

THE UNIVERSITY OF MANITOBA

CHEMICAL AND ORGANOLEPTIC EVALUATION OF MEMBRANE FILTERED
APPLE JUICE PRODUCED FROM MANITOBA GROWN
APPLES AND CRABAPPLES.

by
Vincent D'Souza

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OF MASTER OF SCIENCE

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BY

VINCENT D'SOUZA

A thesis submitted to the Faculty of Graduate Studies of
the University of Manitoba in partial fulfillment of the requirements
of the degree of

MASTER OF SCIENCE

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DEDICATION

TO MY PARENTS, BROTHERS AND SISTERS, AND TO
ROSE AND FILOMENO CARVALHO.

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ABSTRACT

A two year study was undertaken to assess the chemical and organoleptic qualities of one crabapple and nine apple cultivars grown on the Canadian Prairies for commercial production of apple juice. Considerable variation was observed among the cultivars for moisture (82.24% - 88.19%), acids (0.49% - 1.09%), sugars (9.00% - 13.84%) and phenolics (0.31% - 0.53%). A pilot plant scale filtration assembly was utilized in this study. Pectinase concentration of 0.15%(w/v) was most efficient in clarification of raw apple juice. The enzyme clarified apple juice was pumped through a cartridge membrane filtration system ranging from 106 to 0.22u for clarification, polishing and sterilization. The cartridge membrane filters reduced the turbidity of the juice from 106 to 0.3 JTU (Jackson Turbidity Unit). Yeast and mold counts were nil on the final product. Organoleptic studies revealed that perceived sweetness increased as the sugar-acid ratio increased, while sourness increased with a decrease in sugar-acid ratio. Breakey and Collet cultivars had the acceptable sugar-acid ratio required to make single cultivar juice. The other cultivars were less suitable due to wider range of compositional differences. The blended juices prepared from the sample juices compared very well with commercial apple juice.

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INTRODUCTION

Apple production on the Canadian Prairies has received strong support from the various segments of the Horticulture Industry since the 1930's. Unfortunately the rigorous climatic conditions have prevented the utilization of the major apple cultivars. The Western Canadian Society for Horticulture initiated the Cooperative Fruit Breeding Program in the 1940's to select and propagate hardy fruits suitable in the Prairie region. There were three phases in this program: 1) several crosses made at Morden, Manitoba were distributed to other Prairie testing sites (ie. Brooks, Alberta and Melfort, Saskatchewan); 2) selections were made from the crosses based on the hardiness, resistance to disease and overall fruit quality; 3) these selections were then named and released to the commercial trade. The ultimate objective of the program was to have tree fruits in every farmstead. At no time was the program identified with the establishment of commercial orchards on the Prairies.

The per capita consumption of apple juice has increased dramatically in the last decade from 4.18 kg in 1970 to 8.50 kg in 1981. Changes in the life style and improvements in juice quality through developments in juice processing technology are two possible reasons for the increased popularity of apple juice. The apples developed by the Cooperative Fruit Breeding program have become popular in the rural communities on the Prairies for juice because of their unique strong apple flavors. Their potential for this industry is unknown at this time.

The Food Science Department and the Agriculture Canada

Research Station at Morden, Manitoba initiated a program of studies designed to assess the food quality characteristics of some of the apple cultivars selected by the Cooperative Fruit Breeding Program. It has generally been stated that these cultivars will not produce fruit acceptable for the fresh trade because they are smaller in size than normal commercial apples. Prior to this study these cultivars have never been evaluated for the production of processed products. Hence this two year study (1983 and 1984) was the completion of a three year program to characterize the overall quality of one crabapple and nine apple cultivars.

The specific objectives of this study were:

1. to analyze the fruit for moisture, pH, acidity, soluble solids, and phenolic content.
2. to process the fruit into juice utilizing a cartridge membrane filtration system and to evaluate the efficiency of this system for a small juice processing plant.
3. to analyze the juice for pH, acidity, sugars and phenolic content.
4. to assess the sensory qualities of the juices from ten cultivars.
5. to evaluate the potential of the juice from the cultivars used in this study for commercial apple juice.

2. LITERATURE REVIEW

2.1. General:

Commercial production of canned and bottled apple juice started in North America about 1937 (Moyer and Aitken, 1980). It has gradually increased in importance since that time. A strong cottage industry for the production of apple cider had developed in the major apple producing regions of North America prior to the 1950's. Today, apples are processed into a wide variety of products which are classified as liquid, solid, or pureed products (Table 1). According to La Belle (1981), apples may differ in size, shape, specific gravity, color, firmness, soluble solids, acidity, and pH because of variety, maturity, or postharvest conditions. The importance of various quality characteristics for the processing of apple juice is presented in the following section.

2.2. Processing Parameters for Apples

Alston (1979) classified the fruit into four categories: eating quality, appearance, storage quality and processing quality. The fruit used for juice is normally second grade (Smock and Neubert, 1950). According to Moyer and Aitken (1980), juice manufacturers use varieties which are available in surplus or rejected from the fresh fruit market mainly due to their physical injury, size, shape, color and blemishes.

2.2.1. Soundness

In order to produce a good quality juice, the apples should

Table 1. Classification of products processed from apples

<u>Liquid</u>	<u>Solid</u>	<u>Pureed</u>
Cider(fresh)	Baked whole apple	Apple sauce (including baby food)
Juice(fresh or pasteurized)	Slices or pieces	
Juice or concentrate	Fresh	Nectar
Hard (fermented cider)	Frozen	Apple butter
Wine	Dehydrofrozen	
Vinegar	Rings (canned, spiced)	

Adapted from La Belle (1981).

not be wholly or partially decayed because these can impart a "moldy" flavor to the juice. According to Moyer and Aitken (1980), decayed fruits produce Patulin a mycotoxin. Recently, Roland et al. (1984) reported that Patulin is highly toxic and has been shown to be carcinogenic in laboratory animals. Patulin is produced by Penicillium expansum, P. patulum and Aspergillus lavatus (Moyer and Aitken, 1980; Scott and Bullerman, 1975). Other blemishes and defects seem to have little effect on the quality of the juice. For example, Moyer and Aitken (1980) reported that scab, scale spots and aphid injury do not adversely affect the quality of juice. However, Smock and Neubert (1950) reported that a physiological disease known as "cork" seriously injures the quality of apples and results in lower yield without contributing any off-flavor.

2.2.2. Maturity

Maturity of apples is perhaps the most important factor in the production of apple juice. The immature and overmature apples seem to be unsuitable for the production of juice. According to Moyer and Aitken (1980), immature apples tend to have high acid and an astringent taste. They lack in sweetness, apple flavor and impart a "starchy" or green apple taste due to a high percentage of starch. Krishnaprakash et al. (1985) observed that the position of the apple on the tree influenced the rate of maturation. Fruits at the bottom of the tree matured earlier than those at the middle and the top. They suggested that it is desirable to harvest apples from the bottom half of the tree earlier than the

top with a gap of 7-10 days between harvests. Improvement in the quality of the juice is due to the conversion of starch into sugars (Moyer and Aitken, 1980). Sapers et al. (1977) reported that with different harvest times (maturities) there was a decrease in acidity, with an increase in soluble solids and volatile composition in McIntosh apples. According to Bradley and Brown (1969), fruit from the early and midpicking season was unsuitable for juice processing. After a certain period of storage, however, it develops satisfactory processing characteristics. These beneficial storage changes are due to the breakdown of starch which results in an increase in the sugar content of the fruit. Hence, with storage and maturity the quality of the juice is improved mainly due to an increase in sugar content (sweetness), a decrease in acidity (sourness), and an increase in the volatile composition (aroma) of the apples.

On the other hand, overmature apples are unsuitable for juice production because they are difficult to press, filter, and clarify, thus resulting in a low yield. The juice is of a poor quality lacking in flavor. LaBelle (1981) reported that overripe apples have high levels of suspended solids in the juice. Even though the suspended solids are removed during clarification, the filtration efficiency and juice yield are affected by their high level. Overmature fruit is more susceptible to mealiness, fungal disease and breakdown due to senescence (Anonymous, 1974). There is no easy way to make an exact assessment of maturity. Indices of maturity include skin color, flesh color, seed color, starch content, ease of picking and the Magness Taylor pressure

test (Anonymous, 1974).

2.3. Juice Processing

2.3.1. Clarification

North American consumers prefer apple juice free from suspended solids. However, fresh apple juice has a cloudy appearance due to the presence of suspended solids. Suspended solids are colloidal in nature because they consist of mucilaginous, hydrophillic gums and pectic substances (Mian and Bhatti, 1969; Smock and Neubert, 1950). Clarification of apple juice is necessary in order to prevent the rapid clogging of filters during the filtration process (Moyer and Aitken, 1980).

To accomplish clarification numerous methods have been proposed. It is based on the ability to disrupt the colloidal system which can be achieved by chemical or mechanical means. Carpenter et al. (1932) recommended that the juice be flash heated to 82°C for twenty seconds and cooled immediately to coagulate the suspended particles. Probably the best known and most widely used methods are the gelatin-tannin and enzyme (pectinase) methods for clarifying apple juice. Small amounts of pectic enzymes occur naturally in fresh apple juice (Anonymous, 1982). According to Smock and Neubert (1950), clarification must be fairly rapid to prevent fermentation. Therefore, pectinase enzyme must be added to reduce the time requirement for clarification so that fermentation will not occur during treatment.

2.3.1.1. Enzyme Clarification

The filtration procedure is simplified if the juice is subjected to enzyme clarification which is capable of disrupting the colloidal system. Pectin is a protective colloid which retards the settling of particles. Pectin is made up primarily of (1-4) α -D-polygalacturonic acid in which two thirds of the carboxylic groups are esterified (Eskin et al. 1971). Product characteristics affected by pectin include viscosity, color stability, clarity and possibly flavor (Kilara, 1982). Hence it is necessary to depolymerize the pectin with pectinase during clarification. The commercial term "pectinase" is given to an enzyme preparation which is obtained from molds of the genus Aspergillus (Kilara, 1982). It is available in liquid and dry powder form. It consists of a mixture of polygalacturonase and pectin esterase (Smock and Neubert, 1950). The enzyme has the ability to hydrolyze pectin and reduce the viscosity of the juice thus making it easier to filter.

A number of enzyme preparations have been employed to clarify apple juice. Early investigations include the use of protein and starch splitting enzymes (Smock and Neubert, 1950). These enzymes were not able to produce filterable juice due to the fact starch and protein substances are only minor constituents of apple juice. Recently, McLellan et al. (1985) suggested the use of honey and enzyme separately and in combination for clarification of apple juice. They also reported that the combined treatment of enzyme and honey induced rapid flocculation as compared to enzyme alone at cold as well as warm temperatures. McLellan et al. (1985) recommended the use of 1.0% honey for clarifica-

tion. The exact mechanism of clarification with honey or honey plus enzyme is still not clear, but it is assumed to be a protein-tannin reaction which is similar to the commercial gelatin-tannin method (Kime, 1983). Mian and Bhatti (1969) recommended the use of pectinol enzyme (0.1%) and bentonite (0.5-0.6%). However, Moyer and Aitken (1980) suggested that tests be carried out to establish the proper enzyme concentration.

Parameters affecting clarification are pH, temperature, contact time and enzyme concentration. The pH is influenced by variety and maturity. The pH of apple juice is between 3.2-4.0, falling within or slightly below the optimum range exhibited by most commercial enzymes (Kilara, 1982). McLellan et al. (1985) reported that as temperature increased the rate of clarification increased. Denaturation of the enzyme was observed at higher temperature (45-60°C) (Kilara, 1982). Commercially available enzyme contains 0.005 to 0.01% gelatin. It seems that gelatin helps in clarification by forming a gelatin-tannin complex. Gelatin is positively charged, whereas the colloidal material dispersed in apple juice is negatively charged, thus the oppositely charged particles coalesce and precipitate. It is still not clear if gelatin promotes floc formation or speeds up pectinase action by removing inhibitors such as tannins (Hathaway and Seakins, 1958). According to Moyer and Aitken (1980), gelatin cuts the clarification time in half whereas, Smock and Neubert (1950) reported that gelatin stabilizes the enzyme in solution. In general, the time required for clarification is inversely proportional to the concentration of the enzyme used at constant temp-

erature (5-50°C) and treatment time (2-16 hours).

2.3.1.2. Clarification Mechanism

The mechanism of clarification can be divided into three distinct stages:

1. Enzyme Hydrolysis
2. Flocculation
3. Sedimentation

Freshly pressed apple juice has a cloudy appearance due to the presence of suspended particles. Pectin prevents the settling of particles due to its colloidal properties. Baumann (1981) observed that spontaneous clarification was not possible because pectin prevented proteins from reacting with polyphenols. He further stated that each stage of clarification is influenced by viscosity.

In the first stage of clarification the colloidal properties of pectin are disrupted by the addition of the enzyme due to enzymic hydrolysis. Baumann (1981) reported that the second stage of clarification will occur only when the viscosity is decreased. This can be achieved when 5-10% of the glycosidic linkages are split. The floc formed eventually settles down due to the electrostatic interaction between unlike charges (Kilara, 1982).

2.3.1.3. Sedimentation in Clarified Apple Juice

Clouding followed by sediment formation has been a problem ever since apple juice was first manufactured. It occurs during storage of clarified apple juice. Kilara (1982) termed this defect as "After Haze", that occurs in juice processed at temper-

atures higher than storage temperatures.

Formation of sediments in fermented pear juice was first recorded by Kelhofer in 1908 was reported by Johnson et al. (1968). They concluded that the sediments contained pectin, protein and oxidized tannins. However, Neubert and Veldhuis (1944) reported that the sediment was probably a phlobaphene, a polymeric phenolic material. Kilara (1982) suggested that leucoanthocyanidins and catechins were the precursors of the polymeric phenolic fraction of the sediment formed during the milling and pressing operations. Although chlorogenic acid is one of the principal polyphenols occurring in apples, it is doubtful that it contributes towards sediment formation (Johnson et al., 1968). According to Heatherbell (1976), sediments could arise from incompletely degraded pectins and starch-tannin complexes. It is reported that amylose and/or amylopectin fragments eventually precipitate (retrogradation) and remain in the apple juice during clarification and filtration. Apparently amylose/amylopectin polymers can complex with small amounts of protein and phenolic substances during retrogradation (Heatherbell, 1976). Retrogradation is accelerated at low temperatures by the presence of tannins and proteins (Whistler, 1953).

Sedimentation problems in clarified apple juice could be overcome by the addition of nonacidulated, starch-free, liquid pectin just prior to flash pasteurization (Smock and Neubert, 1950). On the other hand, Hulme (1958) recommended the use of only fully matured or stored apples which have a low starch content.

2.3.1.4. Effect of Clarification on Juice Quality

Several investigators have studied the chemical changes occurring in juice as a result of clarification. According to Smock and Neubert (1950) suspended solids contribute flavor to freshly pressed apple juice, which is lost during clarification. However, the removal of suspended solids is advantageous because cloudy juice tends to develop a cooked flavor. Kilara (1982) observed a reduction in viscosity due to the removal of pectin in the enzyme treated juice whereas McLellan et al. (1985) observed just the opposite in the honey treated juice.

Loss of astringent substances including tannins was observed in enzyme treated juice (Smock and Neubert, 1950). This loss cannot be explained on the basis of enzyme action and is probably due to coprecipitation with other suspended material. The gelatin-tannin treatment has been found to have a variable effect on astringency and the tannin content of apple juice. According to Moyer and Aitken (1980) gelatin-tannin clarification yields juice much lighter in color as compared to enzyme treated juice. McLellan et al. (1985) observed an increase in °Brix and total solids due to an increase in the sugar content of the honey clarified and honey-enzyme treated juice. In short, consumers prefer clear apple juice, but they are very critical about the changes caused in apple juice as a result of the clarification process (Smock and Neubert, 1950).

2.3.2. Membrane Filtration

Since the early 19th century, the traditional plate and frame

filter press incorporated mainly with an asbestos sheet was used for the clarification and sterilization of juices (Wale, 1982). However, the plate and frame filter press had its own disadvantages like leakage problems, plate gasket alignment, contamination of the exposed filter sheet, and the long cumbersome set up procedures involved. Fiore and Babineau (1979) reported health hazards like pulmonary fibrosis, bronchogenic carcinoma and gastrointestinal tract cancer with the asbestos material. Asbestos fibers were also found in beer (Biles and Emerson, 1968). In 1976 the FDA restricted the use of filters containing asbestos. Since then the use of membrane filters has greatly increased.

Membranes are made from a variety of synthetic materials including cellulose acetate, cellulose nitrate, polypropylene and polycarbonate. Membrane filters are basically screen-type filters (Rankine, 1983). They have the capacity to remove all particles larger than the pore size of the membrane (Reeves, 1983).

Filtration is defined as the process of separating a solid from a liquid by means of a porous substance through which only the liquid passes. There is no one process which meets all requirements. The first filtration operation is coarse filtration either through a diatomaceous earth filter or a coarse grade of sheet filter. Membrane filters are used prior to bottling in order to ensure complete removal of microorganisms and improve clarity. Filtration is a highly efficient and relatively inexpensive process for product recovery, clarification, and stabilization (Fiore and Babineau, 1979). Stabilization of juice by means of filtration represents: