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Research Article Standardization of Condition for Fermentation and Maturation of Wine from Strawberry (*Fragaria ananassa*) by Isolated *Saccharomyces cerevisiae* Strain

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Abstract

Background and Objective: Wine an alcoholic beverage is prepared by fermenting different fruit juice with appropriate processing and addition. The conventional process of wine making involve the fermentation of fruit juices. For that the present study was planned to standardize of condition for fermentation and maturation of wine from Strawberry (*Fragaria ananassa*). **Materials and Methods:** Strawberry used for wine preparation. The wine was prepared in aseptic conditions and stored at room temperature till 90 days in a tighed container. The physico-chemical parameters namely p^H, Brix, acidity, ascorbic acid, total phenol, flavonoid, total sugar, colour estimation, anthocynine, moisture, alcohol estimation were analyzed at 0, 15, 30, 45, 60, 75 and 90 days in control and experimental sample. **Results:** According to the results moisture (g%), flavonoid (mg%), Anthocynine (mg%), Vitamin-C (mg%) and alcohol estimation (μL%) increasing in control and P^H, acidity (%), colour estimation (mg%), Brix (%), total sugar (g%) decreasing in control. The results of moisture (g%), Alcohol estimation (μL%), Vitamin-C (mg%) increasing in sample and P^H, acidity (%), colour estimation (mg%), Total phenol (mg%), Brix (%), total sugar (g%), flavonoid (mg%), anthocynine (mg%) decreasing in sample and P^H, acidity (%), colour estimation (mg%), Total phenol (mg%), Brix (%), total sugar (g%), flavonoid (mg%), anthocynine (mg%) decreasing in sample. **Conclusion:** The wine was one of the functional fermented foods and has many health benefits like anti-aging effect, improvement of lung function, reduction in coronary heart disease and destruction of cancer cells etc.

Key words: Strawberry (Fragaria ananassa), Saccharomyces cerevisiae, wine, fermentation, maturation

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Fruits are very essential for our health and from ancient time fruits are used for the production of different alcoholic beverages like wine. The term wine is applied to the product made by alcoholic fermentation from fruits or fruit juices using yeast as inoculums. Under the generic term of "wine", there is a diversity of quality which is quite unique among the products and determined mainly by interaction between grapes, yeasts and technology. Wine is considered to be the one of the oldest alcoholic beverages produced by the process of fermentation. Fermentation is relatively low energy preservation process which increases the self-life and decreases the need of refrigeration or any other forms of food preservation technology. Tropical wines are subjectively perceived as inferior in quality on the basis of flavour, aroma, odour and colour¹.

Strawberry (Fragaria ananassa) fruit is healthiest fruit in the world. There are so many reasons for select strawberry fruit of this study. Strawberry boost your immunity, prevent heart disease, prevent constipation, diabetes, help in weight loss and very important function of strawberry fruit is help to fight against cancer producing cells and prevent cancer disease. Strawberry is one of the most perishable fruits, being very susceptible to mechanical injury, water loss, decay and physiological deterioration. The sugars (fructose, glucose and sucrose), organic acids (predominantly citric acid) and Phenolic compounds (Anthocynine and flavanols) give strawberry its characteristic taste, while more than 360 volatile compounds distinguish its aroma. The major constituent like other fruits, however, is water whereas Phenolic compounds such as Queraitin, catechin, chlorogenic, ferulic and ellagic acid are present in Strawberry aroma is mainly determined by a complex mixture of esters, aldehydes, alcohol and sulphur compounds².

Saccharomyces cerevisiae mostly present in sugar containing foods. Sugar mostly present in fruits like banana, apple, papaya, mango, etc. The utilization of isolated strain of Saccharomyces cerevisiae is an important strategy for keeping the quality and assuring the reproducibility of wine features. The utilization of strains isolated from specific regions could be even more interesting because of their high adaptation to their own climate conditions and grapes. Even more these strains are usually associated to particular wine characteristics that frequently identify specific wines and regions³.

Fermentation during this process, the yeast require various nutrients for optimal performance and survival. It should be remember that grape juice is not an optimal growth medium for yeast and yeast subjected to considerable stress during fermentation. Optimal yeast growth occurs under aerobic conditions, with an adequate nutrient supply at temperature of 28-30 °C. A side from glucose and fructose as source of carbon and energy, yeast requires these nutrients during a wine fermentation: Assimilable nitrogen as ammonium [NH⁺⁺⁺⁺] and amino acids, phosphate, growth factors or vitamins, Minerals" survival factors or long chain fatty acids and sterols.

The present study was carried out for standardization of condition for fermentation and maturation of Wine from Strawberry (*Fragaria ananassa*) by Isolated Saccharomyces Cerevisiae strain.

MATERIALS AND METHODS

Collection of sample: Strawberry (*Fragaria ananassa*) were collected from local market of Petlad. These were washed in sterile water.

Preparation of starter culture: The present study one microbial species was used i.e., *Saccharomyces cerevisiae*. The strain was isolated from Papaya fruit. First take loop of Papaya inner surface by sterile wire loop and striking on Yeast Extract Potato Dextrose Agar slant. Incubate for 48 h at 30°C. Then perform gram staining for identification and characterization of colony. Then pure culturing of *Saccharomyces cerevisiae* strain. After this take one colony of *Saccharomyces cerevisiae* strain and inoculate in Yeast Extract Potato Dextrose Agar culture and then incubate for 48 h at 30°C. After this, take O.D of culture. If O.D become 1.0000 at 600 nm so, this is conformation of *Saccharomyces cerevisiae* produced in culture. Purified cultures were routinely maintain every 15 days on Yeast Extract Potato Dextrose agar and kept⁴ at -4°C.

Preparation of wine: Selection of well graded strawberries. Remove the green part of strawberries and then cut into pieces and blended into slurry. This slurry called "must". Take sterile 500 mL capacity jars. For control [C] pour 200 mL of must and 50 g of sugar. For sample [S₁] take 200 mL of must, 50 g of sugar and 10% culture of *Saccharomyces cerevisiae* then close the lids. After this pasteurization procedure was performed. Then labelled the jars and incubate the jars for 90 days. Analyzed the sample in 15 days interval.

Physico-chemical analysis: The samples as well as controls were analyzed various physico-chemical characteristics at interval of 15 till 90 days. Brix [%] was measured using a Abbe

refractometer, titratable acidity and P^H, moisture, total sugar, colour estimation were measured as per the AOAC⁵ method. Alcohol estimation was measured by Gas chromatography-MS. Weight 25 g of sample and make up volume 100 mL with distilled water. Then filter the sample if sample have turbidity centrifuge the sample at 2500 rpm for 15 min. Clear solution of sample take as a Gas Chromatography sample. Inject 1 µL sample in Gas Chromatography. After 20-25 min at 120-140°C the alcohol estimation recorded in graph⁶.

Anti-oxidant analysis: Strawberry is rich source of anti-oxidants like total phenol, flavonoid, anthocyanin and ascorbic acid [Vitamin-C]. These all parameters were analyzed by prescribed methods. Total phenol estimation was analysed by Nazir *et al.*⁷ method. The flavonoid estimated by Lafka *et al.*⁸ method. Ascorbic acid content was estimated by volumetric method. Anthocynine estimation was performed by AOAC⁵ method.

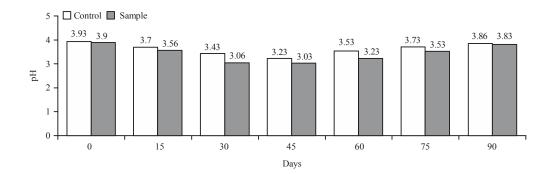
Statistical analysis: All results were presented as Mean \pm SEM difference variable where tested for significant t-test [MS-office Excel] using a level of significant of p \leq 0.01 and p \leq 0.05 level.

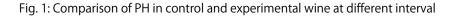
RESULTS AND DISCUSSION

Changes in P^H: Figure 1 showed the mean value comparisons of P^H in control and experimental sample in different days interval. Overall comparison indicated that the p^H was slowly decreasing in control and sample during day 45. After day 45 p^H value was increasing in control and experimental sample. In 0 and 90 days P^H was non significant difference (p>0.05). However in 15, 30, 45, 60 and 75 days P^H was was highly significant difference (p<0.01).

Changes in moisture: Figure 2 showed the comparison of moisture content in control as well as experimental sample at different days interval. The moisture was significant difference in 0, 30, 75 and 90 days and others days sample was non-significant difference.

Changes in brix (%): Figure 3 showed the mean value comparison of brix (%) in control and experimental sample at different days interval. Overall comparison indicate that the brix (%) was decreasing in control and sample during 45 days. After 60 days brix (%) of control was decrease but brix (%) of sample increasing. After 75 and 90 days brix (%) of control was increasing but brix (%) of sample decreasing. Brix (%) was highly significant difference in all days except 30 days.





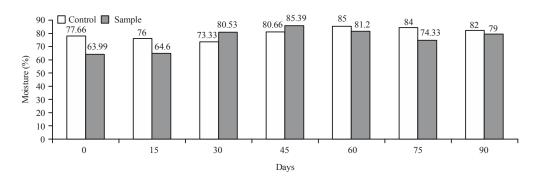


Fig. 2: Comparison of moisture content in control and experimental wine at different interval

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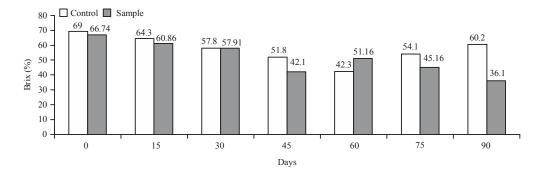


Fig. 3: Comparison of brix in control and experimental wine at different interval

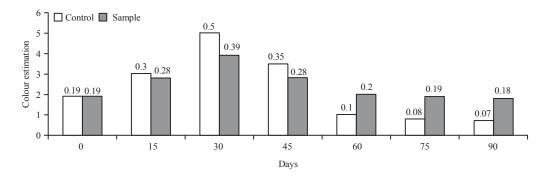
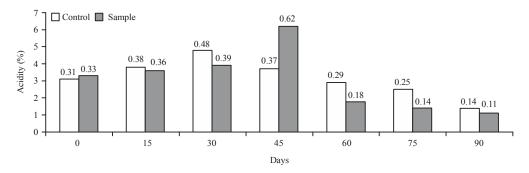
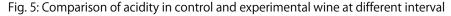


Fig. 4: Comparison of colour estimation in control and experimental wine at different interval





Changes in colour estimation: Figure 4 showed the mean value of comparison of colour estimation control and experimental sample wine at different days interval. The colour estimation was range 0.07-0.5% found during wine production. Overall comparison indicate that the colour intensity increasing at 30 days and after day 45 the colour intensity decreasing in control and experimental sample. All experimental sample was highly significant difference at days interval.

Changes in acidity (%): Figure 5 showed the mean value of comparison of acidity in control and experimental sample wine at different days interval. The acidity range was 011-0.62% found during wine production. Overall comparison

indicates that the acidity values were initially low than after fermentation carried out it was increasing at 30 days and after days gone it was decreased in all control and experimental sample. There was highly significant difference at 30, 45 and 60 days and significance difference at 15, 75 and 90 days except 0 day.

Changes in phenol content: Figure 6 showed the mean value of comparison of total phenol in control and experimental sample wine at different days interval. The total phenol was range 15.16-78.43 mg% found during wine production. There was highly significantly different. Overall comparison indicates that the total phenol was initially higher than after fermentation carried out it was slowly decreasing up to 90 days.

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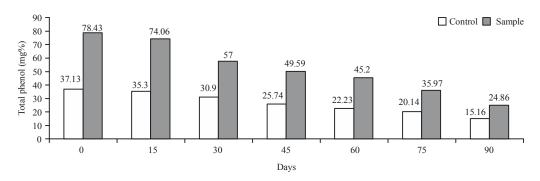


Fig. 6: Comparison of total phenol in control and experimental wine at different interval

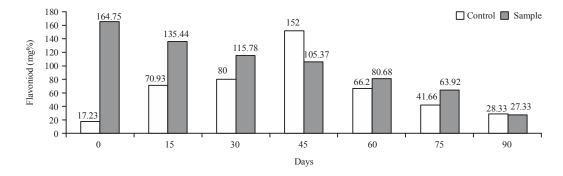


Fig. 7: Comparison of flavonoid in control and experimental wine at different interval

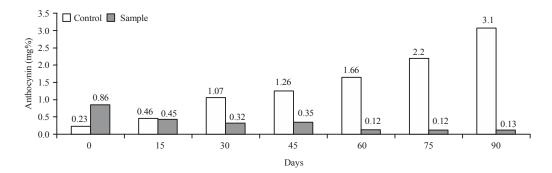


Fig. 8: Comparison of anthocynine in control and experimental wine at different interval

Changes in flavonoid content: Figure 7 showed the mean value of comparison of flavonoid in control and experimental sample wine at different days interval. The flavonoid was range 17.23-164.37 mg% found during wine production. In 60 days there was significantly different. In 75 days there was highly significantly different. After in day 90 the flavonoid content was higher in control 28.33 mg% compare to sample 27.33 mg%. There was non-significantly different.

Changes in anthocynine content: Figure 8 showed the mean value of comparison of Anthocynine in control and experimental sample wine at different days interval. The

anthocyanin was range 0.23-0.86 mg% found during wine production. There were highly significantly different. Overall comparison indicates that the Anthocynine contain in sample decreasing, there was increasing Anthocynine in control.

Changes in total sugar: Figure 9 showed the mean value of comparison of acidity in control and experimental sample wine at different days interval. The total sugar range was 6.07-34.83 gm% found during wine production. There were highly significantly different. Overall comparison indicated that total sugar was initially higher than after fermentation carried out it was slowly decreasing up to 90 days.

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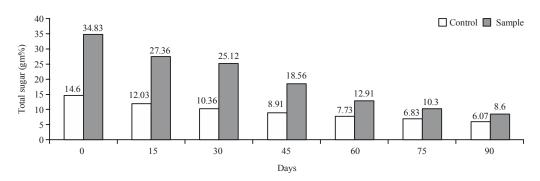


Fig. 9: Comparison of total sugar in control and experimental wine at different interval

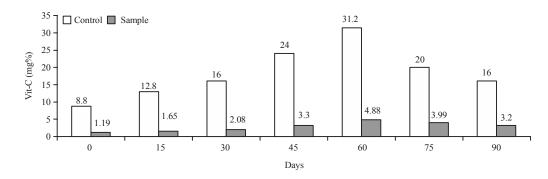


Fig. 10: Comparison of ascorbic acid [Vit- C] in control and experimental wine at different interval

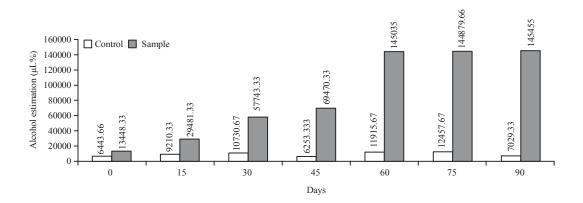


Fig. 11: Comparison of alcohol estimation in control and experimental wine at different interval

Changes in ascorbic acid [vitamin-c]: Figure 10 showed the mean value of comparison of ascorbic acid in control and experimental sample wine at different days interval. There was highly significantly different.

Changes in alcohol content: Figure 11 showed the mean value of comparison of alcohol estimation in control and experimental sample wine at different days interval. The alcohol estimation range was 6253.333-1454.55 μ L% found during wine production. There were highly significantly different. Overall comparison indicate alcohol production in

sample was simantanously increasing but alcohol production in control was decreasing in 45 or 90 days.

CONCLUSION

Wine is natural product resulting from a number of biochemical reaction, which begin during ripening of the grapes and continues during harvesting, throughout the alcoholic fermentation, clarification and after bottling. A large quantity of wine are produced and consumed all over the world. Italy and France being the leading wine producing countries. Although production of wine is largely made by the fermentation of grapes juices yet it has also been practiced widely from fruits such as apples, cherries, currants, peaches, plums, strawberries, etc. In India, the total production of wine was 8.5 million bottles per year indicating a wide scope of production of wine from different fruits.

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