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ORIGINAL ARTICLE

Effect of co-fermentation with *Saccharomyces cerevisiae* and *Torulaspora delbrueckii* or *Metschnikowia pulcherrima* on the aroma and sensory properties of mango wine

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Abstract The effect of pure and mixed culture fermentations by *Saccharomyces cerevisiae* and *Metschnikowia pulcherrima* or *Torulaspora delbrueckii* on physicochemical and sensory qualities of the mango wines were investigated under laboratory conditions. *S. cerevisiae* produced alcohol at 11.9% from sugar, while one *M. pulcherrima* and two *T. delbrueckii* strains (NCIM and IIHR) produced alcohol at 3.8, 7.2 and 6.9% (v/v) in their mono-cultures, respectively. However, in their co-fermentation, they produced similar alcohol content to that of *S. cerevisiae* mono-culture: 11.04, 11.53, 11.35% (v/v) for *S. cerevisiae* + *M. pulcherrima* and *S. cerevisiae* + *T. delbrueckii* strains (NCIM and IIHR), respectively. The formation of major volatile compounds in mango wine was assessed by gas chromatography and the analysis showed that the wines from mixed cultures presented differences in the concentration of volatiles. Further, the wines produced by co-fermentation indicated that these non-*Saccharomyces* strains could be used with *S. cerevisiae* starter cultures to increase glycerol ranging from 5.4 to 7.6 and to reduce volatile acidity from 1.28 to 0.18 as well as the total acidity from 5.5 to 3.8 (g/l) of the final wines. These characteristics positively influenced the sensory qualities of the wines produced with mixed cultures, which was reflected in the preferences of these wines by panelists. The results emphasized the potential of employing indigenous non-*Saccharomyces* yeast strains for the production of mango wines with improved flavor.

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Introduction

Mango (*Mangifera indica* L.) fruit, commonly called "King of fruits", is native to southern Asia. Mango currently ranks fifth in total production among major fruit crops worldwide. The world production of mango fruit is estimated to be over 23.4×10^6 MT per year. India ranks first among world's mango fruit producing countries, accounting for 54.2% of the total mango fruit produced worldwide. Mango fruits are processed into various products at various maturity levels, of which wine is one (Kumar et al. 2009). Wine is produced from mango juice and is mostly fermented by *Saccharomyces cerevisiae*. However, there is limited information in the literature on mango wine co-fermentation with *Saccharomyces cerevisiae* and non-*Saccharomyces* yeast strains.

Spontaneous grape juice fermentation is carried out by a succession of different yeast populations. During the initial stages of fermentation, the low ethanol-tolerant species are predominant. In the course of the fermentation process, they are replaced by high ethanol-tolerant *S. cerevisiae* and related species (Ciani and Picciotti 1995). However, the initial activity of non-*Saccharomyces* yeasts in must fermentation is considered important for the final aromatic profile of wines, because these yeasts are responsible for different enzymatic reactions developing a wide range of volatile and nonvolatile end products, such as higher alcohols, esters, acids, and carbonyl compounds important to the sensory characteristics of wines (Romano et al. 2003; Xu et al. 2006; Lee et al. 2010). Local geography also plays a role in the flora of the vineyard, characterized by specific

Saccharomyces and non-*Saccharomyces* in different areas that are adapted to the local conditions (Querol et al. 1994). This results in the production of unique wines with characteristic flavors, aromas and alcoholic strengths. Furthermore, the contribution of the yeast depends on several additional parameters, such as the fermentation temperature, the quality of the grape juice and the concentration of additives, including sulfur dioxide (Parapouli et al. 2010).

Several authors have reported the influence of non-*Saccharomyces* yeast species like *Hanseniaspora uvarum, Metschnikowia pulcherrima, Torulaspora delbrueckii, Kluyveromyces thermotolerans, Candida stellata* and *Kloeckera apiculata* on wine quality under usual wine-making conditions (Ciani and Maccarelli 1998). Generally, it is accepted that *Kloeckera apiculata* is the predominant non-*Saccharomyces* yeast species found in grape must. This is usually associated with volatile acidity production, and hence its positive contribution to wine quality is low (Gil et al. 1996). However, it was shown that *Candida pulcherrima* also occurs in high numbers in must (Jolly et al. 2003) and is not normally associated with volatile acidity production, but can form relatively high concentrations of esters (Bisson and Kunkee 1991). It was reported that *M. pulcherrima* produces β-glucosidase which is able to release aromatic compounds from odourless grape juice precursors under winemaking conditions (Rodríguez et al. 2007) and have a positive effect on the taste and aroma of alcoholic beverages (Parapouli et al. 2010). *Torulaspora delbrueckii* was also one of the non-*Saccharomyces* yeasts occurring in spontaneously fermented wine (Zott et al. 2008). It was reviewed that the mixed culture fermentation of *T. delbrueckii* was an attractive perspective in reducing volatile acidity and acetic acid in the wine (Ciani et al. 2006, 2010). Suresh et al. (1982) reported the existence of about 15 different groups of yeasts in the fresh mango juice, spontaneously fermenting juice and fermented juice, including *M. pulcherrima, K. apiculata*, some *Candida* and *Pichia* spp. from two varieties of mango *Banginapalli* and *Totapuri* cultivars, and *T. delbrueckii* has also been isolated routinely from stored mango brine (unpublished data from the Indian Institute of Horticulture Research, IIHR).

Although the literature contains numerous reports on using mixed yeast strains for wine making, little has been published about mixed fermentation of *S. cerevisiae* and non-*Saccharomyces* yeast strains in mango wine-making, or on the influence of mixed cultures on the formation of volatile components. In the present work, a laboratory-scale study was performed to investigate the fermentation behavior of *S. cerevisiae* and *M. pulcherrima* or *T. delbrueckii* in mixed cultures with respect to the production of ethanol, glycerol and other byproducts that contribute to the organoleptic characteristics of mango wine.

Materials and methods

Microorganisms and sample processing

A wine strain of *Saccharomyces cerevisiae* was a kind gift from Prof. Roberto Ambrosoli, University of Turin, Italy. Two other yeast strains namely *Metschnikowia pulcherrima* (NCIM 3109) and *Torulaspora delbrueckii* (NCIM 3295) were procured from the National Collection for Industrial Microorganisms, Pune, India. *Torulaspora delbrueckii* (IIHR 85) was procured from the Indian Institute of Horticulture Research, Bangalore, India. These cultures were maintained on MPYD agar slants containing (g/l): malt extract, 3; peptone, 5; yeast extract, 3; dextrose, 10; and agar, 20 (pH 5.0), and stored at 4°C. Ripened mango fruits, var. *Banginapalli*, grown in Andhra Pradesh, South India, were procured from the local market. Fresh edible puree of the fruits was processed according to Reddy and Reddy (2005).

Physico-chemical analysis of the mango wines

The pH of the wine was measured with a hand digital pH meter (Eutech, Japan), pre-calibrated with buffers of pH 4.0 and 7.0. Titratable (total) acidity in wine was determined by titrating with 0.1 N NaOH previously standardized using standard oxalic acid, and the values were expressed as tartaric acid equivalents and volatile acidity in the distillate samples is expressed as g/l acetic acid. Total soluble solids (TSS) was determined using a hand refractometer (0–30) (Erma, Japan) in terms of ºBrix (ºBx). Glycerol was enzymatically determined by using a commercial kit from Megazyme, Ireland. Free and total SO_2 was determined by ripper titrametric method using iodine, and reducing sugars were determined colorimetrically using 3,5-dinitrosalicylic acid (DNS) method (Kumar et al. 2009).

Determination of volatiles by Gas Chromatography (GC)

Cell-free samples were obtained by centrifugation at 5,000*g* for 10 min after the completion of the fermentation and analyzed for alcohols. Ethanol and other major volatiles were determined by GC according to Anthony (1984). Agilent systems GC-FID Model 6890 plus instrument was used for experiments and the conditions were as follows: Carbopack-B 80/120 mesh glass column [2 m (6 ft) with 2 mm i.d.; 1/4 mm], nitrogen gas was used as a carrier gas with a flow of 20 ml/min. Eluted compounds were detected by flame ionization detector (FID). Hydrogen with a flow rate of 40 ml/min was used as the fuel gas and the air was used as an oxidant (with a flow rate of 40 ml/min). For all the samples, 4-methyl–2-pentanol was used as internal standard.

Evaluation of growth and viability

Yeast growth was evaluated by monitoring the culture absorbance at 600 nm on a spectrophotometer, and viable yeast enumeration was determined by plate counting of samples withdrawn throughout fermentation and diluting appropriately in dilution medium (g/l: NaCl 8.5, peptone 1, $Na_2HPO_4 \cdot 2H_2O$ 0.3, pH 5.5) and plating of duplicate 50-μl aliquots onto MPYD agar plates supplemented with chloramphenicol (100 mg/l). The colony count for *M. pulcherrima* was determined in the same manner using Lysine agar medium base (Himedia, India). Lysine medium does not support the growth of *S. cerevisiae*, and therefore permitted the differential enumeration of *M. pulcherrima* in the presence of *S. cerevisiae* and were easily distinguished by their red-brown-colored colonies and *T. delbrueckii* by their white colonies. Plates were incubated at 25°C and colonies were counted after 4 days. A colony count for *S. cerevisiae* was obtained by subtracting the *M. pulcherrima* and *T. delbrueckii* count on Lysine medium from the total count obtained on MPYD agar.

Inoculum preparation and wine fermentation conditions

Fermentations were carried out in triplicate with *S. cerevisiae, M. pulcherrima* and *T. delbrueckii* in a pre-sterilized 2-l flask with 1,000 ml mango juice at 23±2°C. The juice was inoculated with 48-h pre-cultures grown in mango juice medium at 25°C. Fermentation with individual pure culture of *M. pulcherrima, T. delbrueckii* and *S. cerevisiae* were conducted by inoculation of 5×10^6, 5.5×10^6 and 3×10^6 cells/ml, respectively. Mixed fermentation tests were conducted by simultaneous addition of each (*Saccharomyces*: non-*Saccharomyces*) yeast species in the ratio of 1:10.

Sensory evaluation

The sensory characteristics of the final wines were evaluated according to (Dias et al. 2007) with a 20-membered panel. The preferences for taste, acidity, mouth feel, aroma, flavor, color and overall acceptability were determined by 9-point hedonic scale (1, dislike extremely; 2, dislike very much; 3, dislike moderately; 4, dislike slightly; 5, neither like nor dislike; 6, like slightly; 7, like moderately; 8, like very much; 9, like extremely). Randomized refrigerated (10°C) samples, of 50 ml, were served in clear tulip-shaped glasses coded with a random 3-digit code. Potable water was provided for rinsing of the palate during the testing. Evaluations took place in the mornings between 9:00 and 10:00 AM and were conducted at room temperature (22–24°C) under white light. Taste was measured in terms of sweetness, where as flavor and aroma were to mango flavor. The mouth feel was assessed in terms of smoothness of the wine. Acidity was assessed in terms of sourness of the wine in the mouth. Color of the wine was evaluated in terms of its intensity. Overall acceptance was the general preference expressed by the assessor after evaluating the sensory attributes. The mean intensity scores of all the attributes were calculated and plotted.

Statistical analysis

All the experiments were carried out in triplicate and the mean value and standard deviation were presented. Student's *t* test has been used to compare the mean values. The data were analyzed by one-way analysis of variance (ANOVA) using SPSS, v.12.0.

Results and discussion

The mango cultivar *Banginapalli* used in this study was a juicy variety (juice yield ~570±16 ml/kg) and the initial sugar concentration was ranged from 14.9 to 16% (23–25°Bx); however, it was adjusted to about ~20% with commercial glucose (data not presented).

Analysis of wines produced by pure and mixed cultures

It is well known that the most important agent of alcoholic fermentation in wine making is *S. cerevisiae*, capable of very high fermentation power (Zohre and Erten 2002). The main compounds of wines are shown in Table 1. *Saccharomyces cerevisiae* showed the highest fermentative ability in pure culture with the production of 11.9% (v/v) ethanol. Mixed culture fermentations along with the main wine yeast also produced higher concentrations of ethanol ranging from 11.04 to 11.53% (v/v). Fermentations performed with *M. pulcherrima* formed the lowest amounts of ethanol, 3.80% (v/v), and with *T. delbrueckii* (IIHR and NCIM) was 6.90 and 7.2% (v/v), respectively. Herraiz et al. (1990) and Fleet and Heard (1993) reported that *M. pulcherrima* produced up to 4.0% (v/v) of ethanol in monoculture fermentations; however, it was 6.38% (v/v) for *T. delbrueckii* (Ciani and Picciotti 1995) and was in agreement with earlier reports. The wine fermentations were completed successfully to about 1 g/l of sugar in the wines produced with *S. cerevisiae* in pure and mixed cultures, consistent with concentrations for dry wines. The monocultures of *M. pulcherrima* and *T. delbrueckii* could not ferment mango juice to dryness on their own and values are presented in Table 1. Similar results were also reported by other researchers (Fleet and Heard 1993; Ciani and Picciotti

Table 1 General composition of wines produced from pure and mixed cultures

Composition	S.C	M.P	Co-fermentation S.C + M.P	T.D (NCIM)	Co-fermentation S.C + T.D (NCIM)	T.D (IIHR)	Co-fermentation S.C + T.D (IIHR)
pH	4.0	3.71	4.12	3.74	3.41	4.12	3.76
Initial sugar (%)	20.0	20.1	20.1	20.0	20.2	20.1	20.0
Volatile acidity (g/l)	1.28±0.02 d	0.21±0.04 a	0.18±0.01 a	1.17±0.03 a	1.13±0.08 c	0.94±0.03 b	0.98±0.06 b
Titratable acidity (g/l)	5.5±0.06 c	4.7±0.08 abc	3.8±0.09 a	4.5±1.01 ab	3.8±1.0 a	5.1±0.09 bc	4.7±0.03 abc
Free SO_2 (mg/l)	15±2.1 bc	12±2.6 a	15±1.4 c	13±1.7 ab	13±2.1 ab	12±1.1 a	12±1.8 a
Total SO_2 (mg/l)	38±7.1 a	35±5.5 bc	33±3.1 bc	32±4.0 bc	30±3.4 ab	35±2.4 bc	27±2.3 a
Residual sugar (g/l)	1.3±0.3 a	172.4±0.8 b	1.2±0.4 a	159.2±0.6 b	1.4±0.09 a	168.1±0.5 b	1.2±0.2 a
Glycerol (g/l)	5.8±0.8 b	6.7±0.4 c	7.6±0.2 d	6.1±0.31 b	5.8±0.19 b	5.4±0.16 a	5.6±0.35 b

Values not sharing the same letter within the row significantly at $p \leq 0.01$ according to the Duncan's Multiple Range Test (DMRT)
S.C S. cerevisiae, *M.P M. pulcherrima*, *T.D T. delbrueckii*

1995). The production of volatile acidity with *S. cerevisiae* was 1.28 g/l. However, there was no significant difference with respect to volatile acidities in the case of non-*Saccharomyces* yeast monocultures. Total acidity was lower in mixed culture fermentations, whereas the monocultures produced higher levels of total acidity. The monoculture of *S. cerevisiae* produced higher acidity of 5.5% tartaric acid equivalents.

Yeast growth changes during fermentation

Evaluation of yeast populations during pure and mixed culture fermentations of mango juice medium were shown in Figs. 1, 2 and 3. The pure culture of *S. cerevisiae* achieved 8.2 log CFU/ml within the initial 120 h of fermentation and had a long stationary phase after the maximum growth of 8.5 log CFU/ml. However, *M. pulcherrima* in pure culture reached a maximum of 7.2 log CFU/ml and showed a stationary phase similar to *S. cerevisiae* but in low numbers. Both the strains of *T. delbrueckii*, NCIM and IIHR in pure cultures had reached 6.7 log CFU/ml. However, their numbers declined in mixed cultures after the first 15 days of fermentation.

The maximum population of both of the yeasts in mixed culture fermentation was at a lower level than their respective pure cultures. In mixed culture of *M. pulcherrima* and *S. cerevisiae*, *M. pulcherrima* multiplied up to 6.7 log CFU/ml. After maximum growth, it did not show the stationary phase and a decline phase was observed. However, it survived up to 14 days. *M. pulcherrima* proliferated to 3.4 log CFU/ml, followed by a rapid decline and disappearance in the mixed culture. With regard to *S. cerevisiae*, it was the dominant yeast and, within 3 days, the maximum populations reached 7.61–7.85 log CFU/ml. It survived a very long stationary phase and was the only yeast isolated from wines after 14 days. In mixed culture, the maximum viable cell population of *M. pulcherrima* attained was lower than that of its monoculture. The same phenomenon was also observed in other reports on mixed culture fermentations using different non-*Saccharomyces* yeasts (Mendoza et

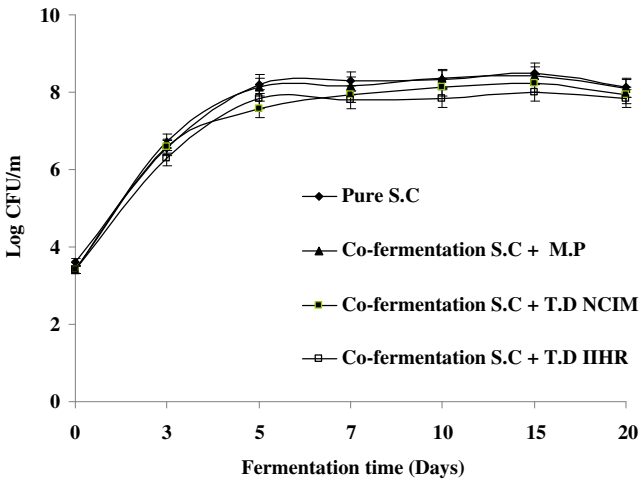

Fig. 1 The growth kinetics of *S. cerevisiae* (*S.C*) during its pure and co-fermentation with *M. pulcherrima* (*M.P*) and *T. delbrueckii* (*T.D*)

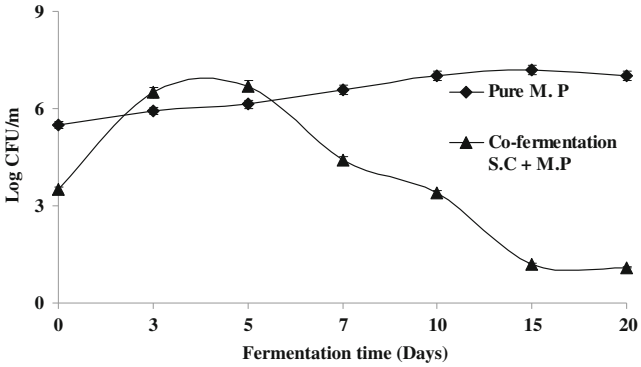

Fig. 2 The growth kinetics of *M. pulcherrima* (*M.P*) during its pure and co-fermentation with *S. cerevisiae* (*S.C*)

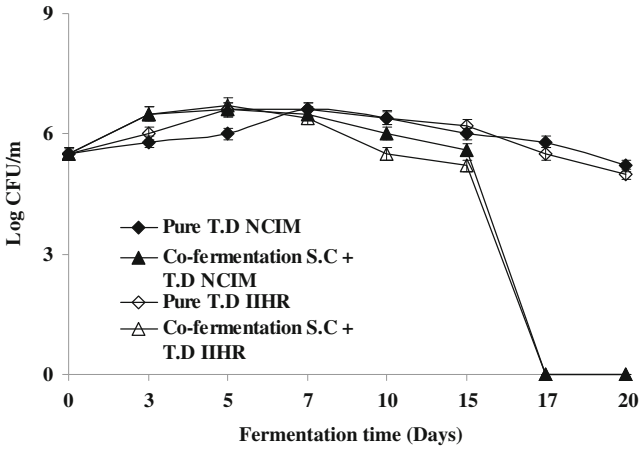

Fig. 3 The growth kinetics of *T. delbrueckii* (*T.D*) during its pure and co-fermentation with *S. cerevisiae* (*S.C*)

al. 2007; Soden et al. 2000). In the mixed culture of *S. cerevisiae* and *T. delbrueckii* (NCIM and IIHR), the cells multiplied up to 6.7 log CFU/ml and the decline phase was observed from the 15th day, whereas, in their respective pure cultures, a stationary curve was obtained at the end of fermentation. Similar results were also obtained by Bely et al. (2008) in grape juice fermentation.

The growth of non-*Saccharomyces* species belonging to genera *Kloeckera*, *Hanseniaspora* and *Candida* is limited to the first few days of fermentation, because of their weak ethanol tolerance ranging from 4 to 6% and from 5 to 10%, respectively. *Metschnikowia pulcherrima* seems to be less tolerant to ethanol and incapable of surviving in ethanol concentrations of 2–3% (Kunkee and Amerine 1970). Combina et al. (2005) and Parapouli et al. (2010) reported that *M. pulcherrima* exhibited the tolerance up to 7 and 6% ethanol, respectively.

The rapid death and disappearance of *M. pulcherrima* including other species like *Rhodotorula*, *Pichia* and *Candida* from fermenting musts was also observed by Fleet et al. (1984). This may be because of their oxidative or weakly fermentative metabolism and sensitivity to ethanol. However, they observed that the slight growth of *C. krusei* and *M. pulcherrima* in musts is noteworthy since these species lead to increased levels of acetic acid, esters, and higher alcohols in wine. The above reasons may be responsible for declining of *M. pulcherrima* growth after the first 5 days of fermentation in our study. In the case of *T. delbrueckii*, Bely et al. (2008) reported that it was a low ethanol producer and able to survive even at high ethanol concentrations (14%).

Aromatic quality and sensory evaluation of wine

Higher alcohols are produced from the Ehrlich pathway in the presence of amino acids and from sugars via biosynthesis by yeasts during alcoholic fermentations. 3-methyl butanol (isoamyl alcohol), 2-methyl butanol (active amyl alcohol), iso-butanol (2-methyl propanol) and n-propanol (1-propanol) are the principal constituents of higher alcohols (Simpson 1979). There was higher production of these alcohols in wines fermented by pure and mixed cultures of *S. cerevisiae* in contrast to those fermented using only single cultures of *M. pulcherrima* and *T. delbrueckii* (NCIM and IIHR) in the present experiments.

Significant differences in the aromatic profiles of the wines were observed for those obtained by mixed culture fermentation, as well as those obtained from mono culture fermentations (Table 2). The amounts of higher alcohols produced in monocultures of *S. cerevisiae, M. pulcherrima* and *T. delbrueckii* strains (NCIM and IIHR) were 359.70, 164.68, 301.58 and 290.26 mg/l, whereas the levels of 350.62, 323.61 and 324.94 mg/l were observed for simultaneous co-fermentations in *S. cerevisiae* + *M. pulcherrima*, *S. cerevisiae* + *T. delbrueckii* (NCIM) and *S. cerevisiae* + *T. delbrueckii* (IIHR), respectively. Both strains of *T. delbrueckii* showed similar capability to produce

Table 2 Analysis of volatile compounds of wines produced from pure and mixed cultures

Composition	S.C	M.P	Co-fermentation S.C + M.P	T.D (NCIM)	Co-fermentation S.C + T.D (NCIM)	T.D (IIHR)	Co-fermentation S.C + T.D (IIHR)
Alcohol (%)	11.9	3.8	11.04	7.2	11.53	6.9	11.35
Acetaldehyde (mg/l)	36.7±1.82 e	12.9±0.9 a	26.5±1.4 d	23.1±1.6 c	18.4±1.1 b	25.4±1.8 cd	23.8±2.0 cd
Ethyl acetate (mg/l)	57.9±2.1 e	29.7±1.5 a	52.5±1.9 d	46.8±1.7 e	41.5±1.5 b	47.2±0.95 c	45.8±1.5 c
n-Propanol (mg/l)	17.8±0.9 e	12.6±0.42 a	15.5±0.8 b	15.2±1.5 b	16.7±1.3 b	14.8±0.84 b	15.6±0.91 b
Isobutanol (mg/l)	41.4±1.2 c	45.1±2.1 d	49.1±2.3 e	27.9±0.8 b	25.2±1.1 a	29.2±0.7 b	24.2±1.2 a
Amyl alcohols (mg/l)	242.4±11.3 d	77.1±7.8 a	233.4±13.2 d	211.5±17.1 c	240.1±12.4 d	198.9±10.1 b	239.2±12.3 d
Total higher alcohols (mg/l)	359.7±15.2 e	164.6±17.3 a	350.6±15.3 b	301.5±20.1 c	323.6±14.5 d	290.2±11.4 c	324.9±13.7 d

Values not sharing the same letter within the row differ significantly at $p \leq 0.01$ according to the Duncan's Multiple Range Test (DMRT)
S.C S. cerevisiae, *M.P M. pulcherrima*, *T.D T. delbrueckii*

higher alcohols in monocultures. But monoculture of *M. pulcherrima* showed significantly ($p<0.01$) lower higher alcohol content, however, they showed similar patterns in simultaneous co-fermentations.

Acetaldehyde, accounting for 90% of the total aldehydes in wines, is a major component that plays an important role in the aroma and bouquet of wine. Among the various yeasts, *S. cerevisiae* has the capability of producing relatively high levels of acetaldehyde (Ciani and Picciotti 1995). However, Barbe et al. (2000) stated that a high concentration of acetaldehyde in wine from botrytized grapes resulted in reduced amounts of free SO_2. It was also observed that *S. cerevisiae* monoculture produced higher levels of acetaldehyde than those of the monocultures of *M. pulcherrima*. In the case of both *T. delbrueckii* strains, it resulted in acquiring the cumulative effects in mixed cultures.

Microorganisms are known to modulate aromatic esters in wine (Sumby et al. 2010). Esters are mainly produced by yeast during alcoholic fermentation in a reaction between alcohols and acetyl-CoA catalyzed by alcohol acetyltransferase and other enzymes. Ethanol is the main alcohol in wine, and therefore, ethyl acetate produced from ethanol and acetyl-CoA is the major ester formed by yeast. Other acyl-CoA compounds also show similar behavior for the production of other esters (Dufour and Malcorps 1995). In the present study, a significantly ($p<0.01$) lower amount of ethyl acetate was observed only with the monoculture of *M. pulcherrima* but not with its co-fermentation with *S. cerevisiae*. Ethyl acetate ester was low for co-fermentations with *T. delbrueckii* when compared to their respective monocultures, which is in agreement with Viana et al. (2008), who reported that the levels of ethyl esters produced by non-*Saccharomyces* yeasts were lower than those detected in *S. cerevisiae* wines. Ethyl acetate imparts a fruity flavor in wines and it produces a solvent-like odor at concentrations exceeding 200 mg/l (Etievant 1991). Unlike ethyl acetate, acetate esters and medium chain fatty acid esters may also have profound effects on wine flavor (Zohre and Erten 2002).

Glycerol is a wine constituent related to yeast metabolism which contributes to the sweetness, viscosity and smoothness of wine (Ciani and Ferraro 1998). In the present study, the production of glycerol was greater by *M. pulcherrima*. Glycerol concentrations in wines fermented with pure *S. cerevisiae* + *T. delbrueckii* did not differ significantly.

In order to evaluate the influence of each starter culture on organoleptic quality of fermented products, the sensory analysis of different young wines was carried out by the tasting panel consisted of 20 judges trained in wine tasting. Intensity ratings of main descriptors were scored on scale from 0 (dislike extremely) to 9 (like extremely) (Fig. 4). It was observed that products obtained from co-fermentation with non-*Saccharomyces* showed higher scores for fruity aroma as compared to wines fermented by pure *S. cerevisiae*. Wine co-fermented with *M. pulcherrima* was acceptable with a high score for overall acceptability (8.5), color (7.1) and taste (6.2) followed by *T. delbrueckii*. There was no significant sensorial difference observed between the two *T. delbrueckii* strains. However, wine fermented with pure *S. cerevisiae* showed the lowest sensory attributes. Rodriguez et al. (2010) reported on wine production using mixed starter cultures of *S. cerevisiae* MMf9 and a β-glucosidase producer *C. pulcherrima* V6 strain, and the results showed a positive impact on the wine by enhancing its fruity and floral aroma. Similar results were obtained for the wines produced by *S. cerevisiae* in co-culture with *Candida stellata*; these products presented the highest total concentration of higher alcohol and esters with strong aroma (Soden et al. 2000). Likewise, *T. delbrueckii* was used to increase the sensory variety of wine made from grape as reported by (Sommer et al. 2007).

Conclusion

The evaluation of non-*Saccharomyces* yeasts used in the present study might be of great value for the mango wine-making industry. Certainly, the results indicated that these can be used in association with *S. cerevisiae* starter cultures to enhance the quality, improve the complexity, and modify some of the undesired parameters of the final wines. However, the results obtained from laboratory-scale assays are not necessarily the same as what might be

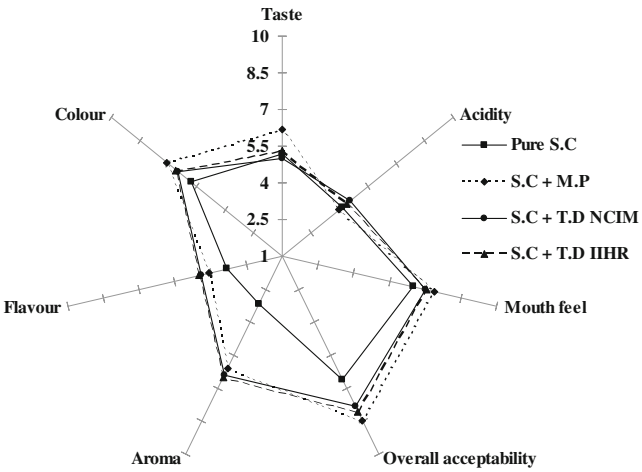

Fig. 4 Sensory profiles of mango wine fermented with pure *S. cerevisiae* (*S.C*), co-fermented with *M. pulcherrima* (*M.P*) and *T. delbrueckii* (*T.D*)

expected in larger-scale fermentations. Thus, larger-scale studies should be performed to confirm the results obtained in this work.

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