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METHODS FOR PREPARATION OF SELECTED CEREALS BASED ALCOHOLIC BEVERAGES PRE-MIXED WITH FRUIT JUICES

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ABSTRACT

The first of its kind study resulted in stable alcoholic beverages using varied starch sources from vegetables, millets, and cereals. The method was green in nature as waste generated was recycled. Fermented beverages have been produced and consumed all over the world and over a very long time span. Man discovered that sugar solutions of different origins, if left standing rather warm, will start fermenting spontaneously into an alcoholic beverage. The requisite microorganism, *Saccharomyces* yeasts is abundant almost everywhere and will do its duty, by producing alcohol. Fermented beverages can be divided into two groups, wines and beers, broadly defined

Key words: alcoholic fermentation, beverages, Saccharomyces

INTRODUCTION

Fermented beverages have been produced and consumed all over the world and over a very long time span. Man discovered that sugar solutions of different origins, if left standing rather warm, will start fermenting spontaneously into an alcoholic beverage. The requisite microorganism, *Saccharomyces* yeasts is abundant almost everywhere and will do its duty, by producing alcohol. Fermented beverages can be divided into two groups, wines and beers, broadly defined. Wines are fermented from various fruit juices containing fermentable sugars. Beers come from starch-containing products, which undergo enzymatic splitting by diastase, malting, and mashing, before the fermentable sugars become available for the yeasts. Vodka is clear distilled liquor composed of water and ethanol. Vodka is made from a fermented substance of grain, rye, wheat, potato, or sugarbeet molasses. There are various different kinds of fermented alcoholic beverages found all around the world *Bouza is* produced in Egypt from malt of grain sorghum that resembles millet, also Wheat beer, Rye beer such as kalja and sahti, Oat beer, Rice beer, Sorghum and millet beer and Maize beer are forms of traditional alcoholic beverages relished in different regions of the world.

Potato is the most widely grown vegetable crop in the country, India ranks fourth worldwide in the production of potato. During its season potato is considered as a glut crop in and hence can be

utilized for production of alcoholic beverages like Vodka which is majorly produced using potato. In the study carried out, an attempt was made to develop fermented wheat, barley, sorghum, rye and potato (in different combinations) based alcoholic undistilled beverages using *Saccharomyces* *cerevisiae*. The fermented product thus obtained was blended with fruit juices (apple juice, grape juice and mixed fruit juice) to prepare novel pre-mixed liquor-fruit juice blends and the consumer acceptability of the new product was analyzed.

It was observed that the total recovery from the potato fermentation mash was very low. To overcome this, commonly available cereals like rye, sorghum, barley, and wheat which are given less importance and are readily available were mixed in different combinations with potato resulting in higher yields. This is so justified by the fact that the potato mash lacks the presence of amylases which convert starch into sugar making it available to yeast for fermentation. While the combination of cereals and potato mash gave higher recovery due to the activation of the amylases present in the cereals during the malting process making it a higher yielding process. The outcome of the study is a novel Potato cereal based undistilled alcoholic beverage which can be suitably blended with selected processed fruit juices masking its haziness and giving rise to alcoholic premixed fruit juice blends. Till date there are no reports of an undistilled alcoholic beverage prepared with different combinations of cereals (rye, sorghum, barley, and wheat) and potato and their blending with selected packed fruit juices. Hence making our product a novel and unconventional from the league available beverages.

The alcoholic product obtained when distilled resulted in very low recovery and hence it was used for the preparation of a novel undistilled potato cereal based beverages which can be blended with fruit juices which masks its haziness and results in development of an unconventional undistilled alcoholic premixed fruit juice blends.

The high nutritive value of fruit juices and the high demand of alcoholic beverages being relished in the society. The product thus developed will meet the consumer need by providing both fruit nutrition as well as alcoholic relishment. The quality testing of the product was carried out by determining the total viable count. The product was found to be shelf stable and could be stored under refrigeration or could be tetra packed.

METHODOLOGY

MATERIALS REQUIRED

Raw Materials:
Cereals: Wheat, Rye, Sorghum, Barley and Potato,
Culture: *Saccharomyces cerevisiae* (Dry Yeast pellets)
Processed fruit juices- Apple juice, Grape juice, Mixed Fruit juice.
Equipments: Shaking Incubator, BOD incubator and Refrigerator.
Glassware: Conical Flasks, funnel, Glass rods, muslin cloth, Glass Bottles.

METHODOLOGY:

Fermentation process was initiated by selecting suitable grains of economic importance, which include wheat, barley, sorghum, and rye rich in complex starches and sugars. They were soaked overnight and then kept for germination for a period of 48 hours. The germinated grains were washed and dried in the oven at a temperature of 60ºC and ground coarsely in a blender to obtain the malted grain powder. This malted grain powder was used to prepare the *fermentation mash.* Prior to mash preparation, all the starches were cooked at boiling temperatures in a pressure cooker for 5-10 minutes in distilled water.

The fermentation mash was prepared in sterile conical flasks (500ml) using different combinations of the malted grains namely: wheat + rye (1:1), wheat + sorghum (1:1), wheat + barley (1:1), rye +

sorghum (1:1), rye + barley (1:1), sorghum + barley (1:1), wheat + rye + sorghum + barley (1:1:1) along with their controls which include wheat, rye, barley, sorghum respectively.

For the set with potatoes fermentation process was initiated by procurement and sorting of potatoes. They were washed and were cooked in a pressure cooker for 5-10 mins. The potatoes were peeled and were mashed well in a blender. This was used as the fermentation mash for potato control. The different combinations prepared using malted grains and potato are: wheat + rye + potato (1:1:1), wheat + sorghum + potato (1:1:1), wheat + barley + potato (1:1:1), rye + sorghum + potato (1:1:1), rye + barley + potato (1:1:1), sorghum + barley + potato (1:1:1), wheat + rye + sorghum + barley + potato (1:1:1:1) along with the control as potato.

To both the sets one with different cereal combinations and the other with different potato and cereal combinations, 10% sugar was added and was inoculated with dry yeast (*Saccharomyces cerevisiae*) @ 2.5% and the final volume was made upto 350 ml by using water and a suitable head space was maintained. Addition of KMS @ 350 ppm was done to avoid risks of bacterial contamination, followed by incubation at 30ºC ± 2 for 24 hours under shaking conditions to allow growth of yeast cells after which next day the flasks were incubated at the same temperature (30ºC ± 2) for 15days without mixing, the pH of the mash was maintained at pH 4.5±2. The flasks were monitored daily to check for development of any contamination.
Once the mash has fermented to maturity bearing clear and some suspended liquids bearing pleasing alcoholic odor, the clear upper layer was decanted and collected in a clean flask which was then filtered to remove any suspended particles. The obtained alcoholic supernatant was inbottle pasteurized at 90ºC for 3-4mins and stored in clean flasks for ageing at 3-4ºC for a period of 15 days. The aged decant was filtered to remove the sedimentation developed with the help of fine filter/double layered muslin cloth. The clear filtrate thus obtained was stored in clean sterile glass bottles. A known volume of the alcoholic filtrate was used for the preparation of blends with selected aseptically packed fruit juices. Thus, pre-mixed liquor-fruit juice blends were prepared (@ 1:1 ratio) using selected processed fruit juices like apple, grape, and mixed fruit juice. Blends were stored in clean and dried glass bottles and were refrigerated for further ageing. The fruit juice unblended filtrate was taken as Control. Sensory Evaluation was carried out of the developed pre-mixed liquor-fruit juice blends on a hedonic scale and pameters of taste, flavor, mouth feel, and OAA of the product. Quality testing of the prepared products was carried out by estimating the Total Viable Count on zero day and weekly basis

The alcoholic decant obtained could also be distilled to obtain corresponding vodka .Distillation was carried out at a temperature of 80 ºC ± 2 the steam rises from decant and was cooled in the collection tube which converts the alcohol back to liquid. This distilled alcohol drips down into a collection chamber and accumulates until the decant has been exhausted of its alcohol.

Flow Chart: Preparation of fermentation mash using different combinations of cereals

Preparation of Malted Grain Powder

Selection of grains (wheat, barley, sorghum, and rye)

↓

Washing (Ambient temperature clean water)

↓

Soaking (overnight at room temperature in clean water)

↓

Germination (48 hrs incubation at room temperature, 24 ± 2)

↓

Washing (Clean luke warm water)

↓

Drying (Hot air oven @ 60ºC for 12hrs)

↓

Cooking (Pressure cooker for 10 mins)

↓

Mashing (Blender)

↓

Malted grains mash obtained

Preparation of Fermentation Mash

Fermentation mash prepared in sterile conical flasks(500ml) using different combinations of the malted grains namely: wheat + rye (1:1), wheat + sorghum (1:1), wheat + barley (1:1), rye + sorghum (1:1), rye + barley (1:1), sorghum + barley (1:1), wheat + rye + sorghum + barley (1:1:1) along with their controls which include wheat, rye, barley, sorghum respectively.

↓

Sugar addition (10%)

↓

Inoculation of Dry yeast (*Saccharomyces cerevisiae*) @ 2.5%

↓

Final Volume made upto 350 ml (using distilled water), head space maintained

↓

KMS addition @ 350 ppm

↓

Incubation at 30ºC ± 2 for 24 hours under shaking conditions

↓

Incubation at 30ºC ± 2 for 15 days under static conditions

↓

pH maintained at 4.5±2

↓

Monitoring flasks daily to check for development of contamination.

Decantation and Filtration

Decantation and Filtration of clear upper layer

↓

Inbottle pasteurization at 90 ºC for 3-4 mins, storing in clean flasks.

↓

Ageing at 3-4ºC for 15 days.

Preparation of pre-mixed liquor-fruit juice blends

Filtration of aged decants (fine double layered muslin cloth.)

↓

Storing alcoholic filtrate (clean sterile glass bottles)

↓

Blending of alcoholic filtrate and selected aseptically packed fruit juices (apple, grape, and mixed fruit juice) in 1:1 ratio

↓

Fruit juice unblended filtrate was Control.

↓

Storing blends (clean dried glass bottles)

↓

Ageing at 3-4ºC

↓

Quality testing (Total Viable Count)

↓

Sensory Evaluation carried on Hedonic scale

Distillation

The decant is distilled at a temperature of 80 ºC ± 2 the distilled alcohol drips down into a collection chamber and accumulates until the decant has been exhausted of its alcohol.

Flow Chart: Preparation of fermentation mash using potato and cereals
Preparation of Potato mash

Procurement and sorting of potatoes

↓

Washing

↓

Cooking (Boiling, 10 mins)

↓

Peeling

↓

Mashing (Blender) to a uniform paste

Preparation of Malted Grain Powder

Selection of grains (wheat, barley, sorghum, and rye)

↓

Washing (Ambient temperature clean water)

↓

Soaking (overnight at room temperature in clean water)

↓

Germination (48 hrs incubation at room temperature, 24 ± 2)

↓

Washing (Clean luke warm water)

↓

Drying (Hot air oven @ 60ºC for 12hrs)

↓

Cooking (Pressure cooker for 10 mins)

↓

Mashing (Blender)

↓

Malted grains mash obtained

Preparation of Fermentation Mash from potato

Fermentation mash prepared in sterile conical flasks(500ml) using different combinations of potato and malted grains namely: potato + sugar (control), wheat + rye + potato (1:1:1), wheat + sorghum + potato (1:1:1), wheat + barley + potato (1:1:1), rye + sorghum + potato (1:1:1), rye + barley + potato (1:1:1), sorghum + barley + potato (1:1:1), wheat + rye + sorghum + barley + potato (1:1:1:1).

↓

Sugar addition (10%)

↓

Inoculation of Dry yeast (*Saccharomyces cerevisiae*) @ 2.5%

↓

Final Volume made upto 350 ml (using distilled water), head space maintained

↓

KMS addition @ 350 ppm

↓

Incubation at 30ºC ± 2 for 24 hours under shaking conditions

↓

Incubation at 30ºC ± 2 for 15 days under static conditions

↓

pH maintained at 4.5±2

↓

Monitoring flasks daily to check for development of contamination.

Decantation and Filtration

Decantation and Filtration of clear upper layer

↓

Inbottle pasteurization at 90 ºC for 3-4 mins, storing in clean flasks.

↓

Ageing at 3-4ºC for 15 days.

Preparation of pre-mixed liquor-fruit juice blends

Filtration of aged decants (fine double layered muslin cloth.)

↓

Storing alcoholic filtrate (clean sterile glass bottles)

↓

Blending of alcoholic filtrate and selected aseptically packed fruit juices (apple, grape, and mixed fruit juice) in 1:1 ratio

↓

Fruit juice unblended filtrate was Control.

↓

Storing blends (clean dried glass bottles)

↓

Ageing at 3-4ºC

↓

Quality testing (Total Viable Count)

↓

Sensory Evaluation carried on Hedonic scale

Distillation

The decant is distilled at a temperature of 80 ºC ± 2 the distilled alcohol drips down into a collection chamber and accumulates until the decant has been exhausted of its alcohol.

RESULTS

The Fermentation mash which was prepared using different combinations of malted grains namely: wheat + rye (1:1), wheat + sorghum (1:1), wheat + barley (1:1), rye + sorghum (1:1), rye + barley (1:1), sorghum + barley (1:1), wheat + rye + sorghum + barley (1:1:1). In each set 100gms of mashed grains in a ratio 1:1 were added along with 10% sugar, 2.5% dry yeast and 300ml water making the final volume of the fermentation mash to 400ml. KMS @ 350 ppm was added to the flasks, followed by incubation at 30ºC ± 2 for 24 hours under shaking conditions. After which the flasks were incubated at the same temperature for 15 days without shaking conditions. pH of the mash was maintained at 4.5±2. Their respective controls of wheat, rye, barley, Jowar were also maintained in the same conditions, containing 50gm of malted grain mash along with 5% Dry yeast and 50 ml water making up the total volume to 100ml.

Flasks incubated at 30ºC for a period of 15 days

After a period of 15 days, the clear upper layer obtained was decanted and collected in a clean flask which was filtered to remove any suspended particles.

Alcoholic Supernatant Decant

The decant was inbottle pasteurized at 90ºC for 3-4mins and stored in clean flasks for ageing at 3-4ºC for a period of 15 days which was then used for preparation of blends.

Flasks Kept for Ageing

The retained biomass containing dead yeast and undigested grain mash was stored and utilized for preparation of media for microbial source.

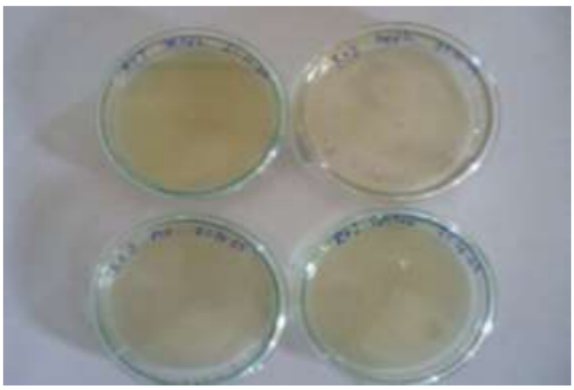
Retained Yeast Biomass

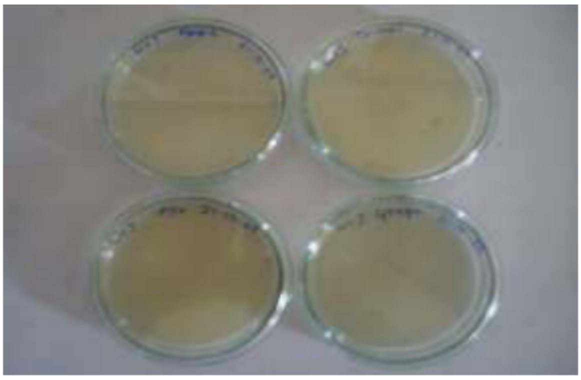
The aged decant was used for the preparation of pre-mixed liquor-fruit juice blends with selected aseptically packed fruit juices like apple, grape, and mixed fruit juice (@ 1:1 ratio). The fruit juice unblended filtrate was taken as Control. Blends were stored in clean and dried glass bottles and were refrigerated for further ageing.

Wheat + Jowar Premixed liquor blends with Apple, Grape & Mix fruit juice

Ragi + Jowar Premixed liquor blends with Apple, Grape & Mix fruit juice

Quality testing of the prepared products was carried out by estimating the Total Viable Count on zero day and weekly basis.





TVC result: Nearly zero bacterial growth during elf life studies

Sensory Evaluation was carried out of the developed pre-mixed liquor-fruit juice blends on a hedonic scale and pameters of taste, flavor, mouth feel, and OAA of the product was analyzed

SENSORY EVALUATION SCORE CARD

Ragi + Jowar Blends

Blends	# People	Colour	Flavor	Odour	Mouth feel	OAA
50:50						
R + J						
Control	**1**	**6.0**	**7.0**	**8.0**	**8.0**	**7.0**
	2					
Apple	**1**	**7.0**	**8.5**	**8.5**	**8.0**	**8.5**
	2					
Grape	**1**	**7.5**	**7.0**	**7.5**	**7.0**	**7.5**
	2					
Mixed Fruit	**1**	**8.0**	**8.5**	**8.5**	**8.0**	**8.5**
	2					

Wheat + Jowar Blends

Blends	# People	Colour	Flavor	Odour	Mouth feel	OAA
50:50						
W + J						
Control	**1**	**6.0**	**7.0**	**7.0**	**8.0**	**7.0**
	2					
Apple	**1**	**7.0**	**8.0**	**7.5**	**8.0**	**7.75**
	2					
Grape	**1**	**7.5**	**7.0**	**6.5**	**7.0**	**6.75**
	2					
Mixed Fruit	**1**	**8.0**	**8.5**	**8.75**	**8.25**	**8.5**
	2					

S.No	Sample	Number of colonies		
		Zero day	1 week	2 weeks
1	R + J			

	Control	ND		
	Apple	ND		
	Grape	ND		
	Mixed fruit	ND		
2	W + J			
	Control	ND		
	Apple	ND		
	Grape	ND		
	Mixed fruit	ND		

Results for Total Viable Count
The results of percentage yield for the process are as tabulated below.

S.No	Sample (100gm)	Additives	Volume of Fermentation Mash	Volume of Decant	Weight of Biomass	% Yield
1	Wheat+ Ragi (1:1)	Water(300ml) + 10%Sugar + 2.5%Dry Yeast	400ml	250ml	145gm	62.5%
2	Wheat+jowar (1:1)			300ml	98gm	75.0%
3	Wheat+Barley (1:1)			285ml	110gm	71.25%
4	Ragi+Jowar (1:1)			285ml	112gm	71.25%
5	Jowar+Barley (1:1)			300ml	93gm	75.0%
6	Ragi+barley (1:1)			290ml	105gm	72.5%
7	Ragi+Wheat+Jowar+Barley (1:1:1:1)			310ml	87gm	77.5%

S.No	Control (50gm)	Additives	Volume of Fermentation Mash	Volume of Decant	Weight of Biomass	Yield
1	Wheat	Water(50ml) + 2.5%Dry Yeast	100ml	55ml	42gm	55.0%
2	Jowar			50ml	47gm	50.0%
3	Barley			40ml	64gm	40.0%
4	Ragi			45ml	65gm	45.0%
5	Wheat+Sugar (10%)			56ml	42gm	56.0%

RESULTS: With Potato

The fermentation mash was prepared in sterile conical flasks (500ml) using different combinations of potato and malted grains namely: potato + sugar (control), wheat + rye + potato (1:1:1), wheat + sorghum + potato (1:1:1), wheat + barley + potato (1:1:1), rye + sorghum + potato (1:1:1), rye + barley + potato (1:1:1), sorghum + barley + potato (1:1:1), wheat + rye + sorghum + barley + potato (1:1:1:1) along with the control as potato. 10% sugar, 2.5% dry yeast and 300ml water were added making the final volume of the fermentation mash to 400ml. KMS @ 350 ppm was added to the flasks, followed by incubation at 30ºC ± 2 for 24 hours under shaking conditions. After which the flasks were incubated at the same temperature for 15 days without shaking conditions. pH of the mash was maintained at 4.5±2.

After a period of 15 days, the clear upper layer obtained was decanted and collected in a clean flask which was filtered to remove any suspended particles.

Filtered Decant kept for ageing

The decant was inbottle pasteurized at 90ºC for 3-4mins and stored in clean flasks for ageing at 3-4ºC for a period of 15 days which was then used for preparation of blends.

The retained biomass containing dead yeast and undigested grain mash.

Retained Yeast Biomass

The aged decant was used for the preparation of pre-mixed liquor-fruit juice blends with selected aseptically packed fruit juices like apple, grape, and mixed fruit juice (@ 1:1 ratio). The fruit juice unblended filtrate was taken as Control. Blends were stored in clean and dried glass bottles and were refrigerated for further ageing.

Potato Premixed liquor blends with apple, grape & mix fruit juice

Quality testing of the prepared products was carried out by estimating the Total Viable Count on zero day and weekly basis

S.No	Sample (100gm)	Additives	Volume of Fermentation Mash	Volume of Decant	Weight of Biomass	Yield
1	Potato(control)	Water(300ml) + 10%Sugar + 2.5%Dry Yeast	400ml	232ml	160gm	58%
2	Wheat+ Ragi+Potato (1:1:1)			290ml	107gm	72.5%
3	Wheat+jowar+Potato (1:1:1)			320ml	78gm	80.0%
4	Wheat+Barley+Potato (1:1:1)			285ml	110gm	71.25%
5	Ragi+Jowar+Potato (1:1:1)			280ml	117gm	70.0%
6	Jowar+Barley+Potato (1:1:1)			300ml	96gm	75.0%
7	Ragi+barley+Potato (1:1:1)			305ml	91gm	76.25%
8	Ragi+Wheat+Jowar+Barley+Potato (1:1:1:1:1)			320ml	78gm	80.0%

CONCLUSION

The first of its kind study resulted in stable alcoholic beverages using varied starch sources from vegetables, millets, and cereals. The method was green in nature as waste generated was recycled.

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