International Journal of Chemical Studies

P-ISSN: 2349–8528
E-ISSN: 2321–4902
IJCS 2017; 5(4): 541-543

Received: 03-05-2017
Accepted: 04-06-2017

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Pectinase enzymatic maceration on colour attributes of jamun (*Syzigium cuminii* L.) wine

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Abstract
The pectolytic enzyme treatments of red grape musts could accelerate the extraction of pigments and phenols. Enzyme treatment produces a brighter, more brilliant colour, and the colour stability is greatly increased. The extraction of phenolic compounds usually occurs during the pulping of the mixture in the course of alcoholic fermentation. The pectinase enzymatic maceration techniques were tried on different *must* like juice, pulp with skin and pulp+skin+seed. The mean values of lightness (*L**) and yellowness (*b**) decreased with increasing storage period whereas, redness (*a**) values showed an increasing trend in jamun wine. Significantly higher *L** value was observed in T_4 (5.80, 2.73 and 0.97, respectively) in fresh, three and six months after storage. Whereas, maximum *b** value was recorded in T_4 (0.25% pectinase with juice) during 6 months after storage. The light colour of the wine may be due to absence of skin and seed in the must during fermentation hence there was lesser extraction of colouring pigments and phenols which may impart dark colour to the wine.

Keywords: Pecinase, Jamun wine, Phenolic compounds, Redness, Yellowness and Storage

1. Introduction
Jamun (*Syzgium cumini* L.) is an evergreen tree belongs to family Myrtaceae, native to India and Indonesia. It is also grown in other areas of Southeast Asia including Malaysia, Myanmar, Pakistan and Afghanistan. The jamun tree starts flowering in February- March. Tree bears fruits in May- June. The berry is oblong, ovoid and shinning crimson black (rich in anthocyanin pigment, an anti-oxidant) when fully ripe. The Jamun fruits are universally accepted for its medicinal properties especially for curing diabetes because of its effect on the pancreas (Joshi, 2001) [5]. The fruit, juice and seed contain a biochemical called 'jamboline' which is believed to check the pathological conversion of starch into sugar in case of increased production of glucose (Amerine, *et al.*, 1980a) [1].
Pectic enzymes are used to increase extraction of colour compounds, but the desired effect has not always been observed (Graham *et al.*, 2004) [4]. Wines produced by enzyme treatment were higher in polymeric anthocyanins, polymeric phenols and catechin than control wines, but not in monomeric anthocyanin content. The enzyme treated wines also had increased aroma and flavor intensity, and enhanced bitterness and astringency characteristics. Because of its high medicinal property, a novel method to utilize the whole fruits for the preparation of wine with pectinase enzyme attempted to maximize quality and health benefits (Neubeck, 1971; 1981) [7, 8].

Material and methods
The present investigation on influence of pectinase enzymatic maceration on quality of jamun wine was carried out during the period of 2012 - 2014.
The three types of *must viz.*, juice, pulp + skin and pulp + skin + seed were exposed to cold soaking of about 8°C for five days and after which the must was normalized for ambient temperature and the yeast culture was added to all the samples at the rate of 0.2% and kept for aerobic fermentation for 24 hours. For enzymatic maceration, the three *musts viz.*, juice, pulp + skin and pulp + skin + seed were ameliorated with 0.25 and 0.50 per cent pectinase for 12 hours.

Treatment details

Treatments	Fermentation with
T_1:Control	Juice
T_2:Control	Pulp + Skin
T_3:Control	Pulp + Skin + Seed
T_4:Pectinase at 0.25%	Juice
T_5:Pectinase at 0.25%	Pulp + Skin
T_6:Pectinase at 0.25%	Pulp + Skin + Seed
T_7:Pectinase at 0.50%	Juice
T_8:Pectinase at 0.50%	Pulp + Skin
T_9:Pectinase at 0.50%	Pulp + Skin + Seed

Note: For all the treatments juice was used as *must.* TSS, pH and aerobic fermentation were maintained at 24°B, 3.2 and one day, respectively.

Instrumental colour (L* a* b* C^o and h^o) analysis

The colour of the samples was measured using a Lovibond colour meter (Lovibond RT300, Portable spectrophotometer, The Tintometer Limited, Salisbury, UK) fitted with 8mm diameter aperture, D65 illuminant and10 0 observer. The instrument was calibrated using the black and white tiles provided. Colour was expressed in Lovibond units *L** (Lightness/darkness), *a** (redness/greenness), *b** (yellowness/blueness), *C**(Chroma) and h^o (Hue). Samples of jamun wine were directly placed under the aperture of the colour meter. Three measurements were performed for each sample in three replicates and values were averaged.

Results and discussion

Significantly higher *L** value was observed in T_4 (5.80, 2.73 and 0.97, respectively) in fresh, three and six months after storage, which contained a must of jamun juice with 0.25 per cent pectinase enzyme. The light colour of the wine may be due to absence of skin and seed in the *must* during fermentation hence there was lesser extraction of colouring pigments and phenols which may impart dark colour to the wine. However, the minimum *L** value was observed in T_8 (2.59, 0.61) at initial and 3 MAS, respectively, and T_3 (0.14) at 6 months after storage, lower *L** value indicating darkness it may be due to contact with skin and seed during fermentation coupled with pectinase would have extracted more colour rendering the wine dark. Similar observations were reported by Kotecha (2010b) [6] and Olasupo and Obayori, (2003) [9].

Maximum values for *a** were recorded in the treatment T_2 (2.09) at initial, T_1 (2.34) at 3MAS and T_4 (2.70) 6 months after storage. On contrary, the lower value for *a** was recorded in the treatments T_5 (0.54) in initial, T_3 (0.45) at 3 MAS and T_8 (0.63) at 6 months after storage. This trend clearly indicates pectinase is an enzyme which is generally used for maximum extraction of juice from fruit and also for clarification of wine. Similar observations were reported by Bhajipale (1997) [2] in karonda and Chikkasubbanna *et al.* (1990) [3] in mulberry wine.

The maximum *b** value of 1.11, 1.16, and 1.08, respectively was recorded in the treatment with 0.25 per cent pectinase and juice *must* (T_4) at 0, 3 and 6 months after storage, respectively. On the other hand the minimum *b** values were recorded in T_1 (-0.80 and -0.68, respectively) at initial and 3 MAS, T_2 (-1.27) at 6 months after storage. This trend is justifiable because the wine in the T_1 contains only juice.

Significantly maximum *C** value was observed in treatment T_1 (1.24, 2.46 and 4.28, respectively) at 0, 3 and 6 months after storage. However, minimum *C** value was observed in T_3 (0.24, 0.48 and 1.01, respectively) at 0, 3 and 6 months after storage. Highest value of *C** indicate saturation or vividness in jamun wine.

Significantly maximum h^o was observed in treatment T_2 (349.58) in fresh, T_9 (355.00 and 363.64, respectively) at 3 and 6 months after storage. However, minimum h^o was observed in T_1 (11.81, 14.43 and 35.94, respectively).

Table 1: Influence of pectinase and *must* type on *L** and *a** values of jamun wine during ageing

Treatments #	*L** value			*a** value			*b** value		
	Ageing in months								
	Initial	3 MAS	6 MAS	Initial	3 MAS	6 MAS	Initial	3 MAS	6 MAS
T_1	4.66^{ab}	2.50^{abc}	0.27^{cd}	2.06^{a}	2.34^{a}	0.98^{bc}	-0.80^{f}	-0.68^{d}	-0.46^{d}
T_2	4.81^{ab}	2.58^{ab}	0.32^{c}	2.09^{a}	2.03^{ab}	1.19^{bc}	-0.53^{e}	-0.59^{d}	-1.27^{e}
T_3	4.77^{ab}	2.28^{abcd}	0.14^{d}	0.23^{f}	0.45^{f}	1.01^{bc}	0.05^{b}	0.12^{bc}	0.29^{bc}
T_4	5.80^{a}	2.73^{a}	0.97^{a}	0.25^{f}	1.65^{bc}	2.70^{a}	1.11^{a}	1.16^{a}	1.08^{a}
T_5	3.82^{bc}	1.77^{abcde}	0.72^{b}	0.54^{e}	1.36^{cde}	1.06^{bc}	-0.36^{d}	-0.24^{cd}	-0.17^{cd}
T_6	3.21^{bc}	1.24^{de}	0.74^{b}	0.77^{d}	1.23^{cde}	1.03^{bc}	-0.33^{d}	-0.23^{cd}	-0.19^{cd}
T_7	3.24^{bc}	1.50^{bcde}	0.88^{a}	1.02^{c}	0.96^{def}	1.29^{bc}	-0.87^{f}	0.62^{ab}	0.54^{ab}
T_8	2.59^{c}	0.61^{e}	0.34^{c}	1.07^{c}	0.69^{ef}	0.63^{c}	-0.34^{d}	-0.26^{cd}	-0.20^{cd}
T_9	3.85^{bc}	1.35^{cde}	0.88^{a}	1.61^{b}	1.55^{bcd}	1.45^{b}	-0.22^{c}	-0.11^{cd}	-0.09^{cd}
Mean	4.08	1.84	0.58	1.07	1.36	1.26	-0.25	-0.02	-0.05
S. Em±	0.57	0.41	0.05	0.04	0.23	0.24	0.003	0.20	0.20
CD at 5%	1.68	1.20	0.14	0.11	0.67	0.73	0.08	0.62	0.58

#Refer methodology for treatment details

*L** = Dark - Light, *a** = Green – Red and *b** = Blue –yellow

Different alphabets within the column are significantly different (p=0.05) according to Duncan's Multiple Range Test

Table 2: Influence of pectinase and *must* type on yellowness (*b**) values of jamun wine during ageing

Treatments #	*C** value			*h°*		
	Ageing in months					
	Initial	3 MAS	6 MAS	Initial	3 MAS	6 MAS
T_1	1.24^{a}	2.46^{a}	4.28^{a}	11.81^{f}	14.43^{d}	35.94^{c}
T_2	1.06^{ab}	2.13^{ab}	3.61^{ab}	349.58^{a}	343.12^{ab}	351.84^{ab}
T_3	0.24^{e}	0.48^{e}	1.01^{c}	345.25^{b}	344.90^{ab}	347.73^{b}
T_4	0.86^{bc}	2.03^{abc}	4.14^{ab}	12.82^{f}	33.50^{c}	41.75^{c}
T_5	0.54^{d}	1.38^{bcd}	3.04^{b}	338.45^{c}	349.49^{ab}	357.56^{ab}
T_6	0.46^{de}	1.25^{cde}	3.07^{b}	342.93^{bc}	349.63^{ab}	358.10^{ab}
T_7	0.45^{de}	1.15^{de}	1.31^{c}	26.10^{e}	31.40^{c}	36.49^{c}
T_8	0.62^{cd}	0.74^{de}	1.04^{c}	321.24^{d}	338.17^{b}	350.28^{b}
T_9	1.03^{ab}	1.56^{bcd}	1.73^{c}	339.08^{c}	355.00^{a}	363.64^{a}
Mean	0.72	1.47	2.58	232.14	239.96	249.26
S. Em±	0.08	0.29	0.38	1.71	4.02	4.26
CD at 5%	0.29	0.84	1.13	5.08	11.94	12.65

#Refer methodology for treatment details

*C** = Chroma and h^{o}= hue angle

Different alphabets within the column are significantly different (p=0.05) according to Duncan's Multiple Range Test

Conclusion

Wines prepared from enzymatic maceration shown higher in anthocyanins and phenols than control wines. The enzyme treated wines also had increased aroma and flavor intensity, and enhanced bitterness and astringency characteristics. These treatments resulted in increases on the organoleptic (colour) characteristics.

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