive alternative. Growing per capita spirit consumption and escalating demand for premium brands are motivating the growth of global spirits manufacturing market. Although the industry was hit hard by the economic downturn during 2009, it has picked up slowly with an average growth rate of 2.2% annually upto 2013. In 2013, global market revenue rose to 9.5% and expected to attain 9.8% by 2018 (Mynews desk, 2013). Kawa-Rygielska et al. (2012) reported the utilization of by-products generated during potato granule processing as feedstock for ethanol production. Ethanol production from starchy biomass generally involves various steps like liquefaction, saccharification and fermentation (Pervez et al., 2014). In the present study, coculture of *A. niger* and *S. cerevisiae* were used to obtain improved ethanol pro-

* Corresponding author.
E-mail address: rb@iitkgp.ac.in (R. Banerjee).

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duction through simultaneous saccharification and fermentation process. Ado et al. (2009a) reported that the synergistic metabolic interactions between *A. niger* and *S. cerevisiae* in a starch medium enhances amylolytic activity and total ethanol yield by preventing accumulation of inhibitory concentrations of reducing sugar.

The solid residue obtained after ethanol production still contains certain amount of nutrients which upon further enrichment can be effectively utilized as biomanure, an efficient way of controlling the solid waste accumulation in the environment. The enriched residue is an excellent organic manure containing all the essential nutrients vital for plant growth and humus to develop the soil structure (Bhattacharyya and Banerjee, 2007).

The deficiency of nitrogen is usually met by supplementing the soil with the chemical fertilizers, whose long term application leads to soil degradation and other environmental issues. The import of the chemical fertilizers as percentage value of consumption for the year 2012–2013 was estimated to be Rs.(₹) 4,04,120 million (Chemicals and Petrochemicals Statistics at a Glance, 2014). In the present study, an attempt has been made to enrich the waste by using blue green algae which are capable of fixing nitrogen, an essential element for plant growth. Ananya et al. (2014) reported excretion of vitamins, amino acids and hormones like auxin, gibberellin etc. by algae which involve in promoting plant growth. The administration of biomanure is not only a cost effective way of enriching soil but also a better solution for environmental problems caused by the excessive usage of chemical fertilizers.

Thus, the present work focuses on a comparative study of ethanol production from potato peel and mash wastes through simultaneous saccharification and fermentation employing co-culture of *A. niger* and *S. cerevisiae* followed by treatment of mixed solid residue with different nutrient enriching microorganisms for its utilization as biomanure.

2. Materials and methods

2.1. Substrate

Potato peel and mash waste generated during processing of potato products were collected from the local plants based in Kharagpur, West Bengal, India.

2.2. Microorganisms

2.2.1. Inoculum for bioethanol production

Aspergillus niger and *Saccharomyces cerevisiae* cultures were obtained from Microbial Biotechnology and Downstream Processing Lab, IIT Kharagpur, West Bengal, India. *A. niger* was maintained on Potato Dextrose Agar (PDA) medium at pH 5.6 ± 0.2 and 25 °C whereas *S. cerevisiae* was grown in medium containing glucose (2%, w/v) and yeast extract (0.5%, w/v) at pH 6.5 and 37 °C.

2.2.2. Inoculum for biomanure production

The blue green algae viz., *Nostoc muscorum*, *Fischerella muscicola*, *Anabaena variabilis*, *Aulosira fertilissima*, *Cylindrospermum muscicola* selected for N, P and K enrichment studies were procured from Vishwabharathi University, Kolkata and cultured in BG11 medium as reported by Rippka et al. (1979) at pH 7.5 and 25–28 °C excluding nitrate or ammonium source as they generate heterocysts in the nitrogen deficient medium (Pankaj, 2008; Pereira et al., 2005; Saville et al., 1987). *Azospirillium lipoferum* and *Azotobacter chroococcum* were cultured in nutrient broth medium at pH 7.3 ± 0.2 and 35–37 °C.

2.3. Methods

Moisture, protein, reducing sugar, cellulose and starch content of potato wastes were determined by following standard methods (AOAC, 1965; Lowry et al., 1951; Miller,1959; Updegraff, 1969; Hodge and Hofreiter, 1962). The estimation of N, P and K content in the potato waste was done by Kjeldahl, spectrophotometric and flame photometric methods (Scan test method, 1986; Chapmann and Pratt, 1961; Hegedus and Pungor, 1955) respectively. The reducing sugar and ethanol content were estimated by dinitrosalicylic acid and potassium dichromate method respectively (Miller, 1959; Seo et al., 2009).

2.4. Experimental setup for bioethanol production

Solid state fermentation process was established with two substrates i.e., potato peel and mash and ethanol production from both the substrates was observed by employing co-cultures. The experimental set up with each substrate consists of 5 treatments each containing 100 g (wet weight) of substrate mixed with 20 ml of modified Czapekdox medium. *A. niger* (1%, v/w) containing 2.5×10^6 spores/ml was added to all the 5 treatments. *S. cerevisiae* (10%, v/w) (Ado et al., 2009b) was inoculated to the first treatment after 24 h of *A. niger* addition. To second treatment, *S. cerevisiae* (10%, v/w) was inoculated after 48 h so on and fifth one after 120 h respectively and incubated at 37 °C (Fig. 1). Inoculum free substrate was used as control. Small aliquots were drawn from each treatment at specific time intervals (24–168 h) and centrifuged at 2000 rpm for 5 min. The clear supernatant was collected and analyzed for ethanol content.

2.5. Experimental setup for biomanure production

After bioethanol production, the liquid portion containing the ethanol was separated from the solid content of both the substrates by squeezing through a cheese cloth. The solid residue (peel and mash) was collected, mixed, characterized and employed for nutrient (N, P, K) enrichment to convert into biomanure. A set of eight treatments, each containing 100 g of mixed residue was arranged and 10% (v/w) inoculum of each organism like *N. muscorum*, *F. muscicola*, *A. variabilis*, *A. fertilissima*, *C. muscicola*, *A. lipoferum*, *A. chroococcum* was added individually and as a mixture in equal proportion and incubated at 25–27 °C. A control was also maintained without the addition of inoculum. The residue was checked for NPK enrichment weekly upto 13 weeks. The schematic representation of integrated production of bioethanol and biomanure was shown in Fig. 1.

3. Results and discussion

3.1. Characteristics of potato peel and mash wastes

The moisture, starch, cellulose, reducing sugar and protein content of potato peel and mash wastes are presented in Table 1. It has been observed that potato peel and mash wastes have considerable starch content (28.52% ± 0.17 and 49.78% ± 1.2 (w/dry weight)), cellulose content (5.69% ± 1.6 and 2.31% ± 1.2 (w/dry weight)), meagre fermentable reducing sugar (0.073% ± 0.0043 and 1.33% ± 0.10 (w/dry weight)) and protein content (0.082% ± 0.002 and 0.16% ± 0.001 (w/dry weight)) respectively. The reducing sugar content in the potato peel was in accordance with that reported by Khawla et al. (2014) but varied in starch and protein content. The probable reason for such variation could be due to the influence of climatic conditions, methods adopted for potato processing etc.

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