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OPTIMIZATION OF PROCESSING PARAMETERS AND STUDY OF THE PHYSICAL- CHEMICAL CHARACTERISTICS OF PLUM JUICE

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OPTIMIZATION OF PROCESSING PARAMETERS AND STUDY OF THE PHYSICAL-CHEMICAL CHARACTERISTICS OF PLUM JUICE

By

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ABSTRACT

OPTIMIZATION OF PROCESSING PARAMETERS AND STUDY OF THE PHYSICAL-CHEMICAL CHARACTERISTICS OF PLUM JUICE

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Five commercial pectinase enzymes were used with Stanley plums to determine which gave optimum juice yield and quality. Among these five enzymes, Clarex L at 0.2% concentration produced the best overall plum juice. After the best pectinase enzyme was identified six new plum varieties (*Prunus domestica* L.) selected from the MSU variety trials were used in conjuction with Clarex L enzyme for the production of plum juice.

Each of the six varieties were processed into press-juice, enzyme-treated juice, High Temperature-Short Time (HTST)-unfined juice, fined juice and HTST-fined juice. Physical and chemical characteristics such as yield, clarity, soluble solids, pH, titratable acidity, Hunter color values, pectin content, total anthocyanin content and total phenolics were determined. The analytical results were combined with sensory evaluation results to aid in the selection of plums for further juice processing and development.

Dedicated to Shaw-Tsu Chang, memory of my father,
and Lee Chu Cheng, my mother, to whom I stand in debt for my
education and knowledge

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Sincere gratitude is extended to Dr. Jerry N. Cash for his guidance and support as major professor, and for his patience and understanding throughout the research program.

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INTRODUCTION

Plums are grown in several areas of the United States and are considered to be a major fruit crop in some of these growing locations. The production of plums in Michigan is not as extensive as in some states, nevertheless, plums are important to the fruit industry in the state. Economically they represent a modest income for the growers and processors at a time in the harvest schedule when other crops are not available (i.e. after cherries and before apples). Also, with the active plum variety program presently underway at the MSU Northwest Horticulture Research Station there are very good possibilities for new cultivar introductions that will further enhance the viability of the plum industry. Presently, one of the major needs within the plum industry is development of new processed products, such as juices, paste, wine and jelly to utilize plums. The purpose of this study was to:

a) investigate the effect of commercially available cell wall degrading enzymes such as Clarex L, Clarex ML, Rapidase Press, Rapidase C80L and Klerzyme L200, on yield and quality of plum juice.

b) optimize processing conditions for plum juice production.

c) analyze plum juice made from plum cultivars grown in Michigan for physical, chemical and sensory characteristics.

REVIEW OF LITERATURE

I. Plum Production and Utilization

The term "plum" includes several species of Prunus. The most commonly grown species are P. domestica L. (the European plum) and P. salicina Lindl. (the Japanese plum). Most research on plums deal with these two species. Of minor importance as a source of edible fruits: P. maritima (beach plum), used for jelly and jam, is grown on the coast from Virginia to Canada; P. americana (De Soto and Hawkeye), grown in Connecticut, Montana, Colorado, Texas, and Florida; the wild goose plums (P. hortulana and P. munsoniana), also used for jams and jellies, are found in the Mississippi valley; and the Pacific or western plum, P. subcordata, native to California and Oregon (Childers, 1973; Magness, 1951 and Seelig, 1969), are grown in all states except Alaska (Weinberger, 1975). Michigan was the 3rd largest producer of plums in the nation in 1991 (Michigan Agricultural Statistics, 1992). The leading varieties grown in Michigan are the Italian purple plums, Blufre and Stanley. Stanley, which was used as the control in this study, is a large, partially freestone plum that has firm greenish-yellowish flesh with a dark blue-black outer skin. As indicated earlier, a number of varieties and selections of plums are

being tested in Michigan and several of these were used in this research.

Plums are mainly used for fresh consumption at harvest but storability at refrigerated temperature is poor because of the soft texture and high moisture content. In Michigan, about 4,000 to 5,000 tons go for the fresh market, while the remainder of the crop (ranging from 2,000 to about 7,500 tons) is processed.

The plum growers have a need for and a genuine interest in alternate outlets for new products. One such outlet may be the beverage industry. Since the mid-1980s, packaged soft drink consumers have increasingly picked up on new, upscale or good-for-you beverages. Some of the most significant representatives include "New Age" drinks such as, Clearly Canadian, sports drink king, Gatorade, and the entire spectrum of fruit juices and drinks. The big winner in the New Age fountain has been juice, with juice consumption increasing approximately 10 percent in the last two years alone. A number of companies are beginning to gear up expressly for the juice business. Because of the popularity of Clearly Canadian, the trend in beverage flavors has begun to shift to more northern fruit flavors such as plum and raspberry (Kortbech-Olesen, 1991, Prince, 1992 and Sfiligoj, 1992a, 1992b, 1993a and 1993b). It looks like juices with plum flavor or plum juices may have a very bright future.

II. Plum Juice

Previous research on plum juice production has examined various methods of extracting and clarifying the juice (Ismail et al., 1981; Ichas et al., 1976; Cejkova, 1977, Grinberg and Kolesnich-enko, 1979; Komiyama et al., 1977; Samsonova et al., 1982; Flaumenbaum et al., 1986; Liou and Wu, 1986 and Wani et al., 1990a and 1990b). As with many other juice products, the most promising process methods seem to involve the use of pectinases and clarifying agents. However, additional research is needed on the effectiveness of other enzymes in addition to pectinases on yield, turbidity and filterability of plum juice. This work is intended as an investigation of the effect of commercial pectinases effect on the qualities of plum juice, and processing conditions affecting the quality of plum juice.

A. Liquefaction of raw plums

Most plums contain very little free run juice so simple extraction procedures (i.e.. maceration and cold pressing) are not effective in producing good yields of high quality juice. A number of operations must be carried out, including heating of macerated pulp to inactive polyphenoloxidase (PPO) enzyme responsible for color loss of plum juice (Arnold, 1992 and Siddiq, 1993) and liquefaction of the heated macerate with pectinase enzyme.

The Pectic Substances

Pectic substances have been investigated from several points of view, and work has been done to define their relation to the metabolic changes which take place in fruits and vegetables during maturation and senescence, their relation to plant diseases, their use in the setting of jams and jellies, their use as emulsifiers and their importance in clarification or retention of "cloud" of juices.

Pectin is the intracellular cement of cell-wall tissue occurring in fruits and succulent vegetables (Bailey, 1938). It is a polymer whose major building blocks are units of galacturonic acid linked by alpha-1,4 glycosidic linkages. The water-insoluble pectic material is usually referred to as protopectin. When the solubilized material retains most of its methyl groups and forms gels under certain conditions, it is commonly referred to as pectin. If all of the methyl groups are removed, the remaining polymer of galacturonic acid units is called pectic acid (Kertesz, 1951 and 1959, and Deuel and Stutz, 1958).

The pectins extracted from most fruits under mild conditions usually have the degree of esterification (DE, % of galacturonic acid monomers which are methyl-esterified) of over 70 (Doesburg, 1965). The molecular weight of the soluble pectin and the extent of methylation of the -COOH

group are subject to large variation. Commercial pectins form colloidal solutions and usually 2/3-3/4 of the -COOH groups are methylated. Molecular weights of 50,000-250,000 have been reported for pectin, so that the polymer may contain several hundred units. However, it is not a homopolymer and has been found to contain various proportions of L-rhamnose in the main chain and arabinose, xylose, and galactose in the side chains (Zitko and Bishop, 1965; Smith and Bryant, 1967).

Methods of extraction of pectins from plant tissue are limited. Most investigators have selected conditions for extractions that would yield the highest quantity of the pectin with the particular properties desired (Bender, 1959 and Kertesz, 1951). Pectins similar to those actually present in plant tissue have not been obtained because of the difficulties of avoiding degradation during extraction (Joslyn and Deuel, 1963). Only Gee et al. (1958 and 1959) introduced procedures for the characterization of pectic substances in situ. Pectic substances can be extracted from fruits with dilute hydrochloric acid, oxalate, Versene, or other extractants. Versene-pectinase is recommended by Owen et al. (1952) after they compared all methods, since heating is unnecessary and the method appears to be specific for extracting pectin substances from fruits.

Galacturonic acid is the fundamental unit in the pectin chain, and all present methods to determine pectin content

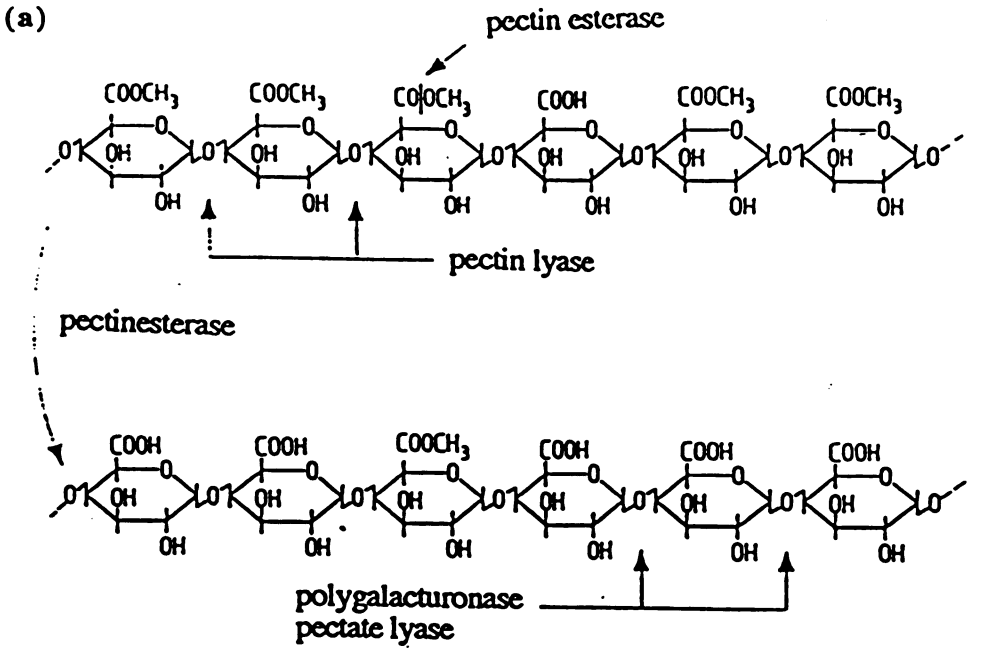
are based on quantification of this acid (Kintner and Van Buren, 1982 and Blumenkrantz et al., 1973). Several types of methods have been proposed: gravimetric, volumetric, and colorimetric. Among them, colorimetric methods are simpler and more selective. They are essentially based on the reaction of uronic acids with sulfuric acid to form 5-formyl-2-furan-carboxylic acid, which may then react with several chromogenic reagents (Scott et al., 1979). Among the possible chromogenic reagents, m-hydroxydiphenyl has the advantage because the color, formed with uronic acids and with their possible accompanying impurities, is stable (Carbonell et al., 1989). Using this method it has been shown that the pectin content of plums or plum jams ranged from 0.37 to 0.70 g monosaccharides/100g fresh weight (Carbonell et al., 1989; Vidal-Valverde et al., 1982).

The Pectic Enzymes

Degradation with purified pectolytic enzymes in apple showed that the side chains of pectin substances are present in blocks called "hairy regions". Enzymatic and chemical degradation of the hairy regions reveals that they consist of arabinogalactan side chains and short xylose side chains. It can be concluded that apple pectin substances are constructed of homogalacturonan, xylogalacturonan and rhamnogalacturonan regions with side chains of arabinogalactan. About 95% of the uronic acid residues are present in the homogalacturonan

regions. The arabinogalactans are highly branched. The methoxyl groups of the uronic acid residues are randomly distributed (Vries et al., 1986).

Pectic enzymes are usually used to extract, liquefy and clarify juice in order to increase yield and reduce viscosity (Cheetham, 1985). The depectinized juice can be concentrated without gelling and without developing turbidity. Using pectinase enzyme with the pulp of red grapes resulted in better release of juice and color pigments. Soluble solids content can also be increased in fruits and vegetable juices through the use of liquefying enzyme. The use of pectic enzymes to improve the quality of juice is very important in many types of juice processing. The protopectinase, pectinesterase, polygalacturonase, pectate lyase and Rhamno-galacturonase enzyme will be studied in this work (Fig. 1). Protopectinase is the name applied to the enzyme activity that converts the natural, insoluble protopectin to a soluble product. Most of the recent workers in the field do not accept the existence of a distinct enzyme which acts on the natural pectin, since thus far no preparation has been isolated which acts in this manner (Reed, 1975). While protopectinases cannot be treated with any clarity or detail, it seems very likely that some enzymes other than the previously identified pectic enzymes play a part in the break down of fruit and vegetable tissues (Josly,1962).



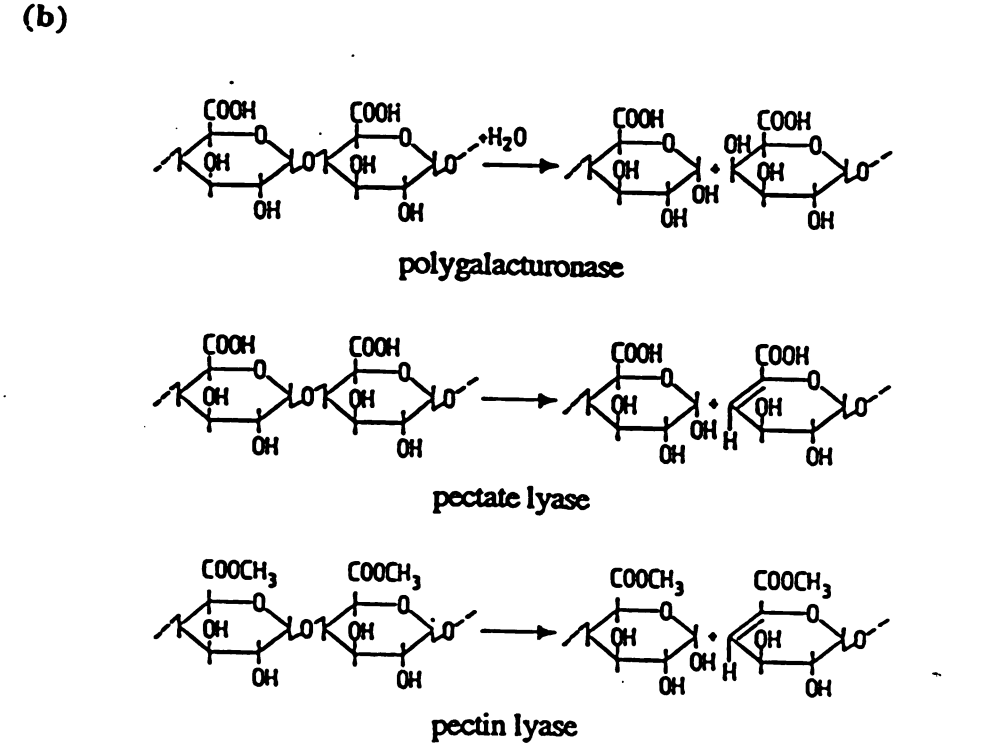


Fig. 1 (a) Fragment of a pectin molecule and points of attack by pectic enzymes. (b) Splitting of glycosidic bonds in pectin by hydrolysis (polygalacturonase) and by ß-elimination (pectate lyase and pectin lyase).

Pectinesterase (PE; EC 3.1.1.11) produces methanol and a free carboxyl group on the galacturonic acid residue by splitting the ester linkage. Irregularities in the galacturonan chain, such as acetylated monomers, ester groups transformed into amides or reduced to primary alcohol, or the occurrence of hairy regions (Vries et al., 1986), inhibit PE activity (Solms and Deuel, 1955). PE is highly specific for the methylester of polygalacturonic acid. Other esters are attacked only very slowly (Manabe, 1973) and the methyester of polymannuronic acid not at all (MacDonnell et al., 1950). The rate of pectin de-esterification depends on chain length; trimethyl trigalacturonate is not attacked at all (McCready and Seegmiller, 1954). Fungal PE differs from plant PE by obeying a multichain mechanism, removing methoxyl groups at random (Ishii et al. 1979).

Polygalacturonases (PG; EC 3.2.1.15 and EC 3.2.1.67) cut the glycosidic linkage between the galacturonic acid units. PG cannot act on the methylated polymer so it is necessary for the esterase to demethylate the pectin to help PG split all the links of the chain and produce free galacturonic acid. The activity measurements by viscosity on methylesters and glycolesters of pectic acid show a rapid decrease in the rate and degree of hydrolysis with increasing esterification (Pilnik et al., 1973).

Rombouts and Thibault (1986) using sugar beet pectin as substrate showed the limitation of degradation by the

presence of acetyl groups and the limit effect of methoxyl groups. The mode of attack is known to differ for PG from different origins. An endo-enzyme may hydrolyze randomly one bond in a single enzyme-substrate encounter, followed by complete dissociation of enzyme and products. This pattern is observed with endo-PG from Kluyveromyces fragilis (Phaff, 1966). It is characterized as multichain attack. In the case of single chain-multiple attack, found for example with PG from Collectotrichum lindemuthianum (English et al., 1972), a single, random hydrolytic scission is followed by a number of non-random attacks on one of the products, resulting in rapid liberation of oligogalacturonates. Many multiple forms and isoenzymes of PG have been described (Pilnik and Rombouts, 1981). Points of difference are dependence on cations and action pattern on oligomers.

Pectate lyase (PAL; EC 4.2.2.2. and 4.2.2.9.) and pectin lyase (PL; EC 4.2.2.10)split glycosidic bonds adjacent to methylester by a beta-elimination reaction, yielding a double bond for each broken glycosidic bond. The best substrate for PL, measured at pH 7.0, is completely esterified pectin as indicated by degradation limits and affinity. At lower pH the affinity for less highly esterified pectins increases and a marked stimulation by Ca^{2+} and other cations is observed. This makes PL a useful enzyme for fruit processing (Ishii and Yokotsuka, 1975). These enzymes need the methylester groups and are inactive on glycol esters and on amidated pectates (Pilnik et al., 1973,1974). PAL does not differentiate

between the methylester and the glycolester of pectic acids. It shows optimum activity on low methoxyl pectins. Changes produced by PAL are conveniently studied since, due to an absolute requirement for Ca^{2+}, enzyme action can be stopped by the addition of a chelating agent (Rombouts, 1972).

Recently, an enzyme has been described (Schols et al., 1990) which splits the galacturonic acid-rhamnose glycosidic linkage in hairy regions of apple pectins with strongly enhanced activity when these have been de-esterified and arabinose has been removed by acid hydrolysis. The enzyme was found in a commercial pectinase preparation and must surely be grouped with pectic enzymes. The end products are oligomers with alternating galacturonic acid and rhamnose units, with rhamnose forming the non reducing end.

Among all these enzymes, PE probably is the most studied of the pectic enzymes. It was discovered in 1840 and was applied as a clarifying and clotting enzyme for orange juice by Cruess (1914). Cruess found that 85º C was a good temperature to destroy the enzyme and prevent juice from clouding. In 1953, Bisset et al. gave the interrelationship between heat treatment, inactivation of PE and clarifying ability. Jansen et al. (1945) applied PE and PG on pectin at pH 4.0 and found it to have a positive effect on the de-esterification rate as compared to the action of pectinesterase alone. The presence of PE in citrus fruit has been studied intensively because of its influence on cloud

loss in orange juice. If PE is not inhibited directly after juice extraction by heat inactivation or by freezing, the pectin will be de-esterified and coagulated by the Ca ion in the juice (Joslyn et al, 1961; Krop, 1974; and Versteeg, 1980b). Dongowski and Bock (1977) did work to determine whether PG could be replaced by cellulases. Voragen et al. (1980) found that a combination of pectinases and cellulases resulted in an almost complete liquefaction of apple pulp.

Commercial Pectic enzymes (Pectinase)

Pectic enzymes are widely distributed in nature but those used commercially are isolated from microorganisms (Bacillus spp., Aspergillus spp., and Saccharomyces spp.) because the quality of pectinase from other sources is usually not good and the price is higher than those from microorganisms. The methods applied in enzyme production have been described by Smythe (1951). Pectic enzymes may be produced commercially by inoculating moist grain or other appropriate material with one of a series of organisms. After incubation for several days, the medium containing the developed culture may be dried and ground to a fine powder. However, the commercial enzymes produced in the U.S. generally come from the purification of filtered extracts of cultures with organic solvents. Recently, liquid forms of pectic enzymes have been introduced to the market.

Commercial pectin reduction enzymes are multi-enzyme complexes consisting of PG, PL, PE, Arabanase, cellulase, hemicellulase and perhaps other enzymes. They break down pectins easily to methanol, saturated monomers and oligomers of galacturonic acid, unsaturated oligomers of galacturonic acid, monomers and oligomers of arabinose, galactose and xylose and low molecular weight arabinogalactans. The activity of the preparation can be adjusted for any specific purpose by genetic technology (Ryu et al., 1980 and Wood, 1985).

Cellulases and hemicellulases (non-pectic enzyme) are usually mixed with commercial pectinase because they aid the liquefaction processes. Cellulase alone had little influence on pectin and solubilized only 22% of cellulose but when combined with pectinase, a synergistic effect has been shown, which has higher solubilization of pulp (Voragen et al, 1980a). Hemicellulases have been used with apple pulp (Voragen et al, 1982) to release arabans of various structures and degrees of polymerization, including oligo- and monomers from cell wall, but they did not show an enhancing effect on pulp liquefaction. When ultrafiltration has been used to clarify fruit juice, hemicellulases have raised the soluble solids level in the juice because the lower-molecular weight degradation products can pass through the membrane (Pilnik and Voragen, 1986).

B. Clarification of the plum juice

Consumers today demand not only good flavor and safety in their fruit juice but also in most juices, a highly clarified products. However, clarification of juice products with unstable colloidal systems can be an expensive and bothersome process. It is necessary to accelerate the formation of haze and sediment, then find a method to remove this sediment. Direct centrifugation or filtration is possible but is often not economically feasible. Not only will the presence of soluble pectin make the juice viscous and the rate of filtration slow but further changes after filtration may lead to the loss of clarity if the juice isn't stabilized by heating. Therefore, fining and depectinizing procedures are often applied before juice is filtered and stabilized by heat. Endo (1964) showed that clarification of apple juice involved both enzymic depectinization and electrostatic neutralization. He divided the procedures into three parts: 1) solubilization of insoluble pectin; 2) decrease in viscosity of soluble pectin and; 3) flocculation of suspended particles.

Scott et al. (1965), and Baker and Bruemmer (1969) analyzed the cloud in orange juice using chemical methods. They pointed out that the cloud consisted of 25% lipid and about 35% of protein encapsulated by pectin. Yamasaki et al. (1964, 1967) found that cloud consisted about 36% protein-pectin. Krug (1969) and Krebs (1971) showed strong evidence

that solubilized starch and starch-tannin complexes are the major part of haze and sediment in apple juice. Later, Heatherbell (1976) summarized that the major classes of compounds which contributed to the formation of haze and sediment in apple juice are pectins, starch, proteins, polyphenolics, tannins, polyvalent cations, acids, and lipids. The tendency of protein, starches and tannins to aggregate resulting in particle formation develops the haze and turbidity. There is no doubt that the lager size (0.1 mm-0.5 mm) particles of turbid juice can settle out quickly and centrifugation and classic filtration can take care of the coarse dispersion material easily (Fig. 2). However, the very minute particles of pulp or colloidal materials, which range in size from 0.001μ to 0.1μ, may not be removed with filters or centrifuges (Heatherbell, 1984). Several methods may be applied to juice clarification. These are:

1. Physical processes: settling, centrifugation and filtration.

2. Biochemical processes: addition of pectinase, amylase and proteases.

3. Chemical processes: fining.

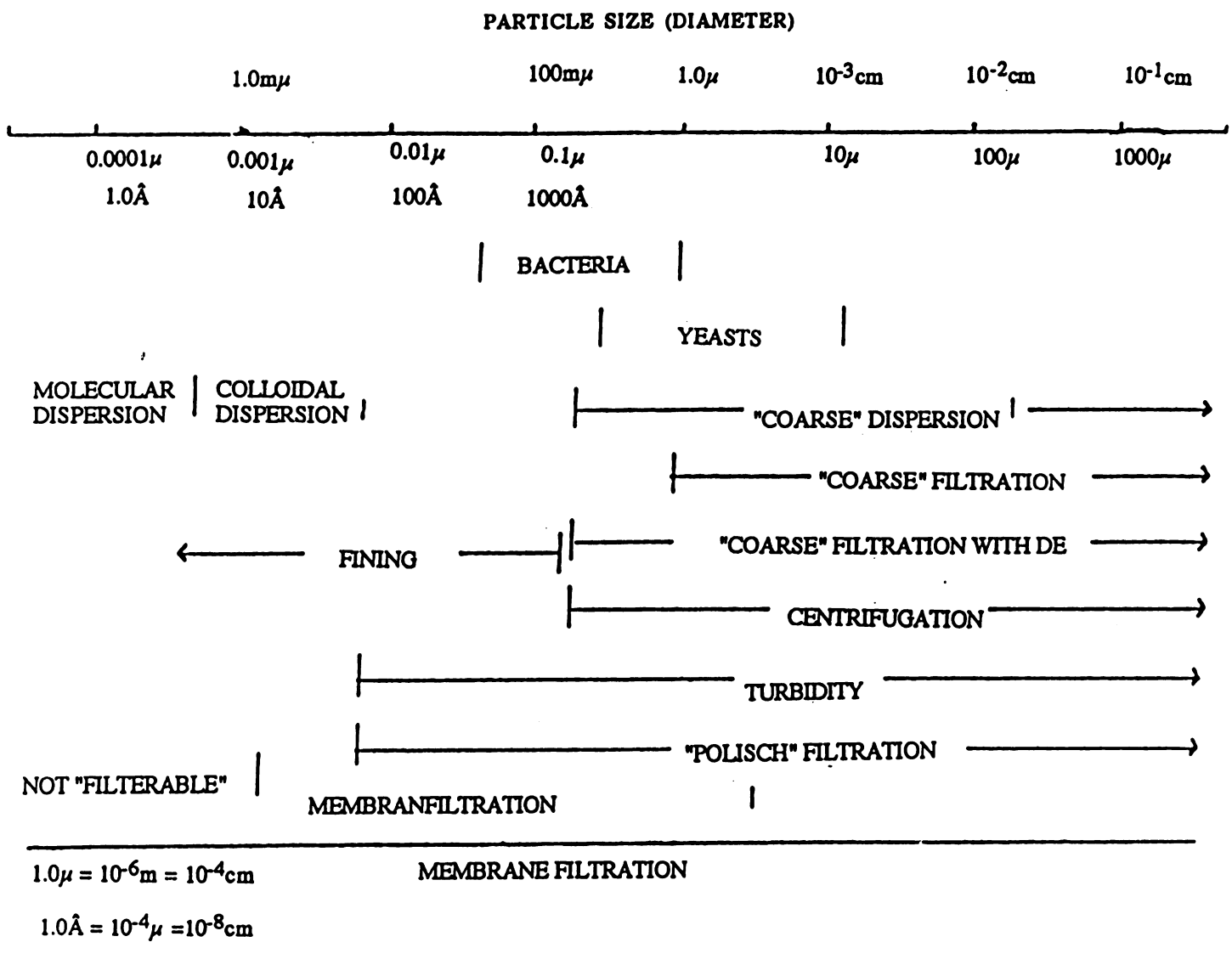


Fig. 2 Clarification Processes and Particle sizes in dispersion

Enzymes for clarification

Enzymes were originally applied to apple juice and grape juice (Kertesz, 1930 and Mehlitz, 1930) in the form of pectinases which were used to clarify these juices. The raw pressed juices obtained from these fruits were viscous and cloudy so enzyme treatment was necessary. The mechanism of enzyme clarification in apple juice at a pH of 3.5-4.0 is dependent on the fact that surfaces of the particles are negatively charged and the core is positively charged protein. Partial hydrolysis of the negatively charged coat leads to exposure of positively charged surfaces so electrostatic attraction will form a floc which is too large to remain in suspension (Fig. 3). As the enzyme works, the reduction of viscosity can be observed visually and the fine haze in the juice begins to agglomerate, forming a floc which settles rapidly. All of these changes can be affected by pH, temperature, amount of enzyme and type of juice. According to Neubeck (1975) and Yamasaki (1964), the higher the temperature, the shorter the flocculation time in apple juice, up to the point of heat denaturation of enzyme. Since the floc formation is due to electrostatic interaction, juices with lower pH's clarify more easily than those with higher pH.

Neubeck (1975) found that haze formed in juice from unripe apples was due to starch which remained in the juice. The haze was removed by adding fungal amylases, heating to

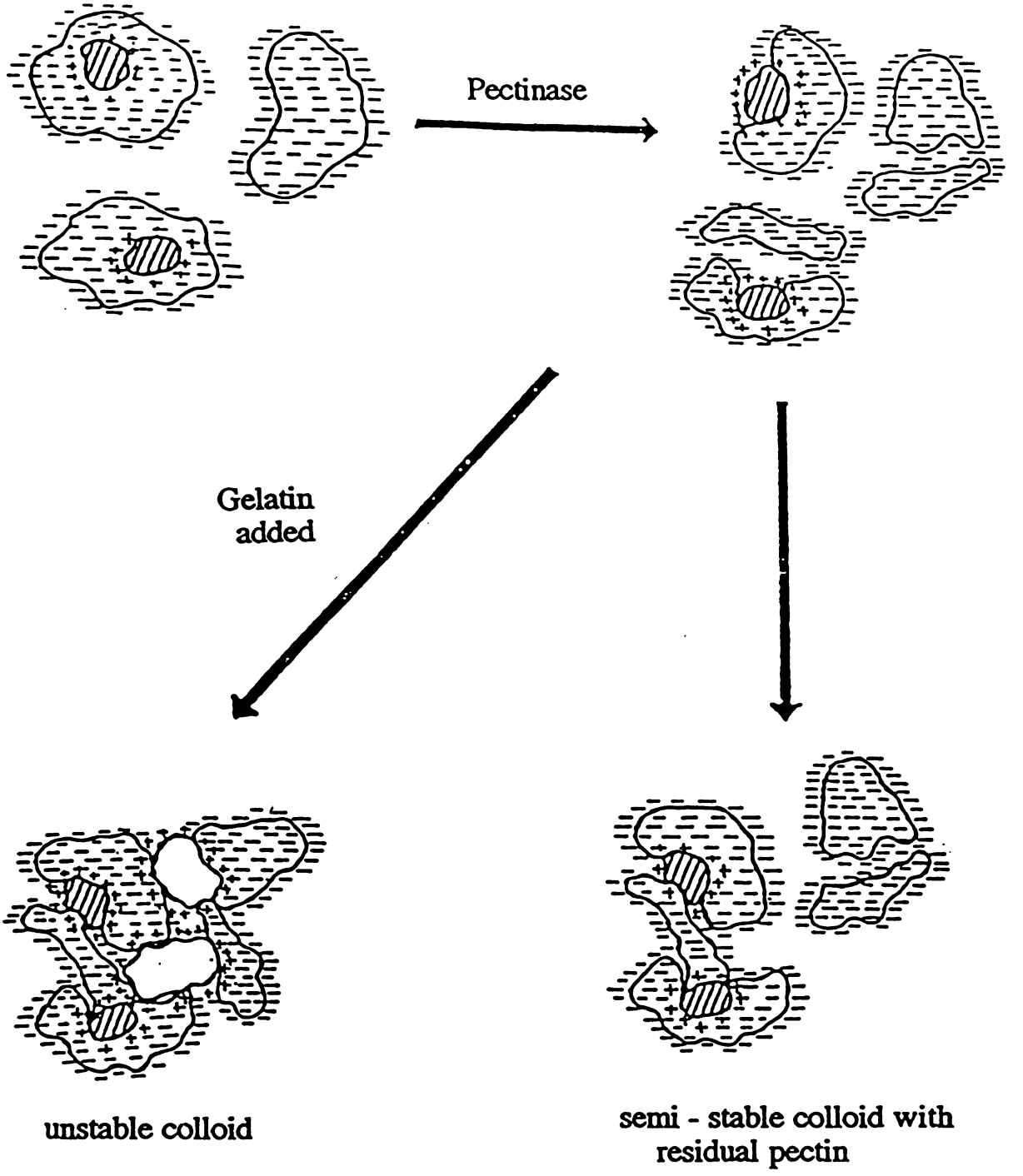
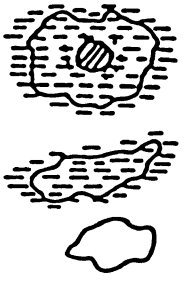


Fig. 3 Theoretical mechanism of clarification with and without gelatin (From: Heatherbell, D.A., 1976)



hazy particles consisting of positively charged nucleus surrounded by pectin

pectin

gelatin

gelatinize the starch, then cooling and adding pectic enzyme and diastase. This "hot clarification" process has been found to reduce apple juice processing time to less than three hours while improving the final juice product (Anon., 1983).

Through the use of enzymes, juice clarification is usually very good but it is possible for reclouding to occur in the juice after processing. Fining and/or filtration to remove flocculated materials after enzyme treatment will reduce this possibility by removing residual short chain pectin and pectin-phenolic-protein complex.

Fining agents

Gelatin

Gelatin is probably the most readily available and one of the most commonly used fining agents. It is obtained by the partial hydrolysis of collagen derived from the skin, white connective tissues, and bones of animals. Two main types are produced: Type A, derived from an acid-treated precursor, and Type B, derived from an alkali-treated precursor. Isoelectric points vary from pH 7 to pH 9 for type A and around pH 5 for type B. At juice pHs, both types will carry positive charges (Bannach, 1984).

Yamasaki et al. (1964) and Zitko et al. (1962) provided the mechanisms for gelatin reaction (Fig. 3). The positively charged gelatin molecules work on the negatively charged polyphenols and aggregate into bonds that are dissolved colloidally. Depending on the degree of condensation and oxidation of the polyphenols, these bonds form together into bigger particles, flocculate, and absorb other constituents that cause turbidity to settle out of solution. The hydrogen bonding between tannin and gelatin combined with an electrostatic reaction between gelatin and cloud particles is the key step. In juice, low levels of gelatin favor the formation of tannin-gelatin complexes while high concentrations of gelatin facilitates the formation of pectin-gelatin-tannin complex. Most of the research on gelatin fining has shown that polyphenols were reduced by gelatin. Therefore, the application of gelatin as a fining agent not only improves the filterability of juice but it may also play a positive role in improving the flavor, color, and odor by removing excess phenolic compounds.

The amount of gelatin to use can be determined by the pH of the juice, the concentration of tannin, the concentration of pectin and the degree of esterfication of the pectin. Preliminary tests are required to establish the procedure and concentration of gelatin but in general 25-150 mg gelatin in 1 liter juice has been suggested by Polyakova et al. (1983) and Zinchenko et al. (1983).

Bentonite

Bentonite is a clay mineral of the montmorillonite type ($Al_2O_3.4SiO_4.nH_2O$). It consists of tiny platelets that carry negative charges that have the ability of binding protein to entrap haze particles and to help remove the particles from suspension. Anthocyanins (Tarytsa, 1988) can be reduced 35-56% (only 5.3% lost by centrifugation) depending on the amount of bentonite used. Dul'neva et al. (1987 and 1988) found that heavy metal content was decreased by use of bentonite and the content of As and F were fully removed after 1 min. exposure.

Because of the slow settling rate of unflocculated bentonite, a combination of bentonite and gelatin is usually applied to remove haze. Polyakova and Filippovich (1983) indicated clarification was also much more rapid and far more economical with the bentonite/gelatin combination (added at 2-3g/L) than with the only gelatin or pectinase/gelatin.

Silica sols

Silica sols, composed of specially prepared colloidal silicon dioxide particles, absorb protein. When silica sol and proteins are mixed in suitable proportions, a light, curdy coagulum forms which may settle out and be filtered.

This has proven to be the most effective way to remove protein from beer and several other protein containing beverages (Hahn and Possmann, 1977). Silica sols may also be used in combination with gelatin (Lehmann et al., 1985; Soto-Peralta et al., 1989 and Hernandez, 1982). Wucherpfennig and Possmann (1972) found that they had good results when they used six times as much SiO_2 as gelatin.

Others

Other materials which have been used as clarifying materials are diatomaceous earth (Jones et al, 1983 and Baumann, 1989), tannin (Kamenskaya et al, 1990), casein (Lodge and Heatherbell, 1976) and honey (Wakayama et al., 1987a and 1987b, and McLellan et al., 1985). Most of these materials utilize the same principles of fining and induced flocculation as the agents already described.

Filtration

In general, solid particles are separated out of liquid by filtration processes. Many techniques are applied to prepare juice for filtration, such as filter aid addition, settling, decantation, and centrifugation. Particles gradually settle to the bottom of the tank when they flocculate during fining treatment. The clear portion is

easily filtered with a minimum amount of filter aid required. A high percentage of the solids in juice can also be removed by centrifugation.

Several types of filters are used successfully in fruit juice filtration (Oechsle, 1984). The most common ones include: pressure filters (plate and frame filter; leaf filter), vacuum filter (tube and leaf vacuum filter; rotary vacuum filter), and membrane filters (ultrafiltration; reverse osmosis).

There are a number of pros and cons associated with each type of filter system but the final choice often depends on economics and energy expense. In recent years membrane technology has advanced to the point that this type of filtration has wide acceptance in the juice industry. Membrane filtration has a number of advantages including: (1) the ability to remove many different types of suspended material (i.e. protein, starch, gums and polymerized tannins) from juice using a single processing step; (2) complete removal of the need for fining agents thus resulting in reduction of production costs and the problem of waste treatment; (3) improved consistency and juice quality; (4) increased juice yield; (5) reduced labor cost; and (6) potential reuse of pectinase (Keefe, 1983; Kim et al., 1989). Heatherbell et al. (1977) was one of the first to clarify apple juice by UF and obtain a stable, and clear product. Recently, a number of other researchers have used this

technology for clarification of juices (Wu et al., 1990; Rao et al., 1987; Elliott, 1990; Chamchong, 1991 and Ben Amar et al., 1990). However, this type of technology is not without problems. In separation processes, the filtration rate can be limited by concentration polarization near the membrane surface, or by membrane fouling. Therefore, the selection of a suitable membrane for the particular juice product and a proper cleaning method are critical for successful use in the processing system.

Heat treatment

Heat treatment of juice product serves several functions, including sterilization, coagulation of protein and inactivation of enzymes. Heating juices may coagulate protein, which can then be filtered out of to clarify the product, however, this process is not effective for all juices because the suspended material is not always protein. A haze caused by starch should be treated with amylase to remove it from juice. In study by Wicher (1988), a temperature of 65º-66ºC was found to be sufficient to kill spoilage organisms, but the juice was treated at 80ºC/2min to inactive enzymes that caused clouding of juice (Wicker, 1988). Siddiq (1993) indicated that heating of plum juice for 5 min. at 75ºC completely inactivated the polyphenol oxidase (PPO) which caused browning.

MATERIALS AND METHODS

I. Plum Samples

Seven plum cultivars, " Stanley", "Au Red", "Abundance", "Pobeda", "Shiro", "Peach Plum" and "Early Golden" from Michigan State University's Northwest Horticultural Experiment Station were brought to the Department of Food Science and Human Nutrition. The plums were harvested in 1992 at optimum maturity and stored at -20°C until processed.

II. Commercial Enzymes for Plum Juice Extraction

The basic plan employed was to determine which commercial pectinase would give the best yield of high quality juice. Several commercial pectic enzymes, including Clarex L and Clarex ML from Solvay Enzymes, Inc. (1230 Randolph St., Elkhart, IN 46514) and Klerzyme 200, Rapidase C-80 Liquid and Rapidase Press from Gist-Brocades Food Ingredients, Inc. (2200 Renaissance Blvd. Suite 150, King of Prussia, PA 19406), were utilized for the initial "Stanley" plum juice processing.

III. Plum Juice

Stanley plums were processed into juice using standard procedures previously developed in our laboratory (Arnold, 1992; Siddiq, 1993). For this study, 300-350g plums held at

-20ºC were thawed for 8-12 hours at 4ºC. The fruits were then crushed, macerated and heated to 82ºC in a stainless steel steam jacketed kettle. The macerated plums were put into 500ml stainless steel pans. Commercial enzyme in concentrations of 0.05%, 0.1%, 0.2%, 0.4% or 0.6% w/w plums were added to aid in liquefaction of plums (Table 1). After holding for 2-3 hours at 49ºC (specific conditions for each enzyme as indicated by manufacturer), the fruit was pressed through several layers of cheesecloth to obtained plum juice. This juice was used to determine the effect of pectic enzyme treatment on juice yield, juice clarity, pH,, total acidity, sugar composition, total phenolics, color and anthocyanins (ACYs). The enzyme which produced optimum yield and quality was used to produce plum juice from the six different varieties previously mentioned. All processing trials were done in duplicate.

IV. Plum Juice Processing Conditions

Plums from each of the six varieties were thawed and crushed. A 0.2% solution of Clarex L pectinase (based an prior results from the preliminary Stanley studies) was added and incubated at 49ºC for 3 hours before pressing. Juice was pressed and filtered through several layers of cheesecloth. Juice samples were then heated in an HTST unit to 85-90ºC for 90 sec and cooled to 4ºC. Bentonite and gelatin were used to clarify the juice. The flow diagram of juice processing is shown in Fig. 4.

Table 1. Enzyme preparation, composition and optimum processing states for extraction trials on Stanley Plums (1992)

Enzyme Preparation	Reported Composition	Optimum pH	Process Condition
Clarex L[1]	Pectinase, cellulase, hemicellulase and protease	3.5-5.0	49°C/4 hrs
Clarex ML[1]	Pectinase, cellulase and hemicellulase	3.5-5.0	49°C/4 hrs
Rapidase Press[2]	Pectin esterase, polygalacturonase, pectin lyase and arabanase	4.0-5.0	49°C/3 hrs
Rapidase C80[2] Liquid	Pectin esterase, polygalacturonase, pectin lyase and arabanase	4.0-5.0	49°C/ 2.5hrs
Klerzyme 200[2]	Pectin esterase, polygalacturonase, pectin lyase and arabanase	4.2-4.6	49°C/ 2hrs

1 Solvay Enzymes, Inc. P.O. Box 4226, Elkhart, IN 46514-0226

2 Gist-Brocades Food Ingredients, Inc. 2200 Renaissance Blvd. Suite 150, King of Prussia, PA 19406

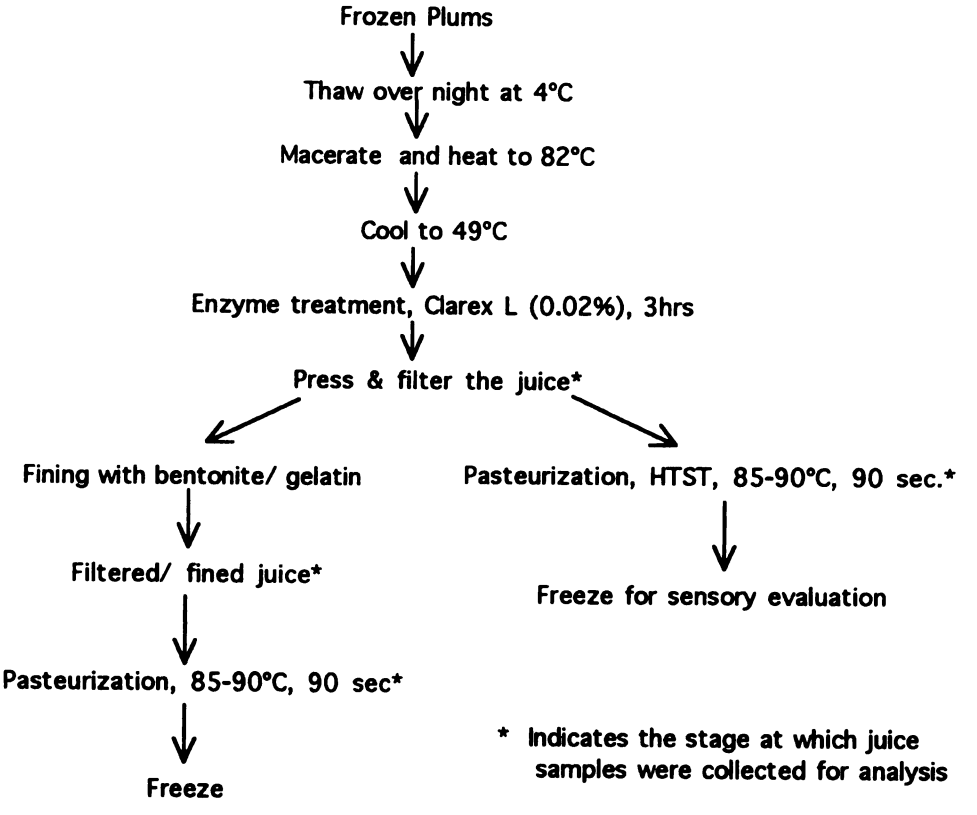


Fig. 4 Flow chart of plum juice processing

V. Analysis

pH

The pH of samples was measured in duplicate using a digital pH meter (Model 601A, Corning Glass Works, Medfield, MA.).

Soluble Solids

Percent soluble solids, expressed as ºBrix, of juice samples were determined with an Abbe-3L (Bausch & Lomb Optical Co., Rochester, NY.) refractometer (sensitivity 0.1%) at 20ºC. The refractometer was calibrated using distilled water.

Titratable Acidity

A 10 ml sample of juice in 100 ml distilled water was titrated to pH 8.0 with a 0.1N NaOH solution using Corning Model 7 pH meter (Corning, NY). Titratable acids of the sample were expressed as percent malic acid (Pangelova, 1979 and Gur, 1986) by volume using following formula:

% malic Acid = ml NaOH x N NaOH x 0.067 mEq x 100/mls sample

Color

Color was measured on the Hunter Color Difference Meter (D25 DP-9000 system, Hunter Associates Laboratory, 11491 Sunset Hills Road, Reston, VA 22090). Fifty mls of each juice sample were placed in a standard optical cell for the

measurement after standardization with a pink tile (L=73.49; a=17.34; b=10.28) for red cultivars and a yellow tile (L=82.09; a=-1.1; b=28.9) for yellow cultivars. This system is based on the Hunter L, a and b coordinates. L representing lightness and darkness, +a redness, -a greenness, +b yellowness and -b blueness.

Sugar Analysis

Glucose, fructose, sucrose and sorbitol are the main sugars in plums (Richmond, M. L. et al. 1981). The separation of glucose and sorbitol by HPLC has been shown to be very difficult because of their structural similarities (Shaw, 1988). This problem was solved by combining the results of two different analytical techniques: HPLC & YSI analyzer (Yellow Springs Instrument Co., Inc., Yellow Springs, OH). The YSI glucose analyzer uses immobilized enzymes bound on a membrane to give accurate determination of the glucose content of samples. In this study, HPLC was used to separate fructose, sucrose and a combined glucose and sorbitol peak. The YSI analyzer was then used to determine the content of glucose in the same sample of juice that had been separated by HPLC. Sorbitol content of plum juice was determined by subtracting the glucose content of juice determined by YSI from glucose & sorbitol measurements made by HPLC. This method was confirmed (Table 2) by enzyme kits of glucose, fructose, sucrose and sorbitol from Boehringer Mannheim Biochemicals (Indianapolis, IN). The correlation

Table 2 Analysis of sugar contents of plum juice by enzymes and combination of YSI and HPLC

Varieties	Glucose	Fructose	Sucrose	Sorbitol	Methods
Au Red	6.16	5.88	0.15	1.65	Enz
	6.03	5.91	0.10	1.61	H & Y
Abundance	1.87	2.41	2.39	0.12	Enz
	1.84	2.50	2.41	0.05	H & Y
Pobeda	2.10	1.11	3.74	0.11	Enz
	1.90	1.21	3.73	0.05	H & Y
Shiro	2.85	1.52	4.79	0.44	Enz
	2.70	1.64	4.73	0.43	H & Y
Peach Plum	2.17	1.24	7.39	0.09	Enz
	2.15	1.29	7.45	0.04	H & Y
Early Golden	3.18	2.55	3.79	0.01	Enz
	3.18	2.67	3.80	0.01	H & Y

Units are g/100ml juice
Enz: Enzyme analysis. Enzyme Kits (Boehringer Mannheim Biochemicals, Indianpolis, IN) for analysis of glucose, fructose, sucrose and sorbitol
H & Y: Combined HPLC with YSI methods to determine the glucose, fructose, sucrose and sorbitol in juice.

between enzyme and combination methods is significant ($r=0.999$, $p<0.01$) so the method developed for this study for sugar determination is acceptable.

a. **HPLC**

For HPLC analysis of sugars, a 10 ml aliquot of plum juice was mixed with 10 ml HPLC grade methanol, followed by centrifugation at 10,000 g for 10 minutes in a refrigerated centrifuge. After centrifugation the supernatant was clarified using 0.45μm Millipore filter paper. The standards for sugar analysis consisted of 250 mg of sucrose, glucose and fructose dissolved in 25 ml deionized water in a volumetric flask. A 10 ml aliquot of standard sugar solution was mixed with 10 ml of methanol, centrifuged and clarified the same as the plum juice.

A Waters Model M-45 solvent delivery system, a U6K injector, and a Model R 401 RI detector (Waters Associates., Inc,. Milford, MA 01757) along with a Shimadzu CR 601 (Shimadzu Scientific Instruments Inc. Columbia. MA) integrator was used. The sugars were separated on All-Tech 600Ch silica based amino bonded phase column with direct connect refillable guard column having pellicular NH2 packing. The mobile phase was acetonitrile-water (75:25) and the operating conditions were: flow rate, 1.20 ml/ min.; attenuation, 4 X; chart speed 10 mm/ min. Samples of 10μl of standard or juice were injected into the HPLC for sugar

analysis. Duplicate samples were analyzed. Quantification was based on the absolute calibration curve method.

b. **YSI : Determination of glucose**

A 10 ml sample of centrifuged plum juice was diluted to 20 ml for YSI analysis. A standard glucose solution was obtained from YSI (Yellow Springs, OH) containing 1.8 g/L glucose. These standards were stable for 6 months at room temperature.

The YSI analyzer equipped with immobilized enzyme membranes was used for glucose determination. In this system, the glucose oxidase is immobilized on a thin microporous membrane. When a substrate containing glucose diffuses into the membrane, the reaction of enzyme and substrate produces hydrogen peroxide. The hydrogen peroxide is then oxidized at the platinum anode, producing an electrical current that is directly proportional to hydrogen peroxide concentration, and hence substrate concentration. Relative precision of replicate analyses is better than 2% and agreement with AOAC methods is very good (Weetal, 1975 and Mason, 1983). The YSI analyzer automatically samples 0.5 ml of liquid for analysis.

Turbidity

Turbidity measurements were done according to the methods of Krop et al. (1974) for citrus juice and Ough et al. (1975) for red grape wine. At specified time, the juice sample was

shaken and 10 ml portions of samples were centrifuged for 10 minutes at 360 X g to remove pulp and coarse cloud particles. Percent transmittance was determined at 660 nm on a Milton Roy Spectronic-70 spectrophotometer (820 Linden Avenue, Rochester, NY 14625) with distilled water as blank. The percent transmittance was considered a measure for the cloudiness and duplicate runs were made for each sample.

Total Anthocyanins

Total ACY in the plum juice was measured spectrotometrically at 535 nm. A 5 ml sample of juice was mixed with 45 ml acidified ethanol (15 ml, 1.5N HCl+ 85 ml, 95% ethanol) left for 5 min. and filtered though a No. 2 Buchner funnel. The pH of the solvent was adjusted as required to obtain a final pH of 1.0 in the plum extract. The diluted extract was stored in the dark for two hours before absorbance measurement on the spectrophotometer (Milton Roy Spectronic-70, Rochester, NY). The total anthocyanin content was calculated with the aid of the appropriate volume, dilution factors and E(=98.2) values (Fuleki, T. et al. 1968a, 1968b; Wrolstad, 1976; Francis, 1982).

Total Phenolics

The tannic acid concentration of each plum juice sample was determined by the method of Singleton and Rossi (1964). Tannic acid was used to assay the total phenolics. For the

preparation of the calibration curve, 0, 1, 2, 3, 5, and 10 ml aliquots of tannic acid stock solution (0.5g of dry tannic acid dissolved in 10 ml of ethanol and diluted to 100 ml volume with water) were pipetted into 100 ml volumetric flasks, and diluted to volume with water. The tannic acid concentrations of these solutions were 0, 50, 100, 150, 250, and 500 mg/L. Analysis consisted of mixing 1 ml sample (or standard) with at least 60 ml of water in a 100 ml volumetric flask. Folin-Ciocalteu reagent, 5 ml, was added and mixed. After about 30 seconds 3 g of anhydrous Na_2CO_3 in aqueous solution.(e.g. 15 ml of 20% solution) was added, and the contents of the flask made to volume. The absorbance was determined after 2 hours at 24ºC, using a 1 cm cell with the spectrophotometer set (Milton Roy, Rochester, NY) at a wavelength of 765 nm. The blank used for zero absorbance was water.

Browning

Tristimulus reflectance colorimetry (usually the measurement of Rd or Hunter values) has been used to follow the extent of enzymatic browning in juice (Smith and Cline, 1984) and apple slices (Ponting et al., 1972). For this study, the method and the time selected was according to Sapers et al. (1987). Juice samples were poured into a glass cylinder, and the "L" value was measured with a Hunter Color Difference meter (Model D25 DP-9000). The degree of

browning was expressed as the L value differences at times 0 and 60 min. All analyses were carried out in duplicate.

Pectins

The procedure for extraction and colorimetric analyses of pectic substance followed was a combination of Mc Cready et al. (1952 & 1970) and Kintner et al. (1982).

A. Versene-Pectinase Extraction of Pectin

10 mls of juice were mixed in a beaker for 5 minutes with 150 ml 95% ethanol. The sample was filtered and the ethanol containing the sugars was discarded. The pulp was washed twice with 75% ethanol then transferred to a 250 ml beaker. Cations were sequestered and the pectin deesterified with 100 ml of a 0.5% Versene solution at pH 11.5 (adjusted with 1N NaOH) for 30 min. The mixture was acidified to pH 5 with acetic acid and 0.1g of pectinase was added, stirred for one hour, diluted to 200 ml and filtered. The first few milliliters of filtrate were discarded before collecting 2 mls of sample for analysis.

B. Colorimetric Determination of Galacturonic Acid

A 1 ml sample containing pectin was pipetted into a 15x180 mm Pyrex test tube and placed in ice-water bath for 5 minutes. Subsequently 6 ml H_2SO_4/ tetraborate solution (0.0125M solution of sodium tetraborate was prepared in concentrated sulfuric acid) was added to each tube in the ice

water and the tube was shaken carefully on Vortex mixer. The mixture was heated in a 100ºC water bath for precisely 5 min. and immediately placed in an ice-water bath to cool. Duplicate samples were developed by adding 0.1 ml 0.15% m-hydroxydiphenyl solution (in 0.5% sodium hydroxide) mixing and allowing to stand for at least 20 min. at room temperature to allow bubbles to dissipate (absorption values were stable for up to 1 hour). A sample blank was prepared by replacing m-hydroxydiphenyl with 0.1 ml 0.5% NaOH, keeping all other additions and treatments similar. The sample blank absorbance was later subtracted from the total absorbance to obtain the absorbance due to m-hydroxydiphenyl. Absorption measurements were taken at 520 nm using a Milton Roy Spectronic-70 spectrophotometer. A reagent blank containing 1 ml distilled water, 6 ml Sulfuric acid/ tetraborate solution and 0.1 ml of 0.5% Sodium hydroxide was used to zero all instruments.

VI. Sensory Evaluation

Preliminary sensory trials were done with a limited number of panelist (8-10) from our lab to determine the optimum level of Brix for each of the juice samples. Then each of the samples was adjusted by sucrose, based on the preliminary trials, and samples were tested by a larger panel for, tartness, bitterness, color, flavor preference and acceptability using an unstructured 10 cm hedonic scale. Judges were asked to mark the horizontal scale at the point

that most closely corresponded to their judgment of the intensity of each attribute. These points were then measured in cm and translated into numerical values for statistical analysis. Each panel consisted of 40-45 panelists from the faculty, staff and students in the Food Science and Human Nutrition department. Panelists ranged in age from 18-55 years old. All tests were conducted in the sensory evaluation laboratory of the Department of Food Science and Human Nutrition, Michigan State University, under cool white fluorescent lighting. All samples were evaluated twice and the data were analyzed by the Analysis of Variance using Super ANOVA (1989-1991, Abacus Concepts, Inc., 1984 Bonita Ave., Berkeley, CA.), with LSD at 5% level used to separate variety means.

VII. Statistical Analysis

In this study, the experiment was designed as a three factor (replication x enzyme x concentration) & (replication x varieties x processes) randomized model with balanced data. All determination values were made in duplicate, except for the value for color, which was determined in triplicate. Mean, standard errors, mean square errors, one factor ANOVA (analysis of variance), two factor ANOVA, correlation and interaction of main effects were done using the SuperANOVA software (Berkeley, CA). Mean separations were performed using LSD with the mean square error term at the 5% level of probability.

RESULTS AND DISCUSSIONS

Effect of Commercial Pectinases on Plum Juice Extraction and its quality

Five commercial grade pectinase enzymes were used in this study to extract juice from Stanley plums. These were (a) Clarex L, obtained from Aspergillus niger, which hydrolyzes both colloidal and soluble pectins, and has been used on apple, pear and grape to clarify and increase juice yield; (b) Clarex ML, derived from selected strains of Aspergillus niger and Trichoderma reesei, this enzyme system having pectinase, hemicellulase, and cellulase activity, is usually applied in fruit juice extraction that employs maceration or liquefaction of fruits. The role of this enzyme is to increase overall juice volume, and decrease mash viscosity. (c) Rapidase Press, extracted from Aspergillus niger is designed to increase juice yield and aid in press efficiency. (d) Rapidase C80L, this enzyme is used primarily by the wine and juice industry to improve clarification and extraction, increase rate of pressing and prevent the formation of pectin gels. It possesses a range of enzymatic (pectin esterase, polygalacturonase, pectin lyase, arabanase, etc.) activities and is especially adapted for depectinization of apple juice. (e) Klerzyme L200, derived from Aspergillus niger, the

functions of this enzyme are for the rapid depectinization and clarification of fruit juices, and the extraction of juice from crushed fruits.

Investigations into use of these commercially available enzymes on plum juice yield, clarity and quality were made. The purpose of this work was to identify the enzyme which gave maximum yield and quality of plum juice. The enzyme which gave the best results in these preliminary studies was then used for the extraction of the various plum cultivars used in later efforts.

Juice Yield (% by weight)

When the enzymes listed above were added to macerated plum in concentrations ranging from 0.05% to 0.6% (w enzyme/w plums), yields of juice increased with increasing concentration of enzyme (Fig. 5). Juice yield of the untreated controls was approximately 38% (weight of juice /weight of plum fruits) as compared to yields that ranged from 60-73% in the enzyme treated samples. The juice yields show the different results that are dependent on the enzymes and use level (Table 3). Clarex L, Clarex ML and Rapidase C80L showed highest yield (about 70%), while the Klerzyme L200, treated samples gave yields of only 56%. The 0.2% to 0.6% Clarex L on Stanley showed the highest juice yield whereas Rapidase Press gave the highest yield at 0.1% levels. The Clarex ML, Rapidase C80L and Klerzyme L200 at 0.4% and 0.6% use levels gave the best yield. Arnold (1992)

Fig. 5 Effect of enzymes on juice yield from Stanley plums

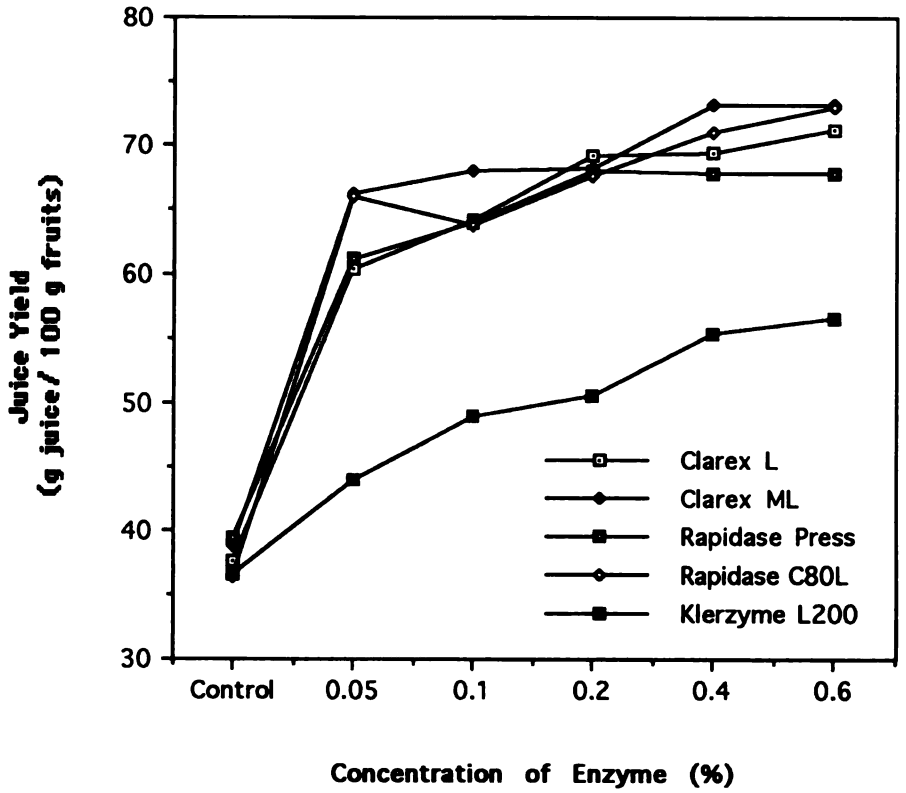


Table 3 Effects of Enzymes on Juice Yield from Stanley Plum

Enzymes	Levels of Enzyme (% by weight)					
	Control	0.05	0.1	0.2	0.4	0.6
Clarex L	37.55a[1]	60.39b	64.29c	69.29d	69.45d	71.29d
Clarex ML	38.83a[2]	66.19b	67.91b	68.12b	73.16c	73.18c
Rapidase Press	39.41a	61.28b	63.95bc	68.03c	67.79c	67.84c
Rapidase C80L	36.36a	66.01b	63.81b	67.70bc	71.03cd	73.00d
Klerzyme L200	36.65a	43.92b	49.01c	50.68c	55.37d	56.70d

1 unit is g juice/100 g fruits

2 Comparisons are between enzyme concentrations. Values with the same letter in horizontal rows are not significantly different at 5% level of significance. This table does not show comparison between enzymes.

reported that use of 0.25% pectinase on Stanley plums (1990) resulted in juice yield increases of 59%. The different results from this study and the one by Arnold may be due to harvest and maturity differences.

The increased juice yield by addition of enzymes is due to their action on the plant tissue. A combination of various exo- and endopectinases (which hydrolyze the pectic substances through the alpha-1-4-glycosidic bond), and hemicellulases and cellulase liberate the pectin through hydrolysis of polysaccharides, resulting in free expulsion of juice (Bielig et al., 1971; Ough et al., 1973). Meischak (1971) and Samsonova et al. (1982) reported that plum juice could not be satisfactorily expressed without the use of cell wall degrading enzymes but when too much enzyme was used, the release of total phenolics made the juice bitter. In this study, higher levels of all five commercial enzymes (beyond 0.2%) tended to make the juice slightly bitter according the the sensory tests. Therefore, the addition of 0.2% enzyme to the macerated plum pulp was considered to be optimal. Even at this enzyme level juice increases were high and quality changes were minimum.

Juice Clarity

The percent transmittance (% T) was taken as a measure of clarity. Juice with 60% transmittance was considered as Clarified (Amir-uz-Zaman, 1985). The enzyme treated juice was free from sediments and had clear consistency as indicated by

significantly higher % T compared to the control juice. Fig. 6 shows the effect of varying concentrations of the different pectinases on plum juice clarity. Beyond 0.1% enzyme concentration, no significant effect on juice clarity (64% T) was obtained for Clarex L and Rapidase C80L (Table 4). Klerzyme L200 showed the highest transmittance values, whereas Rapidase Press treated juice had the lowest transmittance values. Addition of pectinase not only lowered the viscosity but also caused cloud particles to aggregate to larger units which were easily removed by centrifugation and filtration. In grapes these enzymes increased the average juice clarity fourfold and filterability by 100% (Ough and Berg, 1974; Brown and Ough, 1981).

Color

Hunter CDM color values and hue angle are shown in table 5. Color value (L, a, b) of enzyme treated plum juice was significantly different from control juice. Hunter L value represents the lightness and darkness with higher L being lighter and lower L being darker, '+a' is redness, '-a' is greenness, '+b' is yellowness and '-b' is blueness. Addition of enzymes resulted in decreases in all values in this study and varying the concentrations of enzyme had little significant effect on these values. Rapidase C80L treated juice had somewhat higher 'L' and 'a' values as enzyme level increased, resulting in a redder juice than other enzyme-treated samples. Conversely, Clarex L gave the lowest color

Fig. 6 Percent transmittance of juice treated with enzymes

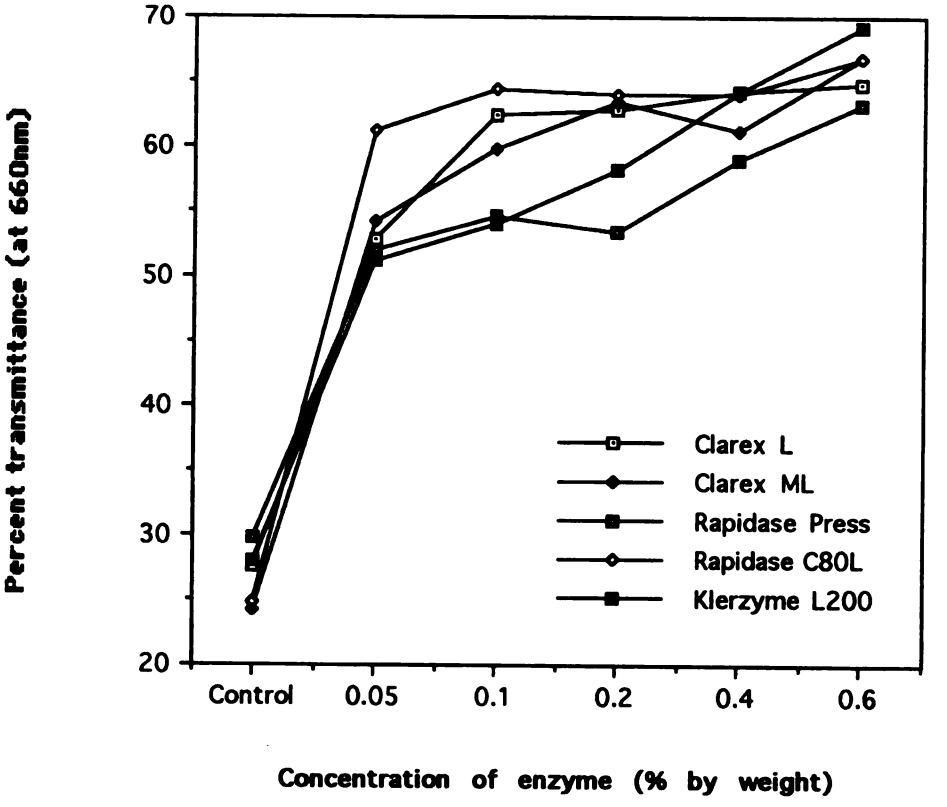


Table 4 Effects of Enzymes on Juice Clarity from Stanley Plum

Enzymes	Levels of Enzyme (% by weight)					
	Control	0.05	0.1	0.2	0.4	0.6
Clarex L	27.65a[1]	52.70b	62.50c	62.90c	64.25c	71.29c
Clarex ML	24.25a[2]	54.10b	59.80bc	63.45cd	61.30cd	66.85d
Rapidase Press	29.85a	51.95b	54.60bc	53.40bc	59.08cd	63.10d
Rapidase C80L	24.80a	61.10b	64.45b	64.00b	64.10b	66.80b
Klerzyme L200	28.05a	51.10b	53.95bc	58.10c	64.15d	69.15d

1 Unit is % transmittance
2 Comparisons are between enzyme concentrations. Values with the same letter in horizontal rows are not significantly different at 5% level of significance. This table does not show comparison between enzymes.

Table 5 Effects of Enzymes on CDM Value of Plum Juice

CDM	Levels of Enzyme (% by weight)					
	Control	0.05	0.1	0.2	0.4	0.6
Clarex L						
L	7.15a[2]	3.25b	2.62c	2.59d	2.57d	2.34e
a	21.78a	9.71b	7.33c	7.13d	6.73e	6.15f
b	6.11a	2.24b	1.70b	1.70b	1.68b	1.54b
Color Hue[1]	1.30a	1.34b	1.34b	1.34b	1.33b	1.33b
Clarex ML						
L	7.16a	2.68b	2.60c	2.18d	1.85e	2.44f
a	21.78a	7.73b	8.00c	6.09d	4.80e	6.85f
b	6.12a	1.76b	1.82b	1.30b	1.02b	1.62b
Color Hue[1]	1.30a	1.35b	1.35b	1.36b	1.36b	1.34b
Rapidase Press						
L	6.12a	2.85b	2.88c	2.82d	2.57e	2.57e
a	19.83a	6.82b	8.90c	9.20d	7.49e	7.96f
b	5.42a	1.50b	1.96b	2.09b	1.64b	1.79b
Color Hue[1]	1.30a	1.35b	1.35b	1.35b	1.36b	1.35b
Rapidase C80L						
L	6.33a	2.98b	2.92c	2.87d	3.36e	2.89d
a	19.08a	8.39b	8.25c	7.74d	10.30e	8.30f
b	5.51a	1.84b	1.83b	1.90b	2.29b	1.70b
Color Hue[1]	1.29a	1.36b	1.35b	1.33b	1.35b	1.36b
Klerzyme L200						
L	6.13a	2.84b	2.76c	2.74c	2.66d	2.55e
a	19.83a	9.06b	8.25c	8.21c	8.08d	7.70e
b	5.42a	1.97b	1.77b	1.88b	1.88b	1.87b
Color Hue[1]	1.30a	1.36b	1.36b	1.35b	1.34b	1.33b

1 calculated as the angle whose tangent equals a/b

2 Comparisons are between enzyme concentrations. Values with the same letter in horizontal rows are not significantly different at 5% level of significance. This table does not show comparison between enzymes.

values of any of the samples and the result was a darker, purple colored juice. Fig. 7 shows the relationship between juice clarity and Hunter 'L' value. In this study, Clarex L, Clarex ML, Rapidase Press, and Rapidase C80L revealed a declining linear relationship between % transmittance and Color 'L' (R^2 for Clarex L, Clarex ML, Rapidase Press, and Rapidase C80L was 0.978, 0.938, 0.927, and 0.974, respectively). This is probably due to release of ACYs with enzyme added because the hunter L value significantly ($r=0.342$, $p<0.05$) related to the amount of ACYs in the samples from this study. The correlation shows that the higher the level of ACYs, the darker the juice.

Little (1975) defined the hunter hue angle as $\tan^{-1}$ a/b. A negative $\tan^{-1}$ a/b indicates greenness and a positive $\tan^{-1}$ a/b shows redness in samples. Ough et al. (1975) found that enzyme treated red grape juice was darker than the untreated control, and was more red as judged by hunter hue. In this study, hue angle of enzyme treated juice was not influenced by enzyme concentration (Fig.8) but hue angles of control juice were smaller than that of enzyme-treated. This indicated that juices became more blue-green (but still red because angle was positive) than untreated juice. The hue angle was significantly negatively correlated to the hunter L value of Stanley juice through an increase of hue angle and a decrease of 'L' value. All these results indicate that Stanley juice became darker and more purple when enzymes were added.

Fig. 7 Relationship between juice clarity and hunter 'L' value for Clarex L treated juice

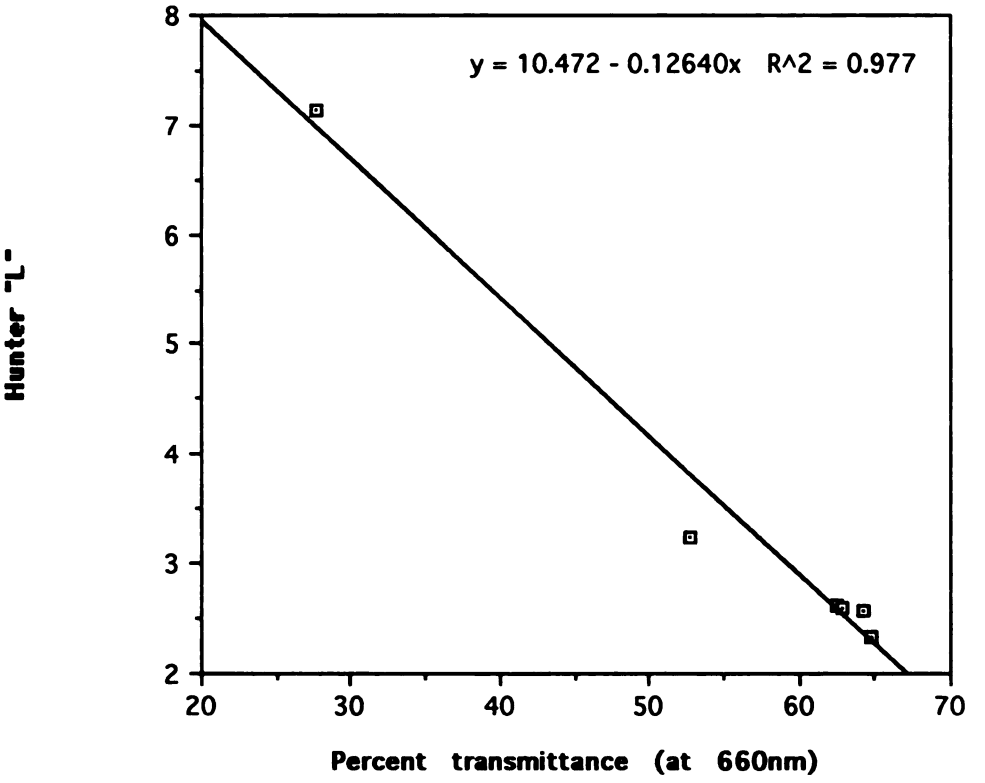
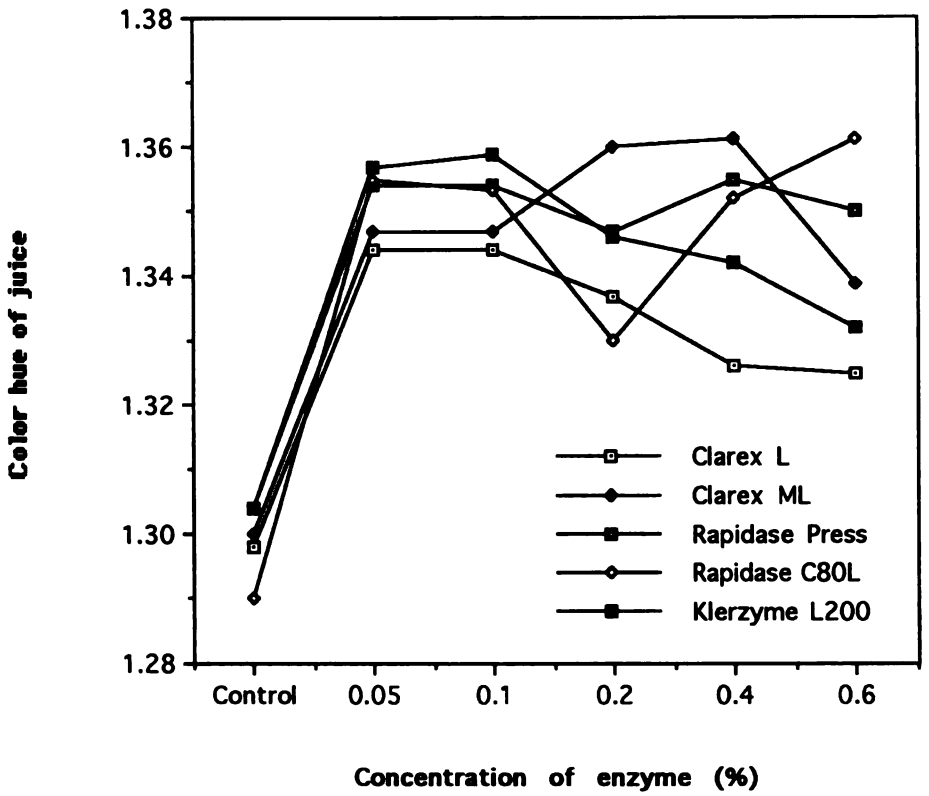


Fig. 8 Effects on color hue of plum juice



Soluble Solids (Brix)

While the average soluble solids content of the enzyme-treated plum juice ranged from 14.5 to 16.7º Brix, untreated Stanley plum juice had soluble solids content of 14.4º Brix (Fig. 9). Clarex ML, Rapidase C80L and Klerzyme L200 did not show significant effect on Brix value of plum juice but Brix values (Table 6) increased significantly as Clarex L and Rapidase Press were added. The greater degree of tissue breakdown released more components which contribute to soluble solids, so higher Brix levels were obtained in these enzyme treated juices. This has also been shown in apple, pears, apricots and carrots (Pilnik et al., 1975 ; Mclellen et al., 1985).

% Titratable Acid as Malic Acid (TA)

Enzyme-treated juice had higher titratable acid than untreated control (Fig. 10). This may be due to enzymatic de-esterification and degradation of pectin resulting in increase of total acid. Enzyme concentration significantly ($p<0.05$) influenced titratable acid, and as enzyme concentrations increased so did TA values for Rapidase C80L-treated and Clarex ML-treated juice, as compared to other enzymes. An increase in malic acid has been reported during enzyme liquefaction of Guava (Aurora et al., 1990).

Fig. 9 Effect of enzymes on Brix of plum juice

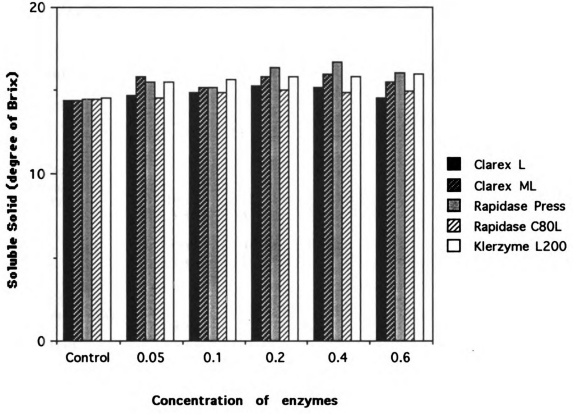


Table 6 Effects of Enzymes on Brix, pH, Titratable Acid and Brix/ Acid of Plum Juice

	Levels of Enzyme (% by weight)					
	Control	0.05	0.1	0.2	0.4	0.6
Clarex L						
Soluble Solid(°Brix)	14.40a[1]	14.73a	14.87a	15.28a	15.17a	14.55a
pH	3.35a	3.28ab	3.25b	3.25b	3.26b	3.21b
Titratable Acid						
(% as Malic Acid)	1.10a	1.23b	1.26bc	1.33c	1.30bc	1.34c
SS/Acid	13.09a	11.99ab	11.76ab	11.51ab	11.66ab	10.90b
Clarex ML						
Soluble Solid(°Brix)	14.42a	15.82b	15.20b	15.85b	16.02b	15.51b
pH	3.31a	3.23ab	3.17bc	3.14c	3.12c	3.12c
Titratable Acid						
(% as Malic Acid)	1.11a	1.26b	1.29bc	1.37c	1.34c	1.35c
SS/Acid	12.99a	12.59a	11.78a	11.54a	11.98a	11.45a
Rapidase Press						
Soluble Solid						
(degree of brix)	14.50a	15.55abc	15.21ab	16.40bc	16.73c	16.11bc
pH	3.35a	3.28ab	3.26b	3.24b	3.27b	3.25b
Titratable Acid						
(% as Malic Acid)	1.09a	1.19b	1.21bc	1.29c	1.23bc	1.25bc
SS/Acid	13.30a	13.09a	12.55a	12.74a	13.63a	12.87a
Rapidase C80L						
Soluble Solid(°Brix)	14.50a	14.55a	14.88a	15.08a	14.92a	14.93a
pH	3.35a	3.23b	3.25b	3.22b	3.20b	3.20b
Titratable Acid						
(% as Malic Acid)	1.08a	1.28b	1.31bc	1.35bc	1.36bc	1.37c
SS/Acid	13.43a	11.41b	11.37b	11.14b	10.97b	10.92b
Klerzyme L200						
Soluble Solid(°Brix)	14.52a	15.55a	15.70a	15.81a	15.82a	15.97a
pH	3.33ab	3.39a	3.37ab	3.34ab	3.32ab	3.30b
Titratable Acid						
(% as Malic Acid)	1.09a	1.13a	1.13a	1.22b	1.24bc	1.31c
SS/Acid	13.32a	13.75a	13.92a	12.96ab	12.78a	12.19b

1 Comparisons are between enzyme concentrations. Values with the same letter in horizontal rows are not significantly different at 5% level of significance. This table does not show comparison between enzymes.

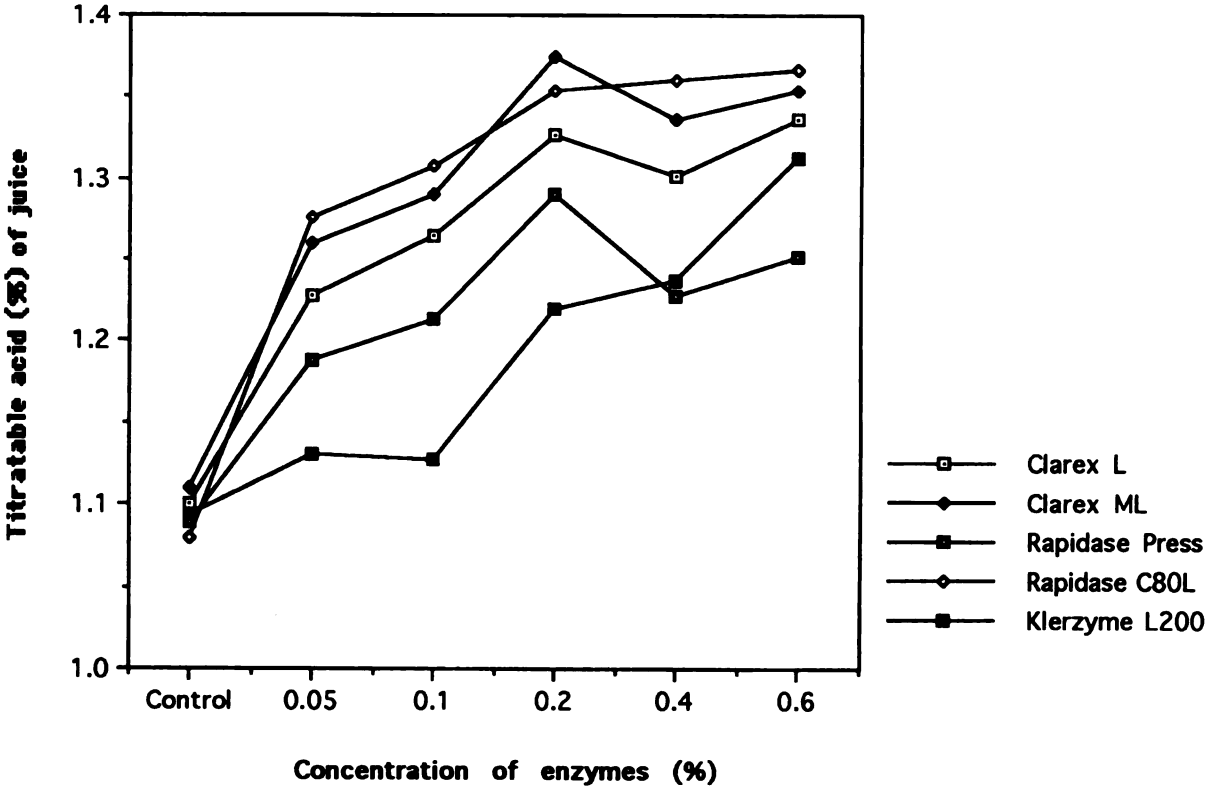


Fig. 10 Effect of enzymes on titratable acid of plum juice

pH

The pH of plum juice ranged from 3.12 to 3.35 and pH was lowered by addition of enzyme. The higher the concentration of enzyme the lower the pH values (Table 6). The pH of juice made by adding 0.1% Clarex L, Clarex ML and Rapidase Press was significantly different than the control juice. Rapidase C80L lowered pH significantly. The Klerzyme L200 did not effect pH at the lower concentration but did show an effect at the 0.6% level.

Brix/Acid Ratio

The Brix/acid ratio is the major analytical measurement for quality in citrus and several other juices. The higher the ratio the better the flavor of the juice (Fellers, 1991 & 1988). With plum juice the Brix/acid ratio was significantly influenced ($p<0.01$) by addition of Clarex L, Rapidase C80L and Klerzyme L200 (Table 6). The higher the concentration of enzymes the lower Brix/acid ratio (Fig. 11). Unlike these three enzymes, the use of Clarex ML and Rapidase Press showed no significant effect on the ratio.

Sugar Concentration

In this research, glucose, fructose, sucrose and sorbitol were analyzed by combination of HPLC and YSI analyzer. The effect of added enzymes on sugar content was apparent. Glucose, sorbitol and fructose increased significantly as the enzyme concentration (below 0.4%) was raised (Table 7). The

Fig. 11 Effect of enzyme concentration on Brix/ acid ratio of plum juice

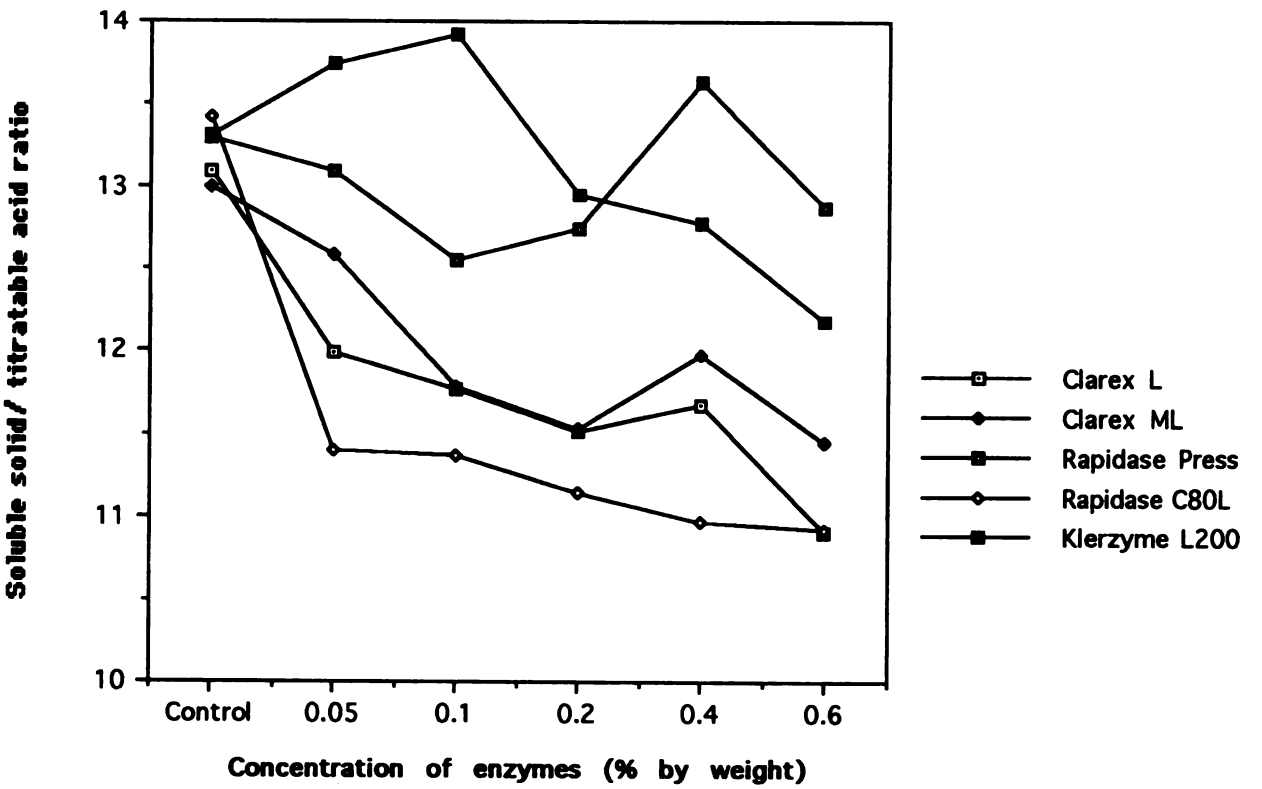


Table 7 Effects of Enzymes on Sugar Contents of Plum Juice

Sugar Content (g/100ml juice)	Levels of Enzyme (% by weight)					
	Control	0.05	0.1	0.2	0.4	0.6
Clarex L						
Glucose	3.17a[1]	3.75c	4.12d	5.32e	4.14d	3.57b
Fructose	1.52a	1.90b	1.93b	3.06d	2.22c	1.74ab
Sorbitol	0.64a	0.81a	0.71a	1.55b	1.63b	0.72a
Sucrose	3.22a	3.83a	3.78a	5.27c	5.26c	3.24b
Clarex ML						
Glucose	3.17a	4.61b	4.16c	5.22d	4.33e	4.63b
Fructose	1.52a	2.68	2.15b	3.00d	2.71c	2.82cd
Sorbitol	0.64a	1.49bc	0.95ab	1.69c	1.57c	1.30bc
Sucrose	3.22a	4.97b	4.35c	6.20d	3.33e	3.58e
Rapidase Press						
Glucose	3.64a	3.76a	4.67b	4.88c	4.96c	5.26d
Fructose	1.87a	1.96a	2.53b	2.65b	3.04c	3.14c
Sorbitol	0.50a	1.06ab	1.05ab	1.15b	1.36b	1.24b
Sucrose	3.87a	4.05b	3.12a	3.09a	2.95a	2.16c
Rapidase C80L						
Glucose	3.23a	3.93b	4.63c	4.79d	6.06e	5.15f
Fructose	1.50a	2.21b	2.60c	2.61c	3.60d	3.16e
Sorbitol	0.36a	0.86ab	1.05b	0.94ab	1.32b	1.18b
Sucrose	3.15a	3.07ab	3.14a	2.57b	1.81c	1.13d
Klerzyme L200						
Glucose	3.64a	4.02b	5.01c	5.51d	4.65e	5.03c
Fructose	1.87a	1.89ab	2.23cd	2.16bc	2.47de	2.59e
Sorbitol	0.50a	0.50a	0.52a	1.20b	0.61ab	0.46a
Sucrose	3.87ab	3.55c	5.06d	4.88d	3.32bc	2.67a

1 Comparisons are between enzyme concentrations. Values with the same letter in horizontal rows are not significantly different at 5% level of significance. This table does not show comparison between enzymes.

greatest concentrations of simple sugars were provided by addition of Rapidase C80L. The concentration of glucose, fructose and sorbitol ranged from 3.23 to 6.06, 1.5 to 3.6, and 0.36 to 1.32 g/100ml plum juice, respectively.

Robertson et al. (1991) reported Au-Rubrum plums contained glucose, fructose and sorbitol but had no sucrose. However, Vangdal (1982), in an investigation of the sugar contents of 11 plum cultivars, revealed that the major sugar in ripe plums was sucrose which accounted for 65% of the total sugar. Wrolstad and Shallenberger (1981) showed that not only varietal differences can change the amount of sucrose but also processing can make sucrose disappear. All these difference are most likely due to invertases in the juice (Wrolstad et al., 1981 and Gorsel et al., 1992). The content of sucrose in Stanley plum juice went up with addition of enzymes but after a period of time the sucrose level dropped (Table 7). This is most likely due to enhanced invertase activity during processing resulting in sucrose hydrolysis. Levels of invertase activity in Stanley plums during ripening, processing and storage is worthy of investigation because of the changes in the physical and chemical properties brought about by sucrose hydrolysis.

Total Anthocyanins (TACYs)

The ACYs located within the flesh and skin of plums are responsible for desirable purple color of Stanley plums. Pectinase enzymes have been reported to aid in release of

pigments from plant cell (Reed, 1975). In this study the effect of various commercial enzyme on TACY release is apparent (Fig. 12). The increase in TACY content is about two-fold by addition of Klerzyme L200, which was in contrast to Rapidase C80L which had little effect on ACY release. Rommel et al. (1992) indicated preferential release of ACYs from blackberries into the liquid phase by pectinases. Arnold (1992) reported TACY content of 0.18 absorbance unit by treating Stanley plums with 0.25 pectinase as compared to the 0.165 units from control juice. In this study, enzyme treatment gave better release of ACYs (6% increase) from Stanley plum then Arnold's results (1.5%) in 1992. The mean ACY content of enzyme treated juice is 0.26 units in Stanley plum juice as compared to the 0.2 units of control juice.

Browning results are shown in Fig. 13. The correlation between ACYs and browning index is detected significantly ($r=0.368$, $p<0.05$) through all data. The higher browning level in the juice the higher degradation of the ACYs (Fig. 14).

Total Phenolics

Total phenolics as tannic acid in plum juice were determined colorimetrically in this study. Pectic enzymes extract not only the pigments but also other phenolic compounds, such as tannic acid which gives a bitter flavor in the fruit juice. In this study, the total phenolics in enzyme treated plum juice ranged from 88 to 142 mg/100ml

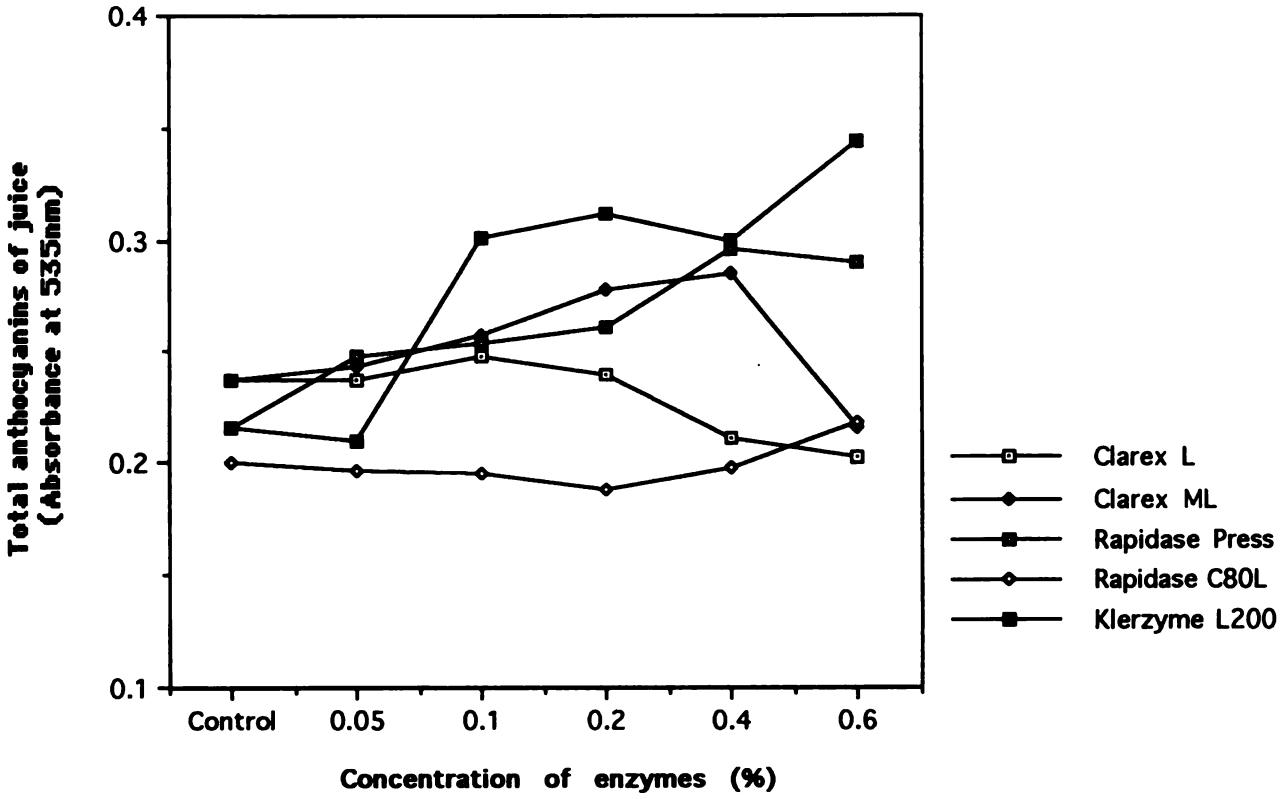


Fig. 12 Total anthocyanins of juice treated with enzymes

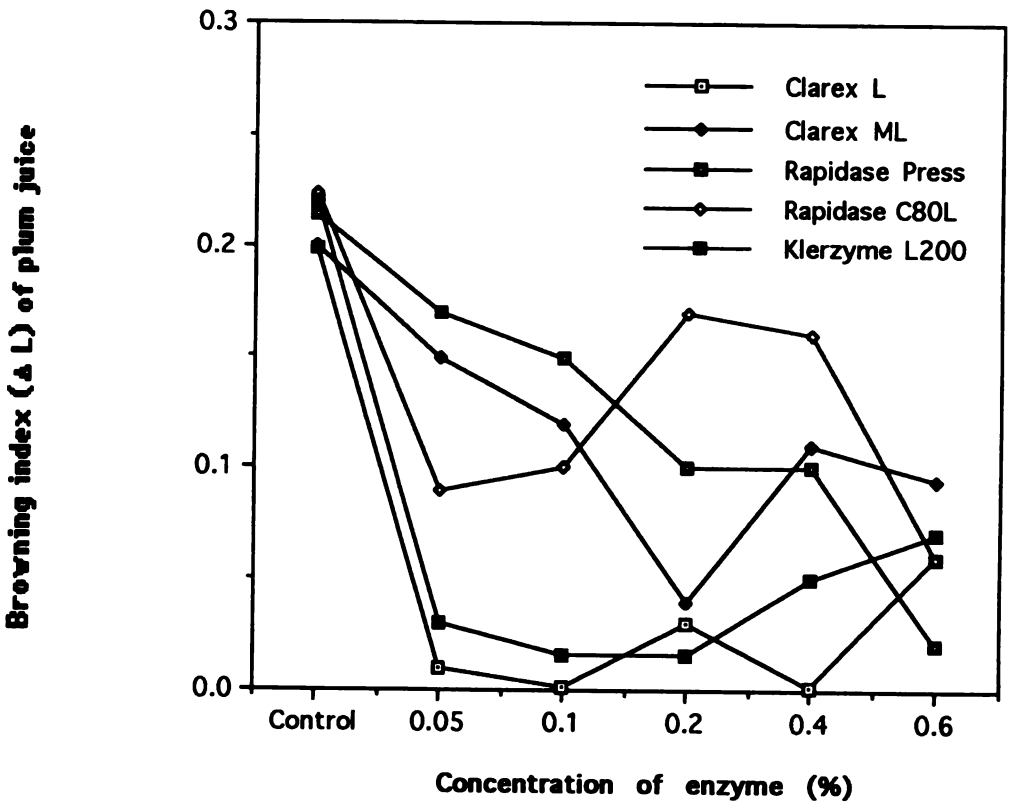
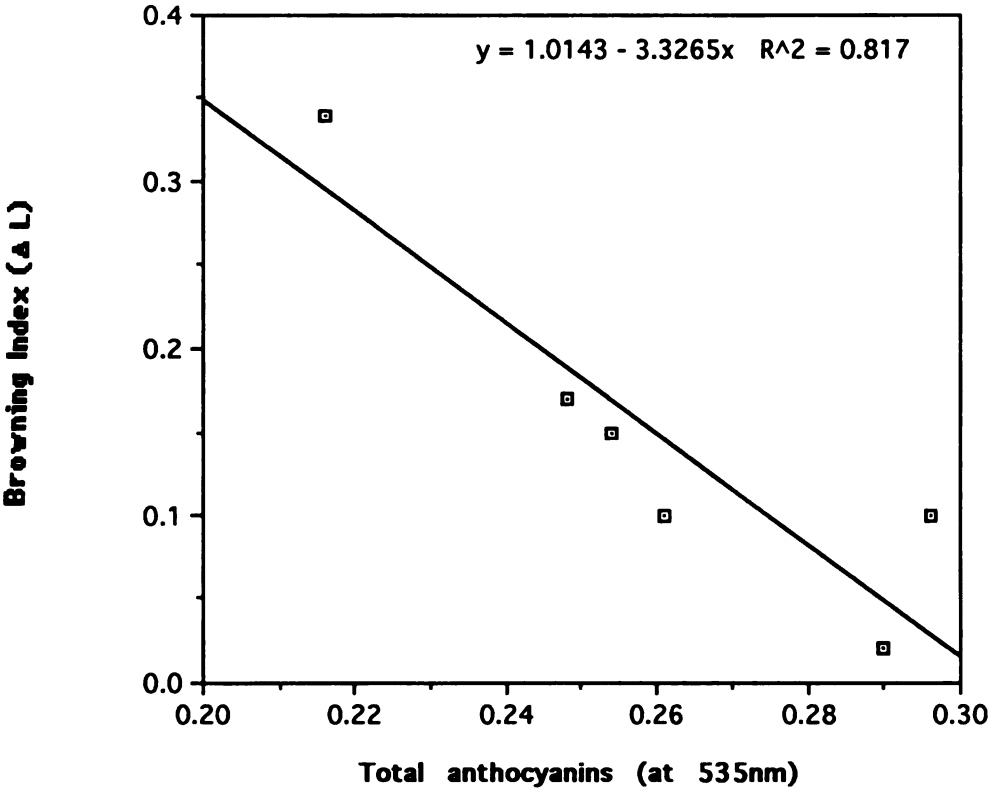


Fig. 13 Effect of enzyme concentration on browning of plum juice

Fig. 14 Relationship between browning and anthocyanins for Rapidase Press treated juice



(Fig. 15) as compared to a total phenolic content in the control of 70 mg/100ml. Brown (1981) and Ough (1979) indicated that pectinases influence on the amounts of total phenolics released in grape juice was related to grape varieties, enzyme types and concentration of enzymes. George et al. (1990) reported on the comparison of total phenolics determined by HPLC and colorimetrically. They found that colorimetric analysis resulted in a 10-fold higher level of total phenolics than the HPLC determinations. Gorsel et al.(1992) measured the phenolics in plum using HPLC. There was an 8 to 12 fold difference between their results (11 mg/100ml plum juice) and the data from this study. Interference of intermediates and final browning products may explain the discrepancy in the total colorimetric quantitation (Van Buren et al., 1976; Spanos et al., 1990a and 1990b). In addition, HPLC is a specific method for quantitation of individual phenolic compounds while the colorimetric procedure is a general assessment of the levels of phenolics. This may explain some of the variation in total phenolics obtained in this study.

Summary

Among the five enzymes investigated Clarex L at 0.2% concentration produced the best overall plum juice. It gave optimum juice yield (compared to Klerzyme 200L), a more stable color (compared to Clarex ML and Rapidase Press), higher ACY content, better flavor, lower phenolics (compared

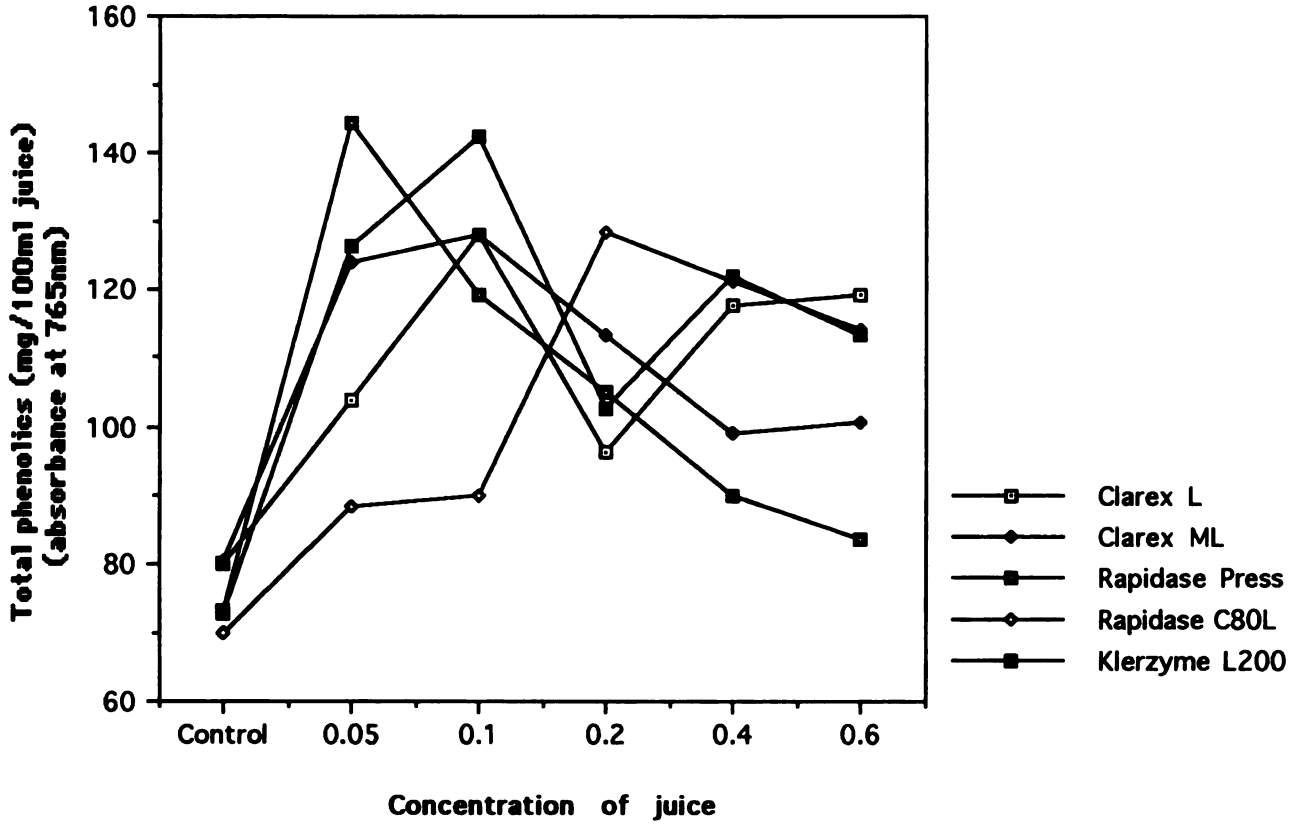


Fig. 15 Effect of enzyme on total phenolics of plum juice

to Rapidase C80L) and a sediment free, clear juice (compared to Rapidase Press). Enzyme concentrations above 0.2% resulted in bitter flavor which were not acceptable though they gave very high yields.

Physico-Chemical and Sensory Characteristics of Plum Juice from Selected Plums Grown in Michigan

Plum juice was extracted and processed from six selected Michigan plum cultivars, Au Red, Abundance, Pobeda, Shiro, Peach Plum and Early Golden. Enzyme Clarex L (0.2% w/w of plums) which produced optimum quality plum juice in phase I of this research (Selection of Enzymes) was used to extract plum juice from these cultivars. This section describes the physico-chemical and sensory quality of plum juice made from the above plum samples.

% Plum Juice Yield

As a result of Clarex L addition, a 26-54% increase in juice yield was obtained in plum samples (Table 8). Highest yield was obtained in Au Red sample (54%) followed by Peach Plum (50%) and Shiro (45%), and the yield of plum juice in these varieties was higher than Stanley plum (32%).

In a study by Wani and Saini (1990), plum juice (extracted from Santa-Rosa, Satsuma and Alubukhara plum varieties) yields of 17% and 23% were reported through the

Table 8 The effect of Clarex L on juice yield in selected six plum varieties

Varieties	Juice Yield (%) Control	Juice Yield(%) enzyme-treated	Increase (%)
Au Red	25.30a	79.48e	54.18
Abundance	55.87c	82.07f	26.2
Pobeda	59.95d	84.76g	24.81
Shiro	36.21b	81.84ef	45.63
Peach Plum	34.61b	84.70g	50.09
Early Golden	53.79c	83.63fg	29.84

Values with the same letter are not significant at 5% level of significance.

use of pectinase. Compared to this, yields of plum juice using Clarex L, are much higher. Although the increase in juice yield is cultivar specific, Clarex L enzyme system which combines the properties of pectic, hemicellulose and cellulose enzymes obviously enhances liquefaction of plums resulting in higher juice yield, which is very important from economic standpoint.

Plum Juice Clarity

Percentage transmittance (%T) of plum juice made from different plum cultivars was used as an indicator of juice clarity (Table 9). As can be seen the %T of Clarex L extracted juice samples was higher than the control in all cases, indicating that the Clarex L enzyme system was effective in the different cultivars in removing constituents which affect juice clarity. A higher %T represents less finely dispersed matter in juice. Meischak (1971) reported separation of plum juice treated with cell wall degrading enzymes into two distinct layers of partially clarified juice and settled precipitates. Combination of fining and clarification can remove the precipitated matter. Juice treated with fining agents in addition to enzymes are usually more clear than juices treated only with enzymes. Hsu et al. (1989) showed that enzyme extraction and fining reduced the concentration of total protein resulting in clearer juices.

Table 9 shows the degree of clarity (%T) of plum juice which had added enzymes, pasteurized (HTST-unfined juice),

Table 9 Turbidity, Pectin Content, Total Phenolics and Total Anthocyanins of Plum Juice Made From Selected Plum Varieties

	Varieties					
	Au Red	Abundance	Pobeda	Shiro	Peach Plum	Early Golden
Turbidity (%)						
Control	25.3a	55.87a	59.95a	36.21a	34.61a	53.80a
A	27.00b	87.55b	75.40c	87.90b	84.90d	94.20c
B	22.70a	86.85b	68.75b	92.85c	78.30b	91.93b
C	38.15d	95.50c	83.85e	96.25d	81.40c	96.00c
D	35.60c	94.70c	81.00d	95.80d	82.65c	95.80c
Pectin (g/100ml)						
Control	0.44d	0.20d	0.15d	0.26d	0.22d	0.35c
A	0.25c	0.07b	0.08b	0.11b	0.09c	0.16b
B	0.14a	0.09c	0.11c	0.14c	0.05b	0.16b
C	0.16a	0.03a	0.03a	0.04a	0.01a	0.13a
D	0.18b	0.01a	0.02a	0.03a	0.02a	0.16b
Total Phenolics (mg/100ml)						
Control	123.7a	116.0a	205.9a	26.76a	67.09a	43.82a
A	350.1d	330.0d	429.2e	156.3d	96.55d	143.1d
B	298.2c	330.7d	417.6d	150.1c	77.94b	128.3c
C	255.5b	282.7c	345.5c	126.8b	94.23d	113.6b
D	259.4b	274.9b	332.3b	122.1b	88.02c	109.7b
Total Anthocyanins (mg/100ml)						
Control	16.53a	3.00b	22.64b	0.33a	0.42a	0.40a
A	58.66d	4.30c	32.18d	0.39a	3.31b	0.93a
B	51.11c	4.05c	30.76c	0.15a	3.18b	0.87a
C	31.07b	0.91a	12.10a	0.07a	0.97a	0.28a
D	30.64b	1.06a	11.67a	0.07a	0.93a	0.29a

Values with the same letter in the column are not significant at 5% level of significance.
Juices were treated by A. Clarex L
B. HTST of unfined juices
C. Fined juices
D. HTST of fined juices
Control juice without treatment.

clarified using gelatin and bentonite (fined juice), and juice which was fined and pasteurized (HTST-fined juice). The clarity of fined juices was highest in all plum samples. This was followed by samples which were fined and pasteurized. The high temperature short time heat treatment of juice always decreased % transmittance for all varieties. Heat induced dissociation of protein-phenolic complex of fruit cell walls during pasteurization has been shown to influence clarity of fruit juices (Hsu et al, 1989).

In this study, Early Golden plum juice had the highest %T values followed by Shiro, Abundance and Peach Plum. Plum juice from Au Red had the least clarity with only 2% transmittance and 10% increase due to enzyme and processing. Clarex L, gelatin and bentonite had little influence in this variety.

The correlation between %T and pectin content is shown in Table 10. As expected, pectin content negatively correlated with %T for all cultivars. The higher the pectin content of juice, the lower was their %T, indicating that clarity of the plum juice was in direct proportion to the degree of pectin breakdown by the Clarex L enzyme system.

Soluble Solids, Titratable Acid and Brix/Acid ratio

Soluble solid (Brix), titratable acid (% malic acid) and brix/acid ratio are some of the major indicators of fruit quality. Soluble solids are used as a factor in determining maturity of fruit, to establish various grades of quality,

Table 10 The correlation coefficients between % transmittance and pectin content of plum juice made from selected plum varieties

Correlation of % T & Pectin	Au Red	Abundance	Pobeda	Shiro	Peach Plum	Early Golden
A	1.00**	0.992**	0.99**	0.994**	0.998**	0.995**
B	0.44	0.97**	0.98**	0.92**	0.88**	0.97**

* significance at 5% level; ** significance at 1% level

%T: percent transmittance

A : only Clarex L treated juice

B : Clarex L, fined and pasteurized juice.

and as a pricing index under some conditions. The brix/acid ratio is also used as an index of maturity of fruit (McAllister, 1980). In the fruit and juice industry, brix/acid ratio indicates the relative tartness or sweetness of juice.

In this study, the brix values of juice from six plum cultivars analyzed ranged from 9.85 to 18.45 (Table 11). Au Red had the highest soluble solid content (18.45 ºBrix) followed by Early Golden (12.98 ºBrix) and Peach Plum (12.70º Brix). Abundance had the lowest brix reading (9.85º Brix). The recent analysis of 160 plum cultivars (Gur, 1986) in U.S. showed soluble solids of this fairly large sampling of plums ranged from 7 to 24º brix.

Addition of Clarex L for plum juice extraction increased the soluble solid of all plums by 2.20% to 20.80% (Fig. 16). Commercial pectinase has been shown to release about 80% polysaccharides from apple cell wall, in addition to degrading the pectic material, thus increasing the soluble solids content (Pilnik and Voragen, 1991).

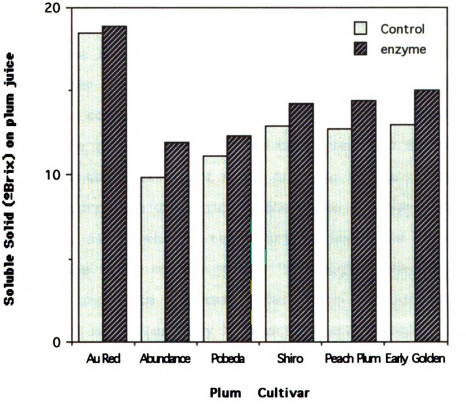
Plum juice has been characterized as having a predominance of malic acid (Meredith, 1992). The % acidity of plums used for juice extraction in this study ranged from 1.10 (Shiro variety) to 1.83 (Pobeda variety). There were no significant effects on acidity from processing. Addition of enzyme for juice extraction increased the acidity of plums by an average of about 24%. These data are in agreement with the work of Jenniskens et al.(1990) who reported 24.2% acid

Table 11 Soluble Solids, % Malic Acid, pH and Brix/Acid Ratio of Plum Juice Made from Selected Plums

	Varieties					
	Au Red	Abundance	Pobeda	Shiro	Peach Plum	Early Golden
Brix						
Control	18.45a	9.85a	11.12a	12.86a	12.7a	12.98a
A	18.85b	11.90c	12.31c	14.28b	14.40b	15.03b
B	18.95b	11.77bc	11.83b	14.15b	14.38b	14.98b
C	18.38a	11.60bc	12.05bc	14.03b	14.08b	14.84b
D	18.50a	11.53b	12.22c	14.20b	14.15b	14.80b
Malic Acid (%)						
Control	1.22a	1.22a	1.83a	1.10a	1.13b	1.52a
A	1.33a	1.59b	2.18b	1.55b	1.17a	2.00b
B	1.37a	1.59b	2.17b	1.57b	1.19ab	1.96b
C	1.33a	1.59b	2.18b	1.55b	1.17a	2.00b
D	1.25a	1.50b	1.96ab	1.44b	1.11a	1.66a
Brix/ Acid Ratio						
Control	15.18a	8.09b	6.09a	11.67b	11.23a	8.53a
A	14.16a	7.49ab	5.66a	9.24a	12.31b	7.51a
B	13.85a	7.44ab	5.45a	9.01a	12.08b	7.63a
C	13.81a	7.30ab	5.54a	9.09a	12.01b	7.42a
D	14.86a	7.68a	6.24a	9.84ab	12.77b	8.92a
pH						
Control	3.37a	3.19a	3.01a	3.24ab	3.37a	3.17a
A	3.37a	3.23a	3.07b	3.23ab	3.39ab	3.28b
B	3.45b	3.22a	3.13c	3.29b	3.41ab	3.30b
C	3.40ab	3.21a	3.11bc	3.23a	3.45b	3.32b
D	3.40ab	3.20a	3.09bc	3.23ab	3.53b	3.27b

Values with the same letter in the column are not significant at 5% level of significance.
Juices were treated by A. Clarex L
B. HTST of unfined juices
C. Fined juices
D. HTST of fined juices
Control = juice without treatment.

Fig. 16 Effect of Clarex L on Soluble Solids Contents of Plum Juice



increase in apple juice, and Rommel, et al. (1992) who found a 22.9% acid increase in blackberry juice. These results are likely due to enzymatic de-esterification and degradation of pectin resulting in increase of total acid (Voragen et al. 1985).

The brix/acid ratios of plum juice samples in this study were not significantly different among enzyme-added, fining agent-added and pasteurized juice (Table 11). Among the plum varieties, Au Red had the highest ratio (15.18) while Pobeda plums gave the lowest values (6.09). This tends to indicate that Pobeda may not be suitable for fresh use in spite of its desirable red color, but it can be processed into acceptable quality juice (or juice drink) by modifying its brix/acid ratio through the addition of sugar or sugar syrup. The same situation occurred in grapefruit juice where a large consumer study (USDA, 1958) showed that tart on sour are directly related to the ratio of the juice. The higher the tartness factors the lower the consumers preference. Adjustment of the Brix/acid ratio (usually by the addition of other fruit juices) was the key to improving this juice.

Barros et al.(1984) found brix/acid ratio to be significantly correlated ($p<0.01$) with flavor in grapes ($n=1039$). Fellers et al. (1988) reported that lower brix/acid ratios of 7.0 in grape juice they tested had the lower score in consumer preference than juice with higher Brix/acid ratios in the range of 11.1.

Color

A bright natural color in food products is essential to the enjoyment of eating. The importance of color is a psychological, as well as a physical fact that has been well established. The Hunter L, a, b and color hue angle are measurements which are for estimating visual color. Determination of color hue angle ($\tan^{-1}$ a/b) was proposed by Little (1975). The function of a/b was described as a hand sweeping counterclockwise on a dial, starting at $0\pi r$ (red), to $\pi r/2$ (yellow), to πr (green), to $3\pi r/2$ (blue) and at $2\pi r$ back to red. The concept follows conventional trigonometric nomenclature, with the yellow-red quadrant (+a, +b) as positive and the yellow-green quadrant (-a, +b) as negative. The measurement of hunter 'L', 'a', 'b' and color hue angle for all plum juice samples are shown in Table 12.

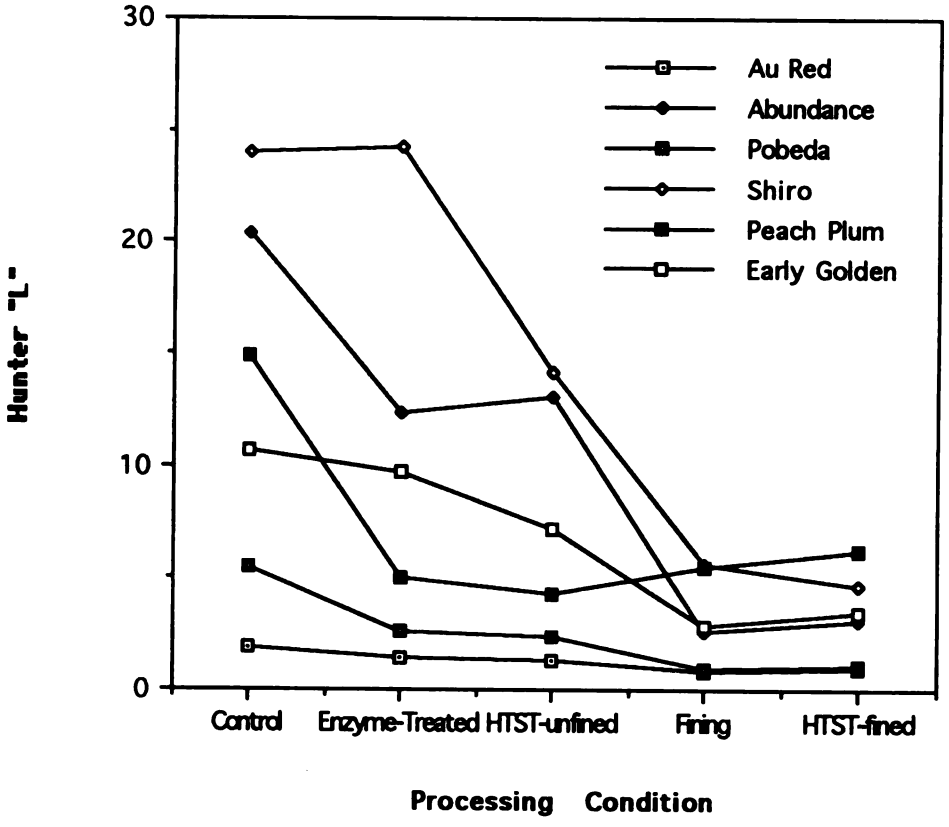
As expected, the L value varied with varieties and processing (Fig. 17). The dark red colored varieties, Au Red and Pobeda, produced juice with lower L values while the yellow Shiro variety gave the lightest juice. Enzyme treatment decreased hunter L value (30%) of juices because more pigment was released from the cells (Pilnik and Voragen, 1991). Shiro color wasn't significantly effected by enzyme due to lack of ACY pigments. However, Shiro was effected by heating, which caused browning in the juice. Browning was also indicated by a decrease in the L value in the unfined juice from Shiro which was heated by HTST. Nonenzymatic browning due to heating may not have been as extensive as

Table 12 Colors (CDM Values) of juice made from selected plums

	Varieties					
	Au Red	Abundance	Pobeda	Shiro	Peach Plum	Early Golden
Hunter L						
Control	1.93a	20.44a	5.51a	23.94a	14.89a	10.72a
A	1.46b	12.40b	2.69b	24.26b	5.02b	9.68b
B	1.28c	13.09c	2.40c	14.16c	4.30c	7.26c
C	0.80d	2.66d	0.99d	5.64d	5.48d	2.83d
D	0.99e	3.08e	1.04e	4.65e	6.27e	3.45e
Hunter 'a'						
Control	3.46a	28.55a	20.28a	-1.04a	3.11a	2.79a
A	2.46b	23.45b	8.36b	-3.44b	9.43b	6.67b
B	1.81c	23.94c	4.74c	-1.36c	7.63c	5.56c
C	0.42d	1.69d	1.45d	-1.04d	3.22d	0.54d
D	0.34e	1.97e	1.71e	-0.18e	3.63e	0.70e
Hunter 'b'						
Control	0.52a	22.43a	5.56a	13.72a	12.53a	3.76a
A	0.35b	10.91b	1.97b	7.90b	2.68b	2.10b
B	0.45c	12.00c	1.08c	2.23c	2.02c	1.78c
C	0.00d	0.62d	0.28d	5.64d	1.57d	0.85d
D	0.02e	0.81e	0.21e	0.68e	6.27e	0.76e
Color Hue Angle						
Control	1.43a	0.91a	1.30a	-0.08a	0.24a	0.64a
A	1.43a	1.14a	1.34a	-0.41a	1.29a	1.27a
B	1.33a	1.11a	1.35a	-0.55a	1.31a	1.26a
C	0.00a	1.22a	1.38a	-0.18a	1.12a	0.57a
D	1.51a	1.18a	1.45a	-0.29a	0.53a	0.74a

Values with the same letter are not significant at 5% level of significance.
Juices were treated by A. 0. 2% Clarex L
B. HTST on unfined juices
C. fined the juices
D. HTST on fined juices
Control means the juice without treatment.

Fig. 17 Changes on Hunter L of Plum Juice



browning from PPO enzyme which was inactivated by heating (Sapers, 1992; Siddiq, 1993). The Hunter L, a and b value was decreased by fining agent. The color value changed less by HTST-fined because fining agent removed the browning factors in the juice. Sapers (1992) reported that the capacity of raw apple, grape and pear juices to undergo browning was associated with particulate fractions that could be removed by filtration with bentonite and diatomaceous earth.

Plum juices from Abundance and Pobeda had higher '+a' values, indicating more redness while Shiro gave '-a' values (Fig. 18). The higher the Hunter 'a', the redder the juice whereas the lower 'a' value, the more green the juice. Juice from Shiro was located in the yellow-green quadrant so it had a slight green tint, although this was not so readily visible to the eye. Abundance juice had the highest 'b' value in control juice while the Hunter 'b' value of Au Red and Early Golden were only 0.52 & 3.76 respectively (Fig. 19). The more blue the juice the lower the 'b' value. The Au Red and Early Golden gave the lowest 'b' value while the Abundance, Shiro and Peach Plum showed the higher 'b' value. The juice was more blue and less yellow when enzyme and fining agents were added to the plum juices. The analysis of variance (ANOVA) of color hue angle indicated that there were no significant differences ($p>0.05$) between juices from the different varieties or from processing. Figure 20 shows the change in hue angle for each of the plum varieties. All

Fig. 18 Change of Hunter 'a' Value in Plum Juice

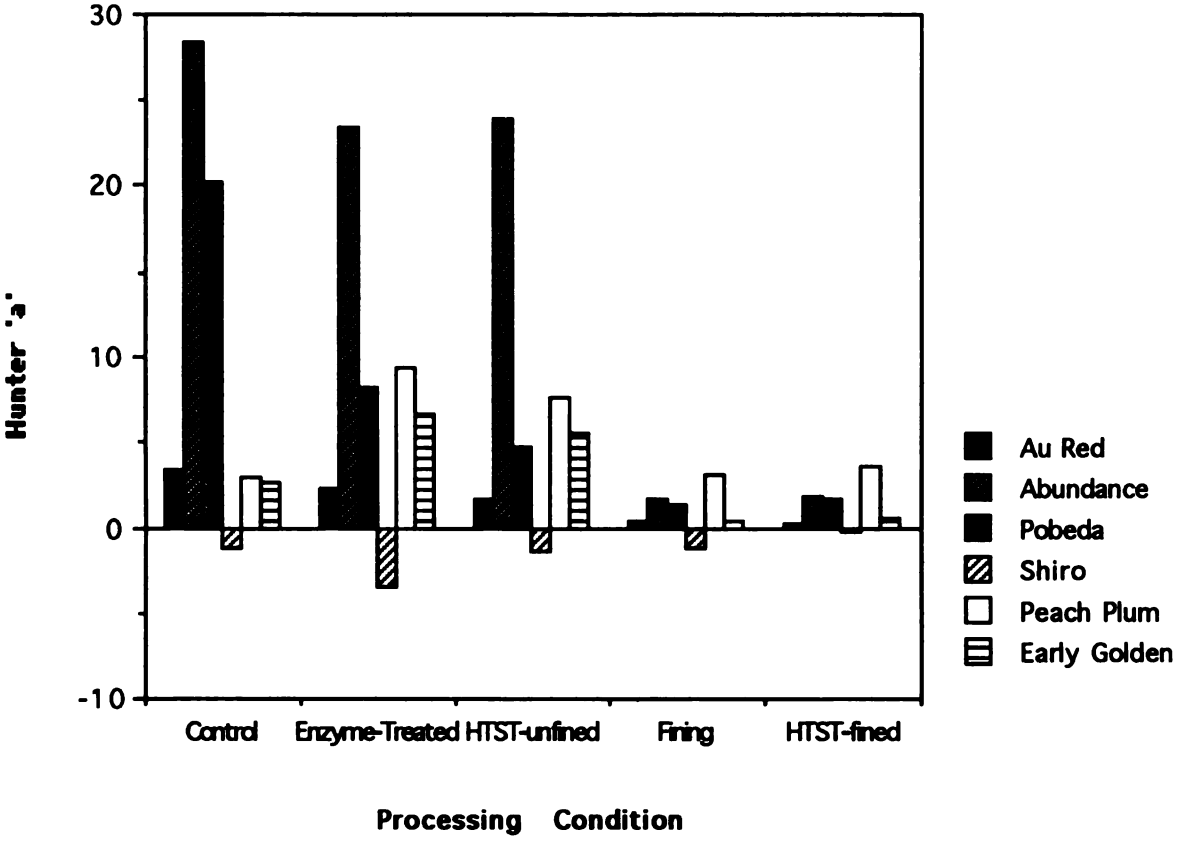


Fig. 19 Effect on Hunter 'b' of juice made from selected plums

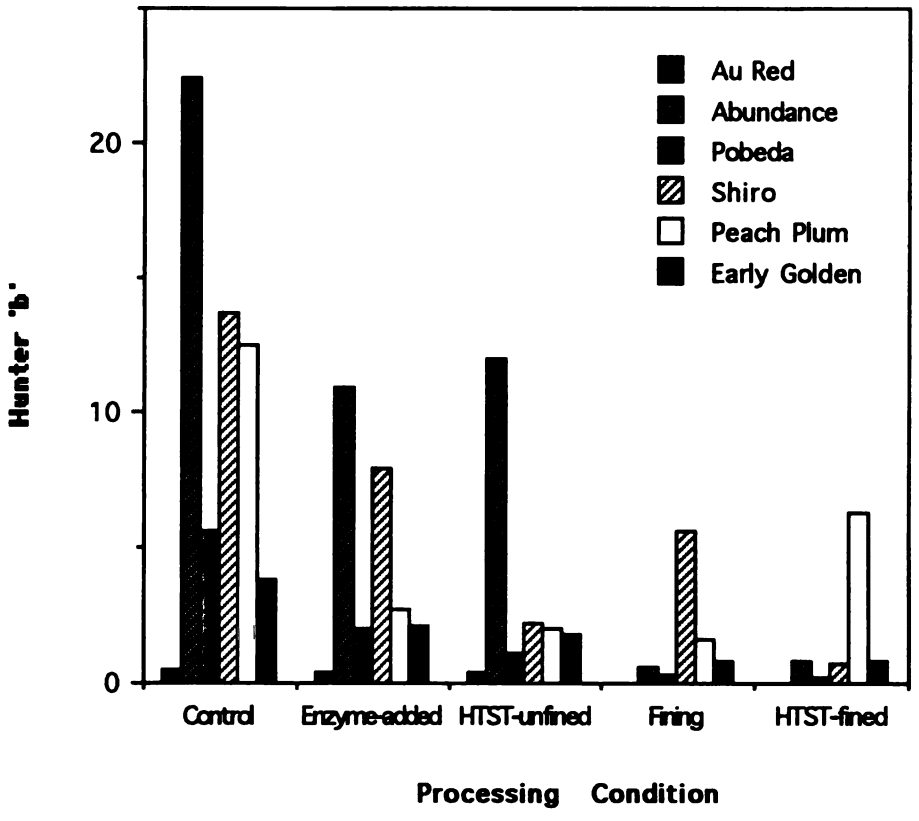
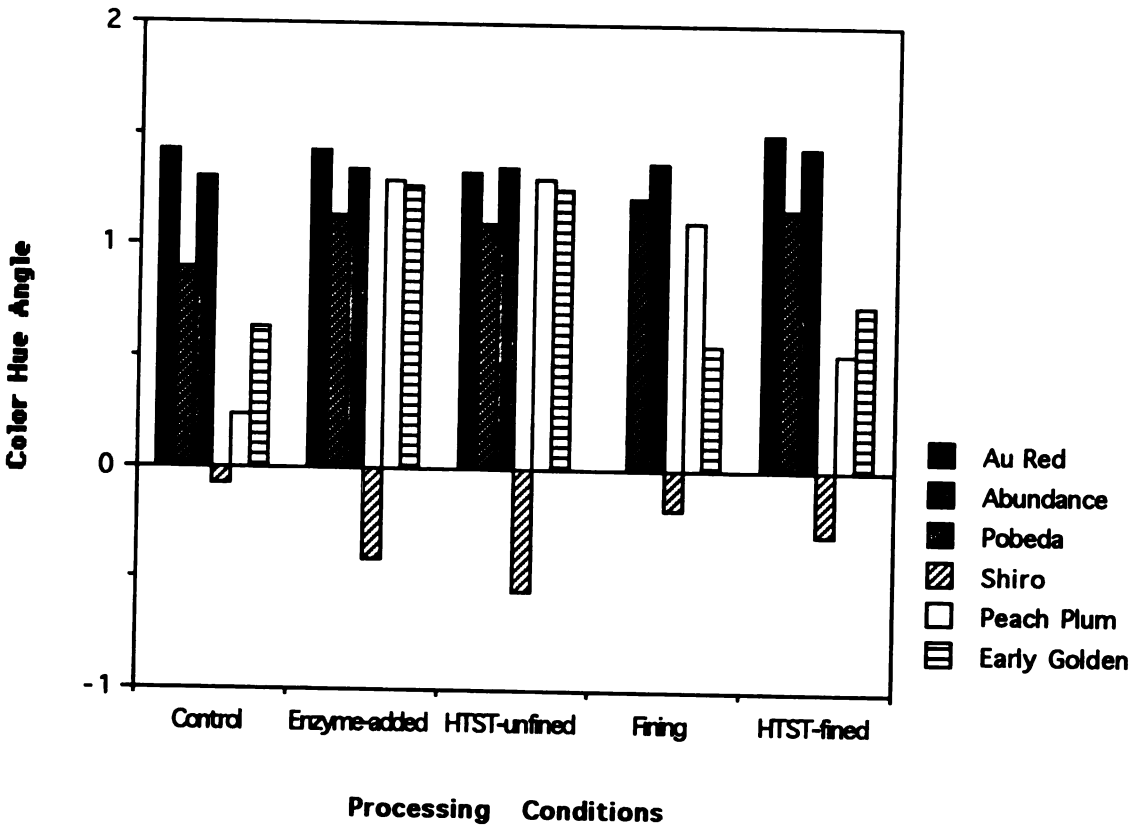


Fig. 20 Change in Hue Angle of Juice Made from Selected Cultivars



color value for Au Red are lower than expected. These results are probably due to the lower transmittance (lower clarity) of light in Au Red juice.

Sugars

Plum fruit contains both non reducing (sucrose) and reducing (fructose and glucose) sugars and sorbitol, a sugar alcohol. Plums contain moderate amounts of sucrose and the presence of large quantities of sucrose in plum juice has been used as an indicator of adulteration (Flynn, 1970) which is similar to the situation with apple juice (Fitelson, 1970).

In this study, juice from Au Red had the most glucose, fructose and sorbitol of any cultivar but was lowest in sucrose content (Table 13). Plum juice from Abundance was lowest in glucose and sucrose, while juice from Early Golden plum had little sorbitol. The variety, geographic, seasonal, maturity, postharvest condition and processing can all affect sugar compositions. The sugar content of plums have been reported by Richmond et al. (1981), Gur (1986), Wrolstad et al. (1981), and Robertson et al. (1991). The range of fructose was from 0.72 to 4.93 g/100g plums, glucose ranged from 1.26 to 5.22 g/100g fresh plums, sucrose content was from 0.02 to 5.68 g/100g fresh fruit and sorbitol content was from 0 to 2.7 g/100g plums.

All varieties but Abundance have glucose to fructose ratios (g/f ratio) over 1. Usually, pear and apple contain

Table 13 Sugars Content of Juices Made from Selected Plums

	Varieties					
	Au Red	Abundance	Pobeda	Shiro	Peach Plum	Early Golden
Glucose						
Control	6.03a	1.84a	1.90a	2.70a	2.15a	3.18a
A	6.57b	3.17b	3.42b	5.13b	3.73b	4.05b
B	6.13c	3.31c	3.97c	5.23c	4.03d	4.22c
C	6.64d	3.27c	3.99c	5.17b	3.99c	4.42d
D	6.42e	3.31c	4.09d	5.55d	3.79b	4.56e
Fructose						
Control	5.91a	2.50a	1.21a	1.64a	1.29a	2.67a
A	5.71b	3.75b	2.73b	4.08b	2.83b	3.55b
B	5.30c	3.87c	3.18c	4.17c	3.09e	3.75c
C	5.72b	3.84c	3.18c	4.14c	3.03d	3.89d
D	5.56d	3.89c	3.31d	4.41d	2.91c	4.07e
Sucrose						
Control	0.10a	2.41c	3.73c	4.73d	7.45e	3.80b
A	0.22a	0.65ab	0.69b	1.22b	4.09d	1.99a
B	0.42b	0.57a	0.75b	1.45c	3.48a	1.99a
C	0.53b	0.77b	0.48a	0.59a	3.88c	1.86a
D	0.88c	0.57a	0.46a	0.59a	3.70b	1.82a
Sorbitol						
Control	1.61a	0.05a	0.05a	0.43a	0.04a	0.01a
A	1.53b	0.05a	0.34d	0.50b	0.14c	0.01a
B	1.58c	0.07a	0.31c	0.50b	0.13c	0.01a
C	1.39d	0.14b	0.21b	0.43a	0.12c	0.02a
D	1.37e	0.14b	0.20b	0.41a	0.11b	0.01a
Total Sugar						
Control	13.65a	6.8a	6.89a	9.5a	10.95cd	9.66a
A	14.03b	7.62b	7.18b	10.93c	10.79bc	9.60a
B	14.28c	7.83c	8.21d	11.35d	10.73b	9.97b
C	14.28d	8.02d	7.86c	10.33b	11.02d	10.19c
D	14.23d	7.91cd	8.06d	10.69c	10.51a	10.46d

Values with the same letter in the column are not significant at 5% level of significance.
Juices were treated by A. 0. 2% Clarex L
B. HTST on unfined juices
C. Fined juices
D. HTST on fined juices
Control = juice without treatment.
Unit is g/100ml juice

much more fructose than glucose, while peach and plum have more glucose than fructose (Wrolstad et al., 1981). The g/f ratio of Abundance was around 0.85 which is similar to the ratio found in pears or apples. This plum cultivar has some rather distinctive flavor characteristics which may be due in part to its sugar composition.

Addition of enzymes had some effects on sugar contents of plum. Enzyme extracted plum juice was higher in total sugar content compared to control sample due to release of soluble solids from the cell wall (Cheetham, 1985). The glucose and fructose content of juice were higher and the sucrose content was lower with addition of enzyme but invertase may be a prime factor in sucrose decrease and simple sugar increase. Probably, invertase hydrolyzed the sucrose with addition of enzyme and then it was stopped by the high temperature short time heating process. Gorsel et al. (1992) reported that fresh plum juice was always higher in sucrose. In contrast, either no sucrose or low levels were found in prunes and processed plum products (Wrolstad et al., 1981; Flynn and Windt, 1970; Gorsel et al., 1992) due to the existence of invertase that caused the reducing sugars to be slightly more dominant. In this study, the juice from Au Red was an exception because it had low concentrations of sucrose even in control juice. The sugar composition of Au Red changed very little ($p>0.05$) during processing but its total sugar was increased slightly by enzymes.

Pectins

Pectin is one of the major cellular structural components. It exists both in the primary cell wall and in the middle lamella, the intercellular cement between cells. In this capacity it contributes significantly to structural integrity of fruits and vegetable (Gur, 1986). As a soluble component of juice and an insoluble component of juice particulate material, pectin affects many facets of juice quality, such as viscosity, gelling ability, and ability to precipitate as pectates, so determination of pectin contents of plums is important.

Pectin contents of control plum juice made from Au Red, Abundance, Pobeda, Shiro, Peach Plum and Early Golden plum were 0.44, 0.20, 0.15, 0.26, 0.22 and 0.35 g galacturonic acid/100 ml fresh plum juice respectively (Table 9). Other studies on plum (Hardinge et al., 1965; Krause and Bock, 1973; Vidal-Valverde, 1982; Southgate, 1991) have reported pectin content of plum to range from 0.23 to 1.00 g/100g fresh plums using similar methods as those employed in this work.

Enzyme extracted juice had an average 54% lower pectin than control sample. This could be due to enzyme break down of the pectin into simpler compounds, such as galactose, xylose, monomers, and oilgomers of galacturonic acid (Ryu, 1980). Fining process further reduced the pectin content by almost the same degree as enzyme treatment. This is probably due to the fact that the fining process removed particles

larger than 150mμ (Fig. 2). In the enzyme extracted, fined and pasteurized plum juice, the pectin content ranged from 0.01 to 0.18 g/100 ml juice. Only Early Golden and Au Red had higher pectin (0.16-0.18 g/100 ml). Other processed juice pectin content was very low (0.1-0.3 g/100 ml).

Pectin content of plums correlated with total phenolics, brix, sugar content and color values (Table 14). Total phenolics, brix, glucose and sucrose had a negative correlation with pectin while sucrose, hunter 'a' and hunter 'b' positively correlated to the pectin content.

The higher the value 'a' and 'b' the higher the pectin contents for Abundance and Pobeda. Sugar content of samples had a direct bearing on the measurement of pectins because sugars tend to interfere with the analytical procedure. The method used to measure pectic substances is subject to interference from the nonuronide carbohydrates associated with pectin samples but removal of these compounds entails considerable manipulation (Selvendran, 1975). This caused difficulty in obtaining reliable uronide measurements from samples containing high levels of sugar substances. Although Blumenkrantz and Asboe-Hansen (1973) found m-hydroxydiphenyl reagent was the more specific for uronic acids and less sensitive to extraneous carbohydrate interference, Kintner and Van Buren (1982) reported that when high levels of sugar are present, the interference was increased. Carbonell et al. (1989) concluded that using m-hydroxydiphenyl as

Table 14 The correlation coefficients between pectin content and totol phenolics, brix, glucose, fructose, sucrose, Hunter 'a' and hunter 'b' for pectin content of plum juices

	Au Red	Abundance	Pobeda	Shiro	Peach Plum	Early Golden
Total phenolics	0.73*	0.73*	0.38	0.70*	0.70*	0.91*
Brix	0.29	0.80*	0.74*	0.83*	0.85*	0.97*
Glucose	0.52	0.90*	0.79*	0.89*	0.95*	0.93*
Fructose	0.77*	0.90*	0.79*	0.89*	0.95*	0.93*
Sucrose	0.68*	0.87*	0.80*	0.95*	0.94*	0.99*
Hunter 'a'	0.81*	0.84*	0.68*	0.14	0.10	0.02
Hunter 'b'	0.56	0.97*	0.86*	0.77*	0.83*	0.90*

* significance at 5% level

chromogenic reagent with previously extracted and purified samples was the most effective method for determination of pectin in plum jams (0.9%<CV<6.4%). Coefficients of variation (CV=standard deviation/mean) were higher (8.6%<CV<26.2%) in samples without extraction and purification. In this study, method for determination of pectins was acceptable because the precision (3.5%<CV<8.4%) was good.

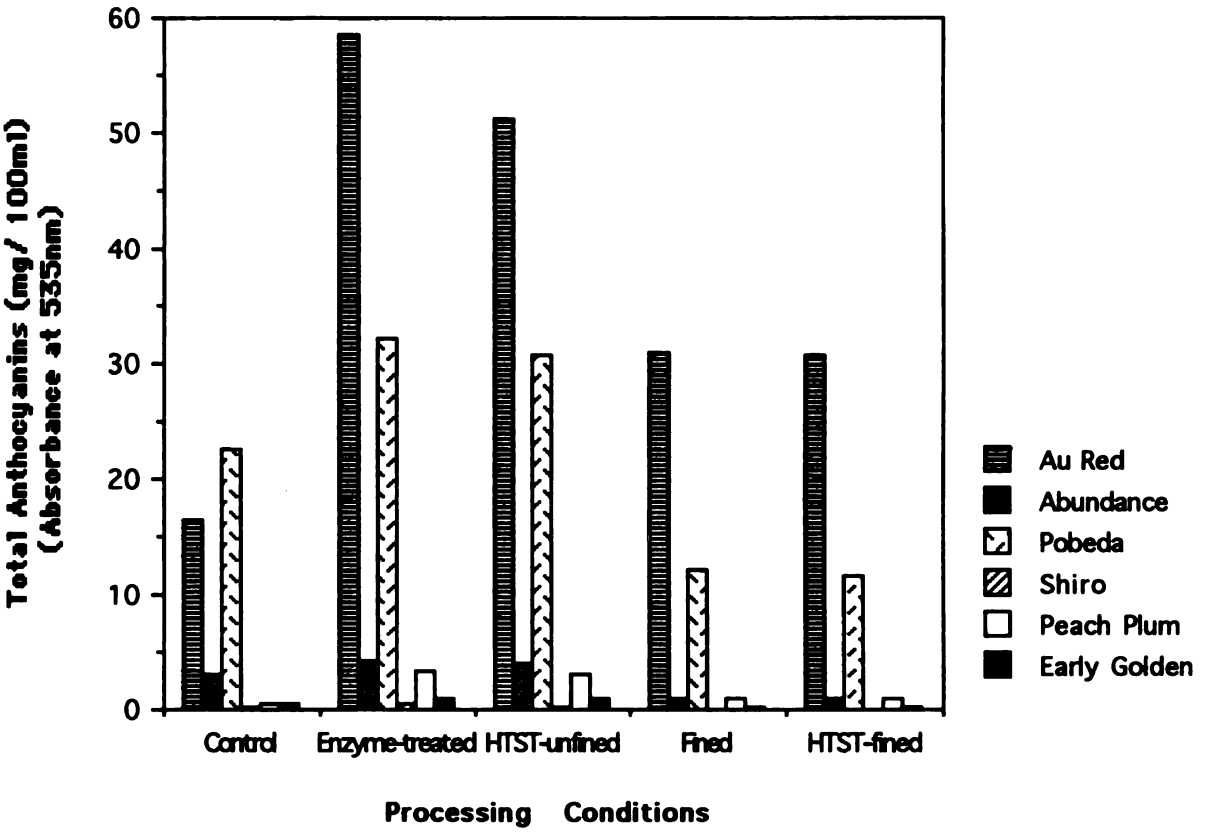
Total Anthocyanins

The major ACYs in plums have been identified as cyanidin 3-glucoside, cyanidin-3-rutinoside, Peonidin-3-rutinoside, and peonidin-3-glucoside by Van Buren (1970) and Gorsel et al. (1992).

In this study, total ACYs were determined according to Zapsalis et al (1965). Red colored cultivars, Au Red and Pobeda, had higher ACY content (16 and 22 mg/100 ml juice) compared to yellow variety, Shiro (0.33 mg/100 ml juice)(Table 9). Draetta et al. (1985) reported ACY content of Carmesim plum to be 29.5 mg/100g.

Enzyme treatment effected release of total ACYs in juice and the enzyme extracted juice had higher total ACYs compared to control juice (Fig. 21). Similar results have been reported by Rommel et al. (1992) who found that enzyme-treated juice had higher total ACYs as compared to only press juice. The increase in total ACYs is believed to be due to

Fig. 21 Effect of Processing Conditions on Total Anthocyanin Content of Plum Juice



preferential release of ACYs into the liquid phase by the action of enzyme.

ACYs are sensitive to heat processing in prune fruits (Raynal et al, 1989) but Siddiq (1993) found ACY pigments were relatively heat stable and only 11 to 16 % losses occurred in Stanley plum juice. In the present study, heating unfined juice produced losses of 3.9 to 12.9% for Peach Plum, Pobeda, Abundance, Early Golden and Au Red. These results show lower ACY losses than the data (37% loss) of Weinert et al. (1990) in canned plums.

The ACYs which are red at pH 2.0 and above change from the orange red of pelargonidin to more purple-red colors with an increase in hydroxylation of the B ring of ACY molecule. As the pH of ACYs increase to alkaline range, the color changes to blue or purple, although sometimes yellow colors are obtained. In this study the pH of plum juice ranged from 3.01 to 3.40 (Table 11) for all plum varieties so the juices with ACY showed the purple-red color.

The change in Hunter L value can be used as a browning index. In this study, the browning (ΔL) was significantly ($r=0.886$, $p<0.05$) correlated to the degradation rate of ACYs during heating treatment. Greater the browning the lower the ACYs. Both enzymatic (Chichester and McFeeters, 1970) and thermic reactions are possible factors that effect disappearance of ACYs from plums and subsequent browning of the juice. PPO is responsible for the enzymatic browning and causes a loss of characteristic color of ACY containing fruit

products (Siddiq, 1993). Arnold (1992) found ACY degradation can occur from high temperatures during processing.

A study of relationship between total ACYs and Hunter color difference measures (CDM) was also made (Table 15). The ACY content of juice made from Abundance, Shiro and Peach plums was significantly correlated with CDM L, a and b values. The higher the ACY content, the lower was the Hunter L, and the higher were the Hunter a and b values in plum juice. The 'a' value is a measure of redness so it can represent ACYs in sample. Au Red was an exception and had the highest ACYs but a lower 'a' value. This may be due to the lower % transmittance of Au Red juice, which interfered with the measure of color meter.

Total Phenolics

Phenolic compounds contribute to the color, flavor, astringency, enzymatic and non-enzymatic browning of horticultural products (Walker, 1975). To consumers, the most evident properties of phenolic compounds are the colors and astringent taste they impart to foods.

The total phenolics (determined as tannic acid) of press plum juice in this study ranged from 26 to 205 mg/ 100 ml. There was considerable difference in total phenolics of plum juice samples which ranged from 96.55 to 417 mg/100 ml (Table 9). Plum juices made from Pobeda and Au Red had higher total phenolics compared to Shiro and Early Golden. Besides cultivar differences, processing factors had an effect on

Table 15 The correlation coefficients between Hunter CDM values and anthocyanins in plum juices

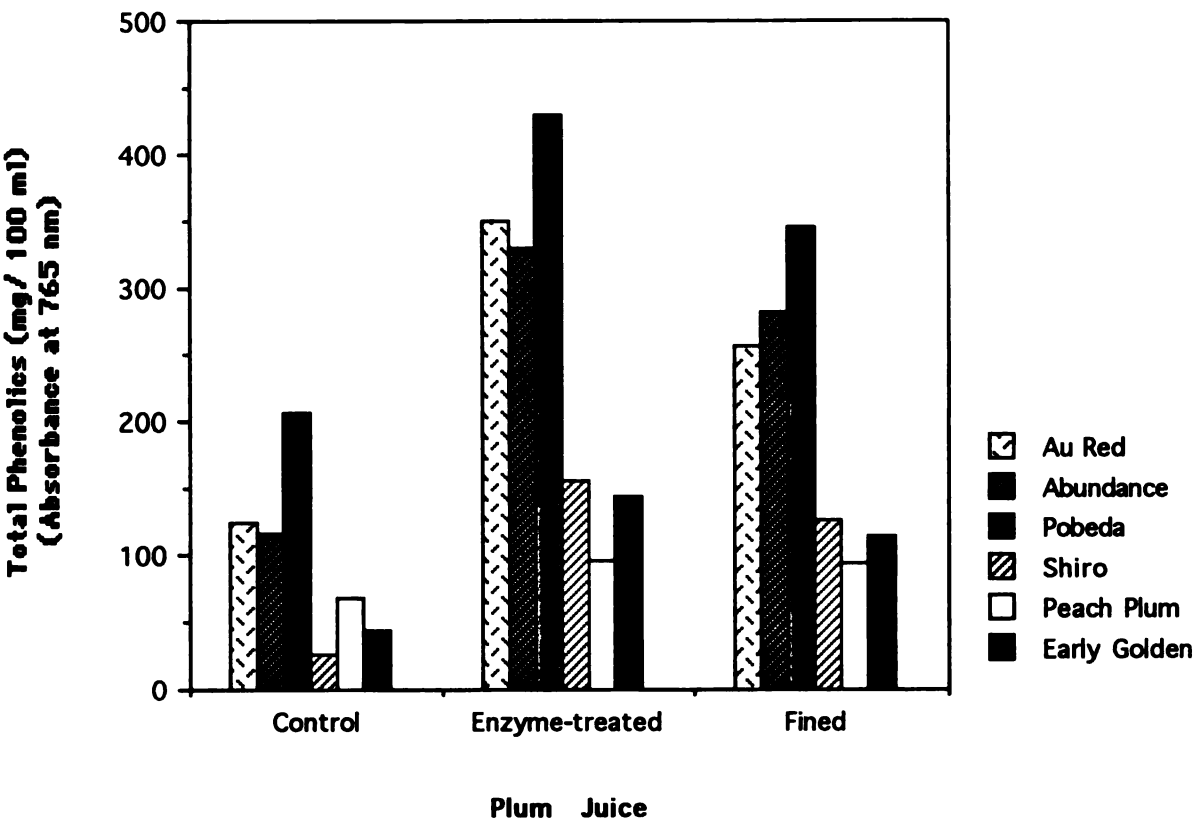
	Au Red	Abundance	Pobeda	Shiro	Peach Plum	Early Golden
L	0.159	0.814**	0.612	0.974**	0.647*	0.566
a	0.019	0.886**	0.651*	0.758**	0.963**	0.980**
b	0.171	0.671*	0.338	0.737*	0.638*	0.216

* significance at 5% level; ** significance at 1% level

total phenolics of juice. Enzyme treatment increased the levels of phenolics while fining decreased phenolic content of juices (Fig. 22). Enzyme treatment effected Peach Plum phenolics less (only 43.9% increase) than other cultivars which had 2-4 fold increases as compared with the untreated juice. Pectic enzyme added to apple juice for clarification has been shown to release phenolics from cell wall so the amount of total phenolics was raised (Spanos et al, 1990). However, addition of gelatin has been related to decrease in total phenolic content in prune juice. In this study addition of gelatin brought about 25% decrease in phenolic content of plum juice samples. Bannach (1984) found losses up to 50% of phenol when using gelatin fining in prune juice.

Methodology is the key to this analysis. The results of this study are two to three times greater than the results of Siddiq (1993) who analyzed the total phenolics in Abundance, Pobeda, Shiro and Stanley by the colorimetric method of Coseteng and Lee (1987). Gorsel et al. (1992) analyzed California plums and prunes by HPLC. Their results are 10 times less than the data from this study. HPLC quantitation in samples that have undergone considerable phenolic degradation would surely give a lower concentration of phenolics. Van Buren et al. (1976) found that the degradation of phenolics occurred during processing and storage and formed brown intermediates, such as enediols and reductones which created significant interference with the colorimetric assay which was used in this study. The

Fig. 22 Changes in Total Phenolics Content of Plum Juice



reactivity of interfering compounds influences the colorimetric measurement of phenolics in plum juice.

Sensory Evaluation

Plum juice made from six plum cultivars by using Clarex L enzyme was pasteurized (90ºC, 90 sec) according to method described by Arnold (1992). The plum juice was adjusted to brix/acid ratio of 13.5-16.6, as was determined in preliminary taste test (Fig. 23).

Table 16 shows the average sensory scores of juice from six plum varieties. The astringency, color and sweetness differed significantly ($p<0.01$) among all juices by judges. Juice made from Au Red and Pobeda were evaluated by the judges as the deepest in red color and the juice from Shiro was the most yellow, with the others intermediate in color. While sensory scores for astringency are least for juice made from Pobeda and Peach Plum, juice from Au Red is evaluated the most astringent. Plum juice made from Pobeda, Abundance and Peach Plum were considered by the panel as the sweetest, least astringent, most red and highest in flavor.

Positive correlations between scores for astringency and tartness were found in Au Red ($r=0.44$, $p<0.01$), Shiro ($r=0.49$, $p<0.01$), Peach Plum ($r=0.39$, $p<0.05$) and Early Golden ($r=0.63$, $p<0.01$). This association of astringency and tartness has been shown in citrus juice to be commonly present (Fellers, 1980). For Peach Plum juice, flavor seemed to have a significant influence ($r=0.44$, $p<0.01$) on the

Fig. 23 The suitable brix/acid ratio on plum juice for sensory evaluation

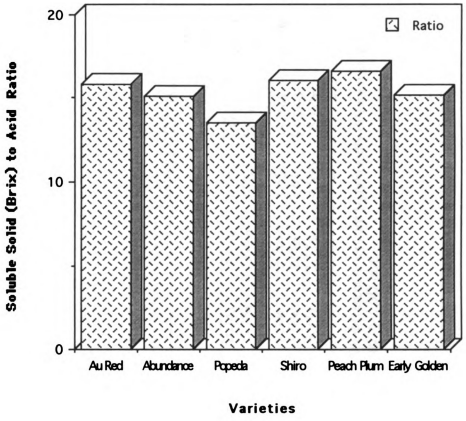


Table 16 Means of sensory attributes for six varieties plum juice

Cultivar	Sensory Attributes					
	Tartness	Astringency	Color[x]	Sweetness	Flavor Preference	Overall Acceptability
Au Red	4.74a	3.95b	9.02d	4.69a	4.61ab	4.61abc
Abundance	5.28a	3.61ab	7.78c	5.69abc	5.88cd	5.39bcd
Pobeda	4.75a	2.64a	8.76d	6.51c	6.07d	5.80d
Shiro	4.48a	3.56ab	1.55a	5.26ab	4.82bc	4.39ab
Peach Plum	4.58a	2.78a	7.63c	5.62abc	5.28bcd	5.54cd
Early Golden	4.94a	3.37ab	4.59b	6.12bc	3.63a	4.01a

Mean in columns with different letters are significantly different at the 5% level.
Means were separated by LSD test.

judges' perception of astringency, with less astringency being related to a better flavor. Although it is difficult to assign a reason for this correlation it may be due to, or associated with , the fact that higher concentrations of phenolics which contribute to astringency may mask aroma volatiles.

Correlation coefficients among the sensory attributes and overall acceptability was determined (Table 17). The overall acceptability showed significant correlation for all juices with flavor preference of judges calculated across groups. From table 16, Pobeda had the highest score for flavor preference, with Peach Plum and Abundance also showing high flavor preference by judges and the overall acceptability of these three juices was high. Sweetness significantly ($p<0.05$) correlated with acceptance for Au Red, Abundance and Early Golden. The higher the sweetness the higher acceptability. This result agrees with LaBelle et al. (1960) who reported positive effects of sweetness and brix/acid ratio on flavor and acceptability of apple sauce. The color, astringency and tartness of Peach Plum had significant correlation ($p<0.05$) with overall acceptability. The astringency of Au Red and Abundance highly correlated with acceptance of these two juices. The juices from Pobeda, Peach Plum and Abundance gave the highest score for overall acceptability (Table 16).

In summary, the laboratory evaluation of acceptable plum juices indicated that these juices could be characterized by

Table 17 Correlation coefficients between overall acceptance and color, sweetness, astringency, tartness and flavor preference of plum juice

Varieties	Flavor	Sweetness	Astringency	Tartness	Color
Au Red	0.745**	0.399*	0.381*	0.104	0.138
Abundance	0.819**	0.334*	0.319*	0.303	0.062
Pobeda	0.730**	0.226	0.083	0.006	0.053
Shiro	0.657**	0.294	0.019	0.107	0.186
Peach Plum	0.724**	0.295	0.501**	0.364*	0.394*
Early Golden	0.794**	0.414*	0.107	0.104	0.283

* Significance at 0.5% level
** Signficance at 0.1% level

sweetness, tartness, color and astringent sensations but the preference rating for plum juice could be adequately predicted by flavor preference ($r=0.75$, $p<0.01$) under the condition that brix/acid ratio was suitable.

SUMMARY AND CONCLUSIONS

In this study, five commercial pectinases were used in plum juice processing. They all had some ability to improve the yield, color (release of anthocyanins) and clarity of juice but at concentration higher than 0.2% they tended to cause a bitter flavor in the juice. Among five pectinases, 0.2% Clarex L was identified as the best enzyme for plum juice yield and quality so it was used with six new plum varieties being evaluated for juice processing.

Of these six varieties, Au Red had the highest content of pectin while Pobeda showed the lowest value for pectin. Press juice from Au Red gave the least juice yield and % transmittance but it was the sweetest of all varieties. Pobeda and Au Red were red colored varieties so they had the highest content of ACYs. Shiro was a yellow plum and had the lowest ACY content. Pobeda was the most acidic variety and had the highest phenolic content. The juice from Abundance showed the lowest amount of total sugar.

The use of 0.2% Clarex L enzyme with the new varieties increased juice yield, clarity (i.e.. percent transmittances) soluble solids, titratable acidity, content of total ACY and total phenolics, but caused decreases in the pectin contents in most instances. The color and pectin content of juice from Au Red variety was least affected by enzyme treatment of

any of the varieties. Fining agents removed pectin, ACYs and phenolics but increased the % transmittance of juice. This almost sediment-free, clear juice was considered to be more like an artificial juice by consumers because the loss of natural color and brilliant clarity made it seem somewhat unnatural in appearance. The fined juice are more stable than unfined juice. The turbidity, total phenolics and total ACYs of unfined juice were decreased by heat but heat didn't significant effect fined juice characteristics.

All cultivars included in this study produced highly acceptable juices. The Brix/Acid adjustment had the biggest impact on sensory ratings, causing greater acceptance of juice from all cultivars. The juice from Abundance, Pobeda and Peach Plum showed the highest flavor preference and acceptability by consumers.

Future areas of research in varietal plum juice should explore the development of juices using blends of several different cultivars. Acceptable apple juice is almost always made by blending two or more varieties of apples and the same principle should be applied to plums. To obtain the best product with a consistent flavor, juice from several cultivars should be blended to obtain the desired balance between acidity, sweetness, aroma, and astringency. Proulx and Nichols (1980) make some suggestions for blending. They recommend sweet, low-acid cultivars such as Au Red for the basic juice. Plum such as Pobeda possess higher acid levels and would add tartness to the juice. Better flavored

varieties, Pobeda, Abundance and Peach Plum, could also be added to the blend. The proper proportions and combinations of these and other varieties needed to achieve the exact blends should be the subject of further work.

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