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Design and Development on Pomegranate Cider Vinegar

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ABSTRACT
Vinegar is the fermented product which consisting about 5-20% of acetic acid, prepared by fermentation of alcohol with the help of Acetobactor species. Vinegar is the food additive & used in ketchup, salad dressing and in pickle. It is also used as food preservatives. The use of vinegar as a medicine is firstly carried out by Hippocrates. He used vinegar as treatment of wound healing. Different types of Vinegar are present in the world. The different possible medicinal uses of Vinegar are reviewed in the present review. Vinegar is used as Anti- diabetic, Antimicrobial, Antioxidant, Antitumor, Antiobesity, it reduces Cholesterol levels, In present Article we have reviewed all previous works which were carried out with Vinegar including method of preparation, Characterization of Vinegar and uses of Vinegar etc. The Vinegar is prepared with the help of different methods like artificial method and natural fermentation method etc. The characterization of Vinegar is mainly carried out with the help of following tests pH, Titratable acidity, Alcohol content, Total soluble solid etc.
Keywords: *Vinegar, Pomegranate, Acetic acid fermentation,*

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INTRODUCTION

Vinegar is prepared by different raw materials and methods. Wine cider fruit musts barley, or pure alcohol is used as a substrate. Vinegar as Antidiabetic acetic acid present in vinegar may prevent the complete digestion of complex carbohydrates by either accelerating gastric emptying time or by increasing the uptake of glucose by tissues resulting in reduction of blood glucose level. The word Vinegar is derived from the French Vin(wine) and Aigre (sour).Vinegar is a sour and sharp liquid used as a condiment and food preservative. Natural vinegars contains small amount of tartaric acid, citric acid and other acids.[1,2,3]

Vinegar is used ancient times and is an important element in European, Asian and other cuisines. vinegar is also used as cleansing agent. Natural vinegar is a superior food additive over synthetic vinegar as it carries essential amino acids from its fruit source and is reported to act as a medicine for aches and gastric trouble. Vinegar can more than an addition to any dish; it can be a source of income, a promising business. In this paper work aims to elaborate the different types of vinegars and major vinegar processing method used for commercial production. As an example a flow chart for the production of cane vinegar is included and possible improvement for cane vinegar production are discussed.[4,5].

Vinegar plays an important role in salad dressings, ketchup, hot sauce and other sauces. This need demands industrial fermentation systems capable of producing a large amount of vinegar. These systems must maintain reliable controls and optimum conditions for acetic acid bacteria fermentation Many techniques have been developed to improve industrial production of vinegar. Most try to increase the speed of the transformation of ethanol into acetic acid in the presence of the acetic acid bacteria today, the most common technology for the vinegar industry is based on the submerged culture with diverse technical modifications which try to improve the general fermentation conditions (aeration, stirring, heating, etc.).[6,7,8,9]

Vinegar bacteria, also called acetic acid bacteria, are members of the genus Acetobacter and characterized by their ability to convert ethyl alcohol, C_2H_5OH, into acetic acid, CH_3CO_2H, by oxidation as shown below:

Anaerobic Aerobic

$2C_2H_5OH \rightarrow 2CH_3CHO \rightarrow 2CH_3CO_2H + 2H_2O$

Most microorganism strains derived from vinegar factories are able to oxidize acetic acid to carbonic acid gas and H_2O (over-oxidation) and thus square measure classified within the genus Acetobacter Common kinds of vinegar embody white distilled vinegar, cider vinegar, wine vinegar, rice vinegar, and malt vinegar.

Main component of formation of vinegar are liquids containing alcohol, AAB, jaggery, dhataki flowers, different microbes etc. ethanolic fermentation process plays important role in formation of vinegar. Depending on physicho-chemical properties like temp, pH and microbes form in the material bio-transformation occurs. Microbial species concerned in fermentations could vary from yeast and carboxylic acidbacterium (LAB) to molds and AAB. The microorganisms concerned within the elaboration of vinegars area unit chiefly yeasts and AAB. The former being responsible for the ethanolic fermentation and the latter needed for the acidification.[10-14].

Objective of the Study

1. To design and develop and characterize Pomegranate Cider vinegar.
2. To understand the process of vinegar preparation.
3. To improve patient compliance.

MATERIAL AND METHODS

Materials: Pomegranate, Dhatki flowers, Jaggery, AAB culture.

Alcoholic preparation of Pomegranate is prepared by following process the plant part was identified and collected from local market, separate the seeds for alcoholic preparation, and then kept it for the period of 45days.

Methods:

The vinegar was prepared in 18 batches. Firstly take the accurately weighed quantity of pomegranate seed, then add dhataki flowers and jaggery in tightly closed amber colored bottle for the process of fermentation in an aerobic process. Then keep that container without disturbing it for 45 days. At the different time interval i.e (0,5,10,15,30,45)in that days we take the evaluation of each batch. In that this period of time due to chemical reactions the alcohol is produced. After the preparation of alcohol in that the AAB is added and kept that container as it is for more than 15days to form vinegar.[14,15]

Vinegar is a diluted acetic acid made by a two-step bioprocess. In the first step, fermentable sugars are converted into ethanol with help of microorganisms. In the next step, AAB oxidize the alcohol to vinegar in an aerobic process. General formula for preparation of vinegar is stated in following table. in that each batch having 6 formulation are prepared.

Table 1: Blank formulation

Sr. No.	Name of ingredients	Quantity taken
1	Jaggery	300 gm
2	Dhatki flowers	20 gm
3	Purified water	q.s. to 500ml

Batch I-(F1)

Table 2: Pomegranate Formulation

Sr. No.	Name of ingredients	Quantity taken
1	Pomegranate	100 gm
2	Jaggery	300 gm
3	Dhatki flowers	20 gm
4	Purified water	q.s. to 500 ml

Batch II-(F2)

Table 3: Aqueous extract Formulation

Sr. No.	Name of ingredients	Quantity taken
1	Pomegranate	100 gm
2	Purified water	q.s. to 500 ml

Batch III-(F3)

Antidiabetic Effect :

Insulin sensitivity has been enhanced through vinegar action in 19% of humans with type 2 diabetes and 34% of humans with prediabetes (Johnston and others 2004). New studies in both animals and humans have revealed that vinegar may be useful for diabetic management. In rats, the result of vinegar on blood sugar in persons, the area under the insulin response curve decreased 20% after consumption of sucrose

co-administered with. Many placebo-controlled experimentations have definite the blood glucose reducing or "antiglycemic" effect of vinegar. Some systems have been studied to clarify the outcome of vinegar on blood glucose concentrations. Acetic acid in vinegar may avoid the complete breakdown of complex carbohydrates by either quickening gastric emptying or by improving the uptake of glucose by muscles resulting in reduced blood glucose stages.

RESULTS AND DISCUSSION

Preformulation Study

Calibration curve of Ellagic acid:

Table No 4: calibration of Ellagic acid

SR NO	CONCENTRATION (µg/ml)	ABSORBANCES
1	0	0
2	10	0.1252
3	20	0.2038
4	30	0.3424
5	40	0.4521
6	50	0.5112
7	60	0.6874

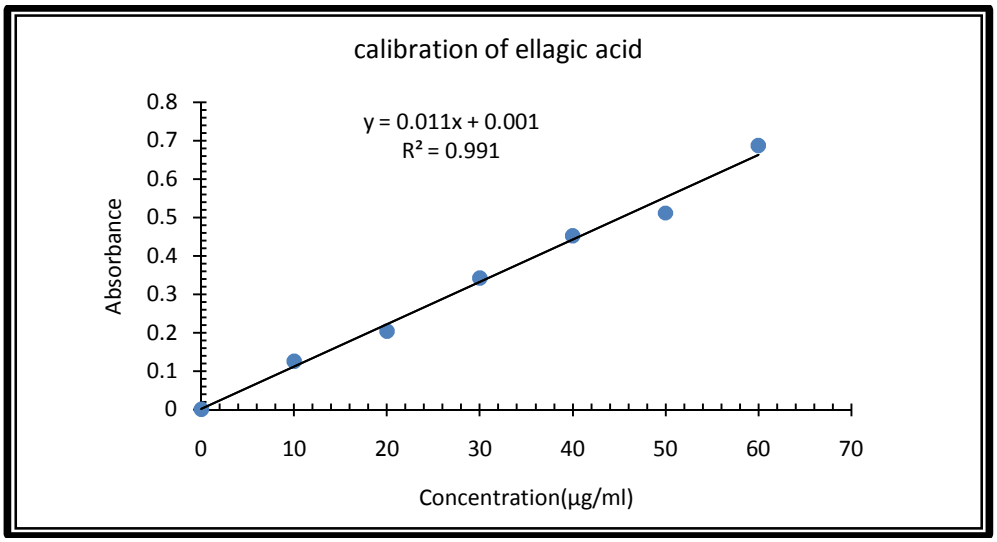

Fig no:1 Calibration curve of ellagic acid

Slope: 0.011, Intercept: 0.001,Correlation coefficient (R2): 0.991 From the calibration curve, equation can be given as Y=0.011x + 0.001 [E.Q.] The drug obeys Beer-Lambert's law in the range of 2-10µg/ml The calibration graph of Ellagic acid was obtained by plotting Absorbance vs. Concentration. Table shows the absorbance value of Ellagic acid. The calibration curve is shown in figure: Calibration curve shows straight concentrations in2-10µg/ml at 254nm. The calculation of concentration of extraction in vinegar studies are based on this calibration curve. Slope: 0.011 Intercept: 0.001Correlation coefficient (R2): 0.991 From the calibration curve, equation can be given as Y=0.011x + 0.0015 [E.Q.] The drug obeys Beer-Lambert's law in the range of 2-10µg/ml.

The calibration graph of Ellagic Acid was obtained by plotting Absorbance vs. Concentration. Table shows the absorbance value of Ellagic acid. The calibration curve is shown in figure: Calibration curve show straight concentrations in2-10µg/ml at 254nm. The calculation of concentration of extraction in vinegar studies are based.

UV Analysis:

Absorbance of Pomegranate Formulation (F2): 0.0452

y= mx+c

y= 0.011x+0.0015

0.0452= 0.011x+0.0015

x= 3.972µg/ml

Absorbance of Aqueous Extract Formulation (F3): 0.0389

y= mx+c

y= 0.011x+0.0015

0.0389= 0.011x+0. 0015
x= 3.4 µg/ml

Total Pomegranate content:

Table No 5: Total Pomegranate content

Total Pomegranate Content		
F1	**F2**	**F3**
----	3.972 µg/ml	3.4µg/ml

IR study:

IR spectra of standard Ellagic acid:

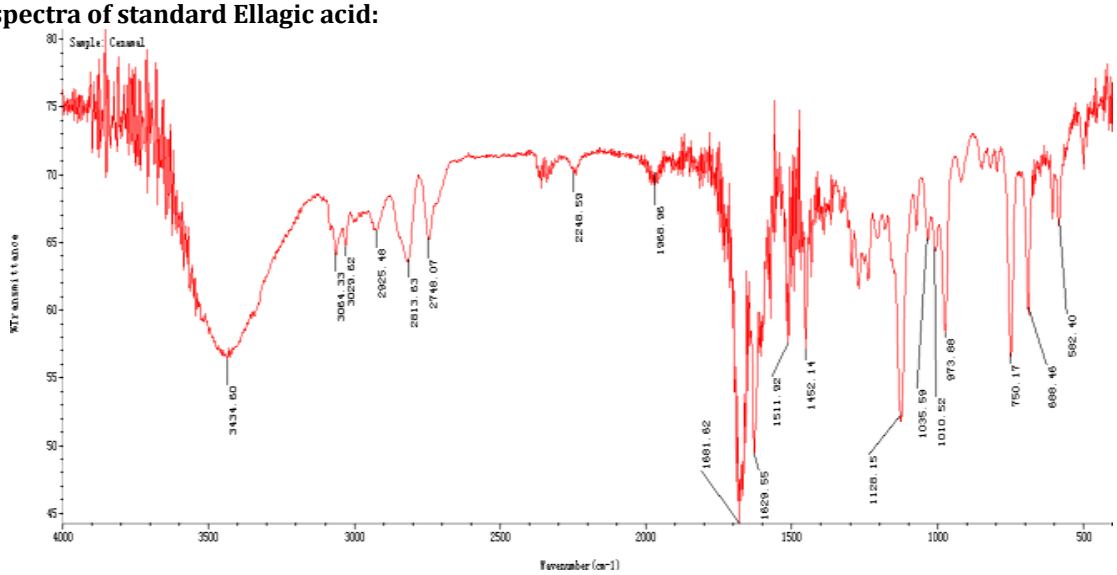

Fig No. 2: IR spectra of standard Ellagic acid.

Table 6: Interpretation of IR of standard Ellagic acid.

Sr. No.	FUNCTIONAL GROUP	INDICATION	OBSERVED WAVE NUMBER (cm^{-1})	REPORTED WAVE NUMBER(cm^{-1})
1	Aromatic ring	C=C stretch	1452.14	1600-1400
2	Aldehyde	-C=O stretch	1681.62	1700-1600
3	Alkane	C-H stretch	1128.15	1300-800

The IR spectrum showed the dominant characteristics peak of Ellagic acid. Especially C=C, -C=O and C- H stretching at 1452.14, 1681.62cm^{-1} and 1128.15cm^{-1}on that stretching vibrations which confirms drug sample was Ellagic acid.

IR spectra of Pomegranate Vinegar:

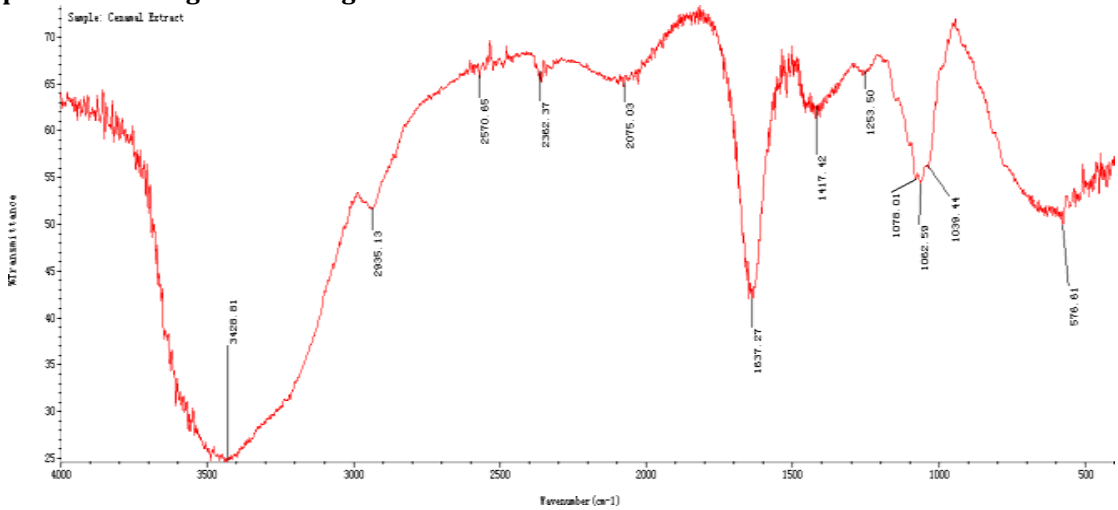

Fig.3: IR spectra of Pomegranate Vinegar

Table No.7: Interpretation of IR of Pomegranate Vinegar.

Sr. No.	FUNCTIONAL GROUP	INDICATION	OBSERVED WAVE NUMBER(cm^{-1})	REPORTED WAVE NUMBER(cm^{-1})
1	Aromatic ring	C=C stretch	1417.42	1600-1400
2	Aldehyde	-C=O stretch	1637.27	1700-1600
3	Alkane	C-H stretch	1253.50	1300-800

The IR spectrum of Pomegranate vinegar showed the dominant characteristics peaks. Especially C=C, -C=O and C-H stretching at 1417.42cm^{-1}, 1637.27cm^{-1} and 1253.50$cm^{-1}$by comparing this interpretation with standard IR Spectra confirms Pomegranate vinegar sample contains Ellagic acid

Evaluation test:

pH:

pH of all batches is measured with digital pH meter.

Table No:8 pH batch of F1,F2, and F3

Period in days	F1	F2	F3
0	5.1±0.34	3.9±0.12	4.2±0.03
5	5.0±0.51	3.8±0.23	4.2±0.03
10	4.8±0.26	3.7±0.21	4.1±0.04
20	4.7±0.34	3.5±0.22	4.0±0.03
30	4.7±0.21	3.4±0.45	3.9±0.65
45	4.6±0.12	3.4±0.15	3.8±0.02

During fermentation pH of formulation F1 &F2is get decreased. During fermentation process the fermentation microorganism produces different organic acid due to that pH is get deceased.

Total Alcohol Content:

Table No:9 alcohol content batch of F1,F2, and F3

Period in days	F1	F2	F3
0	----	----	----
5	1.192±0.42	1.152±0.12	0.032±0.34
10	2.185±0.03	3.511±0.05	0.061±0.23
20	4.851±0.45	5.613±0.43	0.132±0.02
30	5.711±0.11	6.711±0.55	0.141±0.14
45	7.883±0.21	8.312±0.65	0.163±0.01

The total alcohol content is calculated with the help of specific gravity.

The batch 1 and 2 shows increase in alcohol content during fermentation. In fermentation microorganism causes metabolism of sugar molecules and produces the alcohol.

Titratable Acidity:

Titratable acidity is calculated with the help of simple titration method by using 0.1 N NaOH

Table No:10 Titratable acidity of batch of F1,F2, and F3

Period in days	F1	F2	F3
0	----	----	----
5	0.512±0.45	0.541±0.56	0.251±0.11
10	0.713±0.12	0.611±0.43	0.261±0.05
20	0.913±0.34	0.912±0.54	0.271±0.45
30	1.513±0.07	1.651±0.32	0.291±0.53
45	1.751±0.31	1.691±0.04	0.312±0.22

In our work titratable acidity of the batch 1 and 2 get increased. Titratable acidity is nothing but the measure oftotal of all natural acidspresent in the fermented product. In our workwe are dealing with vinegar so after 45 days there is increased value of titratable acidity is observed.

Total soluble solid(^{0}B):

Accurately weigh 50 ml of vinegar sample was transformed into a china dish. Which is evaporated on water bath, and dried at 105^{0}c for 3 hr.(in oven) and then after cooling dish containing residue kept in decsicator for 30 min and weigh immediately.

Table No:11 Solid Content of batch of F1,F2, and F3

Period in days	F1	F2	F3
0	26.12±0.42	20.23±0.35	6.43±0.15
5	21.83±0.22	18.12±0.22	5.34±0.14
10	17.35±0.36	15.36±0.43	4.60±0.12
20	15.12±0.21	12.13±0.32	4.32±0.11
30	13.56±0.61	10.95±0.43	3.02±0.06
45	10.25±0.72	9.23±0.34	2.55±0.03

TSS of pomegranate was found to be decreased. this may due to the converson of sugar into alcohol during the process of fermentation.

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CONCLUSION

Vinegar is mainly used for food flavoring and preservative, recent investigations demonstrate the potent bioactive effects of vinegars which may benefit human health. The vinegar shoes different medicinal activities like reduction in high blood pressure, it prevents oxidation, lovers the increased sugar level and in CVS abnormalities. Person using vinegar everyday shows that vinegar has reduction activity of high blood sugars which is helpful to patients of diabetes.

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