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Full Length Research Paper

Production of bioethanol through enzymatic hydrolysis of potato

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Due to gradual decrease in petroleum resources and impacts of these wastes on the environment, there is a need to utilize the wastes of potatoes to get wealth out of wastes and clean the environment. In this study, potato wastes were investigated as source of bioethanol. 100 g potato powder was mixed with 1 L distilled water in two separate beakers to form potato slurry. Bioethanol production was investigated using effect of pH, temperature and mixture of digesting enzymes after scarification and fermentation. The results show that significant ($p<0.05$) bioethanol was produced at 35°C and at pH 5.5. This investigation also reveals that mixture of enzymes significantly enhanced ($p<0.05$) bioethanol production compared to non treated mixture. Maximum bioethanol productions were due to the presence of sugar in potatoes.

Key words: Bioethanol, potato, ph, temperature, liquefaction.

INTRODUCTION

Due to gradual decrease of fossil fuels, bioethanol has got the attention of many researchers to use it as alternative source of energy across the world. Bioethanol is produced from the fermentation of renewable resources for fuel or fuel additives. Additionally, bioethanol is considered to be produced from biomass-based materials (Grassi et al., 1999), instead of toxic methyl tert-butyl ether (MTBE) and tert-amyl methyl ether (TAMES). Bioethanol is already utilized as additive of gasoline in many parts of the world.

Production of bioethanol as a biofuel is the need of recent time, because in many countries of the world like the United States and Brazil tremendous research are going on this project. The world production of bioethanol has reached up to 51000 million with United States and Brazil getting top position and India on the fourth position. Bioethanol is produced from various agriculture wastes and sugar containing crops, vegetables and fruits like sugar cane (Brazil), corn grain (United States) and starchy agriculture wastes (India). According to the United States Department of energy for every unit of energy consumed for bioethanol production, 1.3 units of energy are returned (Hill et al., 2006).

Sugary substrates and cellulosic materials are either expensive or involves many steps for bioethanol production. Therefore crops which contained greater amount of starch are usually utilized for the bioethanol production such as crops like corn, barley, wheat, rice, tuber crops, potato and sweet potato. Production of bioethanol also depends upon strains of yeast such as *Saccharomyces cerevisiae*, suitable substrate and the methods employed for bioethanol production, greatly increase the efficiency of bioethanol production (Szambelan et al., 2004; Shigechi et al., 2004). For ethanol production, potatoes are cheap substrate because it is rich in starch and require less processing than other grains. After suitable processing, good quality of ethanol can be produced from potatoes which can be used for both fuel as well as potable purpose. Rotten potatoes are used for the

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Figure 1. Flow sheet of bioethanol production from waste potatoes.

production of bioethanol. As by products, rotten potatoes are obtained from 5 to 20% of the crops in potato cultivation (Kimmo and Liisa 1999). In Pakistan, there are millions of potato wastes and rotten potatoes are produced per year. This requires special consideration to utilize these waste potatoes for bioethanol production. In this study, we utilized potato wastes and rotten potatoes obtained from domestic dumping sides and local vegetables market for bioethanol production using different pH, temperature and mixture of enzymes for starch hydrolysis.

MATERIALS AND METHODS

Sample collection and treatment

Potato wastes and rotten potatoes were collected from the vegetable market of District Bannu and from the domestic dumping sides. These waste materials were cut into pieces, washed, dried and ground mechanically (Figure 1).

Formation of slurries and liquefaction

separate beakers to form potato slurry. 30 µl of amylase enzymes was added to one of the potato solution and the other contained only potato slurry. Both solutions were heated at 95 to 110°C for 30 to 40 min in autoclave to liquefy the starch and the pH was adjusted to 5.8.

Saccharification

After cooling, the slurries were mixed with 30 µl of glucoamylase enzyme to convert the starch into the fermentable sugar and also commercial yeast (*S. cerevisiae*) was added to both solutions as described by Oghgren et al. (2006).

Fermentation and distillation

100 g potato powder was mixed with 1 L distilled water in two After proper mixing, both solutions were sealed tightly (anaerobic fermentation) as shown in Figure 1 and incubated at 23°C, and 28 to 35°C to estimate the effect of temperature on bioethanol production. During this period, sugar units were fermented to bioethanol by the yeast and mixture of enzymes. After fermentation, unfermented residues were filtered while filtrate (bioethanol) was distilled with rotary evaporator at 78.5°C. The bioethanol obtained was used as biofuel after the addition of 3 to 5% of gasoline to get anhydrous bioethanol (Kroumov et al., 2006) while the unfermented solid residues (stillage) was used as valuable animal feed stocks

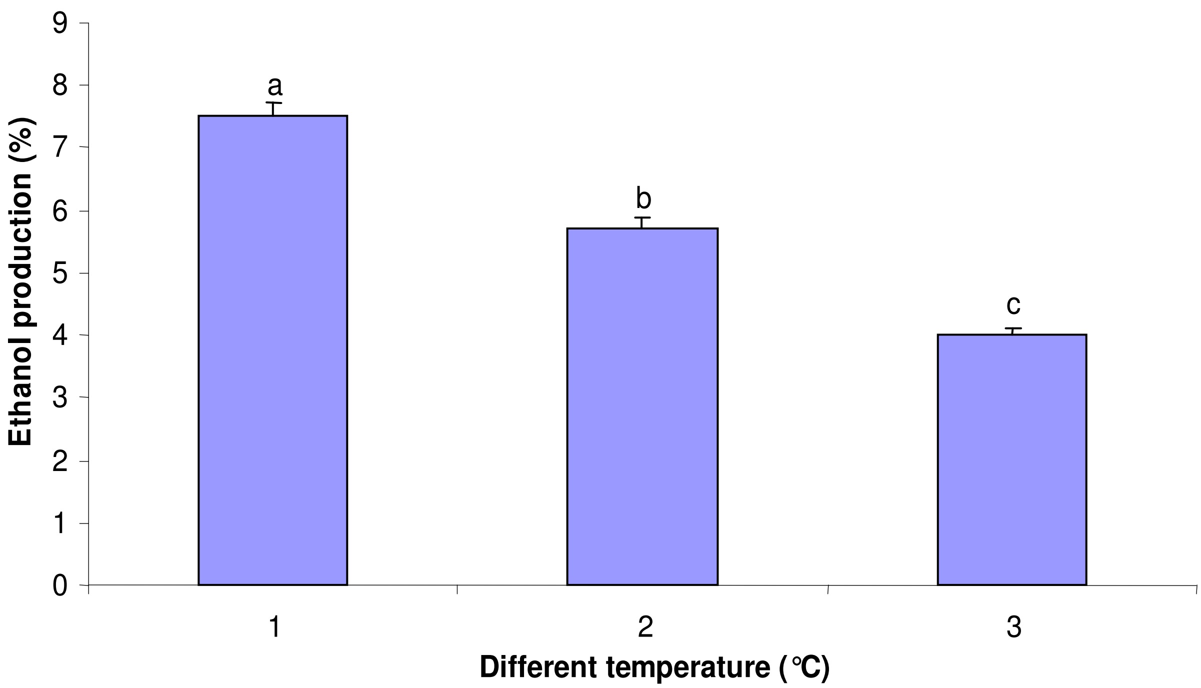

Figure 2. Percentage of ethanol production. 1, 35°C; 2 and 3, 35°C; different letter shows significance at p<0.05.

Statistical analysis

The tabulated data of various treatments were expressed as the mean ± SE for the three replicates in each group. Computer software SPSS 13 was used to determine the differences between groups using one-way analysis of variance (ANOVA). Least significant difference (LSD) was determined using inter group comparisons at probability level of 0.05%.

RESULTS

Temperature effects the production of bioethanol. Data of our this study express that significantly ($p<0.05$) maximum bioethanol was produced at 35°C, as compared to bioethanol produced at 23°C and 28°C respectively as shown in Figure 2.

pH of the media also effect bioethanol production. Data of our study express that considerable ($p<0.05$) bioethanol was produced at pH 5.5, as compared to bioethanol produced at pH 2 and 3 (Figure 3).

The bioethanol production also depends on process. Data of the study show that maximum bioethanol was recovered after treatment with both enzymes and yeast as compared to conventional method proceed with yeast only (Figure 4).

Various treatments of bioethanol production also affect the stillage of the remaining process. Figure 5 reveals that considerable ($p<0.05$) amount of stillage were produced with potatoes treated with a mixture of enzymes as compared to residue obtained from non treated enzymatic potatoes in this process.

DISCUSSION

Recently, due to shortage in natural resources of petrochemical and natural gas, researchers have focused to produce new renewable energy resources. To achieve this goal, scientists take interest on the production of bioethanol using biomass and agriculture wastes. Potato possesses great amount of starch that can be readily hydrolysed into sugars that could be easily fermented to produce bioethanol and found suitable to be used as alternative energy source (Chandel et al., 2007). Temperature effects the production of bioethanol. Three different temperatures; 35, 28 and 23°C were used in this study to find the effect of temperature on bioethanol production through fermentation. Data of this study revealed that significant ($p<0.05$) % bioethanol were produced at high temperature as shown in Figure 1. This might be due to the inappropriate temperature condition contributing to the lack of metabolic activity which consequently gave an effect on the diffusion of substrate and product (Alain et al., 1987). Growth of microbes requires optimum pH and production of bioethanol is effected with different pH values. pH values of bioethanol exhibited significant difference based on ANOVA method at $p<0.05$. The production of bioethanol was maximum at pH 5.5 compared to pH 3 and 2. Process of fermentation also effects bioethanol production from potatoes. Yeast is necessary for fermentations while addition of enzyme increases process of bioethanol production and enhances conversion of starch into sugar. In this study, two sets were used to produce bioethanol from potatoes.

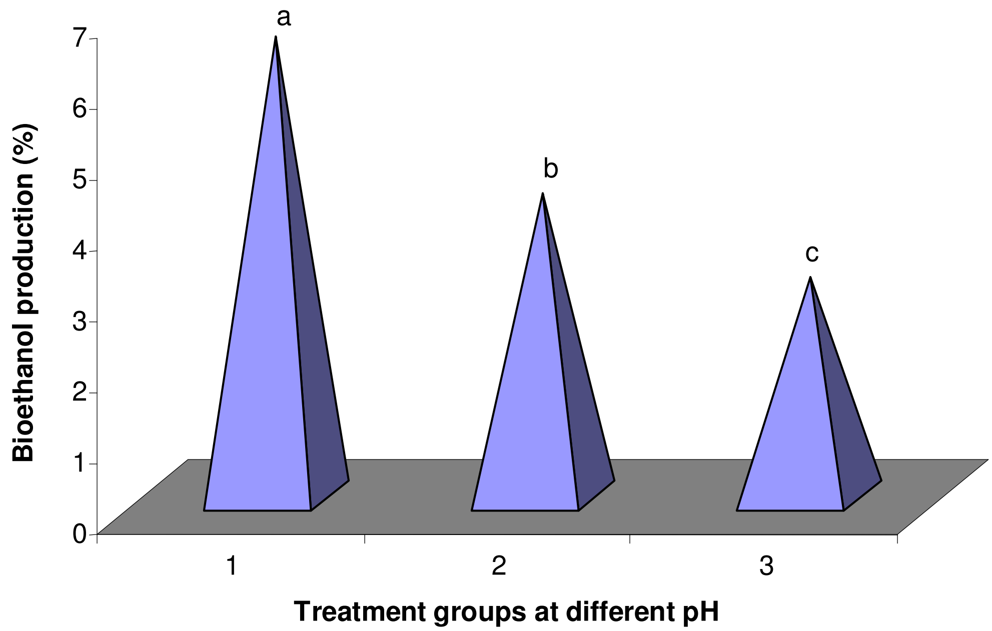

Figure 3. Percentage of ethanol production at different pH. 1, 5.5; 2, 3; 3, 2. Different letter shows significance at $p<0.05$.

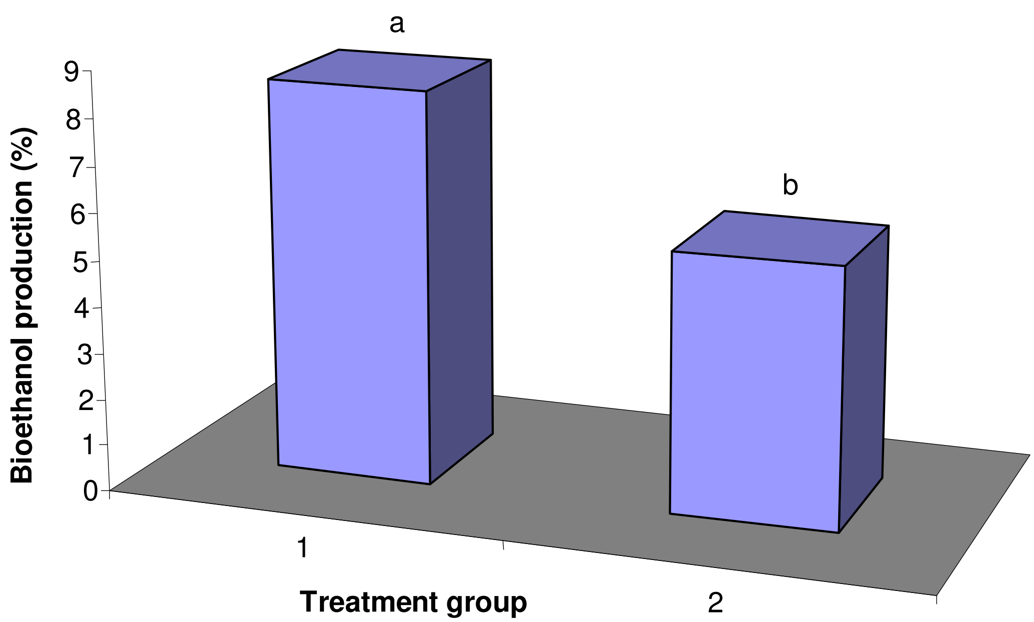

Figure 4. Percentage of ethanol production at different treatment. 1, Treated with mixture of enzymes; 2, treated with conventional method. Different letter shows significance at $p<0.05$.

In one set, we used a mixture of enzymes while the other was treated with yeast alone. Data reveal that maximum production of bioethanol was achieved from enzymatic hydrolysis compared to yeast treated sets. Similar results were documented by other study during bioethanol production from starchy and sugar containing sources (Hossain et al., 2011).

Conclusions

Energy crisis forces researchers to find new and low cost sources for ethanol production. In this study, potatoes were used for ethanol production. Experimental results show that potatoes possesses high amount of reducing sugars. In Pakistan, it is our first attempt to produce

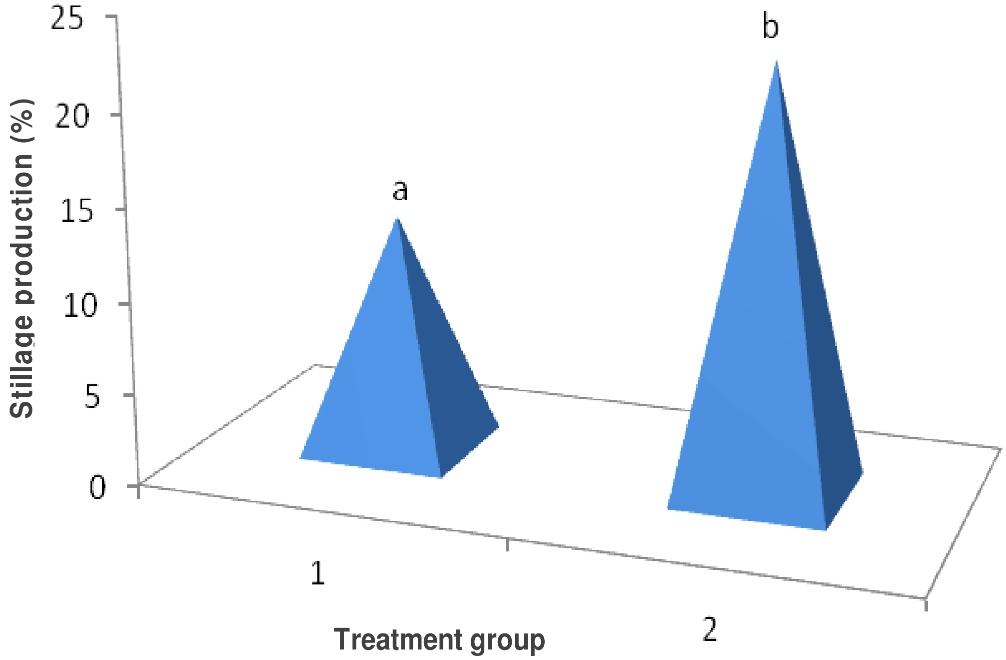

Figure 5. Percentage of stillage production at different treatments. 1,Treated with mixture of enzymes; 2, treated with conventional method. Different letter shows significance at p<0.05.

bioethanol from starchy agriculture wastes which on one side provides bioethanol for biofuel while on the other hand reduce environmental pollution. A comparative study on bioethanol production from the wastes of potato was carried out. It is concluded that potato wastes have greater amount of starch, and produced high bioethanol through enzymatic hydrolysis.

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