THE UNIVERSITY OF MANITOBA

CHEMICAL, PROCESSING AND SENSORY STUDIES FOR THE PRODUCTION OF APPLE JUICE FROM TEN HARDY APPLE CULTIVARS GROWN ON THE CANADIAN PRAIRIES

by
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TO MY ELDERS

ABSTRACT

The present investigation included studies in three major areas related to the utilization of apple cultivars grown on the Canadian Prairies. These areas were: a) raw material analysis , b) a process development and c) final product characterization for the production of apple juice.

Ten apple cultivars were chemically characterized by standard methods. Most of the cultivars had average sugar contents (9.12 - 10.26%) except for Goodland, Heyer #12 and P.F. 36 which were below average (7.34 - 8.84%). P.F. 51 and Norland were found to have medium acid contents (0.48 - 0.55%), while other cultivars had a high level of acid contents (0.60 - 0.99%). All the cultivars also had high phenolic contents (0.28 - 0.42%) except for P.F. 51 which was found to have a medium phenolic content (0.27%). All the cultivars had higher than average moisture contents. On the basis of chemical characterization Norland and P.F. 51 were identified as the most suitable cultivars for the production of liquid and pureed products.

The chemical characterization by chromatographic analysis showed that these cultivars had lower than average concentration of sucrose (0.00 - 1.11%) but average concentrations of fructose (4.40 - 6.96%) and glucose (1.43 - 3.37%). The major organic acids were malic, citric, quinic and shikimic. A comparison of chromatographic analysis and standard method for the determination of total sugar content and acid content

indicated a significant correlation between the two methods.

Preliminary studies on the filtration processing of apple juice were conducted on a pilot plant scale filtration assembly. From these studies turbidity was found to be a good parameter for monitoring the filtration processing of apple juice. Significant decreases in acid (11.54%) and phenolic (16.80%) contents of apple juice were observed during the filtration processing. An increase in the clarity of the juice was also observed. The changes in apple juice during the filtration processing were found to have a positive effect on the quality of the juice by decreasing sourness and bitterness, and by increasing its preference.

Psychophysical analysis of commercial juices was conducted to develop an understanding of the dependence of preference for apple juice upon its sensory and chemical properties. Results of this analysis showed that sensory taste properties ($r^2 = 0.77$) were more important than the taste contributing chemical properties ($r^2 = 0.66$) for the determination of preference for apple juice. The most practical combination of sensory and chemical properties for the determination of juice preference included perceived aroma intensity, sugar content and acid:phenolic balance of apple juice. The equation for preference determination was Y (preference) = -8.11 + 16.99 (log aroma intensity) + 1.28 (sugar content) + 0.76 (acid:phenolic balance), $r^2 = 0.99$.

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TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

APPENDICES

1. INTRODUCTION

The production of apples constitutes the largest volume and greatest commercial value of any fruit in Canada (Statistics Canada, 1983 a). This trend has continued for quite some time. According to Agriculture Canada (1983), from 1977 to 1981, per capita consumption of fresh apples was comparable to oranges and apple juice consumption was second only behind orange juice. However, Canada has been a total importer of orange juice, a net importer of apples, but a net exporter of apple juice (Table 1). A review of fruit and vegetable production and processing statistics indicates that at the present time Manitoba does not significantly contribute to the Canadian fruit industry (Statistics Canada, 1983, a and b).

As early as 1955, it was recognized that in Manitoba the expansion of the fruit and vegetable industry would depend upon the appropriate supplies of raw materials, and that such supplies might not be possible for certain fruits and vegetables (Little, 1955). Since then Manitoba has achieved considerable success in fruit production. The Department of Agriculture, Province of Manitoba reported (Manitoba Agriculture, 1980 - 81 and 1981 -82), good yields of apples in commercial orchards, and a bumper crop of apples and applecrabs throughout the province. During the same period three hardy apple varieties were released by the Prairie Cooperative Fruit Breeding Program from Agriculture Canada Research Station at Morden, Manitoba. These varieties have provided broader range of hardy varieties for commercial development.

Table 1: Production and disappearance of apples, apple juice, oranges and orange juice in Canada (1977 - 81)

Product	Year	Production	Import	Export	Disappearance per/capita
		-Kilotonnes-			Kg.
Apple fresh					
	1977	411.42	66.74	47.89	11.16
	1978	451.99	74.49	56.09	10.75
	1979	434.90	96.08	54.91	11.85
	1980	552.59	78.60	60.31	10.77
	1981[1]	417.38	108.72	75.18	12.96
Apple Juice					
	1977	116.30	---	4.32	6.69
	1978	131.10	---	3.41	6.97
	1979	134.09	---	4.12	7.21
	1980	159.66	---	5.07	8.32
	1981[1]	159.40	---	12.47	8.50
Orange fresh					
	1977	---	340.69	---	14.17
	1978	---	261.26	---	10.79
	1979	---	249.12	---	10.20
	1980	---	294.90	---	11.80
	1981[1]	---	361.99	---	12.02
Orange juice					
	1977	---	220.39	---	17.39
	1978	---	259.83	---	20.35
	1979	---	273.06	---	21.21
	1980	---	286.46	---	21.88
	1981[1]	---	324.31	---	24.49

[1] - Preliminary estimates.

At the present time the potential for expansion within the apple and apple juice industry in Canada is growing. The development of a supply of raw material would create a favorable environment for the expansion of this industry in the Prairie region. Initial expansion would probably be initiated at a smaller level (providing for the local market).

This study was initiated to assess winter hardy apple cultivars for their suitability for processing ,especially for apple juice, and to develop a processing system for the production of apple juice which did not require a large initial capital investment. Overall studies were designed to meet some practical interests of cultivar assessment and process development, and also to meet some academic interests related to the potential utilization of apple cultivars.

Intensive analysis was conducted on nine apple and one crab apple cultivars, which were selected in consultation with Agriculture Canada Research Station, Morden, Manitoba. These apple cultivars were chemically characterized to determine their suitability for the production of processed products. The chemical characterization was further extended to determine the sugar and acid profiles of these cultivars. Chromatographic determination of sugar and acid content together with the determination by standard (A.O.A.C., 1975) methods also provided an opportunity to compare these methods.

A pilot plant filtration assembly was constructed and used to clarify apple juice from the test cultivars. The juice was further polished by filtration through a 1 μm filter then sterilized through a 0.22 μm filter. The juice quality was determined by chemical and physical analysis. The physico-chemical changes in juice during filtration processing were also monitored to assess its effect on the juice quality.

Psychophysical analysis was conducted on five commercial apple juices. The samples were evaluated by magnitude estimation for aroma and several taste properties. The objectives of this study were to understand functional relationships (dose - response) between preference, sensory and chemical properties of apple juice, which could be used in producing an acceptable juice from these cultivars.

2. LITERATURE REVIEW

2.1 Apple Quality Characteristics For The Production Of Apple Juice

2.1.1 General

Apples constitute the largest commercial value of any fruit in Canada. Beside being consumed as fresh fruit, apples are also processed into various products. The suitability of an apple variety for processing into various products depends upon certain quality characteristics. Some of the characteristics in which apples may differ because of variety, maturity or postharvest conditions are: size, shape, specific gravity, colour, firmness, soluble and total solids, acidity, and pH (LaBelle, 1981). The various processed products have different sets of required quality characteristics, which are important for producing acceptable final products. The following section presents a description of the apples that are used in the production of apple juice, along with the importance of various quality characteristics in the processing of apple juice.

2.1.2 Apples for the Production of Juice and Their Important Quality Characteristics

Normally, the manufacturers use the varieties which are available in surplus or rejected from the fresh fruit market because of physical injury, unattractive shape, size, colour or blemishes (Moyer and Aitken, 1980). In selecting from the available supply a manufacturer may check on the quality characteristics of the apples that are important for the production of juice. These quality characteristics can be classified into two groups Physical/Physiological and Compositional quality characteristics (Table 2, LaBelle, 1981).

2.1.2.1 Physical and Physiological Quality Characteristics

2.1.2.1.1 Maturity

The immature and over mature apples have been found to be unsuitable for the production of juice. Moyer and Aitken (1980) reported that immature apples produce a juice which lacks in apple flavour and have a "starchy" taste. The juice from the immature apples is also high in acid and astringent in taste. As the fruit matures improvement in the quality of the juice is observed. The improvement in the quality is due the conversion of starch into sugars (Moyer and Aitken, 1980) and also due to the increase in the pronounced odour in fruits ripened on the trees. Decrease in acidity, increase in soluble solids and increase in volatile composition with different harvest times (maturity) have been reported by Sapers _et al_.(1977) in McIntosh apples.

The improvement in the quality of apple juice could also be brought about by storage of immature apples. Bradley and Brown (1969) found that fruit from early and mid picking seasons were, definitely and slightly unsuitable for the juice processing , respectively. They also observed that apples develop satisfactory processing characteristics after a certain period of storage. These beneficial changes during storage could also be due to the starch breakdown and odour

Table 2: Important quality characteristics for the production of apple juice

Classification	Characteristics
Physical and Physiological	Maturity
	Decay
Compositional	Sugar:acid balance
	Aromatic content
	Phenolic content

Adapted from LaBelle (1981).

changes. Starch breakdown results in an increase in the sugar content of fruits. Sapers et al. (1977) also found a decrease in the acidity and increase in the volatile composition of McIntosh apples upon storage. In short, with the maturity and storage, the quality of juice is improved due to increase in sugar content (sweetness), decrease in acidity (sourness) and increase in volatile composition (aroma) of the apples.

On the other hand overmature apples are unsuitable for the production of juice due to low yield, poor flavour, difficulty of pressing and difficulty of clarifying and filtering. LaBelle (1981) reported that overripe apples are the principal contributors of a high level of suspended solids in juice. Though suspended solids are removed from the clarified juice, filtration efficiency and final juice yield are adversely affected by their high level.

In terms of the effect of maturity on the quality of the juice, apples must be ripe. However, the use of second grade and cull apples because of their low cost for the production of juice makes it difficult to follow the maturity guidelines.

2.1.2.1.2 Decay

The wholly or partially decayed fruits influence the flavour of the juice by introducing a "moldy" flavour. Decayed fruits can also hasten microbial spoilage of the juice and there is also an associated risk of "patulin" (a microbial toxin) with rotten apples (Moyer and Aitken, 1980). Apart from rots, other blemishes and defects do not have adverse effect on the quality of the juice. For example, Smock and Neubert (1950) reported a study that Cork, a physiological disease that seriously injures the quality of apples as fruit, results only in a smaller yield without contributing any off flavour.

These aesthetic considerations and possible effect on the quality of juice require careful sorting to remove decayed and badly damaged fruit. Sometimes, trimmimg is done to remove small decayed spots by machines such as high pressure water-spray rotatory washers. In short care must be taken to remove decayed fruit or decayed parts of the fruit before grinding and pressing.

2.1.2.2 Compositional Quality Characteristics

The differences in chemical composition of the apples plays an important role in the production of a good quality juice. As mentioned in Table 2, sugar:acid balance, aromatic and astringent constituents of the apples are important when manufacturing a good quality apple juice. The sugar:acid balance contributes the sweetness and sourness, the astringent taste is contributed by the tannins and the aroma by the volatile constituents of the apples. Smock and Neubert (1950) reported that a considerable variation may be found in acidity, sugars and tannins of varieties in any group and even among different lots of the same variety because of cultural, climatic and maturity variations to which fruit may be subjected.

Various studies on the effects of varietal, cultural, climatic and maturity variations on the chemical composition of the apples have been reported. The varietal variations in acidity, sugars and tannins among ten varieties of Virginia apples were published by Lopez et al. (1958). The effect of maturity and storage on acidity, soluble solids and volatile flavouring constituents of one variety have been reported by Bradley and Brown (1969) and Sapers et al. (1977). Moyer and Aitken(1980) reported that in eastern regions of North America there is a definite increase in the acidity of apples from south to north. In the south eastern state of Virginia, acidity tends to be low with a range of 0.25 to 0.45%, in Pennsylvania the acidity is usually medium with a range of 0.30 to 0.55%, while in Nova Scotia acidity tends to be high with a range of 0.45 to 0.85%, all calculated as malic acid on fresh weight basis. Also, as a rule apples grown in a sunny season are higher in sugar and acid than those grown in a cool, cloudy season (Moyer and Aitken,1980). These variations in the composition of apples influence the flavour of the finished juice.

The flavour of the apple is carried over into the cider and juice in terms of sugar:acid balance (LaBelle,1981). Similarly, aroma of the juice could also be expected to vary with the volatile composition of the apples. Therefore, flavour (taste and aroma) of the apples could reasonably be expected to be carried over into the juice. As mentioned in previous sections, juice is a by product, mostly prepared from the second grade and cull apples. Therefore, in practice quality characteristic guidelines are not strictly followed. To quote LaBelle (1981) "what is available economically is what is used for these products (juice and cider)". Apart from the cost of raw material another reason for the insufficient followup of the quality characteristic guidelines is the ease with which the blending of the juices from different varieties/lots could be performed to produce a juice of an acceptable uniform quality.

2.1.3 Blending

Moyer and Aitken (1980) reported that most juices from single varieties are not satisfactory and that a good general rule is to make juice from a combination of not less than three varieties that are at approximate "eating" maturity. Blending is done at the time of grinding, as blending the apples is easier than to keep the juice from separate varieties and blend those juices later. Moyer and Aitken (1980) classified apple varieties into the following 5 groups;

I) Acid to Sub acid
II) Sub acid to Mild
III) Aromatic
IV) Astringent, and
V) Neutral .

It is suggested that any of the varieties listed in group II and III makes a very good apple juice without the addition of any other variety but, such juice would be improved by the addition of about 5% of any of the varieties listed in group IV. Group III varieties provide an acceptable flavor and group V apples add astringency. Apples from group I are not suitable for apple juice, but make good juice when blended with 10 -20% of juice from group II or III or both to dilute the acidity. This mixture could be blended with some juice of group IV apples. Group V apples do not makea good juice but can be used for reducing acidity or astringency.

2.2 Apple Juice Preservation

Preservation is basically the prevention of quality deteriorating changes caused by the growth of spoilage microorganisms in the apple juice. Without any control apple juice could be spoiled by moulds, surface yeasts and lactic and butyric acid bacteria (Schelhorn, 1953). Unchecked growth of these microorganisms in apple juice can cause cloudiness of chemical and biological nature, changes in organic acid content, production of alcohols and also production of symbiotically and antibiotically active substances (Luthi, 1959). The methods used to preserve apple juice include heat application, chemical treatment, freezing, use of ultraviolet and X-rays, and sterile filtration.

The application of heat to destroy spoilage microorganisms is the most common method in commercial production of apple juice. Sterile filtration (Filtration Processing) is an effective, though not very common method of apple juice preservation. Marshall and Walkely (1951) reported that not only the filter plates were effective on the first passage of the juice, but that multiple passage reduced the mocroorganisms to practically zero. In using sterile filtration for the preservation of apple juice, microorganisms are removed from the juice by retaining them on a filter while allowing the sterile juice to pass. Two possible advantages of filtration preservation over heat preservation are low energy requirement and production of better flavored juice (Smock and Neubert, 1950) which could be attributed to the absence of heat treatment.

2.3 Filtration Process

2.3.1 Filtration, Filter Cake and Filtration Rate

Filtration is a process of separating a solid from the fluid by retaining the solid on a filter but allowing the fluid to pass. The layers of retained solid on a filter are called filter cake. During filtration, continuous deposition of solids, thickens the filter cake. The filter cake provides increasing resistance to the flow of fluid and reduces the filtration rate. The filtration rate can be expressed by:

$$\text{Filtration Rate} = \frac{\text{Driving Force}}{\text{Resistance}} \quad \text{....(1)}$$

The driving force is usually the total pressure drop throughout the filter area through which fluid is passing (Charm, 1963). Thickening of filter cake results in close packing of deposited particles during apple juice filtration because suspended particles in apple juice are easily deformable. Charm(1963) has reported such packing during the filtration of fruit juices. With continuous filter cake buildup resistance to the flow increases and the filtration rate decreases. The effect of resistance could be compensated by increasing the pressure. However, due to the presence of compressible particles it is not possible to do so in apple juice processing without causing damage to the filters.

Therefore, optimum filtration rate could be better maintained by decreasing the resistance, for example by slowing the rate of filter cake buildup on the filter. The filter cake buildup could be decreased by keeping the amount of retained solids on the filter to minimum possible levels. The amount of solids retained on the filter is analogus to the Non Filtrable Residue of a water sample.

2.3.2 Non Filtrable Residue on the Filters

Non Filtrable Residue (NFR) of a water sample is the weight of the suspended matter retained on a filter of arbitrary size (McGirr, 1975). For a given sample NFR increases with a decrease in filter pore size. NFR is also dependent upon the size distribution of suspended matter in a sample. For example, a sample containing a large proportion of smaller suspended matter of size 0.45 - 0.25 μm would give higher NFR on filters of pore size smaller than 0.45 - 0.25 μm than on the filters of larger pore size.

The retained suspended matter provides resistance to the flow. Therefore, an indirect estimation of the resistance can be obtained by the amount of suspended matter retained on a filter or by NFR. To estimate NFR during apple juice filtration an approach similar to the water analysis could be used. It is a common method in water analysis to establish a correlation between turbidity and NFR of a water body and then use turbidity to control water filtration and water quality (McGirr, 1975).

Though correlation between turbidity and NFR could be used during apple juice filtration, determination of NFR by weighing the retained suspended matter is very difficult on a column type filter such as that used in the present study. To overcome this problem a correlation between turbidity and the number of suspended particles could be used.

2.3.3 Correlation between Turbidity and the Number of Suspended Particles in Apple Juice

2.3.3.1 Turbidity

All the systems through which light can pass show two well known optical effects; namely absorbance and scattering of the incident light. The total effect for a system can be expressed by the following composite relationship (Heimenz, 1977):

$$\frac{I}{I_o} = \exp\left[-(a + t)\,l\right] \qquad \ldots\ldots(2)$$

Where;
I = intensity of transmitted light
Io = intensity of incident light
a = absorbance
t = turbidity
l = light path

This composite relationship also defines the optical property of a sample which causes light to be scattered and absorbed rather than transmitted in a straight lines through the sample, and it is called Turbidity (A.P.H.A., 1975)

2.3.3.2 Measurement of Turbidity

Turbidity is measured by photoelectric instruments in terms of the intensity of light. A photoelectric instrument used for measuring turbidity, known as nephelometer measures the intensity of scattered light at a 90 degree angle to the plane of propagation of the incident light.

Turbidity measured by the nephelometer does not include absorbance effect of the incident light, and dissolved color is not registered as turbidity (Hach, 1968). This characteristic of the nephelometer makes it suitable for measuring the turbidity of apple juice samples. The turbidity measurement by the nephelometer is affected by all the parameters which affect scattering of the light. These parameters are the size, shape and refractive index of suspended particles, wavelength of exciting light and angle of observation (Black and Hannah, 1965). In the presence of many influencing factors experiments to establish a correlation between the number of suspended particles and the turbidity would require controlled instrumentation and some theoretical assumptions.

2.3.3.3 Use of the Number of Suspended Particles in Correlation Experiments

All the variables which affect the turbidity of a sample could not be controlled by instrumentation. The variations in size, shape and number of suspended particles cause the variation in the turbidity of the samples. The relationship among amount, size, shape and the number can be given by:

$$W = \sum w_i n_i \qquad .... (3)$$

Where;
W = total wt. of suspended particles
w_i = weight of individual particle
n_i = number of particles weighing w_i

$$w_i = \text{Volume x Density} \qquad ..... (4)$$

:

Volume of the particle is based on its geometrical dimensions or its size and shape. The amount of suspended particles, as mentioned before, is difficult to measure by weighing and mathematical calculation of the amount from above formulas requires that size, shape and density be known. However, a simpler method could also be used where affects of size and shape on turbidity are considered negligible without any explanation. Now, equation 3 could be written as:

$$W = N\ c \qquad .... (5)$$

Where;
c = density constant
N = total number of suspended particles

or

$$W \propto N \qquad .... (6)$$

Therefore, correlation between the number of suspended particles and turbidity could serve the same purpose as the correlation between the turbidity and amount.

2.3.4 Characterization of Filtration Processing of Apple Juice

Filtration processing involves removal of microorganisms by retaining them on a filter while juice is allowed to pass through the filter. During filtration suspended particles (other than microorganisms) are also removed from the juice. The effects of removal of suspended particles from the juice are important from the regulatory and sensory point of view.

According to the Deparment of National Health and Welfare apple juice:

- " shall have a specific gravity of not less than 1.041 and not more than 1.065 (20°C/20°C); and
- shall contain, in 100 ml measured at a temperature of 20°C, not less than 0.24g and not more than 0.60 gram of ash of which not less than 50 percent shall be potassium carbonate", (Anonymous, 1981 c). Therefore, monitoring of changes in the ash content and specific gravity of juice upon filtration is important to know whether or not filtration processing could make apple juice unacceptable even without any adulteration.

From the sensory point of view, the changes in sugar, acid and tannin content of juice upon filtration and changes in the clarity of apple juice are important. As early as 1932, sugar and tannins were recognized as the constituents of suspended particles of apple juice (Smock and Neubert, 1950). The changes in the tannin content upon removal of suspended particles have been reported by Johnson et al. (1968) and Joslyn et al. (1952). The investigations into sugar and acid changes are however scarce. The changes in the viscosity and appearance of apple juice upon filtration have been reported by Neubert (1943). Changes in the viscosity causes the change in the "body" of apple juice, whose effect on the quality of apple juice has not been studied. The effect on the appearance or clarity of apple juice upon its quality, has also not been previously studied. However the effect of clarity on apple juice quality may be important because in the North American market mainly clear juice is sold.

The major removal of suspended particles from apple juice takes place during clarification and no marked additional losses of the chemical components of the juice upon filtration during heat processing have been reported. However, filtration processing involves more extensive filtration of apple juice than the filtration during conventional processing. Therefore, the effect of this filtration process on the properties of apple juice needs to be investigated.

2.4 Psychophysical Analysis of Apple Juice

2.4.1 General

Psychophysics is the science which relates individual sensory experience (taste, smell, visual percept, etc.) to antecedent physical and/or chemical properties which can be instrumrntally measured (Moskowitz, 1977 a). In psychophysical analysis sensory, chemical and physical evaluation of the food is performed. Sensory analysis provides a measure of sensory responses and their relation with consumer acceptance. The interest to translate sensory responses into objective information, such as chemical and physical properties of a product is steadily increasing because objective information could be used more routinely than the sensory tests to control and improve the acceptability of a product.

An objective three directional approach involving preference rating, objective sensory assessment and analytical data to measure the flavor quality of apples, apple juice and apple cider was discussed by Williams et al. (1977). The three directional approach also detailed by Moskowitz (1977 a) has been used by Poll (1981) in determining the suitability of 18 apple varieties for the production of apple juice.

In the three directional approach, preference rating is performed by a trained panel to obtain the overall quality rating of the juice. The trained panel also provides qualitative and quantitative measurement of the various sensory characteristics which contribute to the overall quality of the juice. The individual sensory score and overall quality rating are examined statistically by correlation analysis to measure the relative dependence of the overall quality on various sensory characteristics. The chemical analysis of the juice is also performed. The statistical examination of the chemical composition of various juice samples with the scores of individual sensory characteristics indicates the chemical components of the juice which are important for sensory characteristics and in turn influence the overall flavor quality of the juice.

Utilizing a three directional approach, Poll (1981) evaluated 18 apple varieties for their suitability for the production of apple juice. In short the results of that study were:

- Overall Aroma (Y)/ Fruit Aroma (X), r = 0.93 **,
- Overall Aroma (Y)/ Off Aroma (X), r = -0.84 **,
- Overall Taste (Y)/ Fruit Aroma (X), r = 0.92**,
- Overall Taste (Y)/ Sweetness-Soueness, r = -0.54*,
- Overall Taste (Y)/ Bitterness-Astringency, r = - 0.72*,
- Overall Taste (Y)/ Off taste (X), r = - 0.71 **.

In the same study, the relationship between perceived sweetness-sourness and measured sugar:acid balance was found to be significant at the 99.9 % level. The samples were served at room temperature. Since, apple juice is normally served chilled and Harold (1981) reported that fructose (the major fruit sugar) exhibited diminishing sweetness with increasing temperature, it is advisable to serve juice samples at refrigerated temperatures during sensory analysis.

2.4.2 Measurement Statistics

As mentioned in the last section, two types of measurements are performed in a three directional approach. The measurement of sensory characteristics and the measurement of chemical and/or physical characteristics, are the subjective and objective measurements, respectively. Measurement in the broadest sense, is defined as the assignment of numbers to objects and events according to rules (Stevens, 1946). Moskowitz (1977 a) discussed the use of various scales in sensory evaluation. These (nominal, ordinal, interval and ratio) scales reported by Stevens (1946) follow certain rules, have different mathematical properties and have different sets of statistical operations applicable to the measurement made by each type of scale.

Of all the scales, ratio scaling provides the most powerful measurement, whether physical or psychological because it can determine all four relations : equality, rank-order, equality of intervals and equality of ratios (Stevens,1946). The objective measurements e.g. sugar content, tannin content etc. are made by well known analytical methods using fundamental and derived ratio scaling. Ratio scaling in sensory evaluation is aimed at assigning numbers to stimuli so that they reflect the ratios among the stimuli for the specified property. The numbers in ratio scaling are assigned by two methods, fractionation and magnitude estimation. Magnitude estimation has been extensively used in sensory evaluation because it eliminates many of the biases which occur in sensory evaluation of food and its measurements can be analysed by virtually all statistical techniques (Moskowitz, 1977 a).

2.4.3 Functional Relations

Functional relations (equations) equate numerical sensory measurements to the physical intensity of the stimulus. The ratio scaling (magnitude estimation) gives an equation known as the Power Function which can be expressed as:

$$\log S = \log K + n \log I \quad .... \quad (7)$$

or,

$$S = K\ I^{n} \quad .... \quad (8)$$

Where;

S - sensory measurement
I - Instrumental measurement
n - slope
K - intercept

Functional relations have two purposes, instruction and application (Moskowitz, 1977 a). Functional relations provide information about the dose - response relation between known physical intensity (sugar concentration, acid concentration etc.) and perceived sensory magnitude. A small or large change in stimulus (ingredient) may be perceptually large or small. For example, an exponent of 1 indicates that a ten-fold increase in the concentration of a stimuli would produce a $(10)^1$ = 10-fold increase in the perceived sensory response. The perceptual change can only be obtained from a functional relation between stimulus and response. This information then can be applied for modifying a product with consideration to cost, labor and expected sensory change for each proposed alteration (Moskowitz, 1977 a).

Normally, in sensory evaluation correlational analysis of the rating as compared to known variation in the individual sensory characteristics or physical characteristics (such as performed by Poll, 1981) to test the significance of a relation is used. Correlational analysis and T - test or analysis of variance to determine the significance of difference between two or more products, are not as powerful as functional relations. Methods other than functional relations do not provide the estimate of the direction and extent of change in perception upon change in stimuli. Therefore, they do not provide information about modification (direction and extent) in ingredients required to prepare a desired product (Moskowitz, 1977 a).

2.4.4 Statistical Analysis

In power function analysis, the panelists are instructed to give a numerical estimate of the perceived subjective impression relative to a standard (or reference). These numerical estimates are then used to derive the power function, and also for analysis of variance for determining panelist performance, differences among samples etc. The handling of raw data for the determination of power function (Malcolmson, 1979) is simple and straight forward. However, a limitation arises when a panelist (s) does not perceive a sensory impression for a given stimuli. This limitation is expressed as the "np" (not perceived) or the zero rating in the literature.

One reason for the limitation of np value is associated with the conventional method of calculating a power function. The method is to standardize raw data by dividing each panelists estimate by the geometric mean of his/her estimates. This standardized data is then analysed to determine linear regression of log values of stimuli and sensory estimates, which yield power function or the slope of the line. The use of geometric mean does not permit a value of $\leqslant 0$ for np. Moskowitz (1977, b) reported that for 200 or more observations, the arithmetic mean can be used to overcome this limitation. For the studies that were smaller in scale, many workers assigned a positive value to np to overcome this limitation.

However, methods of assigning a positive value to np vary significantly. Malcolmson (1978) obtained a number from each panelist which they felt was close to their zero value and substituted it for np. Donaldson (1978) did not assign any value to np for the lowest concentration, and when np occured at middle or higher concentrations, they were replaced with a calculated value if the panelist had scored at that particular concentration in other replications. The value for np was calculated by the regression analysis of raw data. Fabro (1979) assigned eighty percent of minimum value given by a panelist to the np reported by the same panelist for a particular sensory response. The level "eighty percent" was established on the basis that a substitution of eighty percent of minimum value for np did not result in the elevation of normalized data by more than ten percent.

Statistically, approaches used by Fabro (1979) and Donaldson (1978) are better, since they use the estimate provided by the panelist at other levels of stimuli to reach a value for np. In other words, the panelist has already given estimate of his/her perception of a particular stimuli and that information is used to derive a value for np at a level of stimuli where he/she does not perceive the stimuli. The np value obtained by the method of Malcolmson (1978) appears to be dependent upon the way by which a panelist is asked to give a value of np. While explaining value assignment to a panelist, if a value (e.g. 0.01) is mentioned, that could probably become the level around which the value may be given. The use of regression analysis proposed by Donaldson (1978) is disadvantageous because it could derive a negative value, which can not be utilized in the conventional method. Regression analysis of raw data to extrapolate np value is also not justifiable, because raw data from a ratio scale first requires log-normalization. The level of eighty percent of the minimum value is set by the arbitrary level of percent elevation of normalized data. Therefore, present methods of np treatment are not in agreement and these are suitable only for the investigations where method does not yield a negative value.

2.4.5 Quantitative Integration

Apple juice contains approximately 85% water, 10-12% carbohydrates, 1% pectin, 0.5% organic acids, 0.5% of various components such as potassium, amino acids, phenolics, and small amount of volatile flavoring compounds (Ryan, 1972). These compounds determine sweetness (carbohydrates), sourness (organic acids), bitterness-astringency (phenolic) and aroma (volatile flavoring compounds) of the juice. These perceived sensory properties eventually become subjective basis for overall aroma and overall taste of apple juice (Poll, 1981). The integration (or the combination) of aroma and taste, yields the sensation which is called flavor (Power, 1974), on which the overall acceptibility or preference for apple juice partially depends. Overall acceptability is an integrated response (Moskowitz, 1977 a). The prediction of an integrated response could be performed by the multiple linear regression and multiple discriminant analysis (Moskowitz, 1977 a).

In multiple linear regression, a mathematical equation or statement; $Y = B_o + B_1 X_1 + B_2 X_2 \ldots + B_n X_n$ is obtained. Where, Y is the dependent variable (acceptance rating), B_o is the Y intercept, B_1, B_2, through to B_n are regression coefficients for the independent variable X_1, X_2 through to X_n. According to Korth (1982) the model can be specified by selecting predictors (or independent variables). However, Korth (1982) cautioned that selecting predictors should be based on a sound understanding of the phenomenon being studied, and also cautioned against statistical selection of predictors. The statistical selection of a predictor is done by specifying a level of significance (alpha 0.05 or 0.10 etc.) for the inclusion or omission of a variable from the model. This is considered to be a disadvantage of multiple linear regression that some variables though important for sensory perception do not get included in regression equations (Moskowitz, 1977 a).

The prediction of an integrated response by multiple regression analysis requires evaluation of integrated response, and the different independent variables. The required measurements are obtained using various sensory and analytical methods. To understand the interactions and relationships among the sensory and chemical properties, bivariate (x - y) regression is commonly performed (Poll, 1981, Moskowitz, 1977 a) and according to Korth (1982) the working of the model can be understood mostly by observing positive or negative effects of the independent variable on the dependent variable in a graphical presentation of the data.

3. MATERIALS AND METHODS

3.1 Compositional Analysis of Apples

3.1.1. Sample

Approximately 10 kg lots of one crabapple and nine apple cultivars were received from Agriculture Canada Research Station, Morden, Manitoba. These cultivars were hand harvested at their proper maturity and stored at 90 - 95% relative humidity and 1 - 3°C temperature. Harvesting dates and lot particulars are listed in Appendix 1. After lots were received at the Department of Food Science, University of Manitoba, Winnipeg, Manitoba, they were stored at 85 - 90% relative humidity and 1 - 3°C. Except for the analysis of polyphenol content all other determinations were performed within a period of two weeks after the lots were received at the Department of Food Science, University of Manitoba. Unless otherwise mentioned all determinations were performed on four (app. 500g) samples. Four samples were also stored at -36°C on every date of analysis for the determination of total polyphenol content at a later date.

3.1.2 Sample Preparation

All samples were washed in running cold tap water to remove surface deposits and dried by blotting with paper towels. Preliminary sample preparation was done using an Oster blender. Final sample preparation from blended samples was performed according to the A.O.A.C. method, 22.008 (1975). Samples frozen for the determination of total polyphenol content were finely grated by Hobart Mixer, equipped with grater plate in a 9" vegetable slicer. Grating was done in a cold room at 4°C to avoid the browning of samples. Clear grated sample portions were uniformly mixed and stored at -36°C in clear glass bottles.

3.1.3 Analytical Methods and Statistical Analysis

Immediately after preliminary preparation four 20 - 25g samples were weighed in aluminum dishes and frozen at -36°C for the determination of moisture content. Moisture content was determined by freeze drying in a Virtis freeze drier (model no. 10-146 MP-BA, The Virtis Company Inc., Gardiner, N.Y. 12525), without a condenser, at a pressure less than 100 and with the shelf at room temperature. Total soluble solids (T.S.S.) were determined according to the A.O.A.C. method, 22.024 (1975). T.S.S. values were then converted to total sugar content, expressed as percent sucrose, using the Schobinger and Muller Table (Poll, 1981). Total acid content, expressed as percent malic acid, was determined by the A.O.A.C. method, 22.061 (1975). T.S.S. and total acid content determinations were performed on the solution obtained after the final sample preparation.

Total polyphenol content, expressed as percent tannic acid, was determined by the A.O.A.C. methods, 9.098, 9.09 and 9.100 (1975). The determination was made on alcohol extract prepared by blending 10g of grated frozen sample in 60 ml of 75% ethanol for five minutes. Ethanol extractions (at 60, 75 and 90% ethanol) for three and five minutes were conducted for the selection of best extraction procedure. Since 75% alcohol and five minute extraction time gave significantly higher results of total polyphenol content (Table 3 and Appendix 2), all sample determination were conducted by this extraction procedure.

A total of four replications was completed for every sample. Therefore, for every analysis the total number of observations were 16. The means and standard deviations for each chemical component were calculated. Analysis of variance and Tukey's test (HSD) were conducted to test the significance of differences among means of cultivars, samples and replicates.

3.2 Chromatographic Analysis of Sugars and Acids in Apples and Apple Juices

3.2.1 Samples and Sample Preparation

A 15 - 20 ml portion of the sample solutions (used to determine T.S.S. and total acid content, section 3.1.2) were frozen at -36°C in clear glass bottles to be used later for the determination of sugars and acids by High Performance Liquid Chromatography (HPLC). Apple juice samples used in sensory analysis were also frozen at -36°C. Final sample preparation was done by filtering thawed samples through 0.45 μm filter and cleaning it according to the method of Waters Associate (Anonymous, 1981 a).

3.2.2 Apparatus

The Organic Acid column (Aminex Ion Exclusion HPX - 87H) and the Carbohydrate column (Aminex HPX - 87P Heavy Metal), were

Table 3: Statistical analysis of phenolic extraction procedures

Duncan's multiple range test for variables[1]

Alcohol (%)	Total Phenolics mg/100g	Time of Extraction (min)	Total Phenolics mg/100g
60	167.90 a[2]	3	152.71 a
75	174.37 a	5	172.61 b
90	145.71 b		

[1] - Alpha =0.05, Df =12 and MSE = 104.65.
[2] - Means with the same letter are not significantly different.

purchased from Bio - Rad Laboratories, 32nd & Griffin Ave., Richmond , Calif., 90804). Columns were placed in a temperature control oven and attached to a Water Associates (Water Associates Inc., Maple St., Milford, MA 01757). HPLC assembly. The HPLC assembly consisted of a solvent delivery system (model 6000A), sample injector (model U6K), an ultraviolet absorbance detecter (model 440) with extended wavelength (214 nm) module and a differential refractometer (model R401).

3.2.3 Eluent Preparation

Water and 0.25N sulphuric acid with 10% acetonitrile were used as eluent for sugar and acid analysis, respectively. Eluents were filtered through 0.45 µm filter and degassed under vaccum. Water held in reservoir for sugar analysis was kept degassed by continuous stirring at 70 - 80°C.

3.2.4 Operating Conditions

Sugar analysis was performed at a flow rate of 0.70 ml/min and at 85°C. Acid analysis was performed at a flow rate of 0.50 ml/min and at 60°C. Recording and analysis of detector responses were conducted on Vista 401 (Varian Instrument Group, Walnut Creek Division, 2700 Mitchell Dr., Walnut Creek, Ca. 94598). Attenuation of the refractive Index detecter was set at 16X and sensitivity of the Ultra Violet detector at 0.20 aufs.

3.2.5 Expression of Results and Statistical Analysis

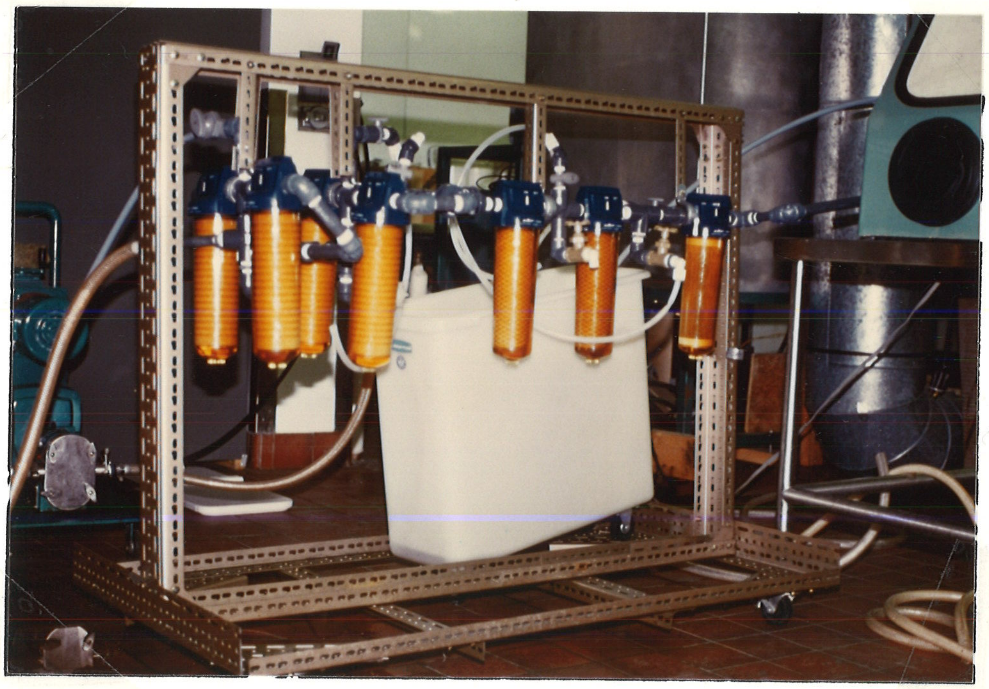
Since no confirmation of the presence of acids was conducted all acid concentrations were expressed as % malic acid. If at a particular retention time more than one acid was eluted, the peak was identified as the most probable acid. A total of two replications was completed for every sample to obtain a total of eight obsrevations per variety. Mean and standard deviation were calculated for every deterimination. Analysis of variance and regression analyis on total sugar and acid content values obtained from chemical analysis and from HPLC were conducted to measure the relation and the difference in values obtained by these methods.

3.3 Filtration Process

3.3.1 Reagent and Apparatus

Pectinase enzyme was used for juice clarification. Pectinase (Clarex P - 150, lot no. 7725 SLA R1080) was purchased from Miles Laboratory Inc., Toronto, M9W 1G6. Bosch Centrifugal Juicer (Type no. 0712 500 007) was used for laboratory scale juice extraction. A multispeed Tri--clover Rotary pump was used to circulate apple juice through filtration assembly. AP100T filtration housings and 1 μm filter (D CCPY nominal rating, CUNO MICRO-WYND II) were purchased from AMF CUNO Divison, Meriden, Conn.. Twenty five, 5 and 0.20 μm filters were purchased from Culligan International Company, Northbrook, Illinois. Filtration housings were assembled with P.V.C. Schedule 80 pipes on a mobile metal frame to construct a filration assembly (Fig.1). A nephelometer (DRT-100, HF Instrument Ltd., 105 Healey Rd., Bolton , Ontario) was used to

FIG. 1. FILTRATION ASSEMBLY FOR THE PRODUCTION OF APPLE JUICE



measure the turbidity.

3.3.2 Filtration Assembly

Pilot plant scale studies were conducted on a filtration assembly (Fig. 1). A flow diagram of the filtration assembly is illustrated in Fig. 2. The filtration assembly consisted of a coarse filtration unit and a fine filtration unit. The coarse filtration unit was equipped with the filters of larger pore sizes ranging from 1 μm to 25 μm and the fine filtration unit with a 0.20 μm filter. In principle, the coarse filtration unit removed suspended particles from enzyme clarified apple juice to prepare clear juice. Clear juice was then filtered through the 0.20 μm filter to prepare sterile juice. Sterile juice was then bottled aseptically in an ultraviolet chamber. In between every filter a bypass/sampling valve was provided.

3.3.3 Juice Preparation

One crabapple cultivar (Kerr), two apple cultivars (Collet and Goodland) from Agriculture Canada Research Station, Morden, Manitoba, and two apple varieties (McIntosh, and Red Delicious) from the local market were used for studies on the filtration process. For pilot plant scale juice preparation, juice was extracted by a hand operated Fruit press. This Fruit press was lined with a nylon net to filter debris and larger particles from extracted juice. Before pressing, apples were thoroughly washed with cold water spray and crushed into small pieces by a crusher. For laboratory scale juice extraction,

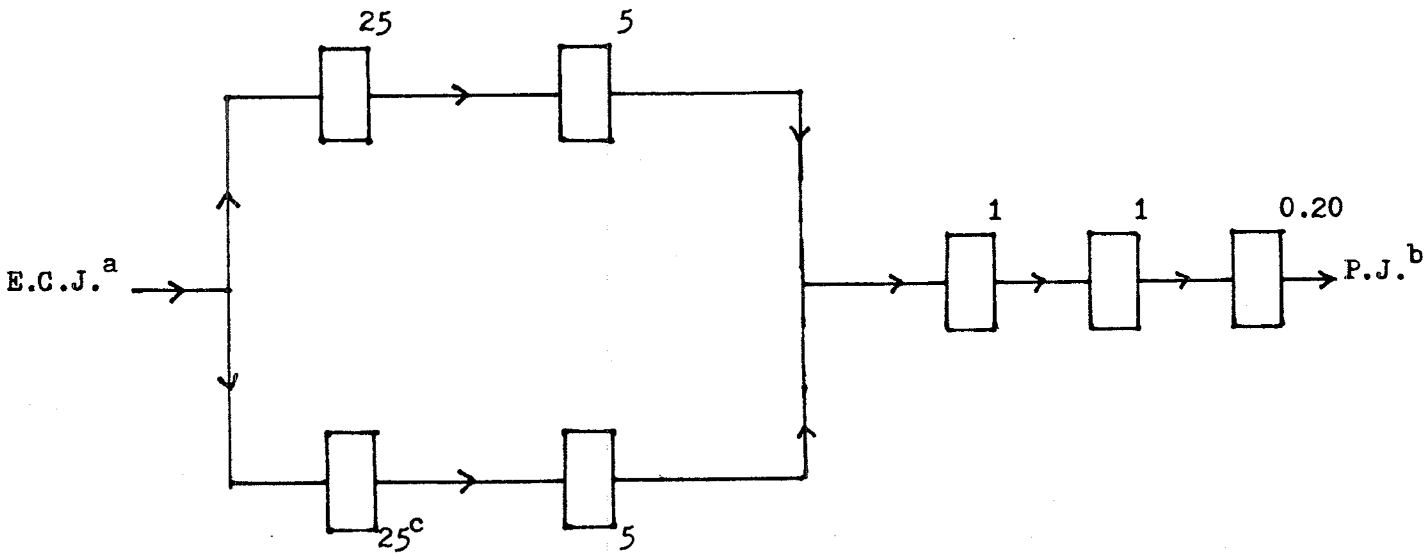


Fig. 2. Flow diagram of the filtration assembly.

a - Enzyme clarified juice.

b - Processed juice.

c - Numbers represent pore size (μm) of filters.

sliced apples were fed into the centrifugal juicer. After extraction juice was clarified overnight at room temperature with a sufficient amount of Pectinase.

3.3.4 Samples, Analytical Methods and Statistical Analysis

For process characterization experiments, approximately 500 ml samples were collected in vacuum cap, clear glass bottles, during processing of apple juice and immediately analysed or frozen at - 36°C for the analysis at a later convenient time. Total sugar, acid and polyphenol contents were determined by the methods described in section 3.1.3. The specific gravity of apple juice was determined by the table conversion (A.O.A.C. Table 52.008, 1975) from the total soluble solids. To establish correlation between number of suspended particles in apple juice and its turbidity an estimate of the number of suspended particles was calculated by a formula based on dilutions (Table 4). Experimental controls of variables affecting turbidity are listed in Table 5. A stepwise method of turbidity correlation experiments is given in Appendix 3. After establishing correlation between number of suspended particles and turbidity of apple juice, turbidity measurements were used to monitor recycling, filtration process and to calculate approximate size distribution of suspended particles in apple juice.

Data obtained from turbidity experiments were analysed by regression. Results were expressed as r (correlation coeffecient) and r^2 (coefficient of determination) at alpha 0.05. Turbidity measurements used for recycling and monitor-

Table 4: Relative numbers of particles in a four times diluted sample

Dilution Series	Actual Number	Relative Number
Undiluted sample (US)	X	16
1st dilution (1D) (US : d[1] = 1:1)	X/2	8
2nd dilution (2D) (1D : d = 1:1)	X/4	4
3rd dilution (3D) (2D : d = 1:1)	X/8	2
4th dilution[2] (4D) (3D : d = 1:1)	X/16	1

[1] - d or diluent.
[2] - Number of particles in undiluted sample in relation to any diluted sample can be calculated from formula:

$$\text{Relative number of particles in undiluted sample} = 2^n$$

where:
n = total number of dilutions

Table 5: Control of variables affecting turbidity in correlation experiments

Variables	Control/Assumption
Size of the particles	Effect assumed to be negligible
Shape of the particles	Effect assumed to be negligible
R.I.[1] of the particles	Effect assumed to be negligible
Wavelength of light	Use of same light source for for all samples
Angle of observation	Use of nephelometer
Absorbance effect	Use of apple juice as diluent
Number of particles	Not controlled

[1] - Refractive index.

ing and for size distribution calculations were expressed as percent reduction in turbidity and also as means. Process characterization data was analysed for variance. Significance of mean differences was analysed by Tukey's test (HSD).

3.4 Psychophysical Analysis of Apple Juices

3.4.1 Samples

Based on their availability in local super markets five brands (Best Pack, Dominion, Sun Rype, Scotion Gold and Bright's) of apple juices were selected for sensory evaluation. Sufficient quantities from the same lot were purchased and stored at 3 - 5°C for a period of one month. Four juices were sold in Cans and 1 juice was sold in tetrapack. Three juices were single strength and two juices were made from concentrate. Code letters A (Scotion Gold), B (Sun Rype), C (Bright's), D (Dominion) and E (Best Pack) were assigned to apple juices for the presentation of results. Two natural apple flavors (nos. 80294 and 86650) and one artificial apple flavor (no. 86662) samples were received from Fries and Fries (Cincinnati, Ohio 45216) and were used in selecting apple aroma reference. The code numbers 1 (no. 80294), 2 (no. 86650) and 3 (no. 86662) were assigned to flavor samples, also for the presentation of results.

3.4.2 Panel Selection, Training and Stimuli Selection

In an introductory panel eight male and eight female panelists were introduced to the magnitude estimation technique by a card game, similar to the method described by Moskowitz (1977,

b) The ballot and card diagrams used in the introductory panel are presented in Appendixes 4 and 5, respectively. A survey of the results indicated appreciable understanding of magnitude estimation by panelists. However, no statistical analysis of panelist performance was conducted. All the panelists were either staff members or students of the Department of Food Science, University of Manitoba. The work load and schedule of sixteen panelists were consulted and based on their availability during the period of sensory evaluation nine panelists, three females and six males, participated in subsequent sensory evaluation sessions.

Panelists were further oriented with the magnitude estimation procedure and sensory stimuli during four orientation panels. Orientation panels were arranged to familiarize panelists with the magnitude estimation technique, and to select stimuli standards. Sweetness, sourness, bitter - astringent taste (BAT) and aroma of standard solutions (Appendix 6) were evaluated during orientation panels. After each session panelists were consulted about acceptable levels (neither too intense nor too weak) of standard stimuli. Since no preestablished standard apple aroma solution was available a question for comparing apple juice aroma and standard (artificial apple flavor) solution was included in the ballot (Appendix 7). The result of aroma evaluation indicated a need for better aroma reference. To select the most suitable aroma reference one artificial and two natural apple flavor solutions were evaluated on a five point hedonic scale (Appendix

8). Based on the Results (Tables 6 and Appendix 9) a natural apple flavor was selected as aroma standard.

After selecting the aroma reference, modifications in sweetness and sourness power function determination were attempted. These modifications involved use of mixed sugar and acid solutions to derive sweetness and sourness power functions. The modifications were attempted, in view of significant correlation between the sugar:acid balance and sweetness - sourness of apple juice (Poll, 1981) and in view of the dependence of sweetness of sugars upon acidic ingredients of a system (Harold, 1981). In short, sweetness and sourness power function evaluation included use of sugar:acid solutions with well defined sugar:acid balance (Table 7). The power functions derived from these evaluations were then expressed in terms of sugar:acid balance. During orientation panels sucrose and citric acid were used as the sugar and acid standards (Appendix 10). However, in the final evaluation fructose and malic acid, the most common apple sugar and acid, were used as the sugar and acid reference.

Once procedures and standards for the aroma, sweetness and sourness were finalized, the fourth important sensory property of apple juice, i.e. stimulus of phenolics, was considered. Phenolics in general contribute to the bitterness and astringency of the apple juice. Bitterness was found to be negatively correlated with overall taste of apple juice and was attributed to the phenolics in apple juice (Poll, 1981). Phenolics also contribute to the astringency of apple

Table 6: Statistical analysis of aroma selection data

Tukeys test for mean sensory score[1]

Apple flavor	Mean[2]
Flavor (code no. 1)	3.83 a
Flavor (code no. 2)	2.11 b
Flavor (code no. 3)	1.89 b

[1] - Alpha = 0.05, Df = 42.
[2] - Means with the same letter are not significantly different.

Table 7: Concentrations of taste stimuli used in the determination of sweetness and sourness power functions

Sugar:Acid Balance	Adjusted by Sugar (% Fructose)[1]	Adjusted by Acid (% Malic acid)[2]
5.00	1.50	1.20
10.00	3.00	0.60
20.00[3]	6.00	0.30
30.00	9.00	0.20
40.00	12.00	0.15

[1] - Acid concentration fixed at 0.30% .
[2] - Sugar concentration fixed at 6.00% .
[3] - Used as reference.

juice, which have been negatively linked with the quality of apple juice (Moyer and Aitken, 1980). Tannic acid, a common phenolic standard, was used to derive bitterness and astringency power functions. Attempts to use tannic acid for the separate evaluation of bitterness and astringency were unsuccessful and a common term bitter - astringent taste (BAT), was agreed upon to represent the stimulus perceived from tannic acid solution. Concentrations of stimuli used in the final evaluations of sweetness and sourness, and BAT and apple aroma are listed in Tables 7 and 8 respectively.

3.4.3 Sample Presentation

Samples were evaluated in individual sensory booths. Red light was used to mask possible color differences in the juice, which might influence a panelist's judgement. Panels were held twice a day with at least two hours interval between each session. During normal working hours panelists were allowed to choose their own convenient times. However, care was taken to avoid sessions immediately after coffee or lunch breaks. A total of 24 panels were spread over 38 days in three groups of one week each. These 24 panels included 10 panels (5 apple juices x 2 replicates) for the apple juice, two panels for the sweetness and sourness, two panels for apple aroma and 10 panels for the BAT evaluation. For the evaluation of BAT only one concentration level was presented in a panel. Therefore, for four concentrations levels and a reference evaluation a total of five panels was required. Thus, including five replication panels a total of ten panels

Table 8: Concentrations of stimuli used in the determination of bitter-astringent taste and aroma power functions

Tannic acid (%)[1]	Natural apple flavor (%)[2]
0.045	0.30
0.090	0.60
0.120[3]	1.20[3]
0.180	2.40
	3.60

[1] - Weight by volume in dustilled water.
[2] - Volume by volume in distilled water.
[3] - Used as reference.

was required for the evaluation of BAT of standard tannic acid solutions. This arrangement for BAT evaluation was made to compensate the "palate block effect" (section 4.6.3) which makes unprejudiced assessment of subsequent samples of astringent compounds difficult (Lea and Arnold, 1978). To compensate the effects of time intervals on the performance of panelists, replications of power function evaluations were spread among panels for the apple juice evaluation. A total of two replications was completed for all the evaluations.

All samples were prepared the night before the panel session. Samples were kept and served at 3 - 5°C. The standard solution samples, 10 - 15 ml, and references for taste evaluations were served in white plastic containers covered with aluminum foil. Apple juice samples, 30 ml, for taste evaluation were served in 45 ml pyrex glasses. Apple juice and standard samples, 10 ml, for aroma evaluations and aroma reference were served in 25 ml clear glass vial with screw cap. All the samples were coded with three digit random numbers, and order of serving was randomized for each judge. A concise list of factors expected to influence panelist judgement and their control is presented in Appendix 11. This list of factors presented as errors is from Larmond (1977). All the required precautions were specified in the ballots (Appendixes 12, 13 and 14). For the evaluation of sensory properties of apple juice and their relationship with the preference a combined ballot for magnitude estimation and a 9 - point hedonic scaling was prepared (Appendix 15).

3.4.4 Normalization, Variance and Power Function Analysis

A procedure for standardization of the data as described by Malcolmson (1979) was applied to magnitude estimates assigned by panlists. Standardized scores were log - normalized and the data analysed by the analysis of variance. Tukey's test (HSD) was conducted to determine significance of differences among means. Using the scores obtained for the standard solutions a power function for each of the four stimuli was derived. Each apple juice was placed on the power function using the method outlined by Malcolmson and McDaniel (1980). Analysis of variance and Tukey's test were also conducted for the preference scores.

3.4.5 Treatment of Not Perceived Responses

As mentioned in section 2.4.4 there are various methods of treating "np" values or the not perceived responses in magnitude estimation data. Depending upon the method of assigning value to "np", slope or the power function has been found to vary, sometimes significantly (Donaldson, 1978). Donaldson (1978) also mentioned that no treatment was provided for np values at lowest concentration.

An approach of no np treatment, at any stimulus level was used to derive power function. In short this method involved calculating geometric mean scores for stimulus levels, without any attempt to balance the data by assigning a positive value to np responses. The log of the geometric mean of the sensory scores was then analyzed with the log of stimulus concentrations to derive a power function. However, if at a given stimulus level more than 50% observations were np, that simulus level was omitted from further analysis.

3.4.6 Regression Analysis

Regression analysis on mean preference scores, mean log- normalized intensity scores for each sensory characteristics and stimuli levels (chemical content concentrations) was conducted to correlate preference with sensory characteristics and chemical contents and also to correlate sensory characteristics with chemical contents. Trant et al (1981) reported that only intensity could be logically related to objective measurements by linear correlation. Therefore, regression models for best fit included linear and curvilinear relationships. The best fit regression model was chosen with the help of a x-y plotting computer program (Anonymous, 1982). The selection of best fit model was based on the coefficient of determination. A regression model which could explain at least 75% variability of "Y" was selected as a best fit model. The 75% level was selected because of its proximity to the correlation coefficient significant at alpha 0.05, for the number of observations in present investigation (For five observations, at df 3, significant correlation coeffecient at alpha 0.05 is 0.878 or the coefficient of determination is 77%). This restriction was relaxed in case of unreasonable relationships (section 4.6.2)

On the basis of the bivariate (x - y) regression (preference vs chemical or sensory properties, and sensory vs chemical properties) various relationships in apple juice were graphically studied. The understanding thus developed was used in the selection of best possible combination of variables for multiple regression analysis. The equation for the prediction of preference was developed without any specified level of variable acceptance in the regression model (i.e. without alpha = 0.05 or 0.10 etc.) by maximum r^2 improvement technique using statistical analysis system stepwise procedure (Helwig and Council, 1979).

4. RESULTS AND DISCUSSION

4.1 Introduction

One crabapple and nine apple cultivars adapted for the Western Canada climate were selected for this study. The purpose of the study was to assess their suitability as processing cultivars. The major focus of this program was;

1. to assess the chemical composition of the cultivars by standard methods,

2. to assess the feasibility of producing apple juice by filtration process and developing basis for monitoring this process, and it's influence on the quality of apple juice,

3. to assess the chemical composition of the basic sugars and organic acids in the cultivars by HPLC,

4. to develop a procedure through chemical and sensory analysis of apple juice for characterizing juices from apple cultivars, and

5. to develop an understanding of sensory properties of apple juice to assist blending operation to produce an acceptable juice from these cultivars.

4.2 Chemical Composition and Classification of Apple Varieties

One crabapple and nine apple cultivars were analyzed to determine their moisture, sugar, acid and phenolic content. These cultivars were also classified into various groups based on this compositional analysis. The classification was provided to estimate their relative suitability for various processed products, especially for apple juice.

4.2.1 Variability due to Sampling and Replication

Analysis of variance of composition data and comparison of mean differences indicated insignificant replication effect, insignificant sampling effect on sugar and phenolic content analysis (Table 9), overall significantly lower value of first sample for moisture content determination (Table 10), and overall significantly higher value of second samples for acid content determination (Table 11). Since different sample numbers represent only the sequence of collection rather than the difference in sampling technique, the significant differences among them indicate only the heterogeneity of the lot.

To overcome the heterogeneity of the lot random sampling was done. However, difference among samples for acid and moisture content indicate that sampling could have been improved. The probable reason of significantly lower moisture content of the first sample could be that the first sample included more apples from the top layers of the lot. Since top layers of the lot are in close contact with the air, more moisture loss in top layers could be expected. This might have resulted in the reduction of moisture content in the first sample. However, in the case of acid content, such conditions did not explain the sample differences. It might be possible that acid content variations in a lot are more prominent than sugar and phenolic content variations, and could not be easily overcome by random sampling. Since replication and sampling effects on the chemical analysis were either insignificant or similar, differences among cultivars could be attributed to the inher-

Table 9: Statistical analysis of chemical composition of apples

Analysis of Variance (ANOVA)

Source	Df[a]				Sum of squares				F[b]			
	M	S	A	P	M	S	A	P	M	S	A	P
Variety	9	9	9	9	157.02	94.40	4.19	0.14	117.02*	42.46*	284.30*	43.17*
Sample	3	3	3	3	2.04	1.14	0.04	0.00	4.56*	1.54	7.44	1.82
Replicate	3	3	3	1	0.15	1.26	0.00	0.00	0.33	1.69	0.12	0.42
Error	143	143	137	66	21.32	35.33	0.22	0.02				

a - M or Moisture, S or Sugar, A or Acid and P or Phenolic content.

b - F values marked * are significantly different at alpha 0.05.

Table 10: Statistical analysis of chemical composition data II

Tukey's test for samples (Moisture content %)[1]

Sample	Mean[2]
B	86.38 a
C	86.35 a
D	86.28 a
A	86.08 b

[1]alpha = 0.05.
[2]Means with the same letter are not significantly different.

Table 11: Statistical analysis of chemical composition data III

Tukey's test for samples (Acid content)[1] [2]

Sample	N	Mean[3]
B	40	0.76 a
C	38	0.73 b
A	37	0.73 b
D	38	0.73 b

[1]Acid content expressed as % malic acid.
[2]alpha = 0.05.
[3]Means with the same letter are not significanlty different.

ent varietal variations and to the difference in the length of storage.

4.2.2 Moisture and Total Sugar Content

The range of moisture content in analyzed cultivars was 85.13 to 88.67% (Table 12). In comparison to the average moisture content of the stored not pared apples (83.9%, Anonymous, 1975) these cultivars have higher moisture content. The sugar content of cultivars ranged from 7.34 to 10.26%. An established average value for the sugar content of apple is not available. However, a review of published results of T.S.S. of apple varieties from various growing areas (Moyer and Aitken, 1980, Osborn, 1964) indicated that the cultivars could be classified into two groups: average sugar and below average sugar cultivars (Table 13). Since the method for the determination of sugar content initially included a measurement of T.S.S., this classification based on the T.S.S. was considered appropriate.

4.2.3 Total Acid and Total Phenolic Content

Classification of cultivars based on their acid content is presented in Table 14. According to Moyer and Aitken, 1980 (section 2.1.2.2), the acid content range 0.48 to 0.99% indicates that except for P.F. 51 and Norland, other cultivars could be considered to have high acid content. P.F. 51 and Norland have medium acid content. The range of phenolic content was 0.27 to 0.42% (Table 15). Using phenolic content of Kerr (a crabapple), 0.29%, as the reference point and compar-

Table 12: Moisture content of apple and crabapple cultivars grown on the Canadian Prairies

Cultivar	Moisture Content (%)		
	N	Mean	±
Breakey	16	85.13	0.35
Collet	16	87.02	0.76
Goodland	16	86.25	0.41
Heyer# 12	16	88.67	0.18
Kerr	16	85.26	0.46
Luke	15	86.25	0.29
Norland	16	85.70	0.57
P.F. 36	16	86.72	0.46
P.F. 50	16	86.00	0.58
P.F. 51	16	85.63	0.11

Table 13: Statistical analysis of chemical composition data and classification of apple cultivars based on their sugar content (% sucrose)

Cultivars	Mean[1]	N	±	Class
P.F. 51	10.26 a	16	0.50	Average Sugar
Norland	10.08 a b	16	0.74	" "
Luke	9.61 b c	16	0.44	" "
Kerr	9.58 c d	16	0.34	" "
Breakey	9.47 c d	16	0.49	" "
P.F. 50	9.44 c d	16	0.57	" "
Collet	9.12 d e	16	0.34	" "
P.F. 36	8.84 e	16	0.37	Below Average Sugar
Goodland	8.62 e	16	0.52	" " "
Heyer# 12	7.34 f	16	0.50	" " "

[1] - Mean differences were tested by Tukey's test at alpha 0.05 and means with the same letter are not significanlty different.

Table 14: Statistical analysis of chemical composition data and classification of apple cultivars based on their acid content (% malic acid)

Cultivars	Mean[1]	N	±	Class
Kerr	0.99 a	15	0.08	High Acid
Luke	0.93 b	15	0.05	" "
Heyer# 12	0.88 c	16	0.01	" "
P.F. 50	0.83 d	15	0.05	" "
Breakey	0.78 e	16	0.01	" "
Goodland	0.68 f	15	0.01	" "
Collet	0.67 f	15	0.01	" "
P.F. 36	0.60 g	16	0.05	" "
Norland	0.55 h	16	0.04	Medium Acid
P.F. 51	0.48 i	14	0.05	" "

[1] - Mean differences tested by Tukey's test at alpha 0.05 and means with the same letter are not significantly different.

Table 15: Statistical analysis of chemical composition data and classification of apple cultivars based on their phenolic content (% tannic acid)

Cultivar	Mean[1]	N	±	Class
Luke	0.42 a	8	0.01	High Phenolic
P.F. 36	0.35 b	8	0.01	" "
P.F. 50	0.34 b	8	0.02	" "
Breakey	0.33 b c	8	0.01	" "
Norland	0.33 b c	8	0.02	" "
Heyer# 12	0.31 c d	8	0.02	" "
Goodland	0.30 d	8	0.03	" "
Kerr	0.29 d	8	0.01	" "
Collet	0.28 d e	8	0.01	" "
P.F. 51	0.27 e	8	0.01	Medium Phenolic

[1] - Mean differences were tested by Tukey's test at alpha 0.05 and means followed by the same letter are not significantly different.

ing phenolic content of all the cultivars (Table 15), these cultivars could also be classified into two groups. All crabapple varieties are considered as high astringent (or high phenolic) varieties (Moyer and Aitken, 1980). Therefore, Kerr was selected as a reference point for classification in stead of the comparison with previously reported phenolic contents of other cultivars.

The need to use Kerr as a reference point arises from the lack of a standard method for the determination of phenolic content of apple. In comparison to the published phenolic content data (Moyer and Aitken, 1980), which is a large, though old collection, these cultivars have very high phenolic contents. However, this variation may only be due to the differences in the methods. The method (Hartman, 1943) used in the study reported by Moyer and Aitken (1980), included loss of phenolics due to oxidation. It also lacked an extensive extraction of procedure. On the other hand, the method used to determine the phenolic content of these cultivars, had largely eliminated those problems by using ethanol extraction at low temperature.

4.3 Utilization of Apple Cultivars for the Production of Processed Apple Products

4.3.1 Suitability Guidelines

The products processed from apples are classified into three groups: solid, liquid and pureed products (LaBelle, 1981). As mentioned in section 2.1.1, the suitability of apple cultivars for the production of a processed product depends upon size, shape, specific gravity, color, firmness, soluble solids, total solids, acidity, pH and phenolic content (La Belle, 1981). However, these characteristics provide only a general basis for suitability and limits of these characteristics required for a particular product could be relaxed by combining two or more apple cultivars for the production of a product where processing permits.

For the production of liquid and pureed products apples are crushed and mixed uniformly, but for solid products a minimum of alteration is allowed. This major difference in processing steps greatly alters the basic requirements for suitability of an individual cultivar for the production of specific products. Since processing of liquid and pureed products includes crushing, the size, shape and firmness requirements are not important for these products. The mixing step provides an opportunity for balancing compositional requirements by blending. Therefore, it is apparent that apple cultivars could be used for processing into liquid and pureed products if their chemical composition and a basis for blending are known. On the basis of this information an evaluation of apple cultivars, which were analyzed in this study, is presented in the following section.

4.3.2 Suitability of apple cultivars

Norland and P.F. 51, which had average sugar and medium acid content, are most suitable for the production of liquid and pureed products, especially apple juice. The relative sugar and acid concentrations of Norland and P.F. 51 provide suitable sugar:acid balance for the production of apple juice. This suitability assessment is based on the works of Poll (1981) and La Belle (1981). P.F. 51 also had medium phenolic content, therefore, it is the most suitable for processing. The other cultivars had high acid and phenolic contents, therefore, they are relatively less suitable for processing. The analyzed cultivars (including Norland and P.F. 51) are small in size and lack the characteristics to compete in the fresh fruit market. Their size, acid content and phenolic content makes these cultivars unsuitable for the production of solid processed products.

4.3.3 Important Requirement for Utilization of Apple Cultivars

Norland and P.F. 51 have been identified as the most suitable of the cultivars for processing into puree and juice. The other cultivars have been down graded due to the high levels of acid and phenolic content. However, it must be recognized that if a wide selection of cultivars with a wide range of composition differences, which could also arise due to seasonal variations and storage conditions, were available for processing then through a proper blending procedure quality differences of these cultivars could be masked. Thus, it is apparent that any attempt to utilize cultivars grown on Canadian Prairies (or any other cultivars) must include study of the basis on which blending could be performed.

4.4 Chromatographic Analysis of Sugars and Acids

4.4.1 Analysis of Sugar and Acid Standard Mixtures

Ten sugars and 13 organic acids were analyzed by HPLC for their retention time (RT), response factor (RF) and minimum detectable concentrations (MDC). The results for sugars and organic acids are presented in Tables 16 and 17, respectively. The representative chromatographs of the sugar and acid separation are illustrated in Appendixes 16 and 17, and Appendixes 18, 19 and 20, respectively.

The analysis of standard mixtures indicated that the predominant sugar of apples and apple juice, fructose (RT 14.74), was retained in the column for the longest time. The predominant acid, malic acid, eluted at 11.67 min. The longest retained acid was lactic acid (RT 15.31). Raffinose and citric acid were retained in the column for the shortest time among all the sugars and acids analyzed. The range of MDC for the sugars was 0.0012 to 0.0016 %. However a wider range of MDC , 0.000004 to 0.00462%, was observed for the acids.

Since the instrumental limits of the detector response and area rejection were constant for all the sugars or acids, MDC for the individual sugars and acids was directly proportional to their response factors. The response factor for malic acid was used to express the total and the individual acid concentrations. This approach was adopted to compare the methods of determining total acid content and also to provide

Table 16

Retention times, response factors and minimum detectable concentrations of various sugars as determined by HPLC

Sugar	Number of Observations	Retention Time (min) Mean	±	Response Factor (ug/10000 Area units) Mean	±	Minimum Detectable Concentration[a] (%)
Raffinose	15	8.36	0.05	1.60	0.13	1.60×10^{-3}
Maltose	2	8.80	0.01	1.19	0.01	1.20×10^{-3}
Sucrose	15	9.08	0.05	1.30	0.10	1.30×10^{-3}
Maltotriose	2	9.50	0.01	1.40	0.04	1.40×10^{-3}
Glucose	15	10.96	0.06	1.17	0.08	1.20×10^{-3}
Xylose	15	11.89	0.06	1.33	0.08	1.30×10^{-3}
Galactose	15	12.74	0.07	1.33	0.07	1.30×10^{-3}
Arabinose	15	13.81	0.08	1.27	0.07	1.30×10^{-3}
Mannose	2	14.25	0.02	1.22	0.03	1.20×10^{-3}
Fructose	15	14.74	0.09	1.17	0.10	1.20×10^{-3}

a - For the calculation of minimum detectable concentration, please refer to Appendix 26.

Table 17

Retention times, response factors and minimum detectable concentrations of various acids as determined by HPLC

Acid	Number of Observations	Retention Time (min) Mean	±	Response Factor (ug/10000 Area units) Mean	±	Minimum Detectable Concentration (%)
Citric	11	9.86	0.09	2.26	0.13	2.26 x 10 -3
Alpha-ketoglu.	8	10.51	0.05	0.12	0.002	1.20 x 10 -4
Tartaric	11	10.81	0.09	1.49	0.11	1.49 x 10 -3
Galacturonic	6	10.82	0.02	4.62	0.86	4.62 x 10 -3
Malic	11	11.67	0.11	2.51	0.17	2.51 x 10 -3
Malonic	8	12.02	0.05	3.20	0.60	3.20 x 10 -3
Quinic	11	12.60	0.10	4.14	0.30	4.14 x 10 -3
Succinic	11	13.40	0.15	2.81	0.20	2.81 x 10 -3
Shikimic	8	13.50	0.03	0.004	0.009	4.00 x 10 -6
Glyceric	6	13.52	0.03	3.06	0.68	3.06 x 10 -3
Glycollic	6	14.94	0.03	1.76	0.51	1.76 x 10 -3
Fumaric	11	15.15	0.23	0.0095	0.005	9.50 x 10 -6
Lactic	8	15.31	0.05	2.30	0.63	2.30 x 10 -3

an estimate of the individual acid concentration, in the absence of the confirmation of their presence by another method.

4.4.2 Determination of Sugars and Acids in Apple Cultivars

4.4.2.1 Sugars

Ten apple cultivars were evaluated for their sugar composition. A typical HPLC chromatograph is presented in Appendix 22. Heyer# 12 (Appendix 23) was not typical for these determinations as it had the lowest sugar concentration and only trace amount of sucrose. The concentrations of separated sugars are presented in Table 18. Sucrose concentration ranged from 0 to 1.11%. The range is quite low compared to the ranges reported by Whiting (1970), 1.28 to 6.64% and Moyer and Aitken (1980), 1.71 to 4.18%. Apart from varietal, and regional differences, the reason for low sucrose concentration could also be the hydrolysis of sucrose during sample preparation. Since sample preparation involved the boiling water extraction of apple constituents, heat and acid hydrolysis of sucrose could have decreased the concentration of sucrose. Hydrolysis of sucrose would also increase the concentration of glucose and fructose.

The concentration of fructose varied from 4.40 to 6.96%. This is comparable to the range reported by Moyer and Aitken (1980). Since the fructose may also have been contributed by sucrose hydrolysis , the range may be an inflated estimate. Therefore, the fructose concentration of these varieties might

Table 18

Sugars in various apple cultivars as determined by HPLC

Variety	Sucrose %	±	Glucose %	±	Fructose %	±	Xylose %	±	Total Sugar %	Total Sugar (% Sucrose)	±
Breakey[a]	.58	.05	2.20	.14	6.88	.14	Trace		9.67	10.68	.34
Collet	.44	.06	1.53	.04	5.97	.13	.03	.05	7.92	8.80	.17
Goodland	.61	.08	2.15	.11	6.41	.27	Trace		9.16	10.18	.50
Hyer # 12	---	--	9.43	.10	4.40	.27	--		5.83	6.48	.44
Kerr[b]	.23	.17	3.37	.27	5.83	.36	.02	.02	9.47	10.52	.87
Luke	.33	.03	2.94	.40	6.05	.56	Trace		9.30	10.33	1.08
Norland[c]	1.11	.07	1.92	.09	6.46	.28	.10	.04	9.50	10.56	.42
P.F. 36	.24	.09	2.56	.36	5.60	.72	.06	.05	8.51	9.46	1.17
P.F. 50[c]	.58	.18	1.88	.14	6.96	.48	.06	.05	9.42	10.47	.76
P.F. 51	.60	.16	3.28	.14	5.54	.22	Trace		9.38	10.42	.50

a - Traces of galactose
b - Mean values of four observations (2 x 2 samples x replicate).
c - Mean values of six observations (3 x 2 sample x replicate); also traces of arabinose and galactose.

be below the average or toward the lower end of the range, The average glucose concentration of apple varieties is reported between 1.34 to 1.95% (Moyer and Aitken, 1980). The range of glucose in Manitoba grown varieties was 1.43 to 3.28%. These values like those for fructose may also be higher than the actual range. However, the glucose range is much higher than the average range. Therefore, actual glucose concentration of Manitoba grown varieties could be in the average range. Even though glucose levels are higher and sucrose levels are lower than other reported values the total sugar content of Manitoba grown apple cultivars is lower than average. It can also be concluded that Manitoba grown varieties contain a lower than average concentration of sucrose but average concentrations of glucose and fructose.

However, the relative sugar concentration profile of major sugars in the Manitoba grown varieties is similar to other apple varieties. The predominant sugar was fructose, followed by glucose and sucrose, in that order. The most frequently occuring minor sugar was xylose. It was quantified or identified in all the varieties except Heyer # 12. Galactose and arabinose were present in at least one variety, in trace amounts. These minor sugars have also been reported by several workers (Whiting, 1970).

Apple juice samples were also analyzed by HPLC. Their sugar profile is presented in Appendix 22. Total sugar contents of apple juices determined by HPLC have been used in comparing methods of determination (section 4.4.3.). Apple juice A, B, C, D and E contained 9.90 %, 9.43 %, 9.82%, 8.89% and 9.83% total sugar respectively.

4.4.2.2 Acids

Ten apple cultivars were also analyzed by HPLC for their organic acid composition. A representative chromatograph for the separation of acids is shown in Appendix 24. The organic acids, identified as the most probable acids, and their concentrations (expressed as % malic acid) for each cultivar are presented in Table 19. It was previously reported that positive identification of some organic acids was not possible because of similar retention times. Once identified the concentrations of identified acids could easily be calculated by their response factors. Pending the confirmation of the presence of acids by another method, at present only the total concentration of acid has been used to compare the methods (HPLC and titrable acidity) of determining total acid content.

The total acid content of apple juices have also been used for comparing methods of determination (section 4.4.3) Apple juice A contained 1.11%, B 0.98%, C 1.00%, D 1.11% and E contained 1.05% acid (expressed as malic acid).

4.4.3 Comparison of Total Sugar and Acid Content Determination Methods

The total sugar and acid content of apple cultivars were determined by standard procedure (section 3.1.3) and by HPLC procedure (section 3.2). A schematic presentation of the differences in sample preparation and the estimation of sugar and

Table 19

Identification and concentrations (% malic acid) of organic acids in various apple cultivars as determined by HPLC

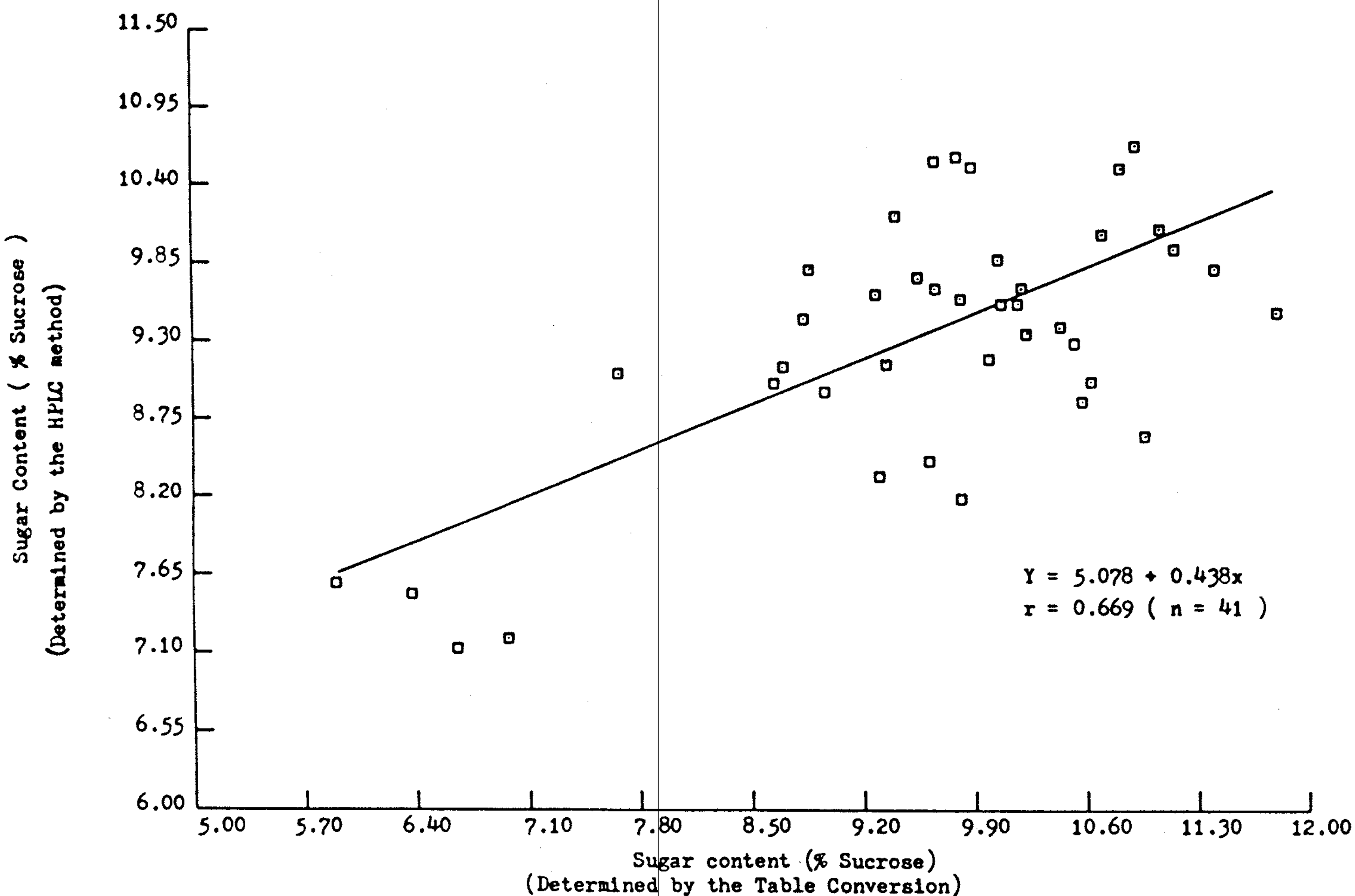
Variety	Citric		Malic		Quinic		Shikimic		Unknown	Total Acid Content	
	%	±	%	±	%	±	%	±	%	%	±
Breakey[a]	.06	.04	.89	.11	.73	.07	.14	.11	.04	1.86	.30
Collet	.02	.01	.71	.03	.48	.02	.32	.07	--	1.54	.11
Goodland	.03	.02	.76	.03	.49	.03	.29	.08	.02	1.59	.16
Heyer #12	.01	.01	.89	.06	.41	.03	.27	.05	.02	1.60	.14
Kerr[a]	.07	.02	1.06	.09	.51	.03	.22	.04	.06	1.92	.18
Luke	.02	.01	1.00	.04	.51	.05	.20	.03	.01	1.74	.11
Norland	.06	.01	.62	.04	.49	.02	.17	.02	.04	1.38	.06
PF - 36	.04	.01	.65	.06	.44	.03	.26	.04	.09	1.48	.14
PF - 50	.03	.01	.94	.07	.56	.03	.26	.03	.02	1.81	.12
PF - 51	.04	.03	.57	.06	.40	.01	.23	.03	.03	1.27	.14

a - Mean of six values (3 samples x 2 replicates).

acid content during compositional and chromatographic analysis is shown in Appendix 25. The chromatographic analysis represents the total sugar and acid contents, which are determined after extensive clarification of sample to remove the interfering components from the sample. This extensive clarification is the major difference between the methods. For the determination of sugar content both methods use refractive index to estimate sugar concentration. However, total acid content was determined by two separate methods including two separate procedure for calculating concentration. In section 3.1.3 acidity was evaluated by titration and concentration as % malic acid was based on the equivalent weight of malic acid. In section 3.2 acidity was evaluated by ultraviolet absorption at 214 nm and concentration expressed as % malic acid was based on the response factor and total peak area.

Since the estimation of sugar content in both methods was performed by refractive index measurement, the differences in the determined concentration could be attributed to the extensive clarification procedure and to the sensitivity of refractive index determination procedures. Linear regression analysis indicated an expected significant correlation (Fig. 3). Total sugar content determined by the chromatographic analysis (9.69%) was significantly higher than the total sugar content (9.27%) determined during the compositional analysis (Appendix 26). A lower refractive index for a sample would be expected in chromatographic analysis due to the clarification of the sample, which would also decrease the estimated sugar content.

Fig. 3. Relationship between sugar contents of apples and apple juices determined by the Table conversion from refractive index and the HPLC method.



However, the observed higher sugar content values might be due to the greater sensitivity of the refractive index detector, used in chromatographic analysis as compared to the sensitivity of the human eye used in compositional analysis. Another effect of the difference in sensitivity of the refractive index measurement is the higher standard deviation obtained in chromatographic analysis than the standard deviation obtained in compositional analysis (Table 13 and Table 18).

Thus, the chromatographic analysis procedure provides the advantage of improving the refractive index determination through the extensive clarification and also through greater sensitivity of the instrumental detector. It provides the advantage of developing the required response factor under conditions which are similar to the sample analysis. It also eliminates the need to approximate using table conversion and the requirement of temperature control or correction. These advantages could provide more reliable results from chromatographic analysis than the method used in compositional analysis. However, the main disadvantage of the chromatographic procedure for the determination of total sugar content is its time requirement. It also includes separation of individual sugars, which is not important for the determination of total sugar content.

The separation of sugars could be excluded from the method by removing the chromatographic column* from the HPLC

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* - Without the chromatographic column the method could not be appropriately called chromatographic procedure. After

system. Removal of the chromatographic column would also considerably decrease the time of analysis. Time required for sample preparation could also be decreased by assembling a series of clarifying columns and placing them between the injector and the detector in a chromatography system. Such a possibility could be observed from the fact that the cations and anions are already exchanged in the guard columns of the HPLC system and do not require separate handling of the sample. Only fine filtration and C18 cartridge cleaning were conducted separately. Of these two, C18 cartridge cleaning could easily be conducted by placing the C18 column along with cation and anion exchangers in the HPLC system.

A similar comparison for procedures used to determine total acid content determination could not be provided. This is due to the difference in the estimation of the acid content by the two methods. As stated above a comparison of the differences in total acid content given by the two procedures was not conducted as acid analysis by HPLC is not complete. Linear regression analysis of total acid content data showed a significant correlation between the methods (Fig. 4). It could indicate that the chromatographic method, if modified like the sugar determination, might be useful for the determination of total acid content.

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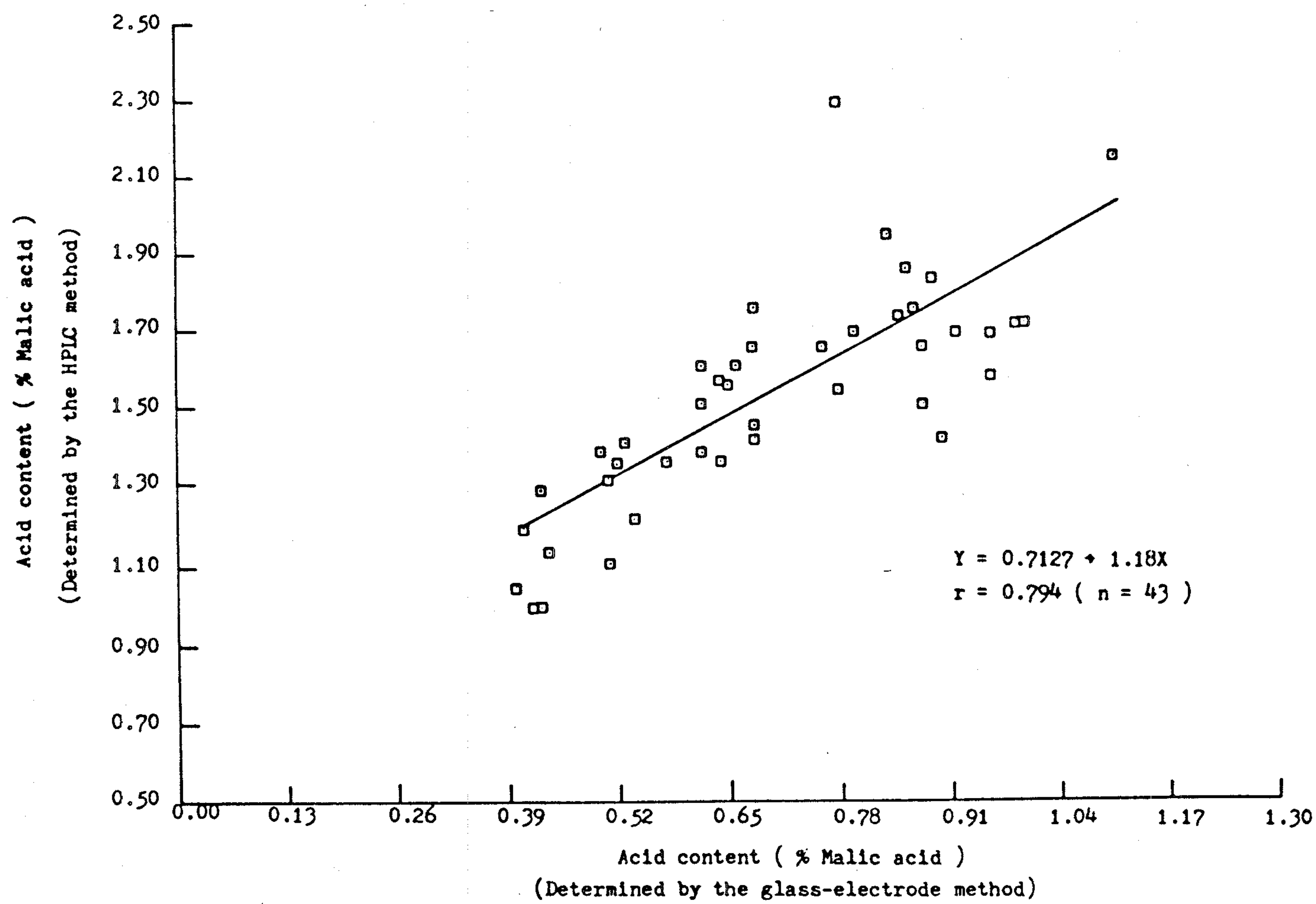
removing the columnthe method becomes an instrumental method for the determination of total sugar content of a sample, which has been extensively clarified to remove compounds which interfere in the determination of refractive index.

Fig. 4. Relationship between acid contents of apples and apple juices determined by the glass-electrode method and the HPLC method



4.5 Filtration Processing of Apple Juice

4.5.1 Correlation between Turbidity and Number of Suspended Particles

As mentioned in section 2.3.1, a slow deposit buildup on filters would maintain optimum filtration rate for a longer period of time. Thus it is apparent that the proper functioning of the filtration assembly would require that the pore size arrangement in the assembly should maintain the narrowest possible range of particles to be removed by every filter. Such an arrangement would maintain the amount of retained particles on every filter to a minimum possible level. It would result in a slow deposit buildup on every filter and would maintain optimum filtration rate for a longer period of time. Since designing and monitoring of the filtration assembly was based on the amount of suspended particles , a parameter to measure the amount of suspended particles retained on every filter was also required.

To develop such a parameter for apple juice filtration, utilization of apple juice turbidity and turbidity changes during the filtration process were proposed as practical methods (section 2.3.3). It should be emphasized that the pressure could only serve as an incomplete parameter to monitor the filtration process. Pressure by definition could not provide a measure of filter performance i.e., a measure of the removal of suspended particles by a filter. To establish turbidity as a practical measure of particle removal, theoretical explanation was presented in section 2.3.3.3. The turbidity observations in the experiments conducted to establish correlation between the number of suspended particles in apple juice and its turbidity are listed in Appendix 27. Statistical analysis of these results (Table 20) indicated that if other variables remain constant, almost 99% (based on the coefficient of determination) of the change in turbidity in apple juice could be explained in terms of variation in the number of suspended particles in apple juice.

4.5.2 Application of Turbidity in Filtration Processing of Apple Juice

Due to a very significant correlation between turbidity and number of suspended particles turbidity measurements were considered to be sufficient to monitor the filtration process. The practical applications of turbidity are discussed in following sections. These applications include determination of approximate size distribution of suspended particles, redesigning of the filtration assembly and testing the recycling of apple juice through the coarse filtration unit.

4.5.2.1 Redesigning of the Filtration Assembly

Turbidity changes observed after every filter during processing of two apple varieties are listed in Appendix 28. Based on these observations approximate distribution of the suspended particles in apple juice is presented in Table 21. This distribution of suspended particles was used to suggest changes in the filter arrangement of thc filtration assembly (Fig. 5). The proposed use of 40 μm, 10 μm and 0.45 μm fil-

Table 20: Correlation coeffecients obtained in regression analysis between observed turbidity and relative number of suspended particles in apple juices

Apple juice	Df	r	r^2
Collet			
Ist batch	6	0.999[1]	0.998
2nd batch	6	0.995	0.990
Kerr			
Ist batch	6	0.996	0.992
2nd batch	7	0.998	0.996

[1] - All values are significant at alpha 0.05.

Table 21: Approximate size distribution of suspended particles in apple juice

Size range	% Particles
25 µm and larger	44.40
25 to 5 µm	18.80
5 to 1 µm	8.03
1 to o.2 µm	25.72
0.2 µm and smaller	2.97

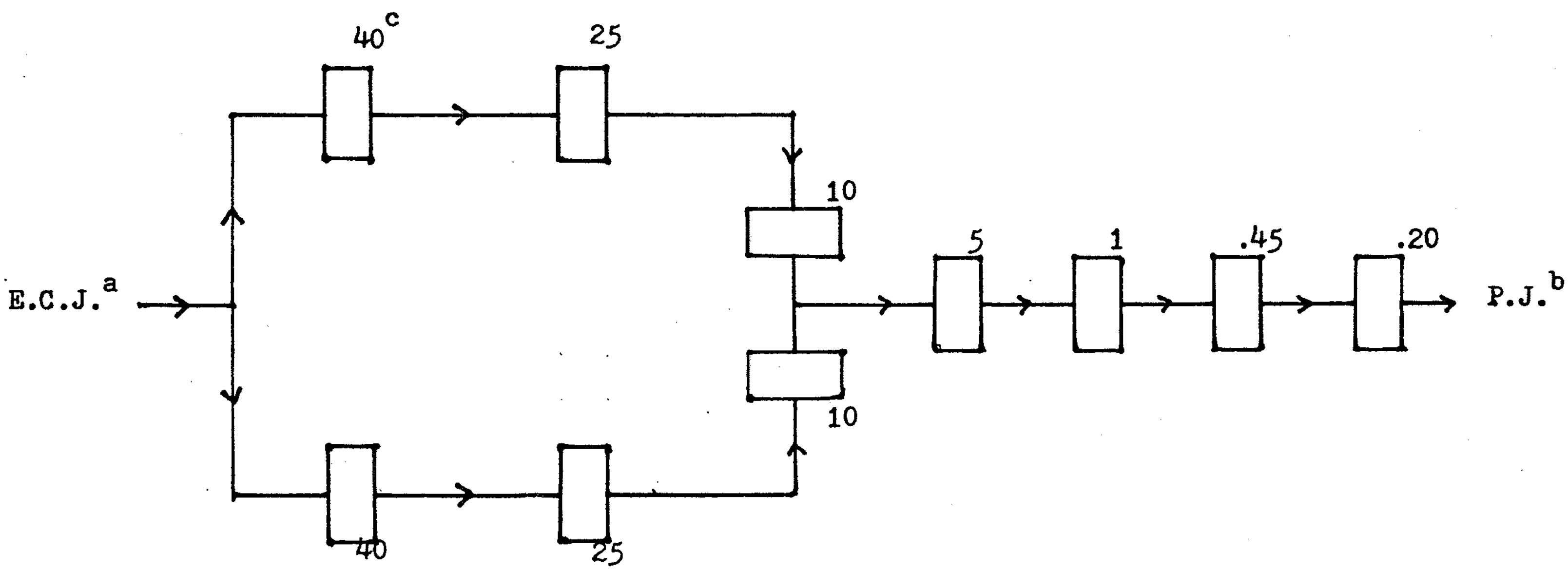


Fig. 5. Flow diagram of the filtration assembly with proposed changes

a - Enzyme clarified juice

b - Processed juice

c - Numbers represent pore size (µm) of filters.

ters would help in reducing filter deposits on the 25 µm, 5 µm and 0.2 µm filters respectively. The reduced resistance to the flow thus achieved would help in maintaining the optimum filtration rate for a longer period of time.

4.5.2.2 Test on the Recycling of Apple Juice

The turbidity changes observed after continuous recycling of apple juice through the coarse filtration unit are illustrated in Fig. 6. The time for one recycle was determined on the basis of total volume to be recycled and the filtration rate after the first passage of juice through the coarse filtration unit.

Approximately, 51% of suspended particles remaining after the first passage of the apple juice through the coarse filtration unit were removed by recycling. It could be easily observed that without recycling these particles would have been deposited on the 0.2 µm filter, and in turn would have reduced the performance of the 0.2 µm filter. During the experiment reported in Fig. 6, a steady turbidity was observed after the 5th and 6th recycles. However, such steady turbidity values were not commonly observed. Therefore, it was suggested that a level of turbidity decrease (1% or 5% etc.) be designated as the point for terminating recycling.

4.5.2.3 Process Monitoring

As mentioned in the last section, the decision to continue or terminate recycling could be made on the basis of observed turbidity decrease. Similarly, turbidity could also be used

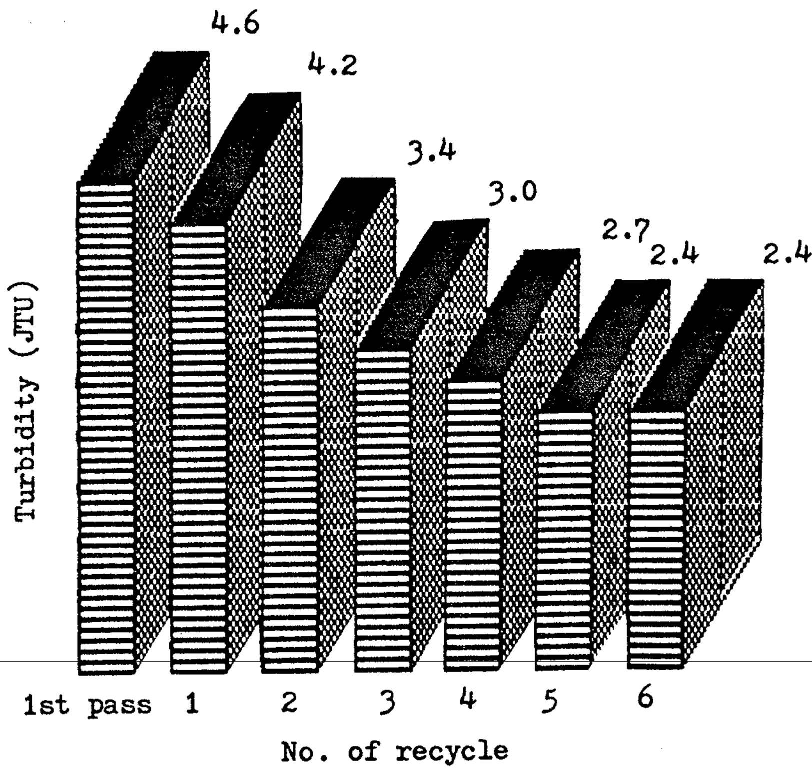


Fig. 6. Change in turbidity of apple juice during recycling through coarse filtration unit.

for monitoring filtration processing of apple juice. Changes in turbidity (Table 22) which were observed at various stages during the filtration of apple juice could be and were used to arrive at operating decisions during process monitoring.

The basis for the decision to continue and to terminate recycling has been presented in the above section. The process was terminated after the 21st bottle, because of the increase in turbidity of apple juice. Turbidity of apple juice after the 21st bottle (0.29 JTU) was higher than the turbidity of the juice after the 14th bottle (0.20 JTU). This increase should be considered against the normal practice of the filtration. Since on the filter there would be more deposits after the 21st bottle than the deposits after the 14th bottle, and under normal conditions it would be expected to retain more suspended particles than it did after the 14th bottle. Therefore, theoretically the turbidity of apple juice after the 21st bottle should always be less or at least equal to the turbidity of apple juice after the 14th bottle.

A possible cause of the turbidity increase could have been the pressure induced deformation of the pores of the 0.20 μm filter. This deformation might have resulted from the increase in speed which was needed to counteract the reduced flow of juice through the 0.20 μm filter. The reduced flow was a result of particle sedimentation on the filter. The net effect was an increase in pressure on the 0.20 μm filter. It might be argued that the process could be allowed to continue until increasing turbidity reaches the level of the initial

Table 22: A list of process monitoring decisions based on the observed Changes in turbidity of apple juice during filtration processing

Filtration Stage	Turbidity (JTU)	Decision
Enzyme Clarified Juice	21.00	.....
Ist Circulation (juice through the coarse filtration unit)	4.60	.....
Ist Recycle	4.20	Continue recycling
2nd Recycle	3.40	Continue recycling
3rd Recycle	3.00	Continue recycling
4th Recycle	2.70	Continue recycling
5th Recycle	2.40	Continue recycling
6th Recycle	2.40	Terminate recycling
First Sample (juice through the fine filtration unit)	0.34	
Sample after 7th bottle	0.24	Continue bottling
Sample after 14th bottle	0.20	Continue bottling
Sample after 21st bottle	0.29	Terminate bottling

sample (0.34 JTU). However, the decision to terminate the process was taken to avoid the irreversible pore deformation, which might have occurred if resistance had increased by continuing the filtration beyond the 21st bottle.

4.5.3 Chemical and Physical Changes in Apple Juice upon Filtration

As mentioned in section 2.3.4 chemical and physical changes in the apple juice that are produced during the filtration processing could be important from sensory and regulatory point of views. It was also elobrated in the same section that such changes could be expected to result from the extensive removal of the suspended particles from apple juice during the filtration process. The statistical analysis of observed chemical changes in juices prepared from two apple and one crabapple cultivars (Table 23) is presented in Appendix 29. The composition of these three apple juices were significantly different. A significant decrease in the sugar, acid and phenolic contents of the apple juice was also observed. Similar results were obtained for the effect of filtration on the specific gravity and clarity of apple juice (Table 23).

Phenolic compounds of apple juice have been established as one of the constituents of the suspended particles of juice (Neubert, 1942). Therefore, the observed decrease in the phenolic content upon removal of the suspended particles was expected. However, the observed decrease in the sugar content of apple juice could not be attributed to the removal of sug-

Table 23: Chemical and physical changes observed in apple juice upon filtration processing

Observation	Apple juice[1]	Collet Mean	Collet N	Goodland Mean	Goodland N	Kerr Mean	Kerr N
Acid Content (% Malic acid)	E.C.J.	0.42	5	0.62	5	0.52	2
	P.J.	0.38	5	0.60	5	0.52	2
Sugar Content (% Sucrose)	E.C.J.	8.28	5	9.17	5	9.58	2
	P.J.	8.07	5	9.16	5	8.57	2
Phenolic Content (% Tannic acid)	E.C.J.	0.07	2	0.10	2	0.10	2
	P.J.	0.05	2	0.09	2	0.08	2
Specific Gravity (20/20)	E.C.J.	1.0329	5	1.0367	5	1.0382	2
	P.J.	1.0320	5	1.0365	5	1.0341	2
Clarity (Clarity Index)	E.C.J.	4.76	-	4.76	-	8.20	-
	P.J.	294.18	-	263.16	-	625.00	-

[1] - E.C.J. or Enzyme clarified juice, and P.J. or Processed juice.

ars along with suspended particles. Since the method to determine the sugar content was based on the refractive index of apple juice, it could have only measured the soluble sugars of apple juice. However, during filtration soluble components are not removed from apple juice, and the observed decrease in sugar content might have resulted only from the decrease in the refractive index of apple juice upon removal of the suspended particles. Therefore, the observed decrease in the sugar content could be classified as an artifact of the method, such observed decrease could not be important for the sensory properties of apple juice. Such limitations, however, were not applicable to the method to determine the acid content of apple juice. Since the pH of apple juice was low enough to maintain the ionic nature of acids in apple juice, an ionic interaction between suspended particles and acids could be expected. The acids could, therefore, be removed from apple juice along with the suspended particles.

The observed decrease in the specific gravity of apple juice also had a limitation of the method, i.e. change in the refractive index upon removal of suspended particles. The increase in clarity of apple juice was observed as expected. The turbidity of apple juice depends upon the suspended particles of apple juice and removal of suspended particles from apple juice decreases its turbidity or increases its clarity. Research workers (Neubert, 1942, Joslyn et al., 1952 and Ishii and Yokotsuka, 1973) have used the transmitted and/or the reflected light measurement to observe the clarity of apple juice. Those studies used clarity of apple juice as a collective term to include color as well as clearness of apple juice. In this investigation, only the clearness of apple juice was considered. Therefore, only the reflected light was measured.

The reflected light was measured at an angle of 90 degree with a nephelometer, and was expressed as Turbidity. Since clarity is inversely proportional to turbidity, expression of clarity in turbidity units, might not be appropriate. Therefore, clarity expressed as "clarity index" listed in Table 23 was calculated by the following formula:

$$\text{Clarity Index} = (1/\text{Turbidity}) \times 100. \quad \ldots\ldots(9).$$

4.5.4 Effects of the Chemical and Physical Changes in Apple Juice upon Its Quality

An overall decrease of 11.54% in acid content and 16.80 % decrease in phenolic content of apple juice was observed during filtration processing of apple juice. These observed decreases also represent 23.53% increase in acid-phenolic balance of the juice.

Since sourness of apple juice is directly proportional to acid content of the juice (section 4.6.4.2), the observed decrease in acid content indicates that by filtration processing the juice would become less sour. In the same section it is also discussed that bitter-astringent taste of apple juice is directly proportional to phenolic content and indirectly proportional to acid-phenolic balance of the juice. Therefore, a decrease in bitter-astringent taste of apple juice would also occur during filtration processing of the juice. According to Poll (1981) decrease in sourness and bitterness would increase the overall taste of the juice. Preference optimization equation discussed in section 4.6.6.1 also indicates that increase in acid:phenolic balance would improve the preference rating of the juice.

This conclusion that filtration processing would increase preference rating of apple juice is based on the assumption that the sugar content and perceived aroma intensity of the juice do not decrease during filtration processing. Since decrease in sugar content could be an artifact of the method and in absence of heat treatment aroma loss may not be observed, it can be concluded that filtration processing would have a positive effect on the sensory properties of apple juice.

At present, effects of the other changes i.e. decrease in specific gravity and increase in the clarity of apple juice, on the quality of apple juice could only be speculated. The Department of National Health and Welfare has established a range of specific gravity of apple juice as a possible check for the adultration (section 2.3.4). Since a decrease in the specific gravity was observed, a cautious monitoring of the specific gravity and related regulatory requirements (ash content and potassium carbonate content) could be suggested. The clarity of some beverages and fruit drinks is commonly regarded as a positive attribute and also mainly the clear apple juice is available in the North American market. These facts could indicate that the clarity of apple juice is also a desirable attribute. Since increase in clarity of apple juice is observed the effect of filtration process on its quality could be considered positive.

4.6 Psychophysical Analysis of Apple Juice

4.6.1 Introduction

The sensory properties that were investigated included perceived sweetness, sourness, BAT and apple aroma of standard solutions and apple juice, at refrigerated temperature (3 - 5°C). The objectives of this study were to establish the relationship between sensory properties and chemical properties of standard solutions. The relationships developed in model systems were applied to measure the sweetness, sourness, BAT and aroma of commercial apple juices . Preference and chemical properties of the commercial apple juices were also determined, and the interrelationships of sensory properties, chemical properties and preference for apple juice were investigated. Finally, based on the understanding of various psychophysical relationships in apple juice, their quantitative integration was conducted to estimate the extent (amount) and direction (increase/decrease) of psychophysical changes in the juice, which could be useful in enhancing preference for apple juice.

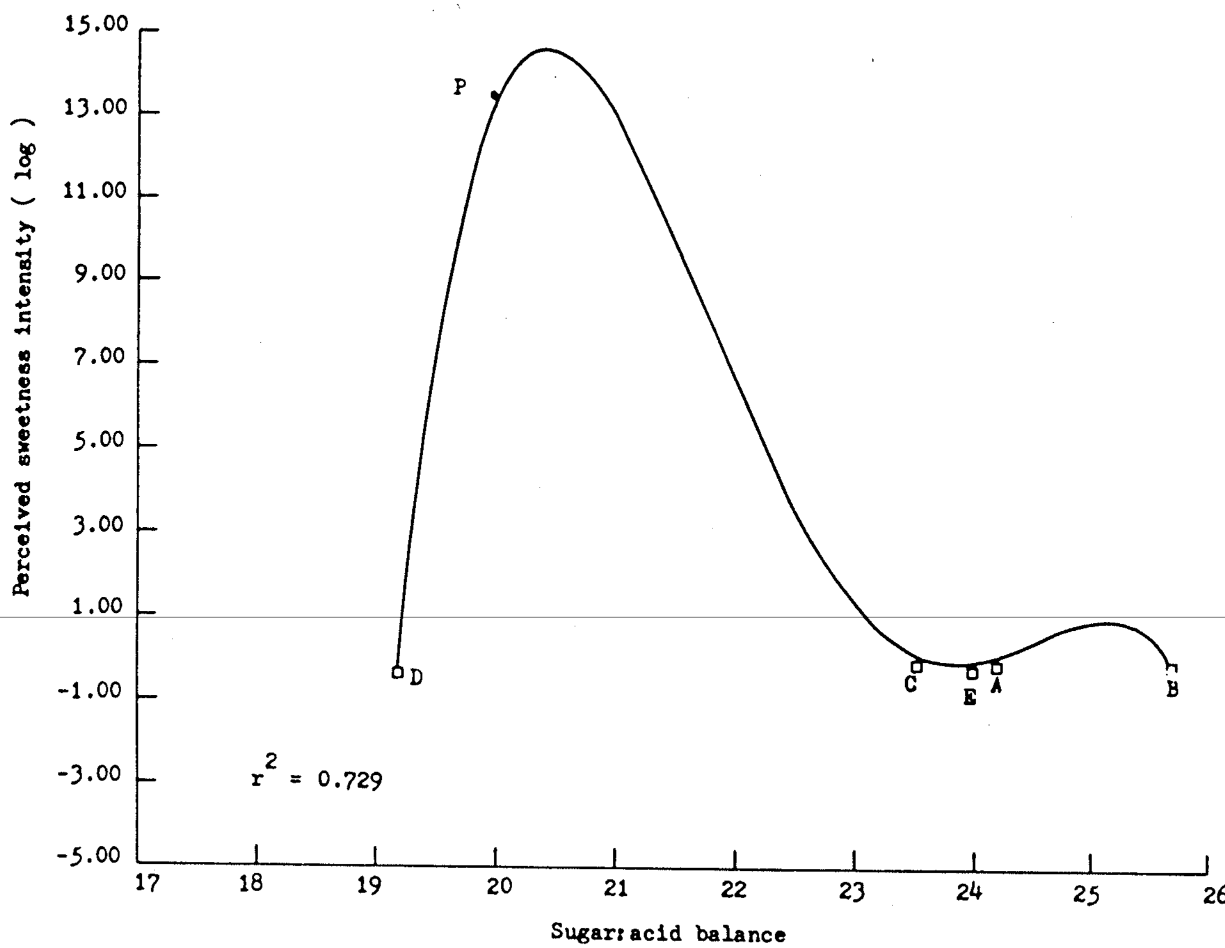
4.6.2 Interpretation of Sensory Data

Trant et al. (1981) reported that at present a set of rules and requirements, which will always relate sensory methods to instrumental methods successfully, are not available. This also holds true for relating one sensory property with another sensory property, such as relating preference to the intensity of perceived sweetness. To deal with the lack of a set of rules and the requirement of a practical application of relationship , some conceptual limits were imposed above the statistical limits . The conceptual limits are "reasonable limits of experience" and " of practical value".

The "reasonable limits of experience" is defined as the limits of perceived sensory characteristics which corresponds to a stimuli concentration likely to be present in apple juice under normal production.

For example, a four degree polynomial relationship is the best fit relationship between perceived sweetness intensity and sugar: acid balance ($r^2 = 0.729$, Fig. 7). However, this curve indicates that for a sugar:acid balance of 20, at point P, perceived sweetness intensity is 13.08 which based on the model system analysis is equivalent to a perceived sweetness intensity of a solution with sugar:acid balance of > 100. A sugar:acid balance of such magnitude is not likely to be present in apple juice under normal production. Therefore, this four degree polynomial relationship is beyond the reasonable limits of experience. The presence of such relationships, which were statistically best but beyond reasonable limits of

Fig. 7. Perceived sweetness of apple juice as a function of sugar:acid balance I.*,**



* - A, B, C, D, and E are the code letters of commercial apple juices.

**- Point P on the curve represents sugar:acid balance 20 and perceived sweetness intensity 13.08.

experience indicated the need of relaxing statistical limits for certain psychophysical relationships in apple juice.

The concept " of practical value" is defined as a relationship, which within reasonable limits of experience yields a simpler/practical solution to indicate sensory properties of apple juice. For example, a simple straight line relationship with less statistical significance may be preferred over a complex but statistically significant relationship, as a practical solution to indicate sensory properties of apple juice.

Another term of interest for the following discussion is "optimization". This term is mainly used to indicate the analysis or results pertaining to the development of the conditions, conducive to the best attainable preference for apple juice.

4.6.3 Studies of Sensory Characteristics in Model System

If other factors remain constant, the perception of a sensory characteristic could be considered to depend upon the sensitivity of the sensory system to the sensory properties and its interaction with all other sensory properties. Sensitivity is the ability to perceive quantitative and/or qualitative differences (Amerine *et al*., 1965 a). Differential sensitivity to taste is considered to be finer than to smell. The power function exponent, which indicates the contraction of physical ratio to sensory ratio (Moskowitz, 1977 a) by the sensory system seems to be a good indicator of quantitative sensitivity, since, like differential sensitivity, the exponents for taste intensities are higher than for odor intensities.

Similar results were obtained in this study. The power function exponent for taste intensities of sweetness (1.30, Appendix 30), sourness (-1.03, Appendix 31) and BAT (0.86, Appendix 32) were higher than the exponent for aroma (0.55, Appendix 33). These results are based on the mean scores presented in Appendixes 34 and 35. The interaction among these sensory characteristics have been widely studied, (Amerine *et al.*,1965 a and b, and Leif, 1981) except for the interaction of taste intensities with astringency. The unique nature of astringency could be expected to reduce other taste intensities.

Astringency, which is a taste sensation resulting from the coagulation of the proteins of saliva and the mucous epithilium of the mouth, and causing reduced lubricant action (Bate-Smith, 1954). It is associated with the cumulative palate block, which makes unprejudiced assessment of subsequent samples of astringent compounds almost impossible (Lea and Arnold, 1978). This effect could be observed from the exponent of 0.86 for BAT, which is the lowest exponent of all the taste intensities. The palate block effect could also similarly, influence (reduce) the perception of other taste intensities for the sample.

4.6.4 Studies of Sensory Characteristics in Apple Juice

Mean sensory scores and preference for apple juices are listed in Appendix 36. These values were analysed by regression analysis together with chemical content and derived ratio values for apple juice (Appendix 37). The coefficients of determinations for various regression relationships between perceived sensory characteristics and antecedent physical properties are presented in Table 24. Polynomial regression analysis always provided the best fit models, however, as mentioned in section 3.4.6, within practical limits simpler regression equations fit enough to explain 75% variability have been chosen for explanations in the following sections. This limit was also relaxed in cases of unreasonable relationships.

4.6.4.1 Sweetness

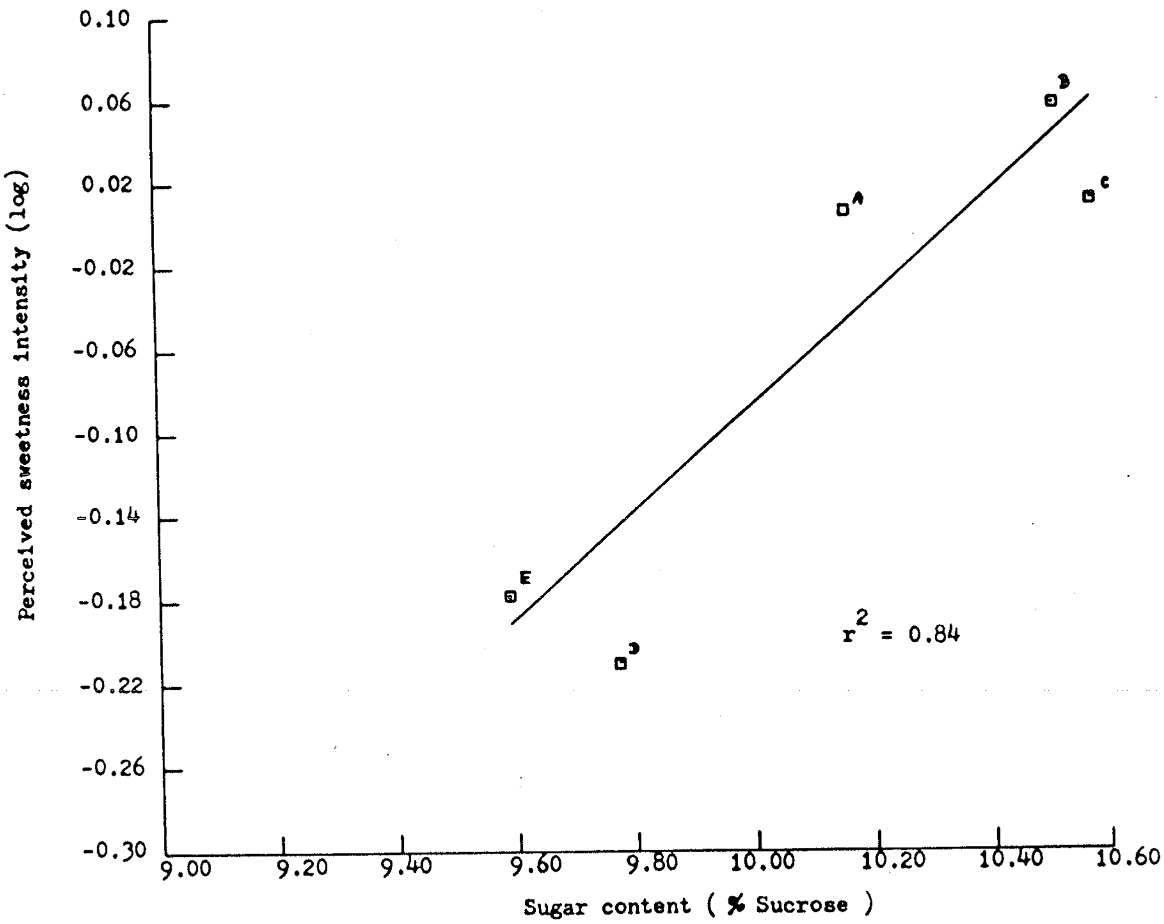
Perceived sweetness intensity of apple juice was found to be log-linearily correlated with the sugar content (Fig. 8). Log-linear (r^2 = 0.559) and four degree polynomial (r^2 = 0.729) relations between perceived sweetness intensity and sugar:acid balance were below the acceptable limit (r^2 = 0.750). The four degree polynomial relationship (Fig 7) though statistically most fit, was beyond the reasonable limits of experience. In the presence of acids sweetness of two major sugars (fructose and sucrose) of apple juice diminishes and the sweetness of glucose is not affected (Harold, 1981). Therefore, sweetness of apple juice may be expected to decrease with the relative (to sugar content) increase in acid content of the juice. Though in this study when relative concentrations of sugar and acids in apple juice were expressed as sugar:acid balance, such effect was not observed. The Log linear relationship (Fig. 9) though far below the practical

Table 24: Coefficients of determination and best fit regression models of relationships between various sensory and chemical properties

Variables[1] Y	X	r^2 Linear	r^2 Polynomial
Sweetness intensity (log)	Sugar content	0.838	----[2]
" "	Sugar:acid balance	0.559	0.729 (4d)[3]
" "	Sugar:phenolic balance	0.015	0.988 (2d)
Sourness intensity (log)	Acid content	0.859	----
" "	Sugar:acid balance	0.848	----
" "	Acid:phenolic balance	0.373	0.909 (2d)
BAT intensity (log)	Phenolic content	0.908	----
" "	Sugar:phenolic balance	0.896	----
" "	Acid:phenolic balance	0.751	----

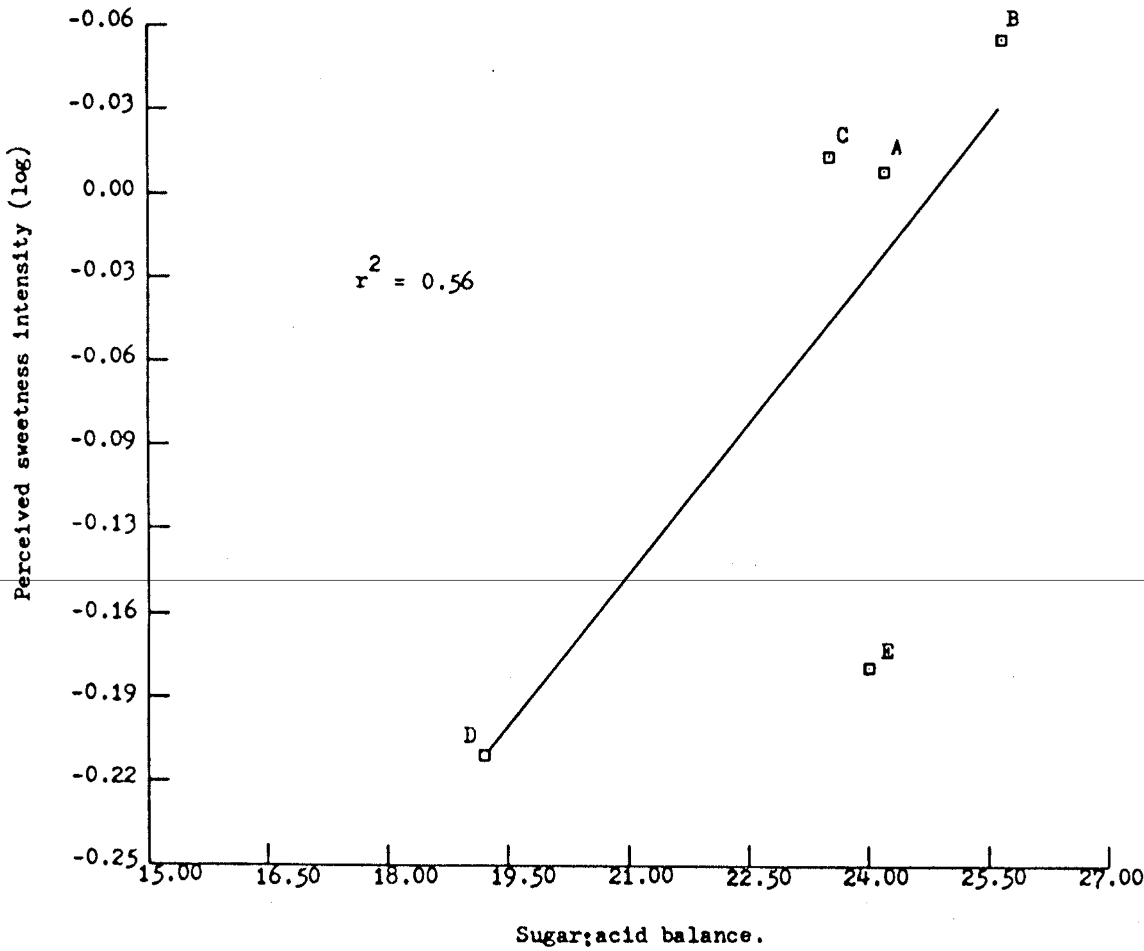
[1] - Y or dependent variable and X or independent variable.
[2] - Not determined, since linear relationship was significant.
[3] - Polynomial degree.

Fig. 8. Perceived sweetness intensity of apple juice as a function of sugar content.



* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 9. Perceived sweetness intensity of apple juice as a function of sugar:acid balance II.*



* - A, B, C, D, and E are the code letters of commercial apple juices.

limit may show that effect. The observed deviations in log linear relationship from such an effect, especially of apple juice "E" may arise because unlike the model system sugar:acid balance in apple juice varies with changes in sugar as well as acid content.

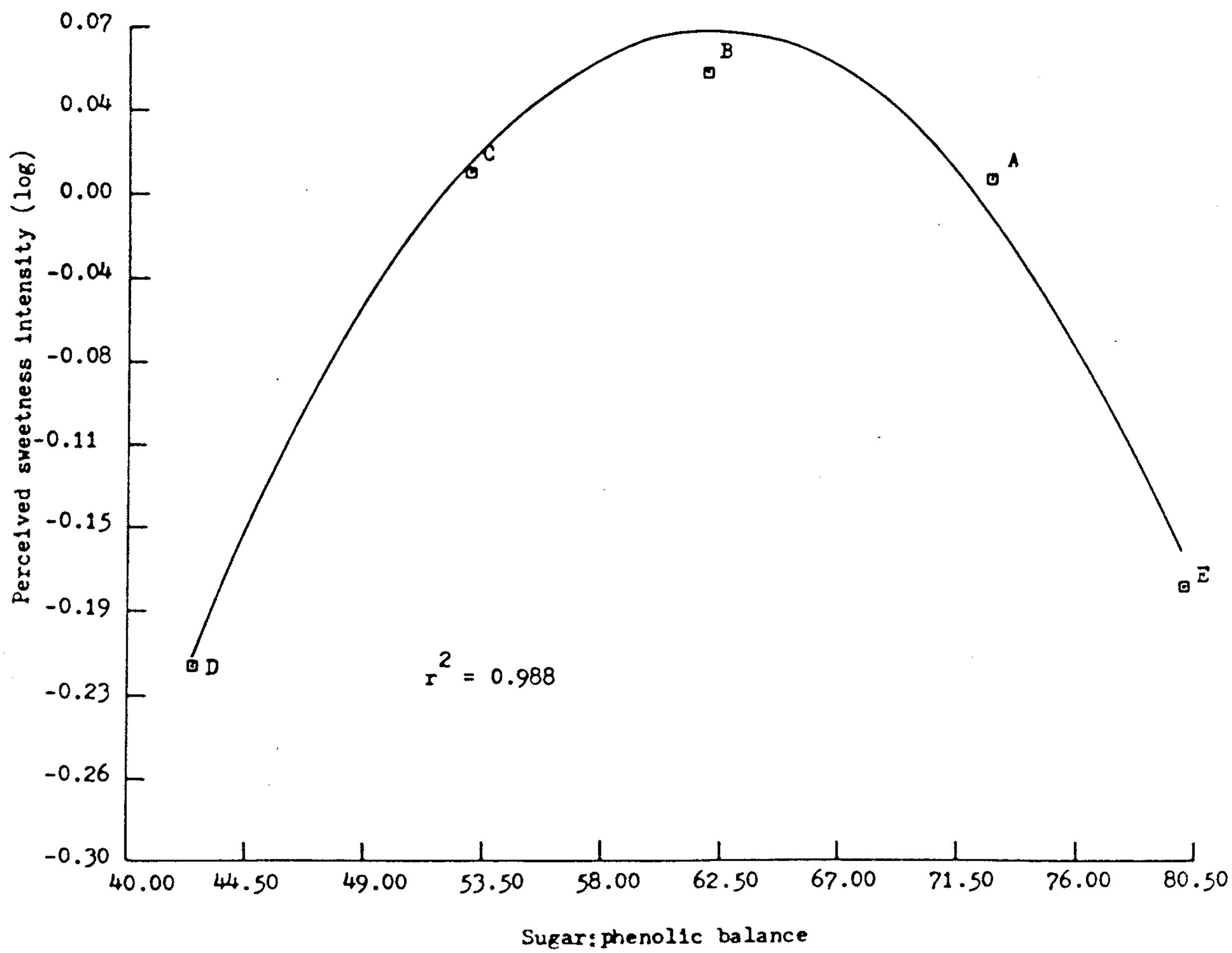
The relationship between perceived sweetness and sugar:phenolic balance, though explanable by a two degree polynomial regression (Fig 10) also indicated similar complexities. As mentioned in section 4.6.3, phenolic content may be expected to decrease the perception of sweetness. However, this parabolic relationship indicated positive as well as negative effects of phenolic content on the perceived sweetness of apple juice.

4.6.4.2 Sourness and Bitter - Astringent Taste

Perceived sourness intensity of apple juice correlated positively with the acid content and negatively with sugar:acid balance of apple juice, Fig 11 and 12, respectively. However, complexities similar to the effect of phenolic content on perceived sweetness were observed in the relationship between sourness and the acid:phenolic balance (Fig 13). Unlike sweetness and sourness of apple juice, the BAT of apple juice was found to log linearily correlate with phenolic content (Fig 14), sugar:phenolic balance (Fig 15) and with acid:phenolic balance (Fig 16). These relationships indicated that there was a negative effect by sugar and acid content on the BAT of apple juice.

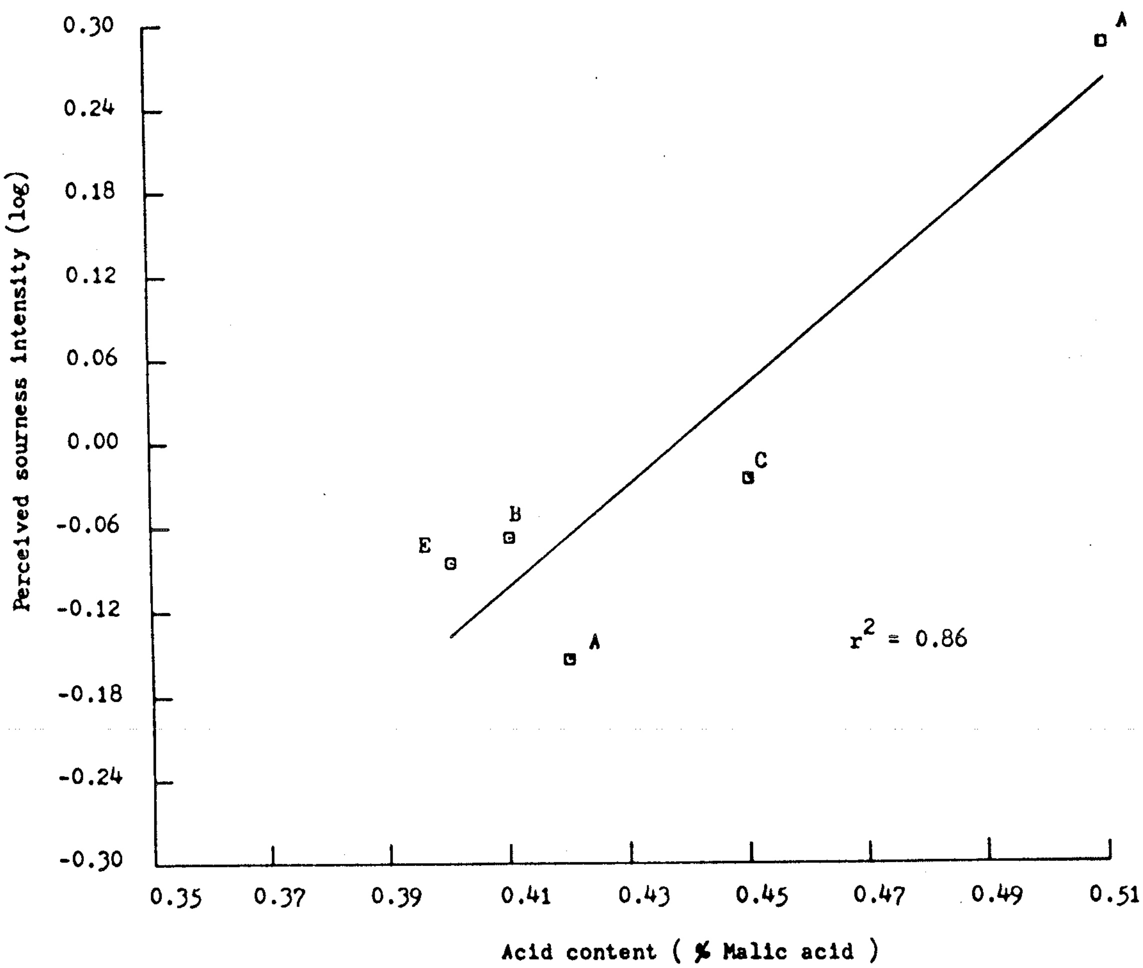
From the above discussion it is apparent that the sweet-

Fig. 10. Perceived sweetness intensity of apple juice as a function of sugar:phenolic balance.*



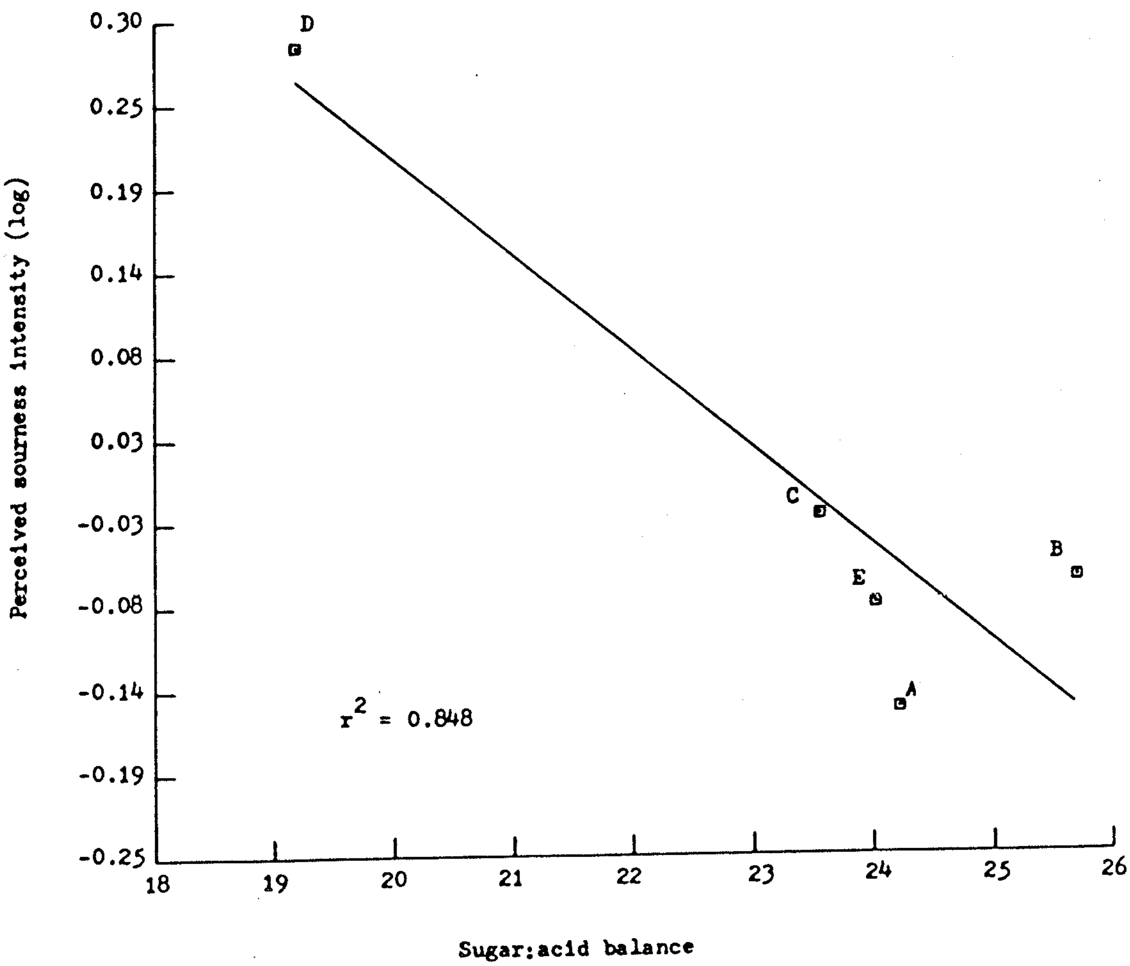
* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 11. Perceived sourness intensity of apple juice as a function of acid content.*



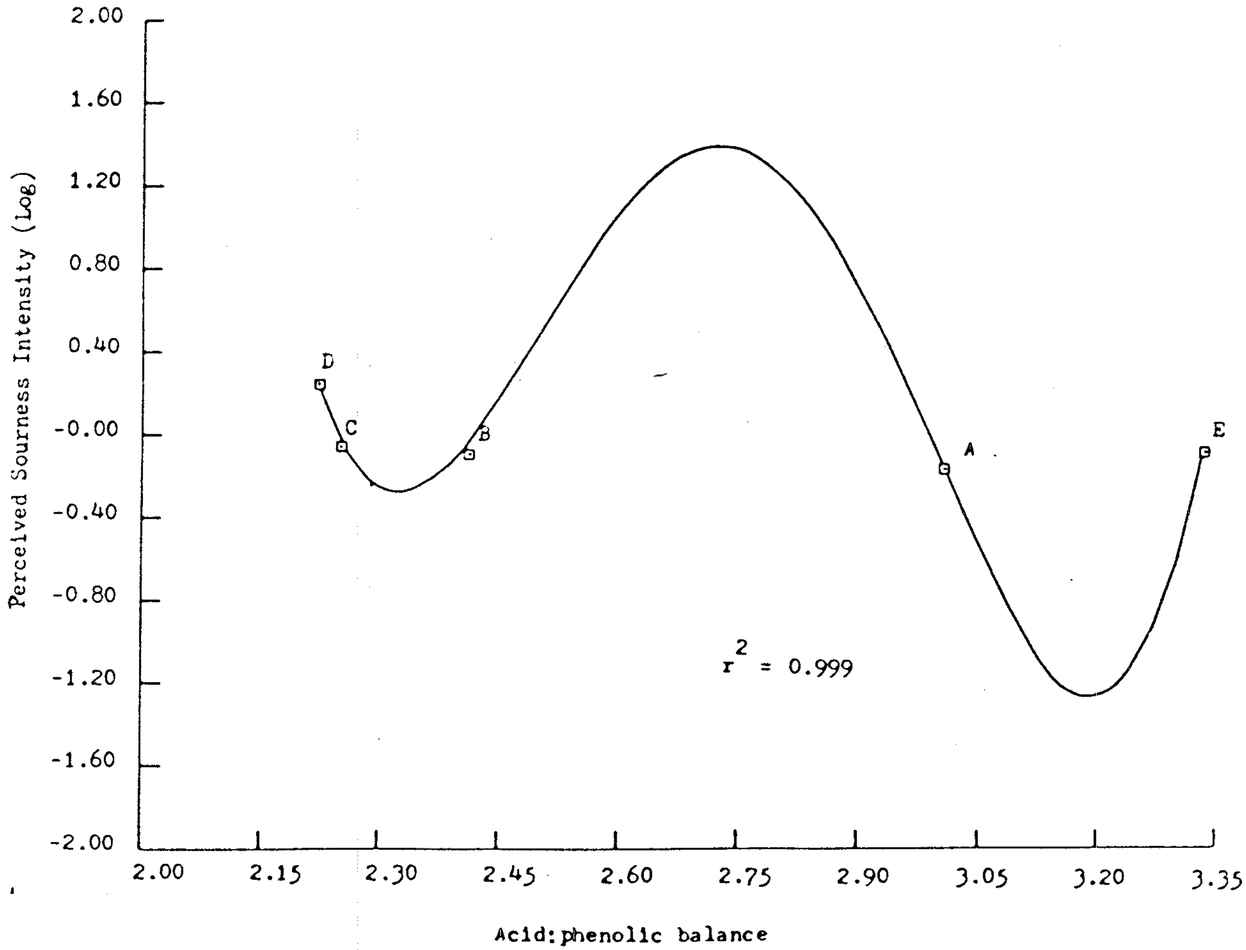
* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 12. Perceived sourness intensity of apple juice as a function of sugar:acid balance.*



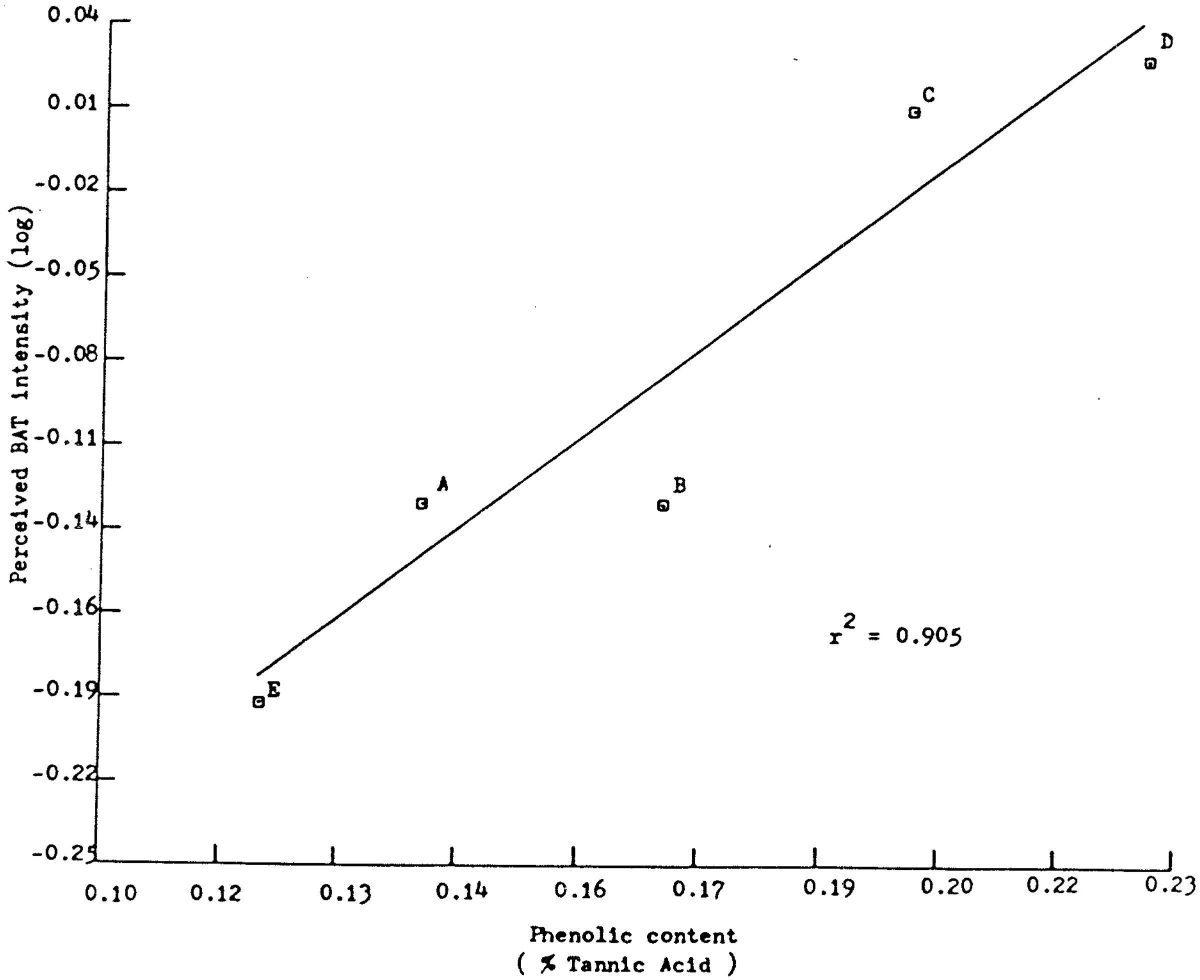
* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 13. Perceived sourness intensity of apple juice as a function acid:phenolic balance.



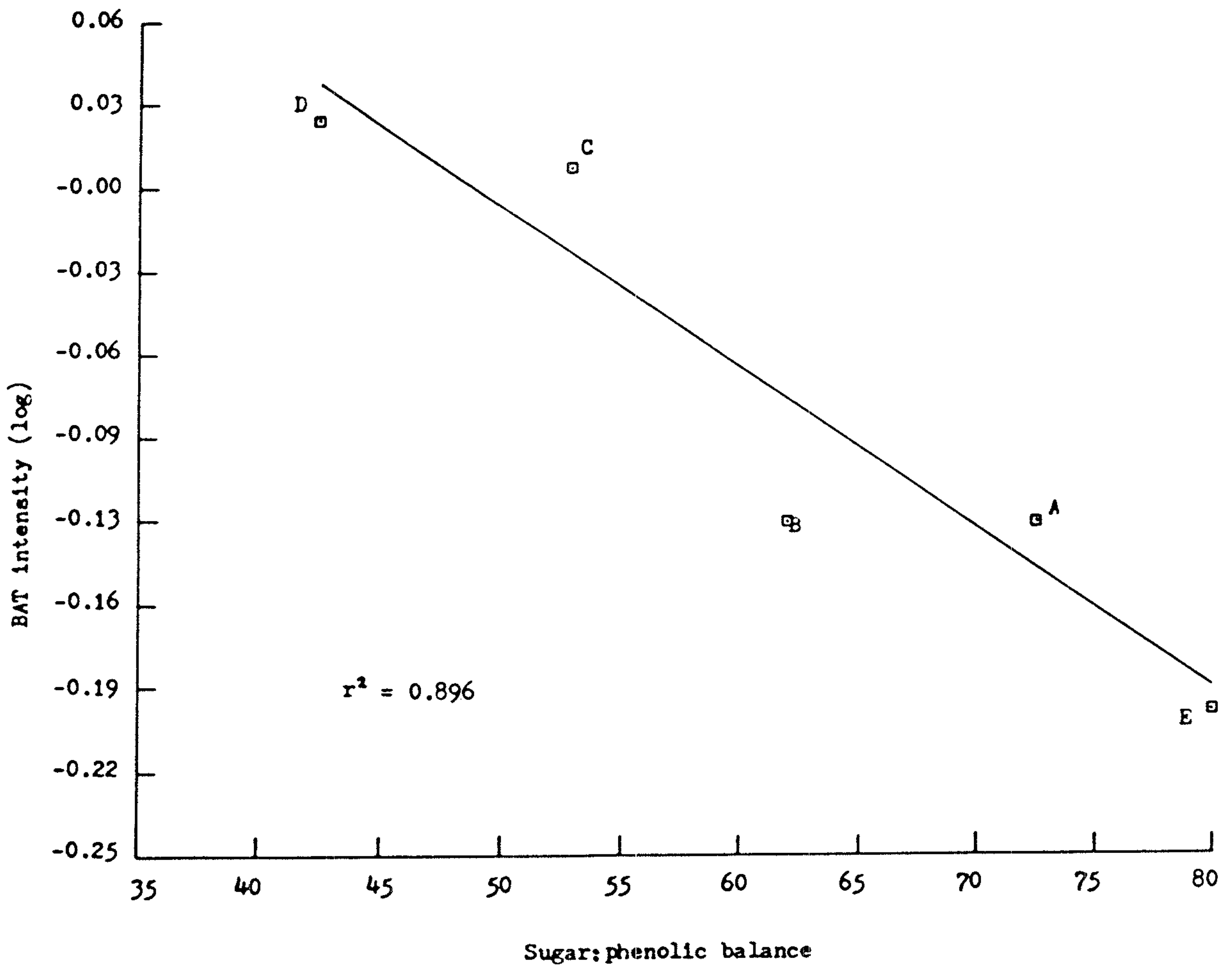
* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 14. Perceived bitter-astringent taste (BAT) intensity of apple juice as a function of phenolic content.*



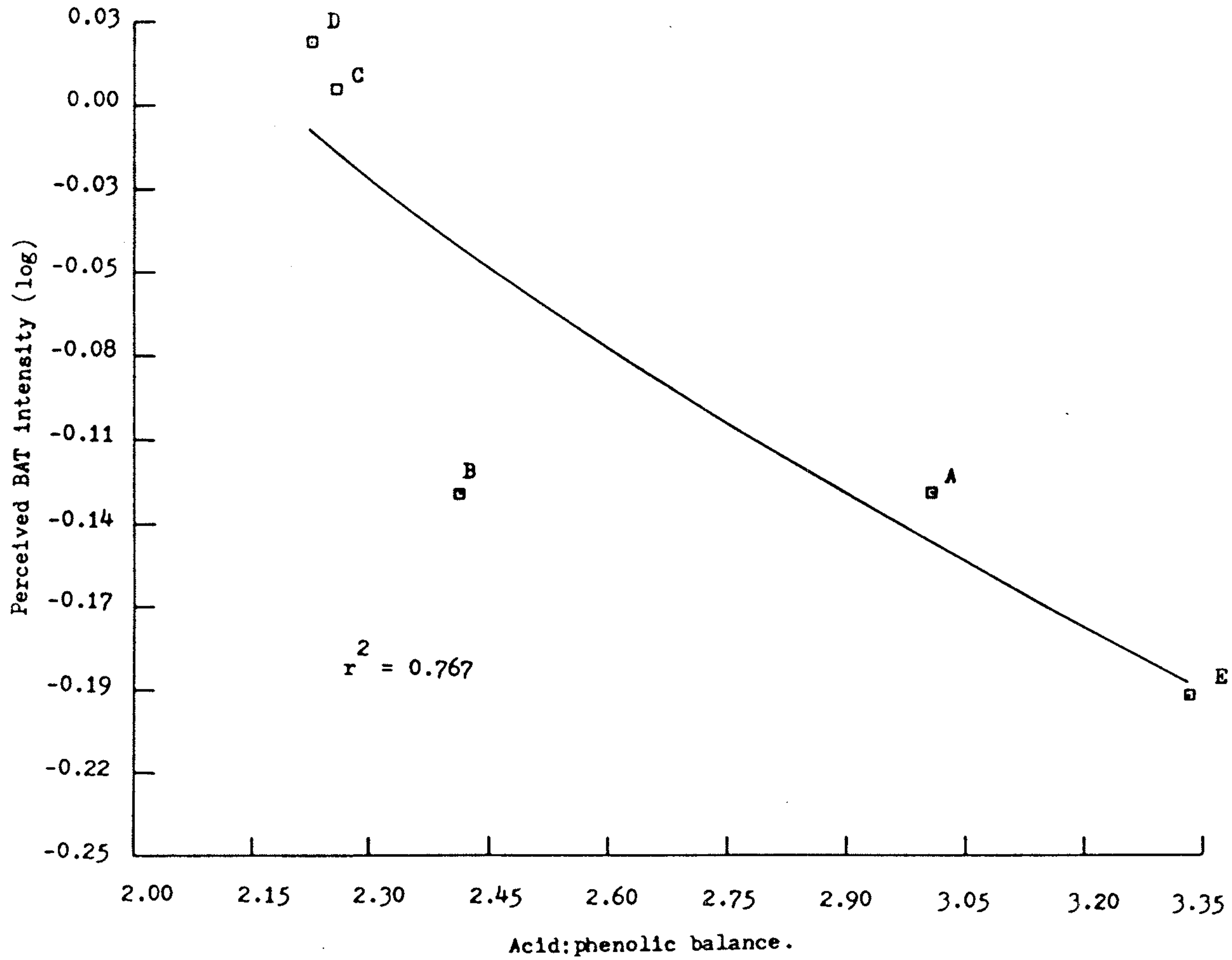
* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 15. Perceived bitter-astringent taste (BAT) intensity of apple juice as a function of sugar:phenolic balance.*



* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 16. Perceived bitter-astringent taste (BAT) intensity of apple juice as a function of acid:phenolic balance.*



* - A, B, C, D, and E are the code letters of commercial apple juices.

ness of apple juice is least affected by other taste contributing constituents of apple juice and BAT is the most affected taste property of apple juice. Since interaction among the taste properties cannot be discounted, the reason for these observed influences or the lack of them may be the relative concentrations of sugar, acid and phenolic contents. The sugar content of apple juice is much higher than the acid and phenolic contents of the juice and, sweetness did not exhibit the expected interactions. Acid content is higher than the phenolic content but lower than the sugar content of apple juice and sourness was affected by sugar content only, and not by the phenolic content. Phenolic content is the lowest of all the taste contributing constituents of apple juice and bitter-astringent taste was affected by sugar as well as the acid content of the juice.

4.6.5 Preference for Apple Juice

Preference scores for five commercial apple juices ranged from 5.67 to 7.88 on a scale of 1 (dislike extremely) to 9 (like extremely). Though all the juices were above the point of "neither like nor dislike" (or considered to be acceptable), significant differences in preference for apple juices were observed (Table 25 and Appendix 38). These differences indicated that the preference for even an otherwise acceptable juice may also be increased.

Preference for apple juice may be considered as an integrated response to the sensory properties of the juice. Since sensory properties are dependent upon the chemical properties

Table 25: Statistical comparision of preference for apple juices

Tukey's test for preferece scores[1]

Apple juice	Mean[2]
A	7.88 a
B	7.81 a
C	7.07 a b
D	6.85 a b
E	5.67 b

[1] - Alpha = 0.05, Df = 59.
[2] - Means with the same letter are not significantly different.

the preference could also be quantitatively integrated in terms of these chemical properties. The relationships for preference with the sensory and chemical properties of apple juice and their quantitative integration for preference optimization are presented in the following sections.

4.6.5.1 Preference and Sensory Properties

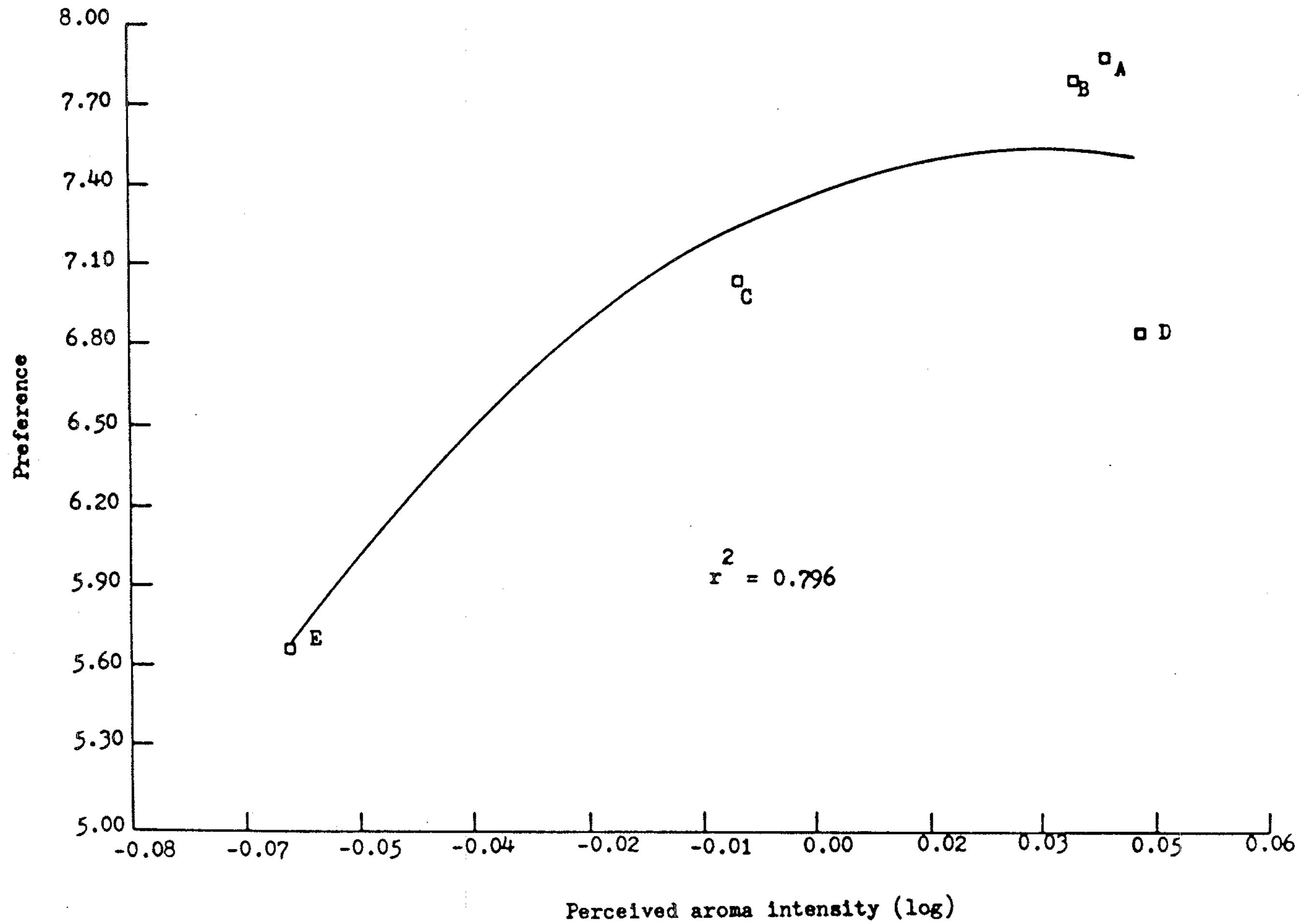
Using the limitations identified in section 3.4.6 and with the assistance of computer analysis, a series of mathematical relationships were investigated between preference and the other sensory properties that were scored by the panel. The parabolic curve ($Y = a + b_1 X + b_2 X^2$) best fit the relationship between preference and perceived aroma intensity (r^2 = 0.80, Table 26, and Fig. 17); and perceived BAT (r^2 = 0.99, Table 26, and Fig. 18). The most fit relationship for preference and perceived sweetness intensity was a three degree polynomial relationship (r^2 = 0.94, Table 26, and Fig. 19). The best fit model for the relationship between preference and the perceived sourness was a four degree polynomial curve (r^2 = 1.00, Table 26, and Fig. 20). Little practical information seems apparent from the relationships of sweetness and sourness with the preference of apple juice. The mathematical relationships between preference and perceived aroma intensity and perceived BAT are less complex and therefore of a greater practical value for preference optimization when all the properties are considered separately.

Table 26: Coefficients of determination and best fit regression models of the relationships for preference, with sensory and chemical properties of apple juice

Variable[1] X	r^2 Linear	r^2 Polynomial
Aroma intensity (log)	0.715	0.796 (2d)[2]
Sweetness intensity (log)	0.594	0.939 (3d)
Sourness intensity (log)	0.038	1.00 (4d)
BAT intensity (log)	0.038	0.985 (2d)
Sugar content	0.528	0.954 (2d)
Acid content	0.000	0.922 (3d)
Phenolic content	0.011	0.974 (3d)
Sugar:acid balance	0.076	0.789 (4d)
Sugar:phenolic balance	0.053	0.996 (3d)
Acid:phenolic balance	0.178	0.998 (2d)

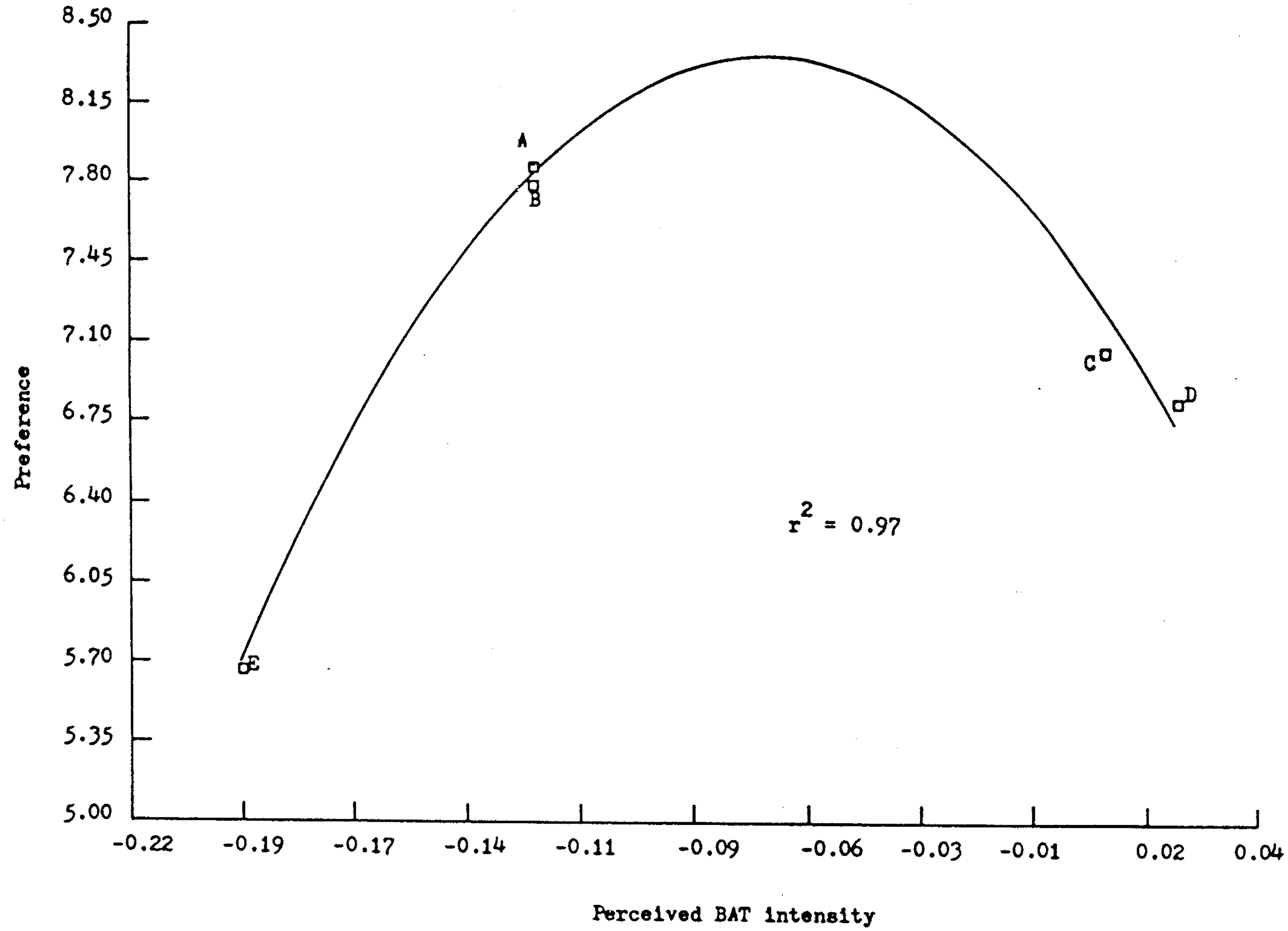
[1] - X or independent variable for dependent variable (Y) or mean preference score.
[2] - Polynomial degree.

Fig. 17. Preference for apple juice as a function of perceived aroma intensity.*



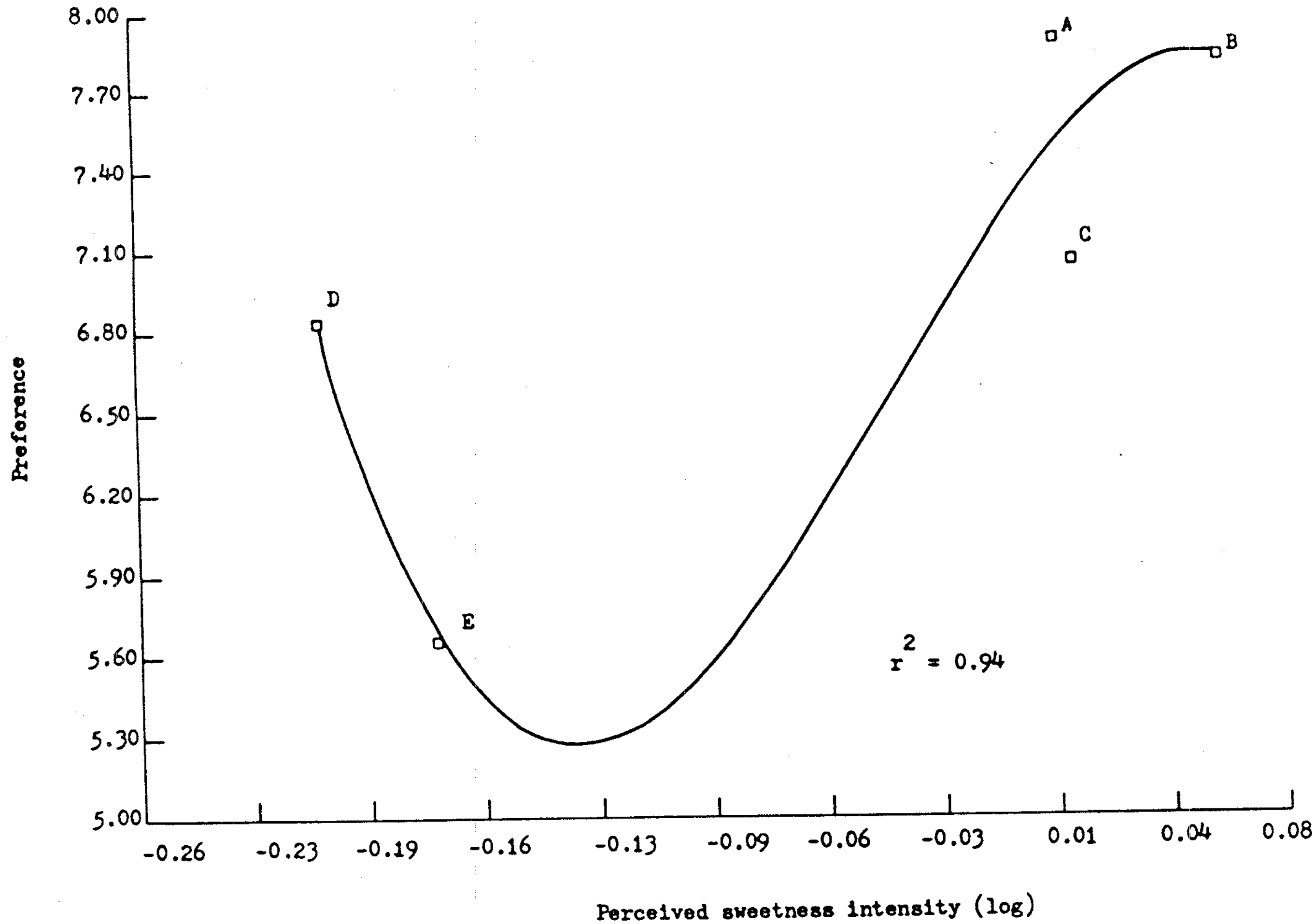
* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 18. Preference for apple juice as a function of perceived bitter-astringent taste (BAT) intensity.*



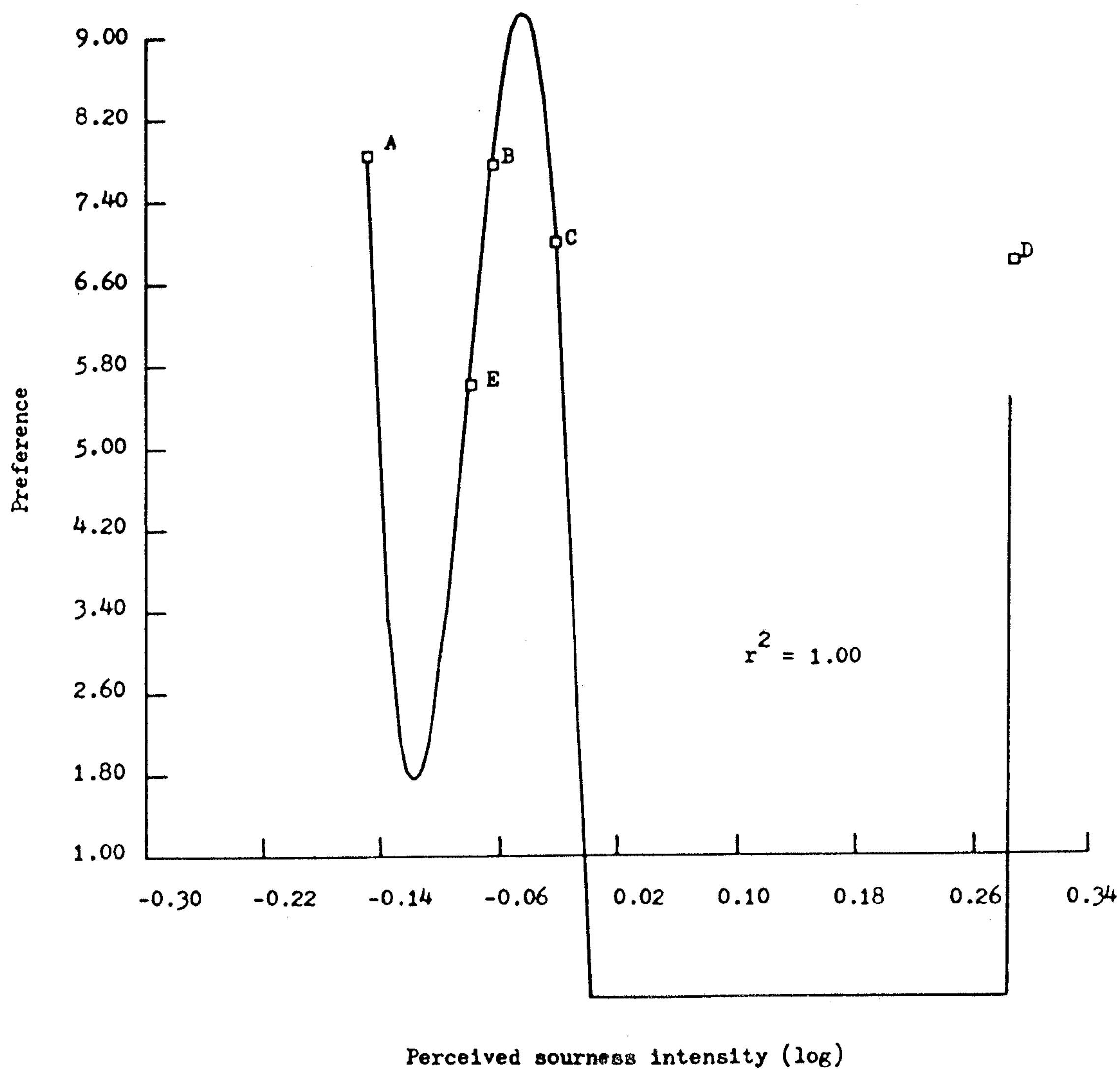
* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 19. Preference for apple juice as a function of perceived sweetness intensity.*



* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 20. Preference for apple juice as a function of perceived sourness intensity.*



* - A, B, C, D, and E are the code letters of commercial apple juices.

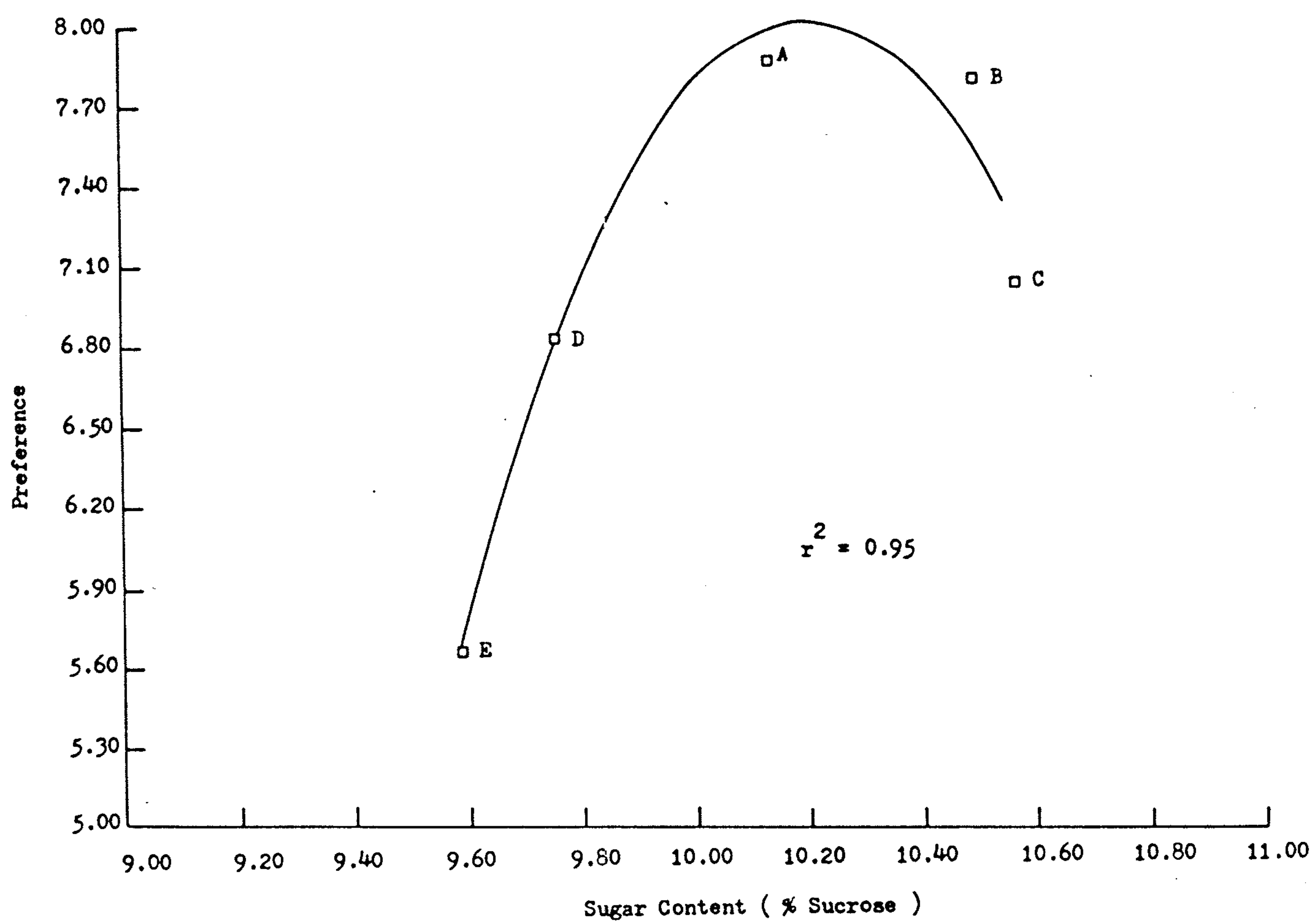
4.6.5.2 Preference and Chemical Properties

The procedure outlined in section 3.4.6 was also used to develop the best fit mathematical models of the relationships between chemical properties of apple juice and preference. The parabolic curves best fit the relationships of sugar content (r^2 = 0.95, Table 26, and Fig. 21), and acid:phenolic balance r^2 = 0.99, Table 26, and Fig. 22) with preference. The three degree polynomial curves best fit the relationships of acid content (r^2 = 0.92, Table 26, and Fig. 23), phenolic content (r^2 = 0.97, Table 26, and Fig. 24) and sugar:phenolic balance (r^2 = 1.00, Table 26, and Fig. 25) with preference. The relationship between the preference and sugar:acid balance could only be explained by a four degree polynomial curve (r^2 = 0.79, Table 25, and Fig. 26).

The significant relationship was observed by Poll (1981) between overall taste and sugar:acid balance of apple juice. That observation was that major factor in using sugar:acid balance for determining power function in the model system and in quantitative integration of the data. The significant relationship of sugar:acid balance with sweetness and sourness was observed in model system, and also in apple juice. However, within reasonable limits of experience the sugar:acid balance was of little practical value for preference optimization. Only it could be concluded that a broad sugar:acid balance range is acceptable to the panelists.

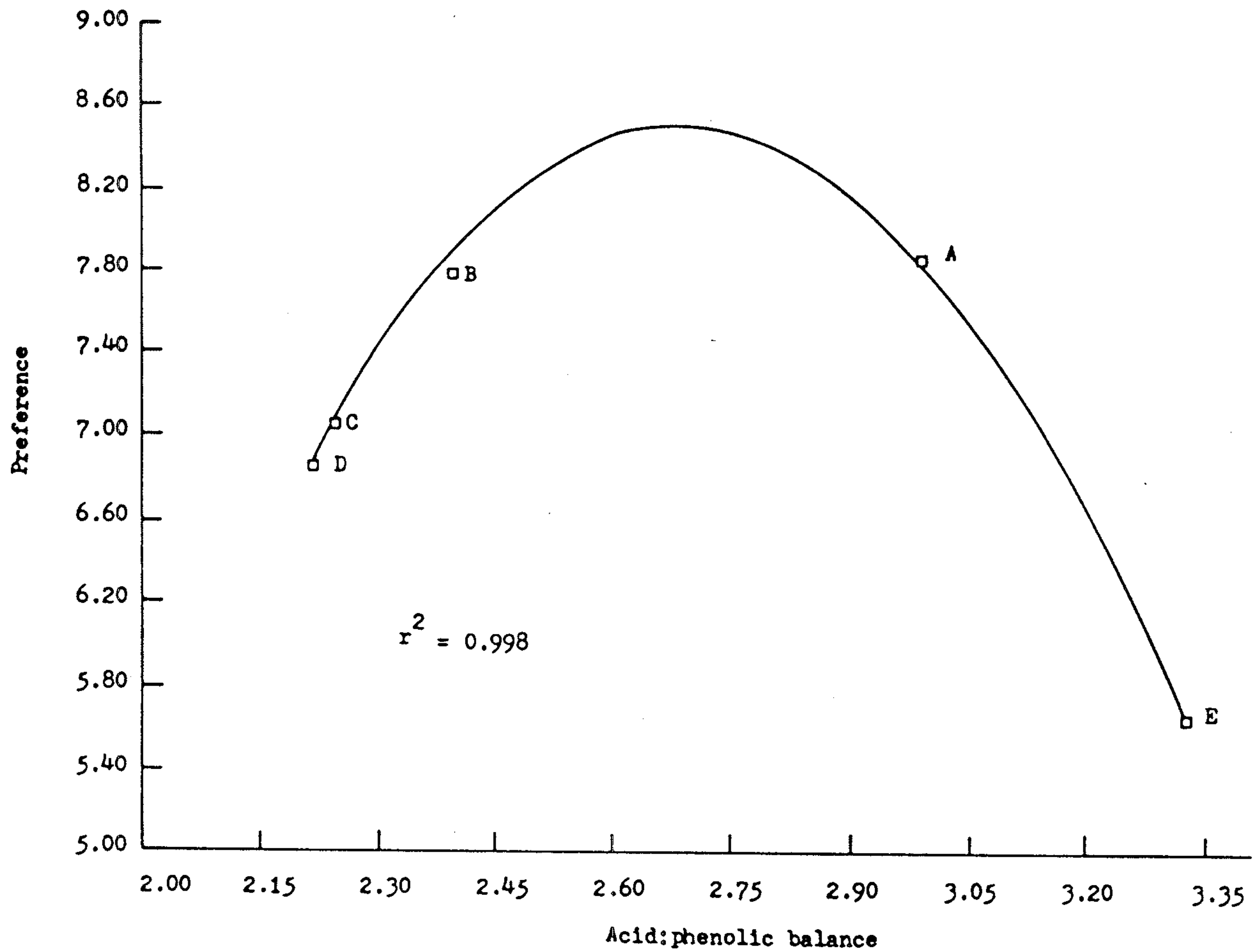
Chemical properties with less complex relationships with the preference, like sensory properties, may be more useful

Fig. 21. Preference for apple juice as a function of sugar content*



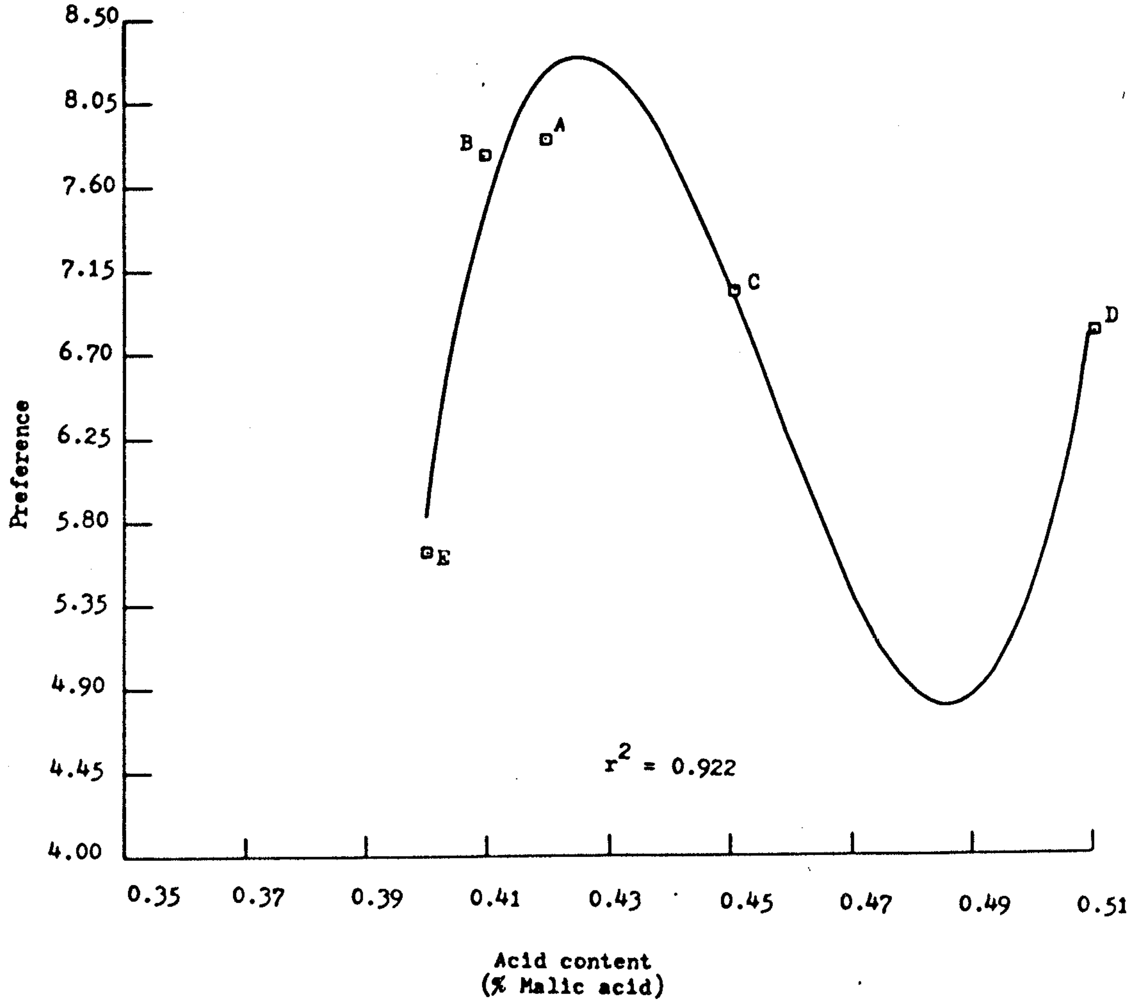
* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 22. Preference for apple juice as a function of acid:phenolic balance.*



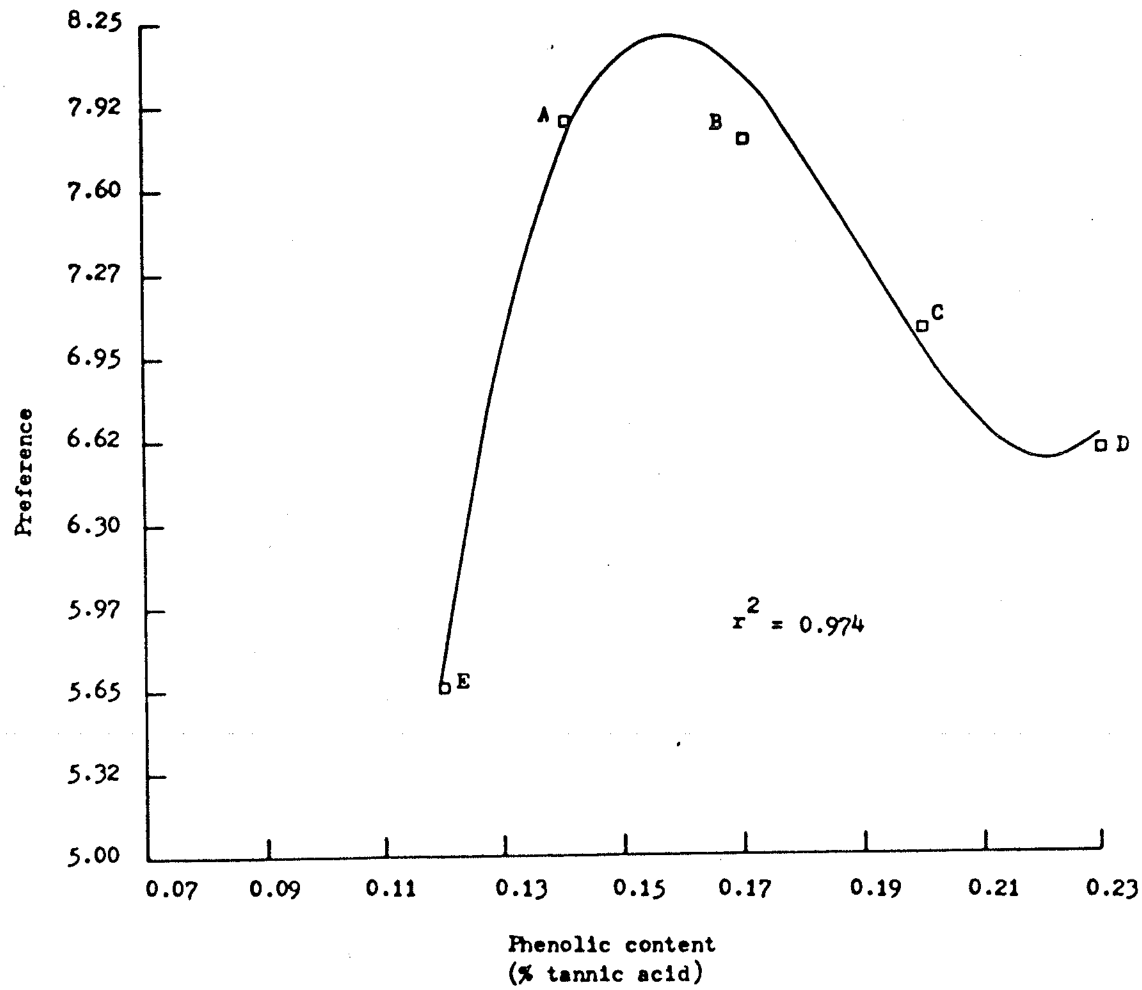
* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 23. Preference for apple juice as a function of acid content*



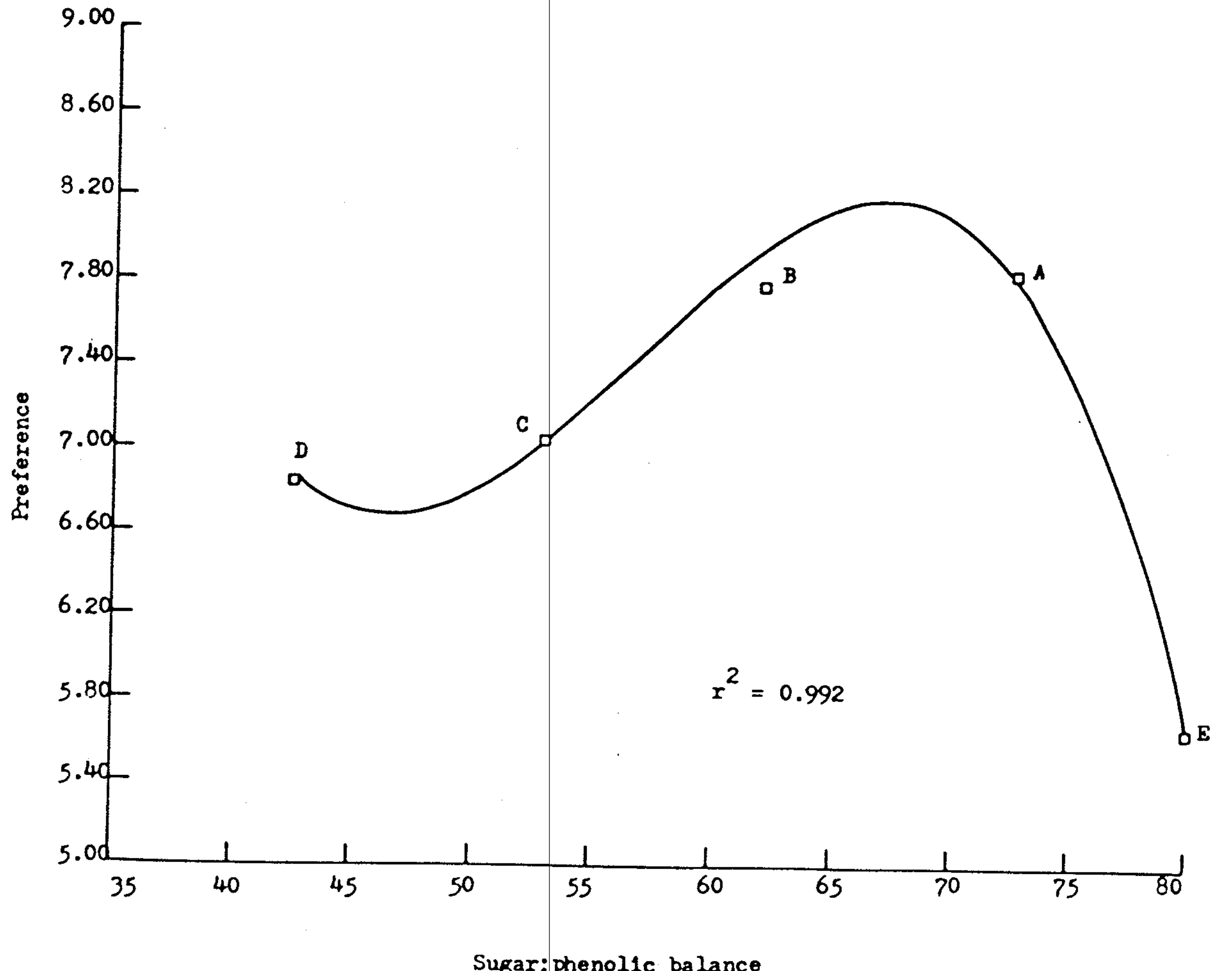
* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 24. Preference for apple juice as a function of phenolic content.*



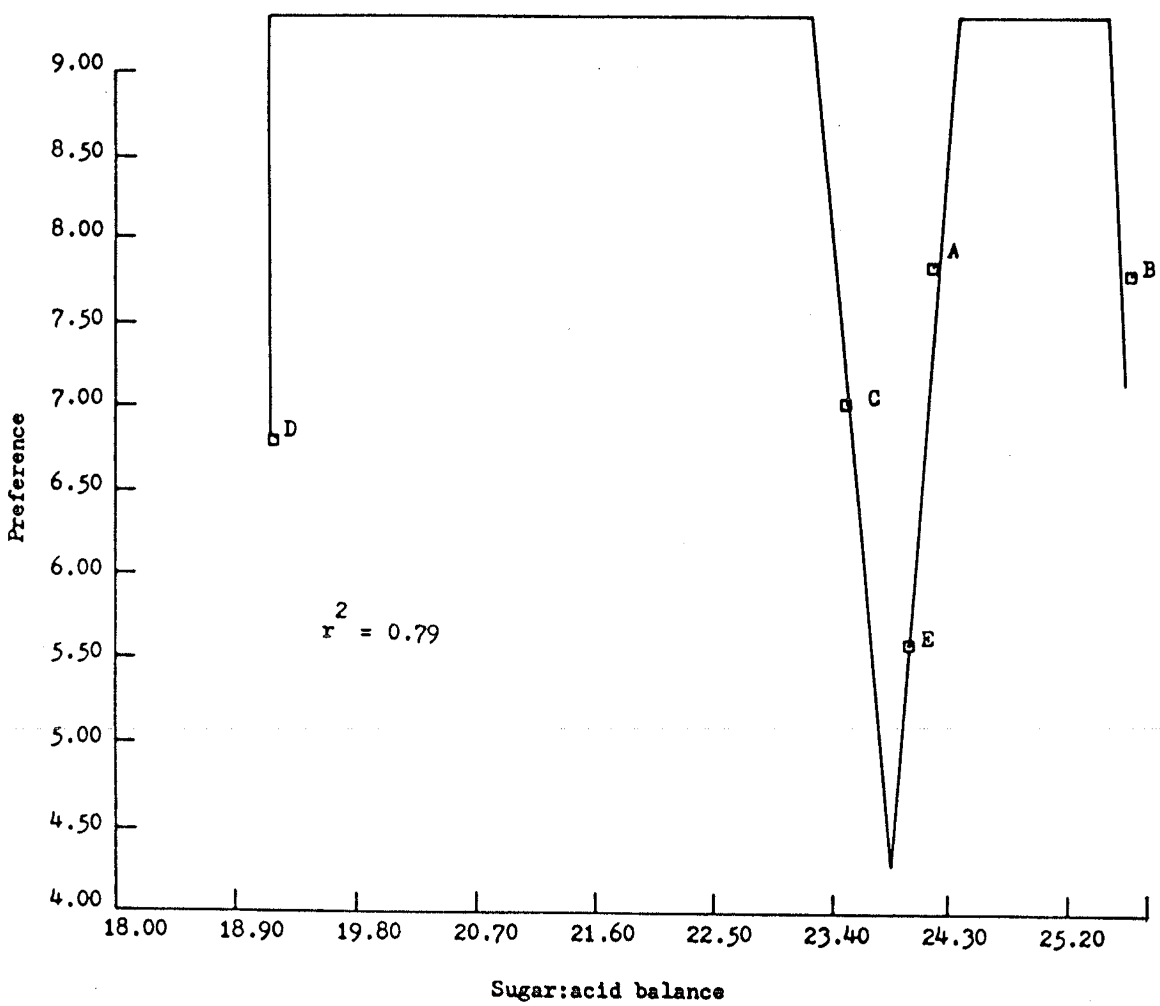
* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 25. Preference for apple juice as a function of sugar:phenolic balance.*



* - A, B, C, D, and E are the code letters of commercial apple juices.

Fig. 26. **Preference for apple juice as a function of sugar:acid balance.***



* - A, B, C, D, and E are the code letters of commercial apple juices.

for calculating preference optimization equations. These chemical properties are the sugar content and acid:phenolic balance.

4.6.6 Quantitative Integration of Psychophysical Relationships in Apple Juice

The sensory and chemical data collected from the five commercial apple juices (Appendix 36 and 37, respectively) were subjected to regression analysis. The polynomial relationships were found to best explain the interrelationships between these properties and preference (Table 26). The preference of apple juice was found to be positively as well as negatively affected by these properties. This indicated that the cause of differences in the preference for apple juices may be the combined effect of all the properties, which resulted from the levels of individual properties in the juices. To investigate this combined effect, relationships for preference with chemical, sensory and chemical and/or sensory properties were investigated by the multiple regression analysis. The objective of this analysis was to determine the contribution of chemical and sensory properties to the preference for apple juice. It also provided an equation which included all the important properties in a relationship, which is practical for preference optimization.

It was mentioned in section 2.4.5 that the preference could be quantitatively integrated in terms of sensory as well as chemical properties. Multiple regression analysis showed that quantitatively integrated taste sensory properties could explain 76% of the variability in apple juice preference (Table 27), and chemical properties explained only 66% of the preference variability (Table 28). Thus for the quantitative integration taste properties are more advantageous than the chemical properties. On the other hand determination of chemical properties is simple and less time consuming than the determination of taste properties. Therefore, considering relative advantages (variability explanation vs simple determination) any set of properties could be used for quantitatively integrating information for preference optimization.

However, when perceived aroma intensity was included in the regression model along with sensory (Table 29) or chemical (Table 30), preference variability could be explained completely. This compensated the advantage of taste sensory properties over chemical properties in explaining variability in preference. It also established that a combined relationship which includes perceived aroma intensity is essential. Considering the advantage of determination of chemical properties it can be stated that a combined relationship, including perceived aroma intensity and chemical properties would be more practical for preference optimization than a combined relationship which includes all the properties evaluated by sensory analysis.

At present, only complex chromatographic methods are available for the determination of aromatic compounds, which unlike the determination of sugar, acid and phenolic contents

Table 27: A stepwise r^2 improvement for preference and preference optimization equations for apple juice. I

Step	Variable[1] x1 - 3	Regression equation[2]	r^2
1	Sweetness intensity (log)	Y = 7.40 + 5.72 X1	0.594
2	Sourness intensity (log)	Y = 7.57 + 8.12 X1 + 2.67X2	0.745
3	BAT intensity (log)	Y = 7.44 + 9.45 X1 + 4.38 X2 - 2.70 X3	0.764

[1] - Variables X1, X2 and X3 entered in stepwise multiple regression analysis.
[2] - Y or dependent variable is preference for apple juice.

Table 28: A stepwise r^2 improvement for preference and preference optimization equations for apple juice. II

Step	Variable[1] x1 - 3	Regression equation[2]	r^2
1	Sugar content (% sucrose)	$Y = -7.91 + 1.48\ X1$	0.528
2	Acid content (% malic acid)	$Y = -9.35 + 1.51\ X1 + 2.46\ X2$	0.543
3	Phenolic cotent (% tannic acid)	$Y = -29.32 + 2.71\ X1 + 33.01\ X2 - 31.89\ X3$	0.664

[1] - Variables X1, X2 and X3 entered in stepwise multiple regression analysis.
[2] - Y or dependent variable is preference for apple juice.

Table 29: A stepwise r^2 improvement for preference and preference optimization equations for apple juice. III

Step	Variable[1] x1 - 4	Regression equation[2]	r^2
1	Aroma intensity (log)	Y = 6.86 + 16.85 X1	0.715
2	Sweetness intensity (log)	Y = 7.15 + 13.25 X1 + 4.11 X2	0.989
3	Sourness intensity (log)	Y = 7.05 + 15.42 X1 + 2.97 X2 - 0.98 X3	0.999
4	BAT intensity (log)	Y = 7.07 + 15.79 X1 + 2.50 X2 - 1.52 X3 + 0.71 X4	1.00

[1] - Variables X1, X2, X3 and X4 entered in stepwise multiple regression analysis.
[2] - Y or dependent variable is preference for apple juice.

Table 30: A stepwise r^2 improvement for preference and preference optimization equations for apple juice. IV

Step	Variable[1] x1 - 4	Regression equation[2]	r^2
1	Aroma intensity (log)	Y = 6.86 + 16.85 X1	0.715
2	Sugar content (% sucrose)	Y = - 2.82 + 13.24 X1 + 0.96 X2	0.905
3	Acid content (% malic acid)	Y = - 1.87 + 16.92 X1 + 0.98 X2 - 7.19 X3	0.996
4	Phenolic cotent (% tannic acid)	Y = -6.21 + 16.13 X1 + 1.24 X2 + 6.25 X3 - 13.00 X4	1.00

[1] - Variables X1, X2, X3 and X4 entered in stepwise multiple regression analysis.
[2] - Y or dependent variable is preference for apple juice.

do not provide a practical alternative to the use of sensory evaluation. Therefore, a combination of sensory and chemical properties which may be most practical for preference optimization is presented in the following section.

4.6.6.1 Preference Optimization Equation for Apple Juice

Aroma, sugar, acid and phenolic contents have been previously reported to have the major influence on overall taste and aroma (flavor) of the apple juice (Poll, 1981). Since overall taste would influence preference for apple juice ,these properties would also determine juice preference. The results of this investigation also support this assumption. Results also indicate that the perceived aroma intensity of apple juice is the most important factor for preference determination followed by sugar, acid and phenolic contents of the juice, in that order. However, the separate effects of these properties on preference were not linear (Table 26). Therefore, to include these properties in a linear equation for preference optimization, a predetermined entry level (i.e. alpha = 0.05 or 0.10) could not be used without ignoring the effects of some of the important properties of apple juice.

A combination of properties was used for developing preference optimization equation. This combination included all of the important properties either directly (perceived aroma intensity and sugar content) or indirectly (acid:phenolic balance in lieu of acid and phenolic contents). This combination was selected based on the maximum possible linearity for preference when factors are considered separately, without excluding any important property. The regression equation for preference optimization was developed by the maximum r^2 improvement technique (Table 31). The equation is Y (preference) = -8.11 + 16.99 (log aroma intensity) + 1.28 (sugar content) + 0.76 (acid:phenolic balance), $r^2 = 0.99$.

4.6.7 Implication

This study was conducted and is presented to show the "in principle" feasibility of psychophysical analysis of apple juice for preference optimization. It has the obvious limitation that it has not been applied in a practical situation to assess the value of the optimization equation for preference optimization. It does however, identified important properties of apple juice, which can be used for preference optimization. It is presented to suggest a basis for blending operations in the production of apple juice. In the production of apple juice where blending is performed during the crushing operation, studies conducted for blending would be required to monitor the changes in the juice which occur during crushing and subsequent enzyme clarification. These changes were not monitored during the present investigation. However, for the production of apple juice from concentrate only reconstitution with known dilution factor is done. Therefore, blending studies conducted on apple juice could be sufficient and equation such as presented in the last section could be employed in the blending of concentrates for the production of apple juice.

Table 31: A stepwise r^2 improvement for preference and preference optimization equations for apple juice. V

Step	Variable[1] x1 - 3	Regression equation[2]	r^2
1	Aroma intensity (log)	Y = 6.86 + 16.85 X1	0.715
2	Sugar content (% sucrose)	Y = - 2.82 + 13.24 X1 + 0.96 X2	0.905
3	Acid:phenolic balance	Y = - 8.11 + 16.99 X1 + 1.28 X2 + 0.76 X3	0.999

[1] - Variables X1, X2, and X3 entered in stepwise multiple regression analysis.
[2] - Y or dependent variable is preference for apple juice.

5.CONCLUSIONS

The present investigation was initiated to characterize ten apple cultivars grown on the Canadian Prairie. These apple cultivars were chemically characterized to evaluate their suitability for the production of processed products. The chemical characterization was further extended to determine their sugar and acid profiles by chromatographic analysis. As a possible avenue for the utilization of apple cultivars preliminary development of filtration processing was carried out. Preliminary development included designing of a pilot plant scale filtration assembly, experiments to suggest design improvement and a parameter for process control, and physico-chemical characterization of filtration processing of apple juice. Psychophysical analysis of five commercial juices was conducted to develop an understanding of sensory properties of apple juice to assist in the blending operation for producing a high quality juice from these apple cultivars.

As mentioned in the introduction overall studies were designed to meet practical as well as academic interests related to the utilization of apple cultivars. Therefore the conclusions in this section are also presented to meet both interests of this study. The findings of this study include:

1. Ten apple cultivars which were analysed in this study had higher than average moisture content.

2. Most of the cultivars had average sugar content except for Goodland, Heyer # 12 and P.F. 36 which had below average sugar content.

3. P.F. 51 and Norland had medium acid content, while other cultivars were found to have a high acid content.

4. These apple cultivars had high phenolic content except P.F. 51 which was found to have a medium phenolic content.

5. P.F. 51 and Norland were most suitable for the production of processed apple products. However, other cultivars could also be used for the production of liquid and pureed processed apple products, if proper blending could be planned.

6. These cultivars contained a lower than average concentration of sucrose but average concentrations of fructose and glucose.

7. The predominant sugar in these cultivars was fructose, followed by glucose and sucrose in that order.

8. The most frequently occuring minor sugar in these cultivars was xylose.

9. The major organic acids in these cultivars were malic, quinic, citric and shikimic.

10. The chromatographic analysis provided higher total sugar content than that determined by standard method.

11. In principle, it could be possible to arrange available chromatographic equipment to provide a faster and reliable method for the determination of total sugar content.

12. Turbidity was found to be a good indicator of filter performance (a measure of the removal of suspended particles from the juice) during filtration processing of apple juice.

13. Using turbidity various aspects of filtration processing e.g. designing of filtration assembly, recycling and process control could be conducted

satisfactorily.

14. During filtration processing significant decreases in acid and phenolic contents, and increase in clarity of apple juice was observed to occur.

15. Filtration processing could be expected to increase the preference for apple juice.

16. The sweetness of apple juice was found to be least affected and BAT most affected by other taste contributing constituents of apple juice.

17. On the basis of simpler relationships with preference for apple juice, perceived aroma intensity and perceived BAT of juice could be considered to have greater practical value for the preference determination than the perceived sweetness and sourness of the juice.

18. On the same basis, sugar content and acid:phenolic balance had greater practical application for preference determination than the acid content, phenolic content, sugar:phenolic balance and sugar:acid balance of the juice.

19. The perceived sensory properties were found to be more important than the chemical properties of apple juice for the determination of preference for the juice.

20. The most practical combination of variables for preference optimization included perceived aroma intensity, sugar content and acid:phenolic balance of apple juice.

21. The most important variables for preference were perceived aroma intensity followed by sugar content and acid:phenolic balance, in that order.

6. RECOMMENDATIONS FOR FUTURE STUDY

To overcome some of the practical limitations (e.g. seasonal variation in chemical composition of apple cultivars), to explore the possiblities raised and to verify the information presented in this study the following recommendations are made for future study.

1. To assess seasonal variations, chemical characterization of apple cultivars should be extended for several seasons.

2. The confirmation of acid presence be conducted to complete acid profile of these cultivars.

3. The available chromatographic equipment should be arranged to test if a fast, reliable method for the determination of total sugar content is practically possible.

4. The suggested alterations in filtration assembly should be made to make filtration assembly more efficient.

5. The pressure recordings (a common parameter for process control) after every filter should also be taken with turbidity, to compare turbidity with pressure as a process monitoring parameter.

6. The sugar content during filtration processing be monitored by a more sensitive method.

7. The specific gravity and ash content changes in apple juice during filtration processing should also be monitored because their levels in apple juice constitute the regulatory guidelines for commercial juice production in Canada.

8. Psychophysical analysis of apple juice be conducted at a larger scale, and using factors identified in this study blending of apple juices from these cultivars should also be carried out to test practical application of such studies.

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APPENDICES

Appendix 1

Sample information

Cultivar	Type	Trees Harvested (no.)	Harvest Date	Analysis Date
Breakey	Apple	1	23-8-82	5-10-82
Collet	Apple	7	8-9-82	17-10-82
Goodland	Apple	5	A	4-10-82
Heyer# 12	Apple	1	B	3-10-82
Kerr	Crabapple	9	24-9-82	18-10-82
Luke	Apple	2	24-9-82	15-10-82
Norland	Apple	1	C	6-10-82
P.F. 36	Apple	2	8-9-82	15-10-82
P.F. 50	Apple	3	8-9-82	14-10-82
P.F. 51	Apple	2	1-9-82	12-10-82

A - First week of August.
B - First week of August.
C - Third week of August.

Appendix 2

Statistical analysis of phenolic extraction procedures

Analysis of variance (ANOVA)[1]

Source	Df	Sum of Squares	Mean Square	F[2]
Alcohol	2	3615.05	1807.53	17.27 **
Time	1	2373.87	2373.87	22.68 **
Sample	1	200.74	200.74	1.92
Replicate	1	187.32	187.32	1.79
Alc x Time	2	667.02	333.51	3.19
Alc x Sample	2	146.12	73.06	0.70
Alc x Rep.	2	139.41	69.71	0.69
Error	12	1255.81	104.65	

[1]Phenolic content expressed as mg Tannic acid/100g.
[2]F values significant at alpha 0.05 are indicated by **.

Appendix 3

A stepwise method for the determination of correlation between number of suspended particles and turbidity of apple juice

1. Prepare 1-2 liters enzyme clarified juice.

2. Filter 250 ml enzyme clarified juice through a 0.22 μm filter.

3. Prepare seven - eight dilutions in duplicate, using filtered juice as diluent.

4. Assign relative number of suspended particles to each dilution using the formula given in Table 4.

5. Determine turbidity (T) of diluted juice samples and also of diluent (t).

6. Calculate dT for each dilution.

   $$dT = T - t$$

7. Correlate dT and relative number of suspended particles for all dilutions by regression analysis.

8. Report r (correlation coefficient) and r^2 (coefficient of determination) and check significance of r at $p < .05$.

Appendix 4

Ballot used in introductory panel

Name ....   Date...............

Please, pick up the card deck which has been given to you. Do not look through the deck, but pay attention only to the top card. The top card has a Circle labeled "REFERENCE CIRCLE". The reference circle has been assigned an arbitrary number 20 to indicate its size, based on its Area.

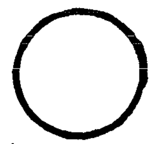
Now, flip to the 2nd card. Give it a number which represents the area or size of the 2nd circle as compared to the reference circle. For instance; if the 2nd circle seemed to have approximately same size or area as the reference circle, you give it a score of 20. If the 2nd circle seemed only one-half as large as reference circle, then you would give it a score of 10. If the 2nd circle appeared to you five times as large as reference circle, then you would give it a score of 100.

Now, flip to the 3rd card and evaluate the size of the 3rd circle in the same manner. Then do the scoring for all the cards in the deck. Please, do not look ahead, but evaluate each card as you come to it.

Card	No
Reference	20
....	......
..2nd..	......
..3rd..	......
..4th..	......
..5th..	......

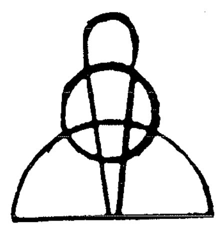
Appendix 5

Ballot diagrams used in introductory panel[1]



No. 1

No. 2



No. 3

[1] - Diagrams no. 1, 2 and 3 were presented separately on cards in five different sizes including a Reference. These diagrams show the actual size of reference circles.

Appendix 6

Concentrations of stimuli used in orientation panels

% Sucrose[1]	% Citric acid[1]	% Caffeine[1]	% Artificial[2] apple flavor
9.00	0.34	0.01	0.01
11.50	0.67	0.02	0.02
14.00[3]	1.00[3]	0.04[3]	0.04
16.50	1.34	0.08	0.08[3]
19.00	1.67	0.16	0.16
		0.34	
		0.64	

[1]Concentration as wt/vol (in distilled water).
[2]Concentration as vol/vol (in distilled water).
[3]Used as reference.

Appendix 7

Ballot used in aroma orientation panel

Name................ Date...........

Evaluate AROMA of each sample in the order indicated below and assign any value to it relative to Reference (R). For each evaluation (both sample and R) remove the cap from the vial and feel aroma by taking three short sniffs. Take few long sniffs to clear your nostril between samples. If you cannot detect any aroma use "NP" to denote not present. Please, do not use zero.

Aroma Intensity

Sample	Magnitude Estimate
Reference	20
....	......
....	......
....	......
....	......
....	......
....	......

Please, feel aroma of covered vial and compare it with R...

- can aroma of R and samples be defined as Fruity aroma - Yes/No

- can aroma of R and sample be defined as apple aroma - Yes/No

Comment: Suggest along with your other suggestions, any improvement in the method, to make it more convenient for aroma evaluation.

Appendix 8

Ballot used for the aroma reference selection

NAME; DATE:

Smell these samples in the order indicated and check hcw close you find Aroma of each sample to Apple Aroma.

For evaluation of aroma, Ist swirl the vial, then remove the cap from vial and smell the sample by taking three short sniffs. Take few long sniffs to clear your nostrils between samples.

Please, do not compare samples, make seperate judgement on each sample.

Samples;

____	____	____	____	____	____
___ Extremely close	___ Extremely close	___ Extremely close	___ Extremely close	___ Extremely close	___ Extremely close
___ Very much close	___ Very much close	___ Very much close	___ Very much close	___ Very much close	___ Very much close
___ Moderately close	___ Moderately close	___ Moderatley close	___ Moderately close	___ Moderately close	___ Moderately close
___ Slightly close	___ Slightly close	___ Slightly close	___ Slightly close	___ Slightly close	___ Slightly close
___ Not at all	___ Not at all	___ Not at all	___ Not at all	___ Not at all	___ Not at all

Comments:

Appendix 9

Statistical analysis of aroma reference selection data

Analysis of variance (ANOVA)

Source	Df	Sum of square	Mean square	F[1]
Aroma Sample	2	40.78	20.39	17.74 **
Panelists	8	9.33	1.17	1.02
Replicates	1	0.46	0.46	0.40
Error	42	48.26	1.15	

[1]F values significant at alpha 0.05 are indicated by **.

Appendix 10

Adjustment of sugar-acid balance by modification of sugar or acid concentration during stimuli selection

Sugar:Acid Balance	Adjusted by Sugar (% Sucrose)[1]	Acid (% Citric acid)[2]
1.25	0.50	3.20
2.50	1.00	1.60
5.00	2.00	0.80
10.00[3]	4.00	0.40
20.00	8.00	0.20
40.00	16.00	0.10
80.00	32.00	0.05

[1]Acid concentration fixed at 0.40 .
[2]Sugar concentration fixed at 4.00 .
[3]Used as reference.

Appendix 11

Lists of measures taken to eliminate or minimize factors affecting panelist judgement

Error	Control measures
Expectation	- Three digit random coding - provide least possible information to panelists
Stimulus and Logical	- use of red light to mask color - uniform pattern of sipping or sniffing from sample to sample
Halo	- preference test after stimuli evaluation to delay formation of a general impression
Suggestion	- instructions to avoid discussion - allowing time convenience, which reduces panelist meetings
Motivational	- treats - time convenience - if possible, allowing panelists to choose their own sequence of stimuli evaluation.
Contrast	- randomization during power function determination - evaluation of only one apple juice in a session

Appendix 12

Ballot used in evaluation of aroma

Name.................... Date...........

Your task is to evaluate Apple aroma of each sample in the order indicated and assign it any value relative to the Reference (R). For each evaluation (both samples and reference) swirl the vial; remove the cap and smell sample by taking three short sniffs. Take few long sniffs to clear your nostril between samples. If you can not detect any aroma in a sample use "np' to denote not present. Please. do not use zero.

Sample	Magnitude Estimate
Reference	20
....	......
....	......
....	......
....	......
....	......
....	......

Comments:

Appendix 13

Ballot used in evaluation of BAT.

Name.................... Date.............

Your task is to evaluate Bitter - Astringent Taste in the sample, and assign it any value relative to Reference (R). For each evaluation, both sample and reference, remove the aluminum foil which covers the cup and sip the sample. Take the same amount for each evaluation. Spit out the sample and rinse your mouth with water and cracker between samples. Do not hurry as there are only two evaluation to do. Finish evaluation of both. the sample and reference in first trial. If you can not detect any Bitter - Astringent Taste in a sample use "np" to denote not present. Please, do not use zero. Do not swallow the sample.

Sample	Intensity Estimate
Reference	20
....	....
....	....

Comments:

Appendix 14

Ballot used for the evaluation of sweetness and sourness

NAME:------------------

DATE:------------------

All the samples are to be evaluated for two parameters.

Evaluate the samples in the order indicated.

The order of evaluation for parameters is of your choice. For example, you may like to evaluate all the samples first for Sweetness and then for Sourness, or you may like to evaluate all the samples first for Sourness and then for Sweetness.

Evaluate the Reference of the parameter which you have decided to take up first and assign it a score of 20. Now evaluate all the samples for the first parameter in relation to the Reference only. For example, if a sample seems half as intense as the reference, give it a score of 10. If a sample seems four times as intense as the reference, give it a score of 80.

Once you have evaluated the samples for the first parameter, evaluate the Reference of the second parameter and assign it a score of 20 also. Now, you are ready to evaluate samples for the second parameter in relation to its Reference.

For each evaluation (both samples and reference) try to sip the same amount, spit out the sample and rinse your mouth with water and cracker between samples. Do not swallow the samples. You may try the reference again if you forget its intensity.

Use 'np' if the parameter is not present in the sample.

Samples:		___	___	___	___	___	___	___
Ist Parameter	___	___	___	___	___	___	___	___
2nd Parameter	___	___	___	___	___	___	___	___

Appendix 15

Ballot used for the evaluation of apple juice samples.

Name: **Date:**

The Apple Juice sample is to be evaluated for four parameters; Aroma, Bitter-Astringent Taste, Sweetness and Sourness.

Evaluate the sample for all the parameters (in the order indicated) in relation to their respective References.

Use 'np' if a parameter is not detectable in the sample. Rinse your mouth or clear your nostril as required using usual porcedures.

After evaluating sample for all the parameters, indicate your preference by making appropriate check mark.

Do not swallow the sample while evaluating it for individual parameters. However, for preference evaluation you can swallow the sample.

Parameter Intensity Score

Parameter	Reference	Sample
Aroma	20	------
Sourness	20	------
Sweetness	20	------
Bitter-Astringent Taste	20	------

Check how much you like or dislike the sample:

------ Like extremely

------ Like very much

------- Like moderately

------ Like slightly

------ Neither like or dislike

------ Dislike slightly

------ Dislike moderately

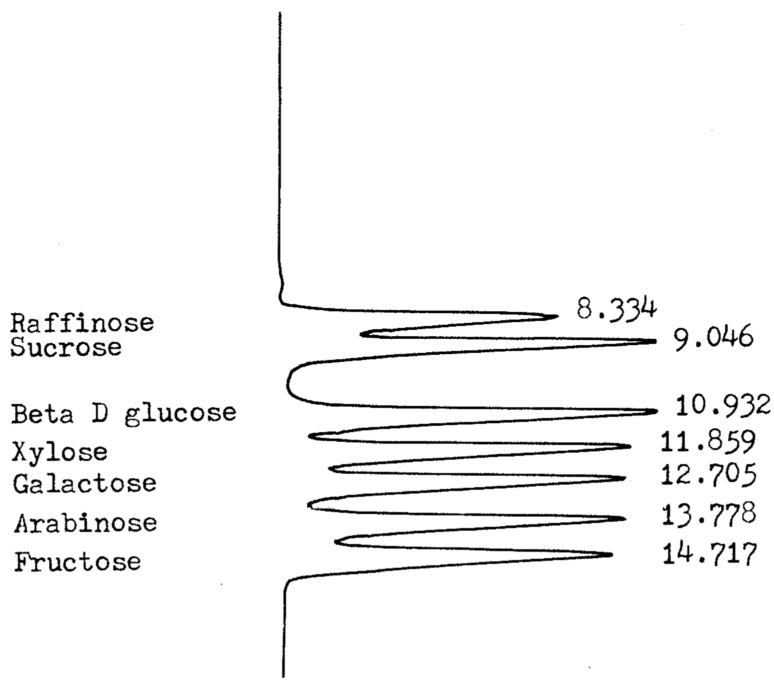
------ Dislike very much

------ Dislike extremely

Comments: List the reasons for your like or dislike, with the help of above mentioned parameters. Please, feel free to mention any other relevant parameter. Use the back side of the page for your comments, if required.

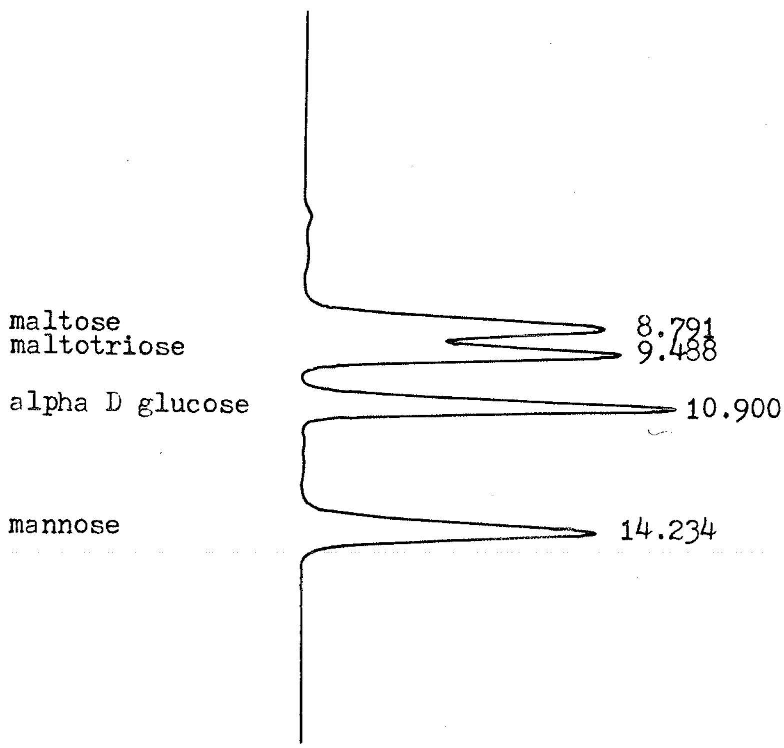
Appendix 16

A representative HPLC chromatogram of sugars
from a standard mixture



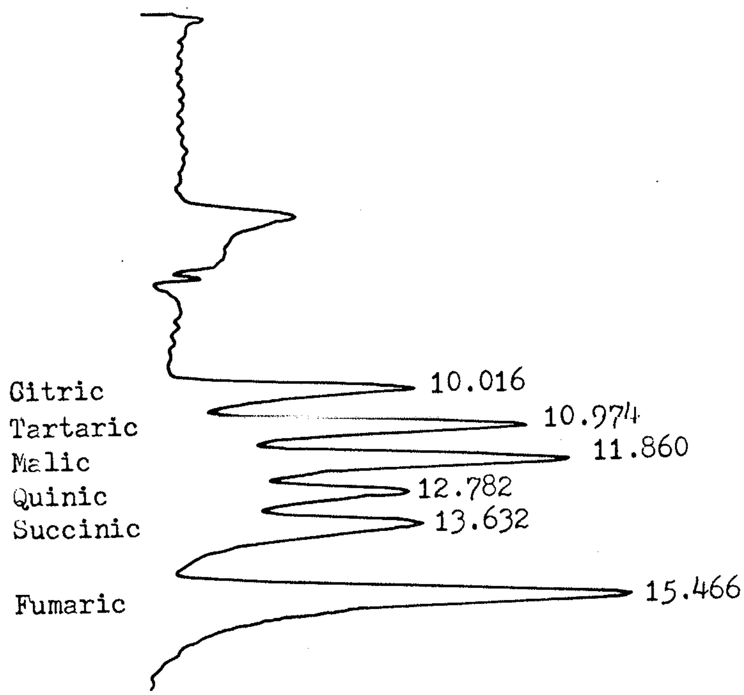
Appendix 17

A representative HPLC chromatogram of sugars from a standard mixture



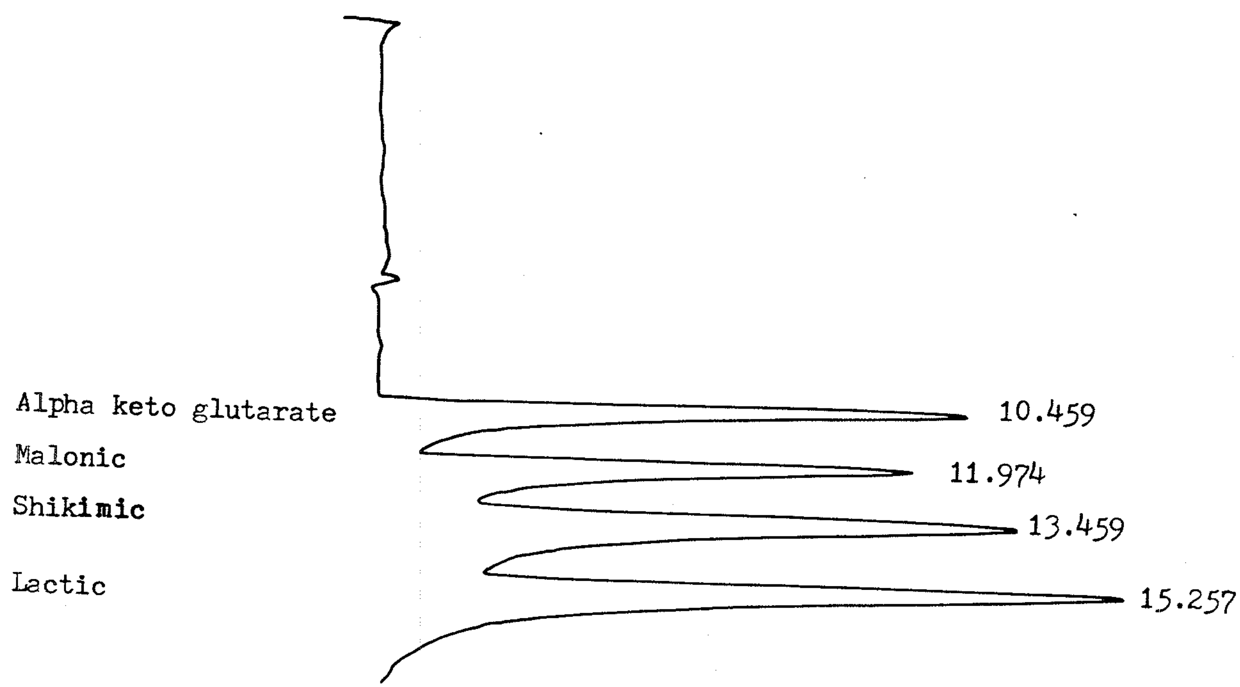
Appendix 18

A representative HPLC chromatogram of organic acids from a standard mixture



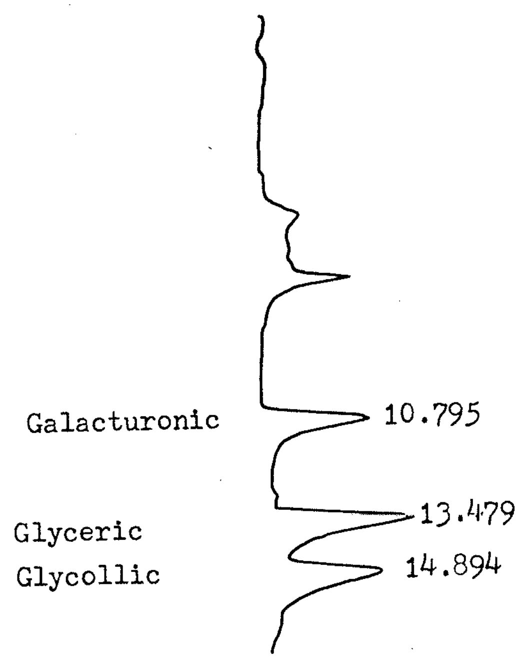
Appendix 19

A representative HPLC chromatogram of organic acids from a standard mixture



Appendix 20

A representative HPLC chromatogram of organic acids from a standard mixture



Appendix 21

Calculations for the minimum detectable concentration

Minimum detectable concentration (%) of a sugar or acid was calculated by the formula used for external standard results during HPLC analysis. Factors which were required for calculation include area rejection limit (area units), response divisor. After including required factors formula for external standard result calculations (Anonymous, 1981 b) could be rewritten as;

$$\text{Minimum Detectable Concentration (\%)} = \frac{500\ Ri\ m}{d\ 10000}$$

Where;

500 = experimental area rejection limit
Ri = response factor
m = experimental multiplier
d = experimental divisor (=1)
10000 = constant scaling factor

Multiplier m was calculated by following formula;

$$m = \frac{0.10\ df}{v}$$

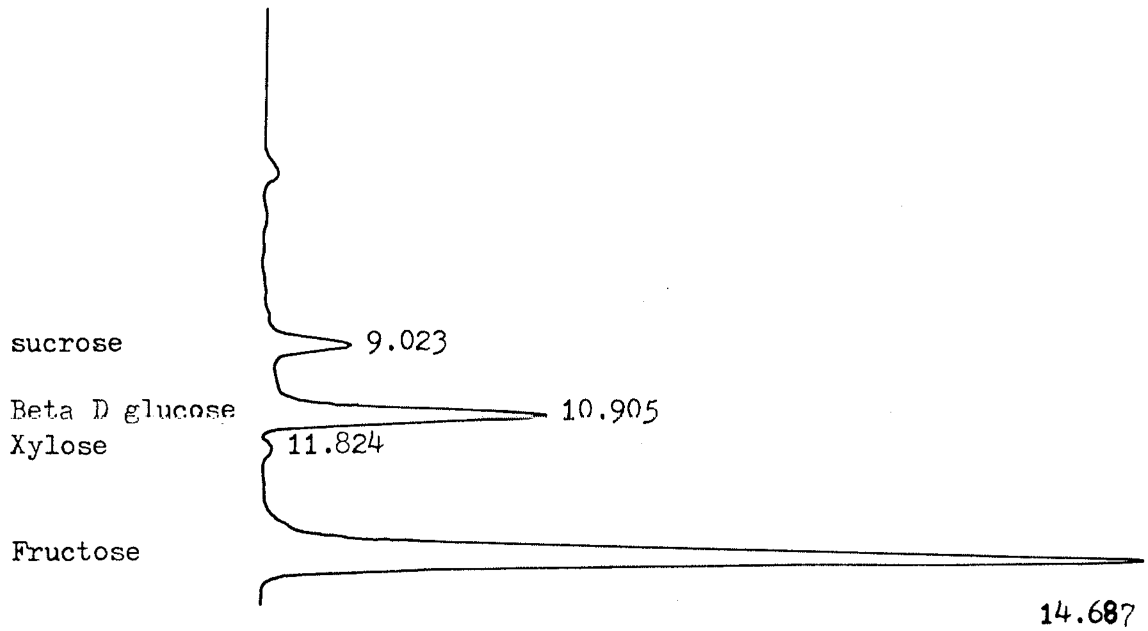
Where;

0.10 = factor to convert g/ l to g/100 ml
df = dilution factor
v = injection volume (μl)

Dilution factors for apple tissue and apple juice samples were 6.67 and 1 respectively. Injection volumes for apple tissue samples in acid and sugar analysis were 33.33 and 35 μl respectively and for apple juice sample injection volume was 5 μl. These dilution factors and injection volumes provided a m value of 0.02.

Appendix 22

A representative HPLC chromatogram[a] of sugars from apple and apple juice samples.

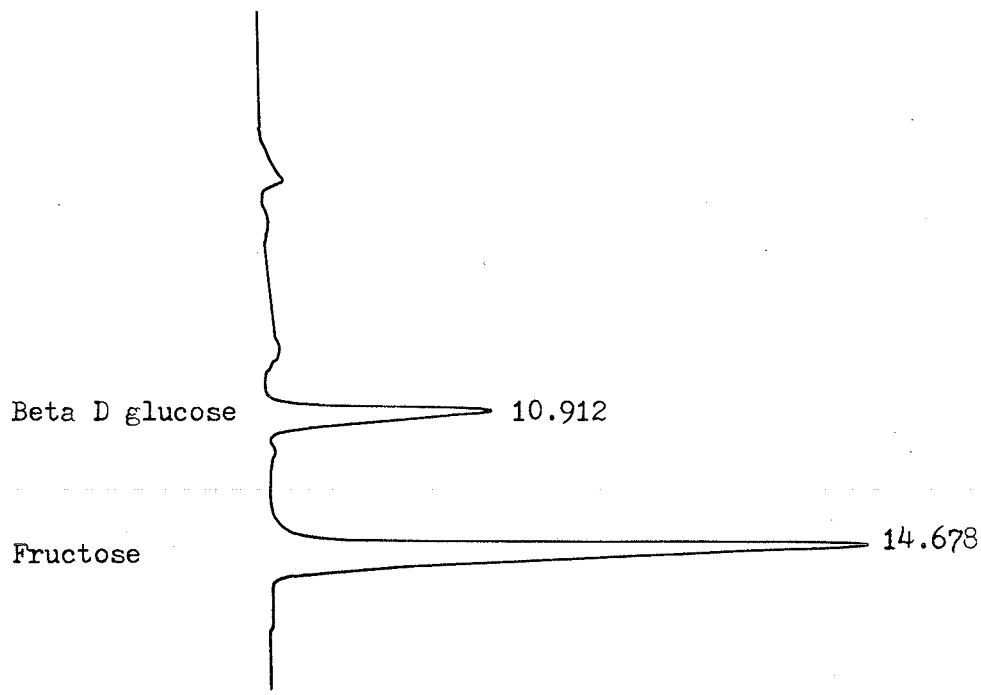


a - sample - apple cultivar, Collet.

Appendix 23

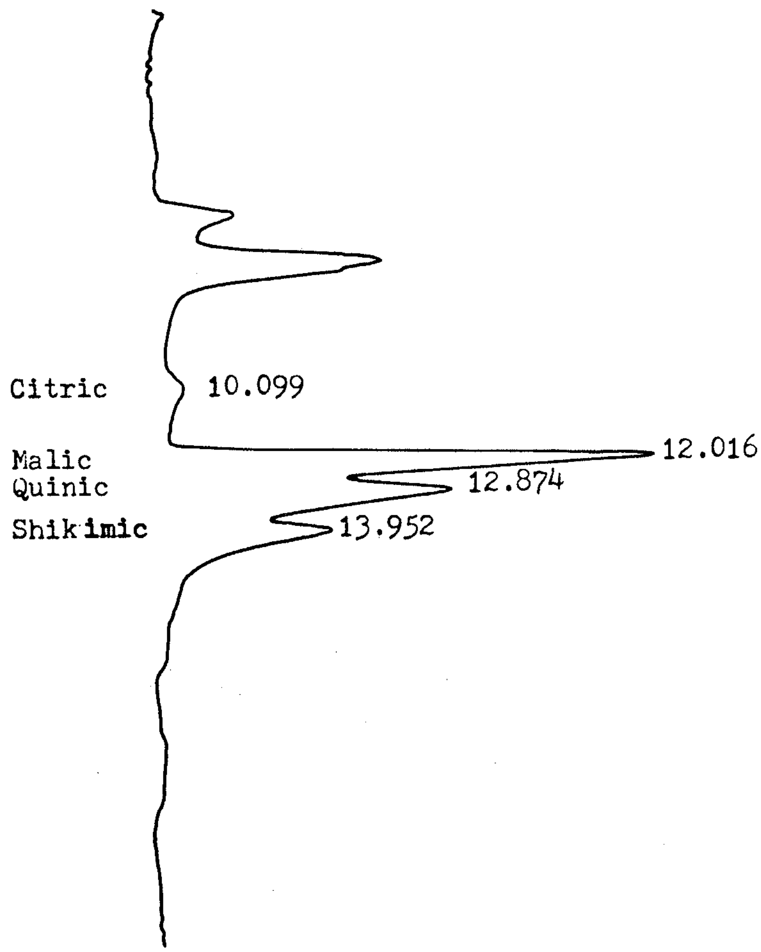
A representative HPLC chromatogram of sugars

from Heyer #12.



Appendix 24

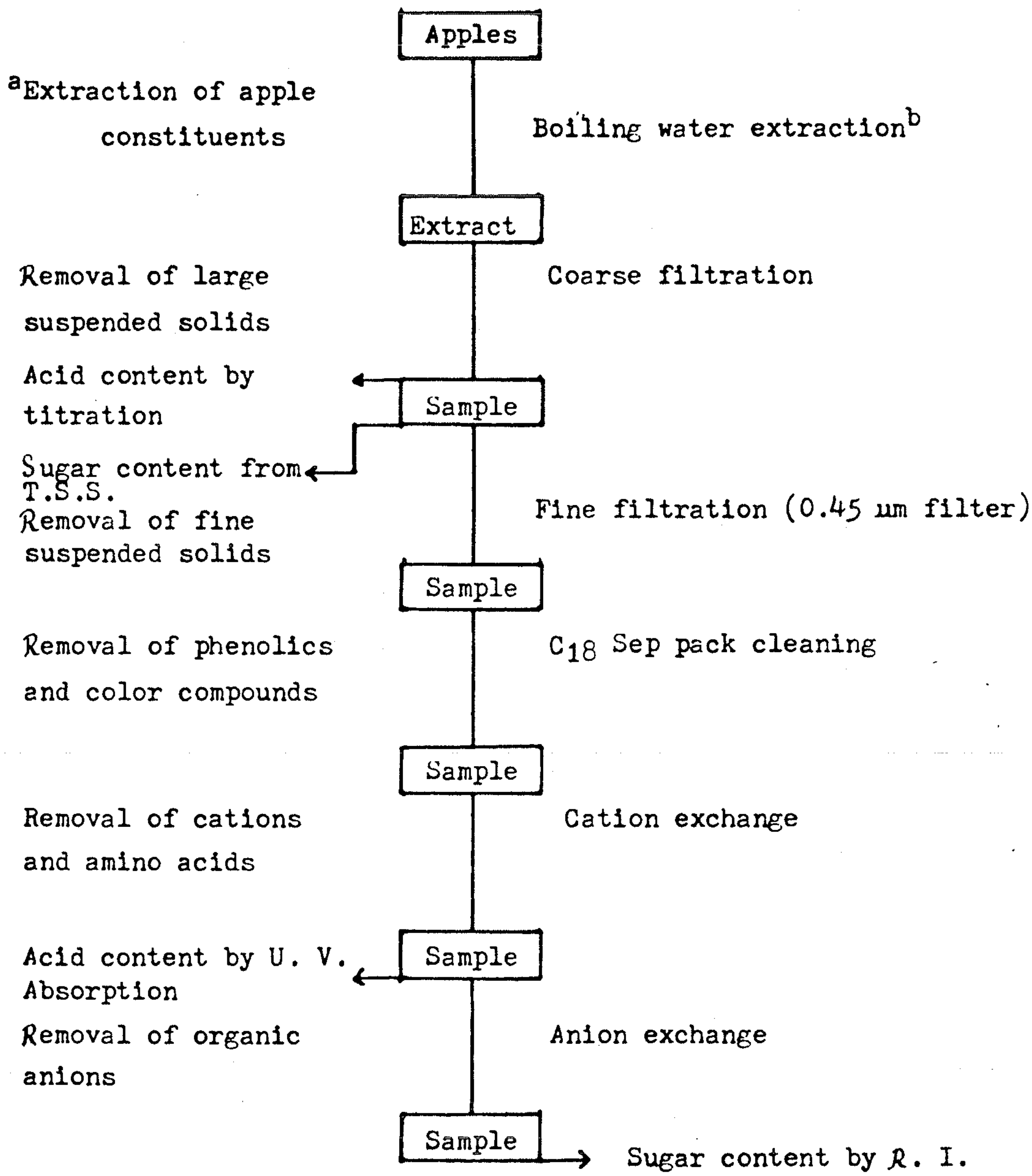
A representative HPLC chromatogram[a] of organic acids from apple and apple juice samples



a - Sample - Apple cultivar P. F. 51.

Appendix 25

Schematic diagram of sample preparation for sugar and acid analysis, including preparation steps and their effects



a - Descriptions on the left side of diagram show the effects of preparation steps.

b - Sample preparation steps.

Appendix 26

Tukey's test for total sugar content (% Sucrose)

Method	N	Mean[1]
Chromatographic Analysis	82[2]	9.69 a
Compositional Analysis	154	9.27 b

[1]Mean with the same letter are not significantly different at alpha - 0.05.
[2]Warning cell sizes are not equal.

Appendix 27

The relative number of suspended particles and turbidity of diluted apple juice samples

Dilution	Relative Number of Particles	Turbidity (JTU) Collet[1] 1	2	Kerr[1] 1	2[2]
Enzyme Clarified Juice	128	10	11	73	32
Ist	64	5.7	5.9	31	14.5
2nd	32	3.0	2.3	16	7.4
3rd	16	1.9	1.8	8.5	4.0
4th	8	1.3	1.3	5.2	2.2
5th	4	1.1	1.0	3.2	1.4
6th	2	0.88	0.90	2.2	0.98
7th	1	0.84	0.80	1.6	0.80
8th	--	---	---	---	0.72
Diluent[3]	--	0.74	0.46	0.80	0.53[4]

[1] - Apple cultivars.
[2] - 1 and 2 are the number for the batches of apple juices.
[3] - To obtain the blank standardization of nephelometer, turbidity values of the diluents were substracted from turbidity values of various dilutions.
[4] - The relative number of particles in the second batch from Kerr juice varied from 256 to 1.

Appendix 28

Turbidity of apple juices after various filters during processing

Filter (um)	Turbidity (JTU) Var. 1[1]	2[2]	% Decrease in Turbidity 1	2
Undiluted Juice	21.00	15.50	---	---
25	14.00	6.90	33.33	55.48
5	11.00	5.20	14.29	10.97
5	10.00	4.00	4.76	7.74
1	8.00	3.00	9.52	6.45
0.2	0.38	0.65	36.29	15.16

[1]Variety 1 - Goodland.
[2]Variety 2 - McIntosh.

Appendix 29

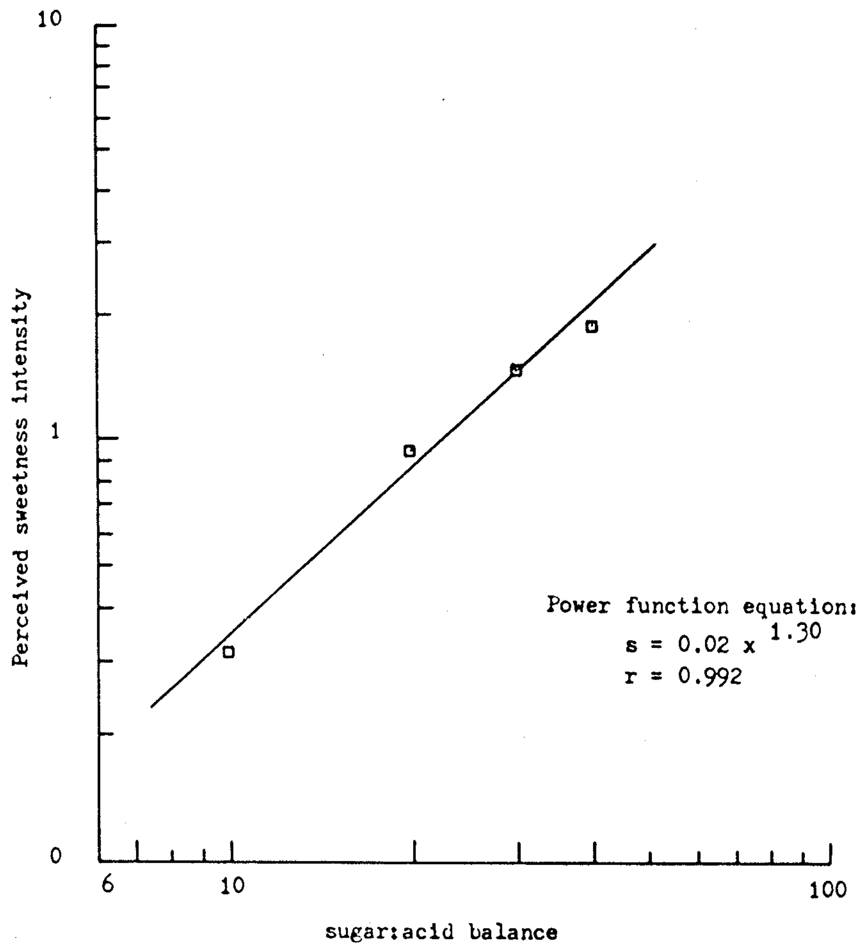
Statistical analysis of observed chemical changes in apple juices upon filtration processing

Observation	Source	Df	Sum of Squares	F[1]
Acid Content (% Malic acid)	Apple juice	2	0.247	208.61 **
	Process	1	0.008	13.60 **
	Replicate	4	0.000	0.00
	Error	16	0.009	
Sugar content (% Sucrose)	Apple juice	2	5.493	56.82 **
	Process	1	0.448	9.29 **
	Replicates	4	0.313	0.16
	Error	16	0.772	
Phenolic content (mg/100 ml)	Apple juice	2	288048.515	57.58 **
	Process	1	62013.75	24.79 **
	Replicates	1	3828.83	1.53
	Error	7	17510.22	

[1]F values significant at alpha 0.05 are indicated by **.

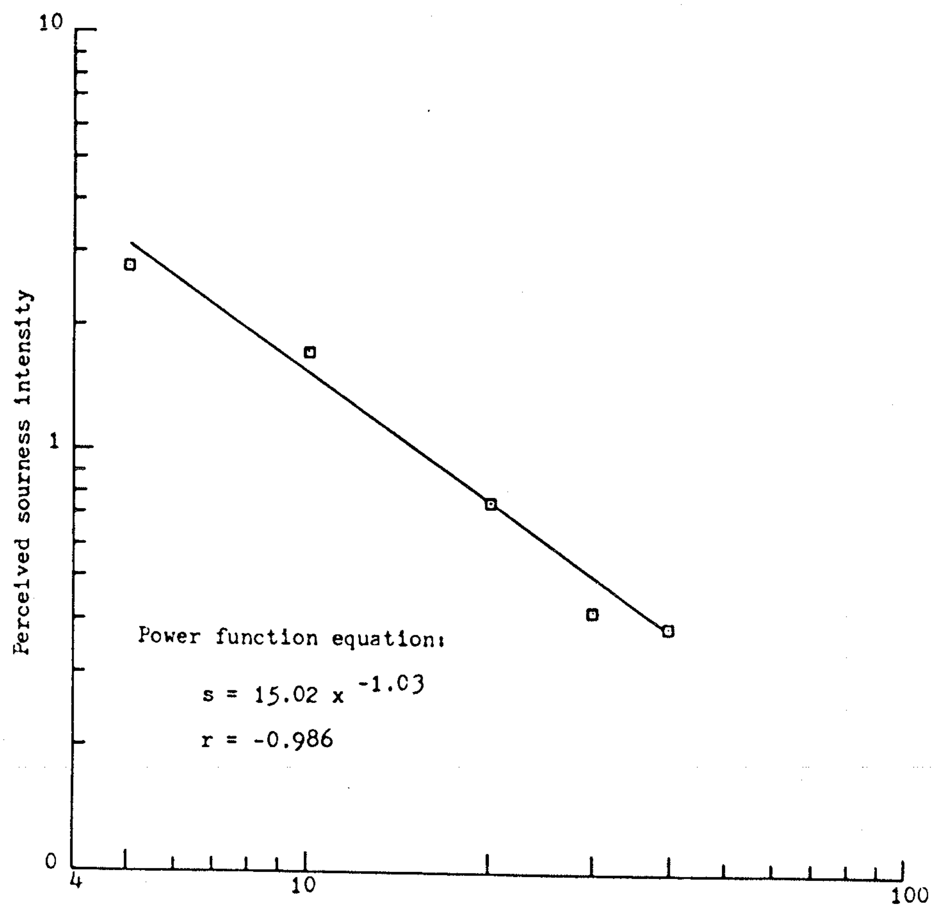
Appendix 30

Perceived sweetness intensity of sugar-acid solutions as a function of sugar:acid balance.



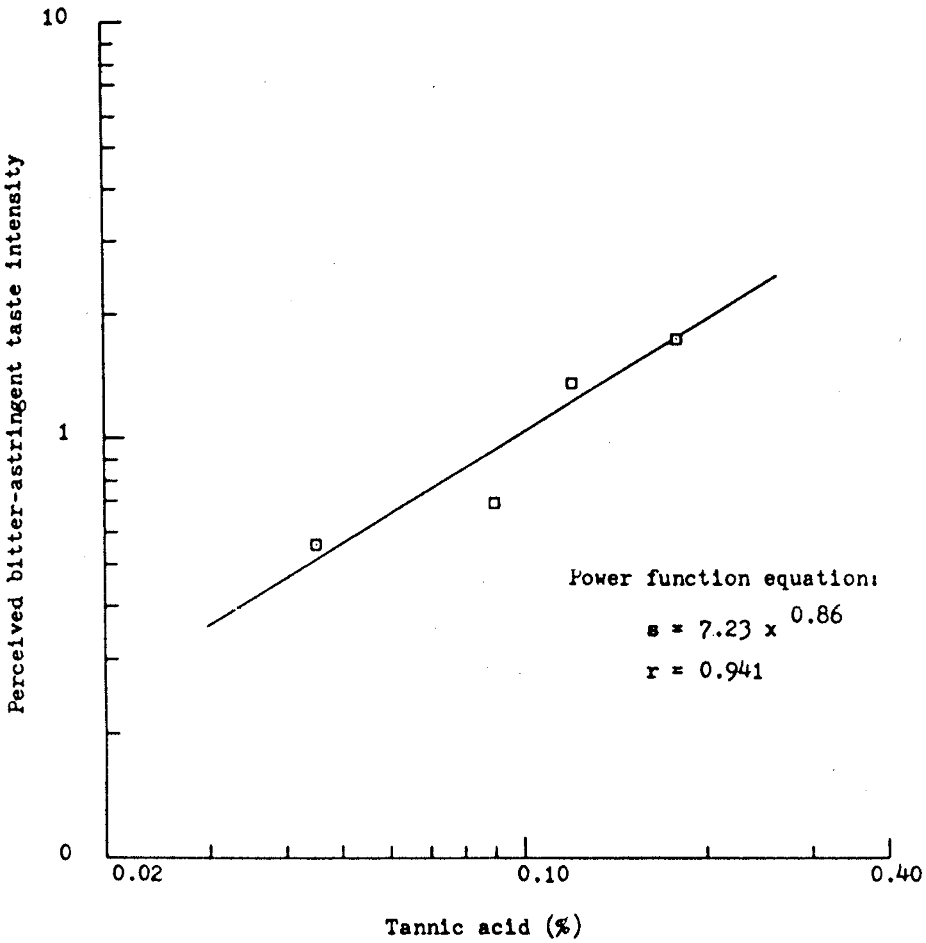
Appendix 31

Perceived sourness intensity of sugar-acid solutions as a function of sugar:acid balance



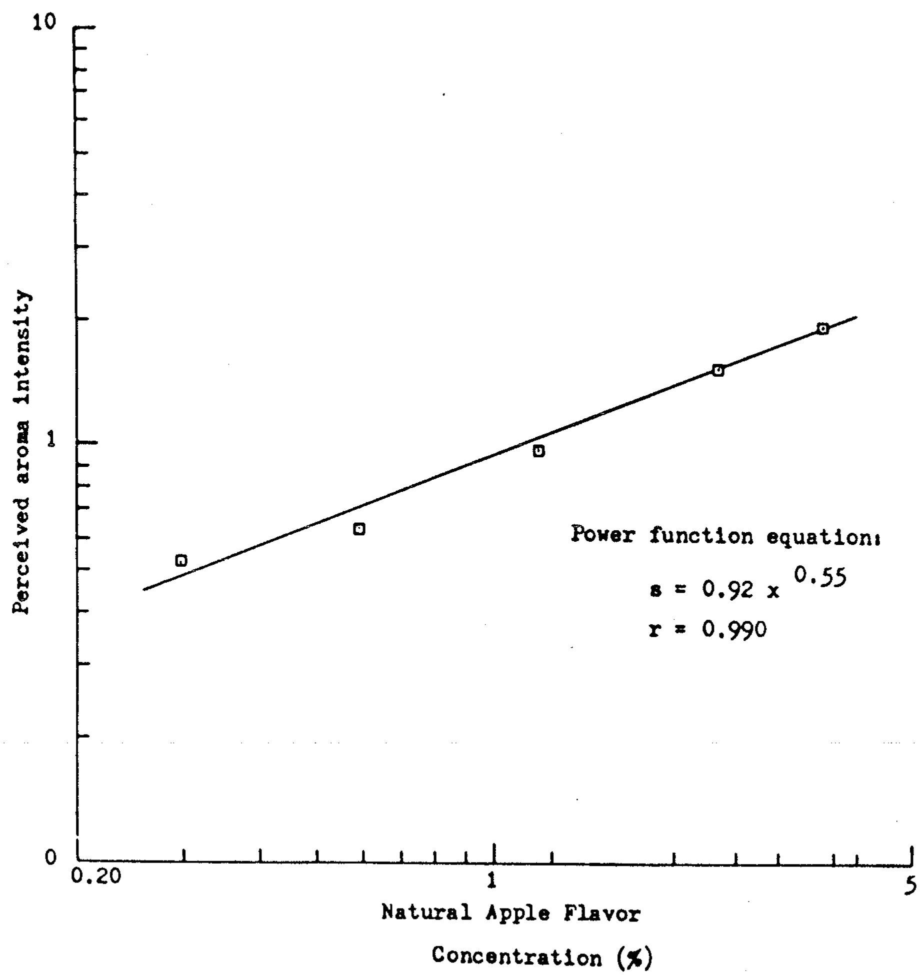
Appendix 32

Perceived bitter-astringent taste intensity of tannic acid as a function of concentration.



Appendix 33

Perceived aroma intensity of a natural apple flavor as a function of concentration.



Appendix 34

Means scores used to derive power function for sweetness and sourness

Sugar:Acid Balance	Sweetness[1] Mean	N	Sweetness[2] Mean	N	Sourness[1] Mean	N	Sourness[2] Mean	N
5	--[3]	--	--	--	0.38	14	0.51	16
10	-.54	12	-.45	14	0.24	14	0.25	16
20	-.07	15	0.02	15	-.08	14	-.18	16
30	0.18	15	0.17	16	-.28	12	-.47	12
40	0.32	15	0.23	16	-.44	10	-.34	10

[1]Sugar:acid balance adjusted by sugar.
[2]Sugar:acid balance adjusted by acid.
[3]More than 50% np judgements.
[4]The averages of two mean values (presented in this appendix) for sweetness and sourness, were used to derive power functions illustrated in the Appendices 30 and 31.

Appendix 35

Mean scores used to derive power function for the aroma and BAT

Conc.[1]	Aroma Mean	N	Conc.[2]	BAT Mean	N
0.30	-.28	13	0.045	-.27	17
0.60	-.20	14	0.090	-.16	17
1.20	-.01	15	0.12	0.12	17
2.40	0.18	15	0.18	0.23	17
3.60	0.29	15			

[1] - Concentration of natural apple flavor (V/V) in distilled water.
[2] - Concentration of tannic acid (W/V) in distilled water.

Appendix 36

Mean intensity scores of sensory properties and preference for apple juices

Apple Juice	Sensory Characteristics				Preference
	Aroma	BAT[a]	Sweetness	Sourness	
Reference[b]	1.02	1.15	.84	.74	
A	1.10	.75	1.02	.71	7.88
B	1.09	.75	1.14	.86	7.82
C	.99	1.02	1.03	.95	7.07
D	1.11	1.06	.62	1.93	6.85
E	.87	.64	.67	.83	5.67

a - BAT or bitter-astringent taste

b - References:

Aroma - 1.2% Natural Apple Flavor Solution
BAT - 0.12% Tannic acid solution
Sweetness - 6% Fructose and 0.3 Malic acid solution with sugar-acid balance of 20
Sourness - Same as sweetness

Appendix 37

Chemical composition of apple juices and derived chemical balances

Apple Juice	Sugar Content (%sucrose)	Acid Content (% malic acid)	Phenolic Content (% tannic acid)	Sugar:Acid Balance	Sugar:Phenolic Balance	Acid:Phenolic Balance
A[a]	10.16	0.42	0.14	24.19	72.57	3.00
B	10.52	0.41	0.17	25.66	61.88	2.41
C	10.58	0.45	0.20	23.51	52.90	2.25
D	9.77	0.51	0.23	19.16	42.48	2.22
E	9.59	0.40	0.12	23.98	79.92	3.33

a - Codes for commercial apple juices.

Appendix 38

Statistical Analysis of Preference for apple juice

"Analysis of Variance"

Source	Df	Sum of sq.	F value[a]
Apple juice	4	43.74	4.66**
Panelists	8	24.61	1.31
Replication	1	2.95	1.26
Error	59	138.58	

a - Significant differences at the 0.05 level are indicated by '**'.