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PRODUCTION OF CLEAR APPLE JUICE USING COMMERCIAL PECTINASES

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ABSTRACT

Clear apple juice was produced from Anna apple cultivar (*Malus* spp.) cultivated in Egypt by two stages, pulp enzyming and juice clarification using commercial pectinases. Apple flavour and cramel were added during preparation of clear drinks. Clear apple drinks stored at refrigerator temperature (4°C), for 5 months were analyzed periodically for chemical, physical, sensory and microbiological characteristics. Data indicated that clear apple juice was processed successfuly using commercial pectinases. Addition of cramel and flavour improved colour, taste, aroma and appearance of natural clarified drink. In respect to chemical chracteristics of the drink, ascorbic acid enrichment is recommended.

Keywords: Clear apple juice, commercial pectinases, pulp enzyming, juice clarification, chemical and physical characteristics .

INTRODUCTION

Technical enzymes preparations are widely used in the fruit –processing industry to facilitate juice release, increse juice yield and to clarify juices (Schols *et al*. 1991). A variety of sparkiling clear juices and cloudy juices production depends on pectic enzyme treatment (Pilnik and Rombouts, 1981; Whitaker, 1984 and Voragen and Pilnik, 1998).

Pectic enzymes account for about one-quarter of the world›s food enzyme production and most of pectic enzyme preparations are used in the fruit processing industry to increase juice yield and to clarify juices (Rombouts and Pilnik,1980 and Jayani *et al*. 2005).

The oldest and still the largest use of pectinases is the fruit juice clarification (Pilnik and Voragen, 1993). Addition of pectinases lowers viscosity and causes cloud particles to aggregate to larger units "break" which sediment and removed easily by centrifugation or (ultra)filtration (Pilnik and Voragen, 1993 and Kashyap *et al*. 2001)

Commercial pectinases performing well in juice clarification are also suitable for enzyme treatment of the pulp, and in the case of apple, any combination of enzymes depolymerizes highly esterified pectin can be used successfully (Voordouw *et al*. 1974).

"Anna" apple is cultivated in Egypt. The whole production of apple is displayed in supermarkets for fresh consumption only. Processed apple products should be developed for longer shelf-life and diverse apple products.

The aim of this work is to evaluate new product of "Anna" apple. Clear apple juices using commercial pectinases were produced and evaluated for their

sensory, chemical, physical and microbiological characteristics during storage for five months at refrigerator temperature.

MATERIALS AND METHODS

Materials

Apple fruits were collected at mature stage, from Racheed (El-Behara governorate), then stored at refrigerator temperature (4°C) untill use. Commercial pectinolytic enzymes; pectinase 444L and macer8^{TM}FJ were obtained from Biocatalysts Limited, Wales, UK. Apple esseence (green apple, 0.017 %, R9410G) from Delta Aromatic company, El-Geeza, Egypt.
Cramel (E15098001, 0.0125%, for colouring of apple juice and drink), from Delta Aromatic company, El-Geza, Egypt.

Microbiological media

Trypton glucose extract agar for standard plate count, was prepared according to Marsall, (1992 a). Standard methods agar for antibiotic plate counts of yeast and mold, was prepared according to Marsall, (1992 b).

Methods

Production of clear apple juice: Clear apple juice was produced using pectinase 444L (300 ppm for mash fermentation and 20 ppm for clarification) and macer8^{TM}FJ (250 ppm for mash fermentation and 20 ppm for clarification), as the following:
Mature apple fruits were washed, cored and mashed using moulinex mincer. Apple mashes were treated by pectinases for 30 min at 45°C. Then raised temperature of the mash to 85°C for enzyme inactivation then cooled. Mashes were squeezed through double layer opaque cloth. Juice clarification was done using 20 ppm enzyme for 6 hours at 45°C. Juice filtered through four layer opaque cloth and centrifuged. For preparation of clear apple drinks, clarified juices were adjusted to 14°Brix with sugar and modified to 1: 1 dilution with 14°Brix sugar solution.

Addition of cramel and apple essence:

Cramel was dissolved in a littel water (w/v %) with gentle heating, then added to the clear apple drinks to reach a final concentration of 0.02 and 0.035(v/v%). Drinks were bottled in a 100 mL- bottles. Apple essence was added to drinks at concentrations of 30ppm, during pasteurization(85°C/5min), before sealing, then cooled. Procedures and enzyme treatments were optimized upon preliminary experiments depending on juice yield (v/w%), TSS, clearance and Sensory evaluation. Seven clear apple drinks were prepared according to the previous flow sheet to be stored at refrigerator temperature (4°C), for five months:

1. apple juice produced without enzyme treatment (Con).
2. clear apple juice produced by 250 ppm macer8^{TM}FJ for mash treatment and 20 ppm macer8^{TM}FJ for juice clarification (Em).
3. clear apple juice produced by 250 ppm macer8^{TM}FJ for mash treatment and 20 ppm macer8^{TM}FJ for juice clarification + 0.02% cramel (Em+Cr1).

4. clear apple juice produced by 250 ppm macer8^{TM}FJ for mash treatment and 20 ppm macer8^{TM}FJ for juice clarification + 0.035% cramel (Em+Cr2).
5. clear apple juice produced by 250 ppm macer8^{TM}FJ for mash treatment and 20 ppm macer8^{TM}FJ for juice clarification + 0.02% cramel + 30 ppm apple essence (Em+Cr1+F).
6. clear apple juice produced by 250 ppm macer8^{TM}FJ for mash treatment and 20 ppm macer8^{TM}FJ for juice clarification + 0.035% cramel+30 ppm apple essence (Em+Cr2+F).
7. clear apple juice produced by 300 ppm pectinase 444L for mash treatment and 20 ppm pectinase 444L for juice clarification + 0.035% cramel+30ppm apple essence (Ep+Cr2+F).

Clear seven drinks were analyzed for TSS, acidity (as citric acid %), pH, clearance (absorbance at 660 nm), reducing sugars % and total sugars % at zero-time and at one month intervals for a total of 5-months storage at refrigerator temperature (4°C). Sensory and microbiological evaluations were carried out at zero-time and at one month and half intervals, to the end of the storage period.

Methods of analysis

Chemical analysis

Total titratable acidity, pH and reducing and total sugars were determined as described in the AOAC (1990).

Physical anslysis

Total soluble solides (° Brix) were measured according to the AOAC (1990) by Abbe refractometer at 20°C. And clearance was determined according to Staelehamatschek (1989) with little modifications (Mostafa, 2004). Apple juice was centrifuged at 4000 rpm for 25 min. The clearance of the supernatant was measured by a spectrophotometer at 660 nm.

Sensory evaluation

Juice samples were sensory evaluated for colour, taste, aroma and appearance. Sensory assessments were carried out by ten semi-trained panelists on a 7-point hedonic scale from excellent, very good, good, satisfactory, poor, very poor, to extremely poor, which for analysis were scored from 7 to 1 (McBride and Richardson, 1983).

Microbiological analysis

Microbiological quality of clear apple drinks were periodicaly examined for bacteria; mold and yeast. For total count of bacteria, trypton glucose extract agar was used. Counts of molds and yeasts were carried out by antibiotic agar method using standard methods agar. Antibacterial buffer (2%) was added to sterile media befor use, for inhibition of bacterial growth. Dilutions up to 1: 10^3 of tested treatments were cultivated. Counts of plates were taken after incubation of 24 hrs/37°C and 72 hrs/25°C for bacteria and (molds & yeasts) respectively.

Statistical anslysis

The analysis of variance (ANOVA) and LSD were performed by CoStat 6.311, Copyright(c) 1998-2005, CoHort Software, at probability ≥ 0.05

(Snedecor and Cochran, 1967). Statistical analysis of sensory evaluations depended on ten replicates of degrees by the ten panelists.

RESULTS AND DISCUSSION

Preliminary experiments

Preliminary experiments were carried out to select the best treatments depending on juice yield, TSS, clearance and sensorial evaluations. Results are presented in Tables (1), (2), (3) and (4). Best treatments were selected for cold storage experiment.

Table (1) shows that juice yield, TSS and clearance values (absorbance at 660 nm) of clarified apple juices increased significantly with increasing of enzyme concentration. Hence high concentration enzyme treatments will be considered in ongoing experiments.

From sensory assessments of clear apple drinks recorded in Table (2), the high concentration enzymatic treatments gained higher degrees of colour evaluation. It is obvious that the colour degree was affected by the clarity of the sample while the hazy control was refused. Also the taste of the higher enzymatic treatments were significantly better than other treatments especialy control. This could be due to the relative higher acidity caused by higher levels of enzyme treatments which balance the sweetness of the sugar and show good taste as the solubilization of cell wall polysacchrides effects various characteristcs of the juice obtained, like °Brix, pH/acidity, contents and composition of phenolics, aroma and also the contents of solubilized polysaccharides (Voragen *et al.* 1992).

Treatment Macer at 250 ppm showed significantly the best aroma, and other treatments were insignificant. Appearance quality of treatments followed the potential of enzymatic clarification and the higher enzymatic concentrations showed the best appearance. Josh *et al.*(2008) evaluated partially purified pectinase enzyme produced by *Aspergillus niger* for juice extraction and clarification of plum, peach, pear and apricot. The overall sensory evaluation of enzyme extracted juice using hedonic test showed a significant improvement in their clarity scores. The flavour of the extracted juice however remained unaffected by the addition of enzyme at all concentration.

Table (1): Juice yield, TSS and clearance of clarified apple juices produced using different pectinase concentrations.

Character / Treatment	Juice yield % (V/W)	Final TSS	Clearance (absorbance at 660 nm)
Control	40c	10.2c	0.219a
Pectinase at 300 ppm	66a	12.7a	0.062d
Pectinase at 150 ppm	58b	11.7b	0.154b
Macer at 250 ppm	68.4a	13.0a	0.054d
Macer at 125 ppm	62.8ab	12.5a	0.125c

Abbreviations: Pectinase: pectinase 444L; Macer: macer8^{TM}FJ and Control: apple juice produced without enzyme treatment.

Mean in the same column with different superscripts are significantly different ($p < 0.05$).

Table (2): Sensory assessment of colour, taste, aroma and appearance of clear apple drinks produced using pectinase enzyme preparations.

Treatment	Character				Overall acceptance
	Colour	Taste	Aroma	Appearance	
Pectinase at 300 ppm	5.8 a	6.0 a	3.7 b	5.9 b	21.4 b
Pectinase at 150 ppm	4.1 b	3.8 b	3.2 b	3.9 d	15.0 c
Macer at 250 ppm	6.3 a	5.9 a	5.4 a	6.9 a	24.5 a
Macer at 125 ppm	5.0 ab	5.1 a	3.9 b	4.7 c	18.7 b
Control	2.8 c	2.7 b	3.3 b	2.0 e	10.8 d

Mean in the same column with different superscripts are significantly different (p < 0.05).

In preliminary experiments, homogenization insignificantly increased crude juice yield from 78.3% for non-homogenized mashs to 83.7% for homogenized mashes (Table 3). Moreover, homogenization resulted in inefficient percipitation during the clarification process and loss more juice. Hence homogenization was ignored in the following production.

Table (3): Percent of juice yield from homogenized and non-homogenized apple mashes.

Treatment	Crude juice yield % (V/W)	Final clarified juice yield % (V/W)
Clear apple juice from non-homogenized apple mash fermented by 250 ppm macer	78.3 a	63.0 a
Clear apple juice from homogenized apple mash fermented by 250 ppm macer	83.7a	63.7a

Mean in the same column with different superscripts are significantly different (p < 0.05).

According to the data of preliminary sensory evaluation presented in Table (4), cramel treatments had significantly attractive colour and appearance, ranged between "very good" and "excellent" for 0.02% and 0.035% cramel in comparison with poor colour/satisfied appearance for control. Taste and aroma improved significantly by falvouring as flavoured treatments gained significantly very good characters and other treatments ranked good and satisfied.

The best treatments depending on juice yield, TSS, clearance and sensory evaluation were selected for further studies.

Table (4): Sensory assessment of colour, taste, aroma and appearance of clear apple drinks modified with cramel and flavour.

Treatment	Character				Overall acceptability
	Colour	Taste	Aroma	Appearance	
Control	4.4 c	4.3 c	3.6 b	4.5 b	16.8 c
0.02% cramel	5.5 b	4.8 bc	3.8 b	6.0 ab	20.1 b
0.035% cramel	5.8 ab	5.0 bc	4.2 b	5.6 ab	20.6 b
0.02% cramel+ flavour	5.6 b	6.4 a	5.9 a	6.7 a	24.6 a
0.035% cramel+ flavour	6.6 a	6.0 ab	5.6 a	5.7 ab	23.9 a

Mean in the same column with different superscripts are significantly different (p < 0.05).

Physical and chemical characteristics of clear apple drinks during cold storage

Means of TSS of the treatments over the storage period ranged insignificantly ($p > 0.05$) between 13.75 and 13.92°Brix for Ep+Cr2+F and Em+Cr+F, respectively as shown in Table (5) .

Table (5): TSS of clear apple drinks stored at refrigerator temperature(4°C), for five months.

torage (months) / Treatment	0	1	2	3	4	5	Mean
Con	13.8	13.8	13.8	13.8	14.0	13.5	13.78 a
Em	13.8	13.8	13.8	13.8	14.0	14.0	13.89 a
Em+Cr1	13.8	13.8	14.0	13.8	14.0	13.8	13.89 a
Em+Cr2	13.8	13.8	14.0	14.0	14.0	13.7	13.89 a
Em+Cr1+F	13.8	13.8	14.0	14.0	14.0	13.8	13.92 a
Em+Cr2+F	13.8	13.8	14.0	14.0	14.0	13.7	13.89 a
Ep+Cr2+F	13.8	13.8	13.7	13.8	13.7	13.7	13.75 a
Mean	13.8 ab	13.8 ab	13.90 a	13.89 a	13.95 a	13.74 b	

Mean in the same column with different superscripts are significantly different ($p < 0.05$).

Brix of the treatments was influenced significantly by the storage period where its mean decreased significantly from 13.8° at zero-time to 13.74° by the fifth month of storage. Such reduction in the TSS could be due to the microbiological activity in the drinks (Ekoli and Ezenweke, 1990).

Enzymatic treatments exhibited insignificant acidity (Table 6). The acidity means of the treatments over the storage period reached the approximate value of 0.28 %, in comparison with the control which had significantly lower acidity of 0.199%. Following the acidity mean of all treatments at each measurment, the mean increased significantly from 0.26% at zero-time to 0.273% at the fifth month. The increasing in acidity could be attributed to the microbial activity through the storage period (Okoli and Ezenweke, 1990).

Table (6): Acidity % (as citric acid) of clear apple drinks stored at refrigerator temperature (4°C), for five months.

torage(months) / Treatment	0	1	2	3	4	5	Mean
Con	0.192	0.196	0.201	0.201	0.205	0.199	0.199 b
Em	0.271	0.282	0.277	0.286	0.284	0.286	0.281 a
Em+Cr1	0.273	0.282	0.279	0.277	0.286	0.286	0.281 a
Em+Cr2	0.271	0.282	0.280	0.286	0.286	0.282	0.281 a
Em+Cr1+F	0.273	0.282	0.284	0.286	0.284	0.288	0.283 a
Em+Cr2+F	0.267	0.277	0.280	0.288	0.284	0.286	0.280 a
Ep+Cr2+F	0.273	0.278	0.275	0.277	0.286	0.286	0.279 a
Mean	0.260 c	0.268 b	0.268 b	0.272 ab	0.274 a	0.273 a	

Mean in the same column with different superscripts are significantly different ($p < 0.05$)

The pH means of the enzymatic treatments over the storage period were equal (3.55), while the control was significantly higher (3.73). The mean of pH of the treatments reduced significantly ($p < 0.05$) with progressing of

the storage period from 3.59 at zero-time to 3.51 at the fifth month as shown in Table (7).

According to Schols *et al.*(1991), apple juices of straight pressing and pulp enzyming showed the same pH and total acidity of 3.4 and 0.29 % , respectively. Will *et al*. (2002) reported that the total titratable acidity (as citric acid) of apple juice was 0.45 , 0.44 , 0.46 and 0.47% when treated the apple mash with pectinex Smash at 18, 40, 50 and 60° C, respectively. Regarding that, 50 °C is the optimum temperature of pectinases.

Table (7): pH of clear apple drinks stored at refrigerator temperature (4°C), for five months.

Storage(months) / Treatment	0	1	2	3	4	5	Mean
Con	3.78	3.76	3.72	3.73	3.71	3.70	3.73 a
Em	3.55	3.56	3.59	3.61	3.52	3.48	3.55 b
Em+Cr1	3.56	3.56	3.60	3.59	3.53	3.48	3.55 b
Em+Cr2	3.56	3.56	3.60	3.56	3.52	3.49	3.55 b
Em+Cr1+F	3.55	3.58	3.60	3.57	3.53	3.48	3.55 b
Em+Cr2+F	3.56	3.57	3.59	3.57	3.55	3.48	3.55 b
Ep+Cr2+F	3.55	3.55	3.60	3.56	3.54	3.49	3.55 b
Mean	3.59 a	3.59 a	3.61 a	3.60 a	3.56 b	3.51 c	

Mean in the same column with different superscripts are significantly different ($p < 0.05$).

From the first glance at Table (8) which shows the clearance of clear apple drinks, it was found that, the treatments exhibited different significant degrees of clarification. The order of the absorbance means of the macer treatments over the five months storage period were; Em was the best clarified treatment which gave absorbance mean of 0.010, then Em+Cr1 and Em+Cr1+F showed insignificant absorbance of 0.028 and 0.027, respectively and Em+Cr2 and Em+Cr2+F showed insignificant absorbance of 0.042 and 0.04, respectively. The results indicated that, the interference of cramel treatments with the absorbance at 660 nm. Ep+Cr2+F had the highest absorbance of enzymatic treatments (0.044) which indicated the efficiency of macer preparation for clarification of Anna apple than pectinase. The control treatment which was visually turbid had significantly the highest absorbance mean of 0.118 as it is not possible to remove the soluble pectin and obtain efficient clarification without the addtion of pectic enzymes or use alternative techniques (Araya-Farias *et al.* 2008) .

With progressing storage there was significant decreases of the absorbance mean of the treatments at each measurment from 0.055 at zero-time to 0.036 at the fifth month, this could due to fine particles that eventually settled out by centrifugation and the action of the gravity (Benitez and Lozano, 2007).

Table (8): Clearance (absorbance at 660nm) of clear apple drinks stored at refrigerator temperature (4°C), for five months.

Storage(months) / Treatment	0	1	2	3	4	5	Mean
Con	0.136	0.129	0.127	0.119	0.102	0.096	0.118 a
Em	0.016	0.012	0.009	0.008	0.006	0.006	0.010 e
Em+Cr1	0.034	0.029	0.027	0.027	0.025	0.025	0.028 d
Em+Cr2	0.054	0.047	0.043	0.039	0.033	0.034	0.042 c
Em+Cr1+F	0.032	0.027	0.026	0.027	0.024	0.023	0.027 d
Em+Cr2+F	0.049	0.046	0.042	0.037	0.034	0.032	0.040 c
Ep+Cr2+F	0.059	0.048	0.042	0.039	0.035	0.034	0.044 b
Mean	0.055 a	0.048 b	0.045 c	0.042 d	0.037 e	0.036 e	

Mean in the same column with different superscripts are significantly different ($p < 0.05$).

Table (9) shows that, at zero-time, reducing sugars contents were 1.9 to 2.09 % for enzymatic treatments in comparison with 1.71 % for control treatment. The means of the treatments over the five months were the same significance for macer treatments (1.62-1.63%). Pectinase treatment showed significantly ($p < 0.05$) lower mean (1.45%), then the control treatment showed significantly the least mean of 1.37%. Contents of all treatments decreased significantly within the storage period. The means of reducing sugars of all treatments decreased significantly at each month, from zero-time to the fifth month, the corresponing values were 1.97 to 1.2%, respectively.

Total sugar percent of clear apple drinks are presented in Table (10). At zero time, the total sugars of the enzymatic treatment ranged between 12.46-12.71% while the control treatment gave the lowest content of 12.06%. Means of total sugar contents of each treatments over the fifth months storage period descended significantly from macer treatments which ranged between 12.46 to 12.53% , to control and Ep+Cr2+F which had the same significance means of 12.12 and 12.05 % respectively.

Over the five months storage period, the mean of the total sugar contents of all treatments decreased every month. It decreased significantly from 12.63 at the first month to 12.39 % at the second month and again decreased significantly to 12.05 % at the fifth month.

Resultes showed that enzymatic treatment affected significantly the acidity, pH, reducing sugars and the total sugar of clarified apple juice which are in agreement with results stated by many researchers, that, the solubilization of cell wall polysaccharides effects various characteristcs of the juices obtained like Brix, pH/acidity and also the content of solubilized polysaccharides. The amounts and types of polysaccharides released in the juice depend on the juice winning system, the conditions of enzyming, the enzyme preparations used and the method of clarification of the juice. It can be concluded that the enzyme preparation affects the intrinsic quality of the obtained juice which stated by Voragen *et al.* (1986). Also Voragen *et al.* (1992) found that the retentate of dialysed, conventionally clarified, straight pressed apple had a neutral sugar and uronide contents of 150 mg/L while for pulp enzyming juice this content was found to range from 1100 to 2510 mg/L, depending on the enzyme preparation used. Moreover, enzyme treatments

increased the total amount of uronide content in clarified apple juice by a factor ranging between 4-15. It was found that in most juices between 50 and 90% of the uronides were dialysable (Voragen *et al*. 1992).

Table (9): Reducing sugars (%) of clear apple drinks stored at refrigerator temperature (4°C), for five months.

Storage(months) / Treatment	0	1	2	3	4	5	Mean
Con	1.71	1.51	1.49	1.27	1.21	1.00	1.37 c
Em	2.00	1.98	1.61	1.55	1.38	1.27	1.63 a
Em+Cr1	2.05	1.97	1.55	1.45	1.40	1.29	1.62 a
Em+Cr2	2.09	2.00	1.57	1.47	1.40	1.23	1.63 a
Em+Cr1+F	2.02	2.06	1.55	1.47	1.40	1.21	1.62 a
Em+Cr2+F	2.05	1.95	1.6	1.5	1.45	1.21	1.62 a
Ep+Cr2+F	1.90	1.88	1.51	1.38	1.38	1.18	1.54 b
Mean	1.97 a	1.91 b	1.55 c	1.43 d	1.37 e	1.20 f	

Mean in the same column with different superscripts are significantly different (p < 0.05).

Table (10): Total sugars (%) of clear apple drinks stored at refrigerator temperature (4°C), for five months.

Storage (months) / Treatment	0	1	2	3	4	5	mean
Con	12.06	12.28	12.29	12.26	11.99	11.90	12.12 b
Em	12.53	12.76	12.46	12.44	12.38	12.2	12.46 a
Em+Cr1	12.72	12.68	12.59	12.45	12.37	12.30	12.52 a
Em+Cr2	12.62	12.83	12.59	12.58	12.37	12.2	12.53 a
Em+Cr1+F	12.60	12.68	12.38	12.37	12.37	12.09	12.42 a
Em+Cr2+F	12.68	12.68	12.60	12.58	12.39	12.09	12.50 a
Ep+Cr2+F	12.46	12.52	11.87	11.95	11.93	11.59	12.05 b
Mean	12.52 a	12.63 a	12.39 b	12.37 b	12.26 b	12.05 c	

Mean in the same column with different superscripts are significantly different (p < 0.05).

Sensory evaluation of clear apple drinks stored at refrigerator temperature.

Table (11) presents means of periodical sensory assessments for clear apple drinks carried out at one month and half intervals for five months at refrigerator temperature (4°C). Results could be summarized in the following comment:

Table (11): Means of periodical sensory assessment of clear apple drinks stored for 5 months at refrigerator temperature (4°C).

Treatment	Character			
	Colour	Taste	Aroma	Appearance
Control	3.475 d	4.08 c	3.55 c	3.375 c
Em	4.45 c	4.65 b	4.28 b	4.70 b
Em+Cr1	5.33 b	4.75 b	4.43 b	5.78 a
Em+Cr2	6 a	4.93 b	4.48 b	5.95 a
Em+Cr1+F	5.48 b	6.15 a	5.7 a	5.90 a
Em+Cr2+F	6.15 a	5.875 a	5.6 a	6.15 a
Ep+Cr2+F	6.05 a	5.65 a	5.38a	6.15 a

Mean in the same column with different superscripts are significantly different (p < 0.05).

Cramel treatments of 0.035% were significantly ($p < 0.05$) the best colour treatments. Other treatments were significantly less attractive while the control was refused. Color of the drinks did not influenced by the storage time.

Flavoured treatments showed "very good" taste quality. Other unflavoured enzymatic treatments were only "good" taste while the control lowered significantly to "satisfied". In respect to the effect of the storage period, taste improved significantly after one month of storage which could be explained by the good combination of added flavour, natural apple taste and sugar that occured after a month of storage. After four and half months, taste quality decreased which could be attributed to weakness of added flavour.

Aroma of flavoured treatments was significantly better than other treatments. Other enzyme treatments had "satisfied" aroma while control was significantly poor. Aroma of all treatments decreased significantly with progressing of the storage period.

Cramel treatments showed very good appearance. The turbid control was refused. But appearance of the treatments did not influenced significantly by the storage time.

Microbiological evaluation

At zero-time and untill the middle of the second month, the microbial load was under detectable levels (Tables 12 and 13), for the rest of the storage period, the log numbers of total bacterial count was less than one (< 1) and less than five (< 5) for molds & yeasts.

Table (12): The log numbers of total bacterial count for clear apple drinks stored for 5 months at refrigerator temperature (4°C).

Treatment	Storage (month)			
	0	1.5	3	4.5
Control	ND	ND	< 1	< 1
Em	ND	ND	< 1	< 1
Em+Cr1	ND	ND	< 1	< 1
Em+Cr2	ND	ND	< 1	< 1
Em+Cr1+F	ND	ND	< 1	< 1
Em+Cr2+F	ND	ND	< 1	< 1
Ep+Cr2+F	ND	ND	< 1	< 1

ND: not detected

Table (13): The log numbers of yeast & mold count for clear apple drinks stored for 5 months at refrigerator temperature (4°C).

Treatment	Storage (month)			
	0	1.5	3	4.5
Control	ND	ND	< 5	< 5
Em	ND	ND	< 5	< 5
Em+Cr1	ND	ND	< 5	< 5
Em+Cr2	ND	ND	< 5	< 5
Em+Cr1+F	ND	ND	< 5	< 5
Em+Cr2+F	ND	ND	< 5	< 5
Ep+Cr2+F	ND	ND	< 5	< 5

ND: not detected

CONCLUSION

Clear apple juice was processed successfuly using commercial pectinases. Addition of cramel and flavour improved colour, taste, aroma and appearance of natural clarified drink. In respect to chemical chracteristics of the drink, the enzyme treatment increased acidity, reducing sugars and total sugars of the clarified apple drink, so it can be concluded that enzyme treatment affects the intrinsic quality of the obtained juice.

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إنتاج عصير التفاح الرائق بإستخدام الإنزيمات البكتينية

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تم تصنيع عصير التفاح الرائق من صنف (*Malus spp.*) Anna المزروع فى مصر خلال مرحلتين، معالجة لب التفاح بالإنزيمات البكتينية (تخمير اللب) ثم إجراء ترويق العصير و ذلك بإستخدام المستحضرات التجارية للإنزيمات البكتينية.

أضيف الكرامل بتركيزات مختلفة و أسنس التفاح عند إعداد مشروب التفاح الرائق. خزنت مشروبات التفاح لمدة خمسة أشهر على درجة حرارة الثلاجة، أجرى خلالها التحليلات الكيميائية و الطبيعية للمشروبات شهريا و كذلك التقييم الحسى و الميكروبيولوجى كل شهر ونصف حتى نهاية مدة التخزين.

أوضحت النتائج إمكانية إنتاج العصير الرائق من التفاح آنا بجودة عالية و ذلك بإستخدام الإنزيمات البكتينية التجارية وإضافات النكهة واللون التى ساعدت على تحسين الجودة الحسية للعصير من حيث اللون و النكهة و الرائحة و المظهر.