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Bioethanol Production from Rotten Fruit

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Abstract: Production of Ethanol from fermented renewable sources for fuel or fuel additives are known as bioethanol. Since the need of bioethanol has been increasing, the production of bioethanol must be increased using cheaper and eco-friendly raw materials. On the basis of these characteristics fruit wastes can be considered as cheaper and eco-friendly. In this study different fruit wastes were used as a raw material for the production of bioethanol by using Saccharomyces cerevisiae. The results of this work show that the rate of ethanol production through fermentation of grape fruit waste by Saccharomyces cerevisiae (baker's yeast) yield is good at pH 5.6, temperature 30°C, specific gravity 0.876, concentration of about 5.82% than other fruit waste The results of this study suggest that wastes from fruits that contain fermentable sugar should not be discarded into our environment, but should be converted to useful products like bioethanol that can serve as an alternative energy source.
Keywords: Saccharomyces cerevisiae, Ethanol production, Fruit waste, fermentation.

I. INTRODUCTION

Ethanol is a critical industrial ingredient, used as a base chemical for organic compounds and utilized as a part of restorative wipes and most usually as anti-microbial hand sanitizer gels and as an antiseptic. Ethanol from bio-mass based waste materials is most often used as a biofuel additive for gasoline [Akin-Osanaiye et al 2005]. World ethanol production for transport fuel somewhere around 2000 and 2007 raised from 17 billion to more than 52 billion liters. Ethanol is mostly used in Brazil and in the U.S., and both nations were in charge of 89 % of the world's ethanol fuel production in 2009. Most autos today in the U.S. can run on mixes of up to 10% ethanol, and the utilization of 10% ethanol fuel is commanded in a few U.S. states and urban areas. Since 1976 the Brazilian government has made it compulsory to mix ethanol with gasoline, and since 2007 the legitimate blending is around 25% ethanol and 75% gasoline. Also, by 2010 Brazil had dart of more than 10 million adaptable fuel vehicles frequently utilizing perfect ethanol fuel [Niger J Technol Res.vol.1,1989]. Alcohol is a natural intensify organic compound that has one or more hydroxyl (OH) group joined to a carbon atom. In dilute aqueous solution, it has a sort of sweet flavor, but in more concentrated solution it has a blazing taste [R., Shankar, T et al. 2011]. Ethanol manufactured from renewable sources for fuel or fuel added substances is known as bioethanol. Bioethanol, not at all like petroleum, is a type of renewable energy source that can be produced by using agriculture feedstock [M V.Ranade et al 2012]. The first generation of ethanol production utilized corn as a substrate, later corn was considered as a feedstock lead to the second generation of production of ethanol which utilized microorganisms and different wastes as substrates [A. Goettemoeller (2007)].

The least expensive and easily available raw material for the production of bioethanol is fruit waste. It is a potential source, from which ethanol can be produced. Fruit waste which is discarded has great antimicrobial and cell reinforcement potential. In this study, looking at of the ethanol efficiency produced by maturation process from mix source of diverse fruits, apples, grapes, and bananas, and study the effect of various parameters like pH, temperature, specific gravity and concentration. The yield of ethanol added up to more noteworthy than 80% of the fermentable sugar consumed [Grassi, M., et al. 1999]. Saccharomyces cerevisiae is utilized as a part of the fermentation process since it changes over sugar with oxygen to give carbon dioxide. As indicated by the International Energy Agency, cellulosic ethanol could permit ethanol fuels to assume a much greater part later on than previously suspected [O. R. Inderwildi, et al. 2009].

The ethanol efficiency produced by fermentation process from different fruit wastes such as grapes, apples and bananas. The efficiency of the ethanol can be calculated by determining the various parameters such as pH, temperature, specific gravity and concentration. Saccharomyces cerevisiae is used in the process since it converts sugar with oxygen to give carbon dioxide.

Grape wastes can be used as a substrate for the production of ethanol. The yield of ethanol amounted to greater than 80% of the fermentable sugar consumed. Although waste bananas (Musa paradisiaca) are commonly discarded as waste they represent a potential energy feedstock which may be especially suited for ethanol production. Waste bananas are low cost, concentrated biomass feedstock. Apple (*Malus pumila*) is the oldest fruit known to man and is grown extensively throughout the temperate zones of the world. Apple is also one of the good sources for the production of ethanol. The main objective of this study is compare the ethanol efficiency obtained from different fruit waste.

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II. MATERIALS AND METHOD:

A. Chemicals and Composition

5% potassium permanganate ($KMNO_4$) ,Beaker yeast, Sucrose, *Saccharomyces cerevisiae*, Conical flask, filter paper, Jones reagent: (2% potassium dichromate(K2Cr2O7), Sulfuric acid (H2SO4)) ,Potassium dichromate reagent, S-diphenylcarbazide , Ethanol solution, Test tube with cap, Paraffin film, Water bath, 40% potassium sodium tartrate (Rochelle salt), KI solution ,Sodium thiosulphate, Starch indicator.

B. Collection of Fruit Waste (Banana, Apple, Grapes)

500 gm of apple, grapes pomace, and banana were collected from fruit market of Ahmedabad. These were washed in 5% potassium permanganate ($KMNO_4$) & rinsed well in distilled water.

C. Routine Culture Maintenance

10gm of Saccharomyces cerevisiae was added into warm water and mix it well.

D. Preparation of the Substrate

The protocol of *akin-Osanaiya B.C.* was used for the process of fermentation with some modification. The apple, grapes pomace and the rotten bananas were weighed separately and were taken in different beakers, which was washed with 5% potassium permanganate solution and then rinsed with distilled water. Apple waste, banana waste and grape waste, were crushed separately in a mixer and collected in beakers. Breaker 300 ml of slightly warm distilled water was taken and 50 gm of sucrose and 10gm of *saccharomyces cerevisiae* was added separately and mixed well. The mixture was transferred into a 2liter conical flask and made the final volume up to 1000ml with slightly warm distilled water.

E. The Fermentation Process

The flask is covered with paper and the mouth is sealed and kept in a shaker incubator and allowed to incubate for 36hrs at 36°C with the speed of 100 rpm.

F. Qualitative Estimation of Bio Ethanol

The qualitative estimation of bioethanol was done. For this we have to prepare I ml of 2% $K_2Cr_2O_7$ and 5 ml of H_2SO_4 (jones reagent) were added in 10 ml sample for the qualitative estimation of bioethanol and observe the result.

G. Quantitative Estimation of bio Ethanol

Ethanol concentration was determined according to the method of *Williams and Darwin (1950).* The 100 ml of potassium dichromate reagent solution was prepared by dissolving 1g of potassium dichromate in concentrated sulfuric acid. saturated s-Diphenylcarbazide solution was prepared by dissolving 1 g of s-Diphenylcarbazide to 1 ml of 95% ethanol and the supernatant was collected. The 1 ml of ethanol solution was added to the glucose sample into the capped test tube. The test tube was covered with a paraffin film to avoid the loss of liquid due to evaporation. The mixture was then heat up using water bath at 90°C for 5 to 15 min until it looks like red-brown color. Then in this mixture added with 1 ml of a 40% potassium sodium tartrate (Rochelle salt) solution to stabilize the color. The ethanol absorbance values were measured at 575 nm after cooling to room temperature in a cold water bath.

H. Distillation Process

Specific gravity of the sample was noted frequently by using a hydrometer. When the specific gravity reaches a steady value, it indicates the end of the fermentation process. The incubation period varies for each fruit waste sample.



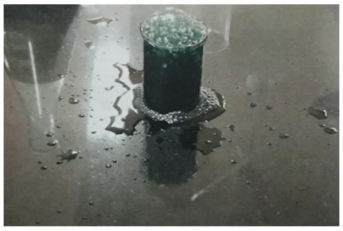
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After the fermentation process a small amount each sample was taken and centrifuged. The supernatant was collected and the volume of the alcohol was determined by the specific gravity method. Then the rest of the sample was distilled using simple distillation unit to collect the ethanol from different fruit wastes. For distillation, the batch distillation method was adopted. The distillation unit consisted of three components: a reboiler, condenser pipe and a distillate or receiving flask. The filtered sample was transferred into the reboiler and the sample was boiled. The vapors started to rise into the still head and passed through the condenser pipe. The continuous circulation of cold water around the condenser pipe assisted in cooling the alcohol rich vapors back to the liquid state. The condensed liquid enters the still receiver and is then collected in the distillate. The final tests such as pH, temperature, specific gravity and concentration of bioethanol were determined. The amount of ethanol obtained from different fruit wastes was recorded and the solution was subjected to iodine test to confirm the presence of ethanol.

III. RESULTS

Ethanol was produced from different fruit wastes. the comparative study has been carried out to check the efficiency of ethanol produced from these fruit wastes by determining various parameters. The influence of various parameters on the production of ethanol is presented as follows.

The following are the pictorial representation of method and result of bio ethanol production from banana, grapes and apple.



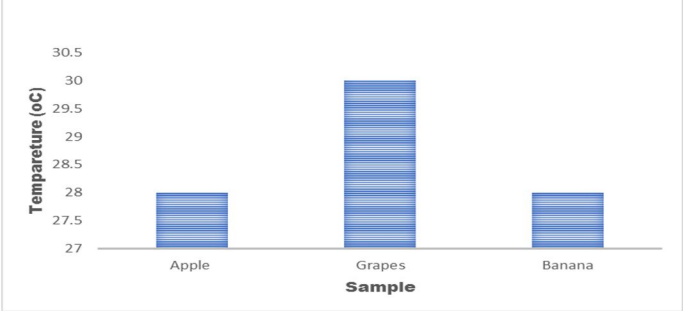
A. Effect of Temperature

Temperature plays a major role in the production of ethanol, since the rate of alcoholic fermentation increases with the increase in temperature. The optimum temperature of ethanol ranges between 25°C to 40°C which depends on room temperature. Bioethanol produced from both apple and banana have same temperature of about 28°C, and ethanol from grapes has 30°C temperature. When temperature goes below the optimal range, their ability to catalyze the intended reaction slows down (Table 1; Fig2). On the other hand, when the temperature increases, enzymes begin to denature or unfold and thus become inactive. Each enzyme will have a different temperature range where it becomes inactive. Even if one essential enzyme stops working, the organism fails to grow. Hence, the first essential enzyme that gets deactivated defines the maximal temperature at which that organism can grow. At the lower end, it gets more complicated. Usually, the enzymes are not inactivated but rather just slow down [*Sanchez, C., et al 2007.]*

Table No1. Production of bioethanol at different temperature.

Sr. NO.	Sample	Temperature
1	Apple waste	28°C
2	Grapes waste	30°C
3	Banana waste	28°C

Fig.2 Comparison of bioethanol production of different temperature obtained from different fruit samples



Conclusion: Bioethanol produced from grapes has 30°C more than apple and banana.

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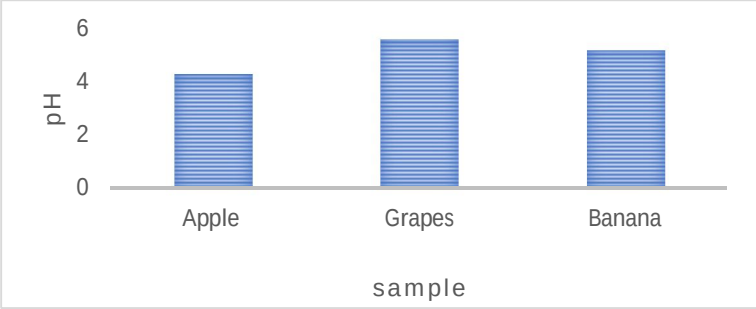
B. Effect of pH

pH value has significant influence on alcoholic fermentation. pH of bioethanol produced from different fruit wastes were determined. The pH value of ethanol produced by the process of fermentation ranges from 4 to 6. Yeast survives in a slightly acidic environment that is with pH between 4 to 6. The pH value of ethanol obtained from fruit wastes was determined as, grape at (pH=5.6), apple (pH=4.3), and banana (pH=5.2). Among these ethanol produced from grape wastes had on higher alcoholic content (Table 2; Fig.3)

Table 2: production of bioethanol at various pH

Sr no.	Sample	Ph
1	Apple waste	4.3
2	Grapes waste	5.6
3	Banana waste	5.2

FIG 3: Comparison of bioethanol pH obtained from different fruit samples.



Conclusion: Bioethanol produced from grape wastes had on higher alcoholic content.

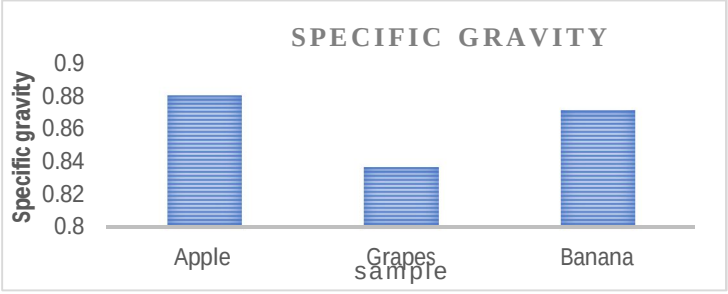
C. Effect of Specific Gravity

Specific gravity is used to measure the sugar content. As the fermentation progressed, the specific gravity considerably decreased and reached a value of 0.881 at 48hrs and remained constant. The specific gravity of apple after fermentation was reduced to 0.881 at 36hrs and remained constant, The specific gravity of grapes after fermentation was reduced to 0.837 at 72hrs, whereas the specific gravity of banana was reduced to 0.872 at 72hrs. The decrease in specific gravity is clear indication of yeast fermenting the sugar resulting in ethanol production. The specific gravity reaching a constant value after incubation period is the indication of end of fermentation (Table 3; Fig.4).

Table 3; Specific gravity of bioethanol obtained from different samples

Sr.no.	Sample	Specific gravity
1	Apple waste	0.881
2	Grapes waste	0.837
3	Banana waste	0.872

Fig 4: specific gravity from different sample



Conclusion: The specific gravity considerably decreased and reached a value of 0.881 at 48hrs

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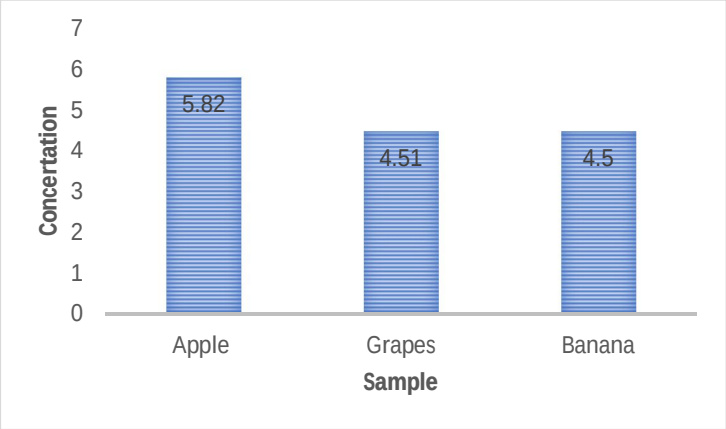
D. Effect of Concentration

Concentration is the measure of ethanol content present in the distillate. Concentration of ethanol was expressed in terms of percentage (%). The concentration of ethanol present in the grapes sample was 4.51%, whereas the concentration of ethanol present in the apple sample was 5.82% and ethanol present in banana was 4.50%. With the increase in sugar concentration, the ethanol production increased significantly (Table 4; Fig.5)

Table 4: Concentration of bioethanol obtained from different samples

Sr.no	Sample	Concentration (%)
1	Apple waste	5.82%
2	Grapes waste	4.51%
3	Banana waste	4.50%

Fig 5: comparison of production of bioethanol concentration.



Conclusion: The concentration of ethanol present in the apple waste had higher concentration.

IV. DISCUSSION

After 36hrs of incubation a total volume of 200ml of 48% ethanol was obtained from a total volume of 1000ml of substrate after distillation. Using higher grade distillation assembly a more concentrated product can be recovered by re distillation. The higher concentrated ethanol can be used as a bio fuel and it reduces no toxic gases to the environment. The substrates used are very cheap raw material and the process was found to be very easy and less cost effective and a common man can involve himself develop it as a small-scale industry. As it releases no toxic residue so it is ecofriendly and hence the left outs can be disposed in the soil. agar utilization during fermentation is one of the important physiological features of yeast strains used for industrial ethanol production. The present study was carried out to check for the sugar tolerance of the yeast isolated from rotten banana waste, because tolerance of yeast strains to higher sugar concentration is necessary for optimum ethanol production. All strains were able to withstand the highest sugar concentration (25%) the growth observed in 25% sugar containing medium was much less than that containing 15% sugar concentrations and growth similar that containing 20% sugar concentrations. At high sugar concentration osmotic pressure increases in the fermenting medium. and this is inhibitory to many yeasts. High osmotic pressure is associated with increased stress on the organisms that grow in the medium. This rapid change in the physiological condition during fermentation imposes water and ethanol stress on yeast cells. Moreover, sugar concentration is also critical in fermentation process and influencing the rate of production and the yield in addition to physiological growth of yeasts.

V. CONCLUSION

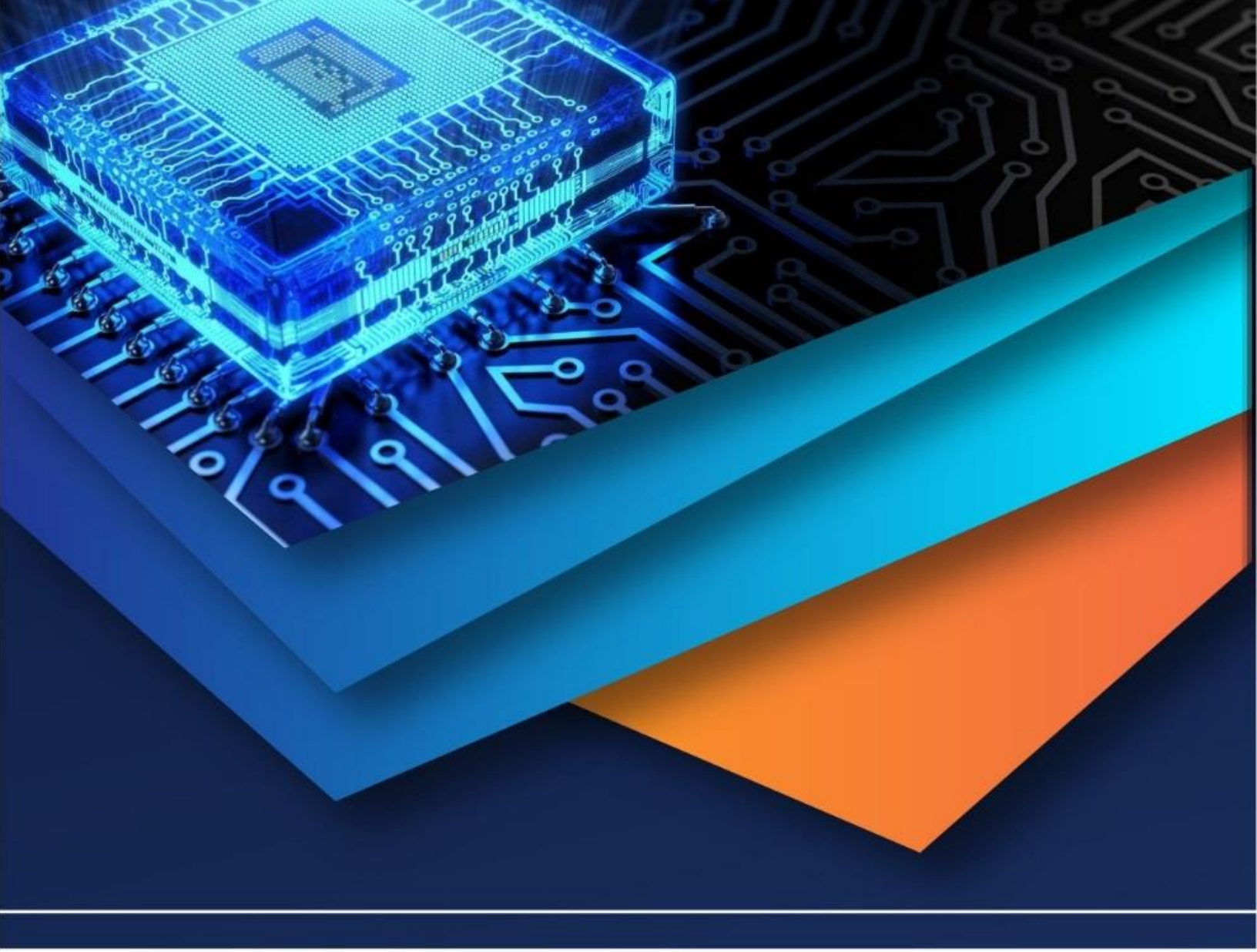
The result of this work has shown that different fruit wastes can serve as raw material for the production of bioethanol. From this comparative study, it is clear that the maximum yield of ethanol was obtained from grape wastes at pH 5.6, temperature 30°C, specific gravity 0.876, and concentration of 5.82% which is appropriately close the constant value of ethanol. Finally, comparative studies of bioethanol production from different fruit wastes shows that apple waste has higher efficiency than other fruit wastes such as grapes and banana or mixed fruit wastes. This process is cost effective and does not yield any toxic residues. Hence, it can be run as a Small-Scale industry.

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REFERENCE

[1] Akin-Osanaiye B.C. Nzelibe H.C., Agbaji A.S. (2005). Production of ethanol from Carica papaya (pawpaw) agro waste: effect of saccharification and different treatments on ethanol yield. Afr. J. Bioethanol. 4(7): 657 659
[2] Association of Official Analytical Chemists (A.O.A.C (1970 and 1980).Official Methods of Analysis 13th Ed. Horwits, W., Ed. A.O.A.C., Washington.
[3] O. R. Inderwildi, D. A. King (2009). Quo Vadis Biofuel. Energy &Environment science. Cimpeanu, C., Campeanu G., Begea, M., Vladescu, M., Cornea, C.P
[4] Bioethanol production by new thermotolerant Romani strains.'' Romanian Biotechnological Leters, vol. 15.no.3, 2010, pp. 5310-5316.an yeast
[5] Mir Naiman Ali and Mohammed Mazharuddin Khan "Screening identification and characterization of alcohol tolerant potential bioethanol producing yeasts," Current Research in Microbiology and Biotechnology, vol.2, no.1, 2014, pp. 316-324.
[6] Chandra J. Panchal. Yeast Strain Selection", Marcel Dekker, Inc..1997, pp 349.
[7] Savitree Limtong, Chutima Sringiew and Wichien Yongmanitchai, Production of fuel ethanol at high temperature from sugar cane juice by a newly isolated Kluyveromyces marxianus." Bioresoure Technologv, vol.98, no.17, 2007, pp. 3367-3374 http:/dx.doi.org/10.1016/j.biortech.2006.10.044
[8] Wickerham, L.J. "Taxonomy of yeasts." Tech.Bull, U.S. Dept.Agr. Washington DC, USA, 1951 pp. 1029
[9] Kurtzman CP, and Fell JW..."The Yeasts. A Taxonomic study. North-Holland, Amsterdam, 1999, 1055
[10] Brooks, A.A.. "Ethanol production potential of local yeast strains isolated from ripe banana peels," African Journal of Microbiolog Research, vol.7, no.20. 2008, pp. 3749-3752.
[11] Ameh JB, Okagbue RN, Ahman AA, "Isolation and characterization of local yeast strains for ethanol production, Niger J Technol Res.vol.1,1989, pp. 47-52
[12] Sathees Kumar, R., Shankar, T. and Anandapandian, K.T.K. Characterization of alcohol resistant yeast Saccharomyces cerevisiae isolated from Toddy." International Research Journal of Microhiology.2011. pp. 399- 405.
[13] M V.Ranade. "Isolation. Identification and Screening of the Yeast Flora from Indian Cashew Apple for sugar and Ethanol Tolerance."International Journal of Biotechnology for Wellness Industries, vol.1, 2012, pp. 259-265.
[14] J. Goettemoeller, A. Goettemoeller (2007). Sustainable Ethanol: Biofuels, Biorefineries, Cellulosic Biomass, Flex-Fuel Vehicles, and Sustainable Farming for Energy Independence (Brief and comprehensive account of the history, evolution and future of ethanol). Prairie Oak Publishing, Maryville, Missouri. ISBN 9780978629304.
[15] Reddy, V., "Production of ethanol from mango (Mangifera indica L) fruit juice fermentation", Research Journal of Microbiology, vol.2, (10), pp.763-769, 2007.
[16] Singh, A., and Jain, V.K., "Batch fermentation of cane molasses for ethanol production by Zymomonas mobilis",Journal of Indian Chemical Engineering, vol.37, pp.80-94, 1995.
[17] Grassi, M., "Modern Bioenergy in the European Union", Renewable Energy, vol.16, pp.985-990, 1999.
[18] Asli, M.S., "A study on some efficient parameters in batch fermentation of ethanol using Saccharomyces cerevisiae extract from fermented siahe sardasht pomace", African Journal of Biotechnology, vol.9 (20), pp.2906-2912, 2010.
[19] O. R. Inderwildi, D. A. King (2009).Quo Vadis Biofuel. Energy & Environmental Science 2: 343.

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