SOME PHYSICAL CHEMICAL AND HISTOLOGICAL CHARACTERISTICS OF RIPENING BANANAS

bу

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The University of British Columbia Vancouver 8, Canada

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ABSTRACT

A study of changes in bananas during ripening at $16 \pm 1^{\circ}$ C and $25 \pm 1^{\circ}$ C is described. Peel color was evaluated subjectively and by reflectance spectrophotometry; rheological properties by parallel plate compression and viscometry; selected chemical properties by appropriate tests and histochemical and histological properties by light microscopy.

The rate of peel color change at the higher temperature was roughly twice that at the lower. Higher temperature-ripened fruits did not develop a full yellow color due to chlorophyll retention in the peel. Also pulpto-peel ratio for such fruits tended to be lower than that of fruits ripened at the lower temperature.

The pulp of high temperature-ripened fruits became progressively softer and was reflected by a linear increase of deformation under 1 kg force. For a given peel color index, maximum force and linear limit of the tissue as well as a power-law consistency coefficient of the puree were generally lower during ripening at the higher temperature. Reducing sugars increased linearly throughout ripening at the higher temperature while at the lower temperature the reducing sugar content was essentially constant beyond color index 6. On the basis of peel color index, total

sugar and moisture content were higher while starch and AIS levels were lower in fruits ripened at the higher temperature. Ripening temperature therefore influences the relations of color index to mechanical and chemical properties.

Ripening was characterized by a gradual loss of rigidity as well as an apparent thickening of the cell wall in over-ripe pulp tissue. Tannins decreased during ripening but did not disappear completely. Esterified pectins were not detected in hard green fruit; however, substantial amounts appeared at peel color index 3, then decreased steadily during ripening.

Peel color was the best overall index of stage of ripeness for both ripening temperatures. Although rheological and chemical properties at a given color index differed for the two ripening temperatures, these intercorrelations remained higher ($P \le 0.01$). It is recommended, that ripening temperatures be taken into account when the color index chart is used to estimate the stage of ripeness of bananas.

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INTRODUCTION

Bananas are grown exclusively in the tropics but they are consumed in practically every country. In addition to being a major item of international trade, the banana is the most important tropical fruit on the world market.

Arthur et al. (1968) stated that without doubt the banana is the world's most widely consumed fruit.

In 1969, total banana exports amounted to 5.93 million tons (FAO 1971). This exceeded the amount of citrus fruits and apples traded during the same year, leaving the banana as the most important fresh fruit in international trade. Preliminary FAO estimates indicate that 199,000 tons of bananas were imported into Canada in 1970. For the period commencing January 1969 to January 1972, the total import of bananas into Canada was valued at \$7.11 million, (Statistics Canada 1972). In British Columbia, bananas account for a substantial amount of all imported fresh fruit. Banana imports through B.C. customs in 1970 amounted to 28.3 million tons, (B.C. Dept. Ind. Dev. Trade & Comm. 1971).

Bananas are marketed throughout the year, but because of their perishable nature, prolonged storage is not possible. Weekly shipments of green bananas ensure a continuous supply of fresh fruit at retail outlets. However, fluctuations in supply and demand make it necessary for commercial ripening establishments to adopt various ripening schedules in order to meet the market requirements.

The ripening period can be varied from 4 to 10 days (Seelig 1969). The principal factors used in the regulation of ripening are temperature, humidity, ventilation and ethylene gas. Two of those factors -- temperature and ethylene -- are perhaps the most important. Most commercial ripeners use ethylene to stimulate or "trigger" the ripening process while the rate of ripening is controlled by temperature regulation.

Fruit quality is known to be affected by storage and ripening temperatures (Simmonds 1966). Prolonged exposure to high temperatures during ripening may lead to "boiled" fruit, while low temperatures give rise to chilling injury (Hall 1967). Under normal commercial conditions, such forms of injury rarely occur. However, there have been several reports (Hall 1967; Dalal et al. 1969; Sanchez Nieva et al. 1969; Murata 1970) of differences in fruit quality, obtained within the range of normal ripening procedures.

Many of the studies involving bananas ripened under different conditions, have focussed on major chemical constituents and used panel evaluation of quality. The changes in physical properties of the fruit have been considered largely in descriptive terms, although their importance in fruit quality is recognized (Seelig 1969).

This study was undertaken to investigate some physical, rheological and chemical properties of the banana

during ripening. A study was also made of the histochemical and histological changes in the ripening fruit, using light microscopy. The overall aim was to obtain a better understanding of the relationships among the major chemical and rheological properties of the ripening fruit and their dependency upon the temperature of ripening.

LITERATURE REVIEW

Banana Ripening

Commercial methods

The controlled ripening of bananas is carried out by various methods. In temperate countries, ripening is carried out in specially constructed ripening rooms in which temperature, humidity and air circulation are carefully controlled.

Modern ripening methods have been reviewed by vonLoesecke (1950), Haarer (1964) and Simmonds (1966).

Seelig (1969) reviewed the requirements for ripening rooms with particular reference to those adapted for boxed fruit. The major companies involved in the banana trade provide customers with ripening recommendations which allow the ripening period to be varied from 4 to 8 days (United Fruit Sales Corp. 1970; Standard Fruit and Steamship Co. 1964).

Factors which affect ripening.

Temperature: During ripening, bananas produce a considerable amount of heat as a result of respiratory activity. The amount of heat given off by green bananas at 54°F (12.2°C) is approximately 140 Btu/ton hr (USDA 1954). Simmonds (1966) calculated the heat production of preclimacteric fruit at 53°F (I1.7°C) to be approximately 150 Btu/ton hr. At 68°F (20°C) heat production increases from 348 Btu/ton hr at preclimacteric to 386 Btu/ton hr

during the climacteric (USDA 1954). These calculations are based on the amount of carbon dioxide evolved.

To obtain a ripe fruit of excellent quality the pulp temperature should be kept between $58^{\circ}F$ (14.3°C) and $64^{\circ}F$ (17.6°C) depending on the rate of ripening desired (United Fruit Sales Corp. 1970). The maintenance of constant pulp temperatures requires that heat be withdrawn from the box at least as fast as it is produced by the fruit. The air temperature must be lower since the cardboard acts as an insulator. In order to maintain the pulp temperatures listed above, air temperatures between $52^{\circ}F$ (11.1°C) and $58^{\circ}F$ (14.3°C) are recommended (United Fruit Sales Corp. 1964).

At the climacteric, pulp temperature tends to increase very rapidly and room temperature is usually lowered to minimize the increase. After the fruit has "sprung" -- i.e. in the post climacteric period -- the air temperature may again be raised gradually as heat evolution subsides. Changes in air temperature result in a gradual change in pulp temperature, and because of this it is recommended that pulp temperatures be recorded at least twice a day (United Fruit Sales Corp. 1970).

Temperature affects the rate of ripening as well as the quality of ripe fruit. The precise effects are difficult to evaluate because of interaction with factors

such as ventilation, humidity and the physiological age of the fruit. Other factors such as variety, conditions of production and pre-ripening storage may be of considerable significance.

Humidity: The skin of a mature banana contains a large number ($\sim 480/\text{cm}^2$) of stomata and transpiration is very active (Palmer 1971). Simmonds (1966) summarized the results of earlier research which showed that during ripening transpiration increases rapidly at the climacteric. This is followed by a steady state in which transpiration is higher than in the preclimacteric stage. Increased water loss occurring at advanced ripening is related to fungal attack. The rate of transpiration is largely dependent upon temperature and humidity.

In order to ensure proper water relations during ripening a high relative humidity must be maintained. In general, relative humidity of 85 - 95% is recommended at the beginning of ripening. As the fruit "breaks" color the humidity is reduced to 75 - 85%.

Ethylene: vonLoesecke (1950) reviewed the developments which led to widespread use of ethylene in banana ripening. Since then other workers (Biale et al. 1954; Burg and Burg 1965) have examined the relationship between endogenous ethylene and the respiratory climacteric. The role of ethylene in fruit ripening has been recently analyzed by Pratt and Goeschl (1969) and by McGlasson (1970),

while Palmer (1971) has discussed the significance of ethylene in commercial transport and ripening of bananas.

The mechanism of ethylene action in fruits is still unresolved, despite extensive research. McGlasson (1970) observed that several hypotheses have been presented which attempt to explain the action of ethylene in terms of its effects on enzyme activities, interactions with nucleic acids and metallo-enzymes and effects on lipoprotein membranes. These hypotheses can be supported adequately, but they fail to establish the nature of the primary action of ethylene in fruit tissue.

Biale et al. (1954) reported that ethylene was a by-product of ripening. Burg and Burg (1965) have since found that preclimacteric bananas contain 0.1 - 0.2 ppm ethylene in their tissue, and that this level increases dramatically a few hours before the climacteric. Endogenous ethylene will induce ripening in the banana, but due to the differences in physiological age among fruits in a lot, uneven ripening is frequently encountered in the absence of ethylene treatment. Supplementary ethylene enables all the fruit in a lot to attain the threshold level, for ripening at about the same time. Ethylene production in the banana is temperature sensitive (Palmer 1971), thus low temperature ripening is not possible in the absence of applied ethylene.

In commercial practice ethylene is applied at the

rate of one cubic foot per 1000 cubic feet of ripening space (United Fruit Sales Corp. 1964). Higher concentrations do not provide additional benefits. The gas is usually applied as soon as the green fruit is stacked in the ripening room. Treatment may be single or multiple during a 24 - 36 hour period (Hall 1967).

Changes Associated with Ripening

An extensive review of biochemical changes in the ripening banana was carried out by vonLoesecke (1950). Simmonds (1966) summarized the more conspicuous biochemical features of ripening. These are presented in Figure 1. Recently Palmer (1971) has reviewed the compositional changes during ripening, as well as the enzymes involved. In this section only those changes which are relevant to the study will be discussed.

Color

During ripening the banana peel changes in color from green to yellow. Yellowing of the peel begins at or follows the climacteric peak (Palmer 1971), while the rate of yellowing is dependent upon the ripening conditions. Peel color is probably the most widely used index of the stage of ripeness.

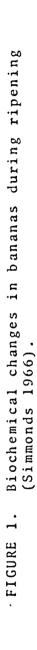
vonLoesecke (1950) summarized information on the pigment composition of banana peel. Chlorophyll, xanthophyll and carotene are the major pigments in green banana peel. During ripening chlorophyll is reduced from 50 - 100 $\mu g/g$

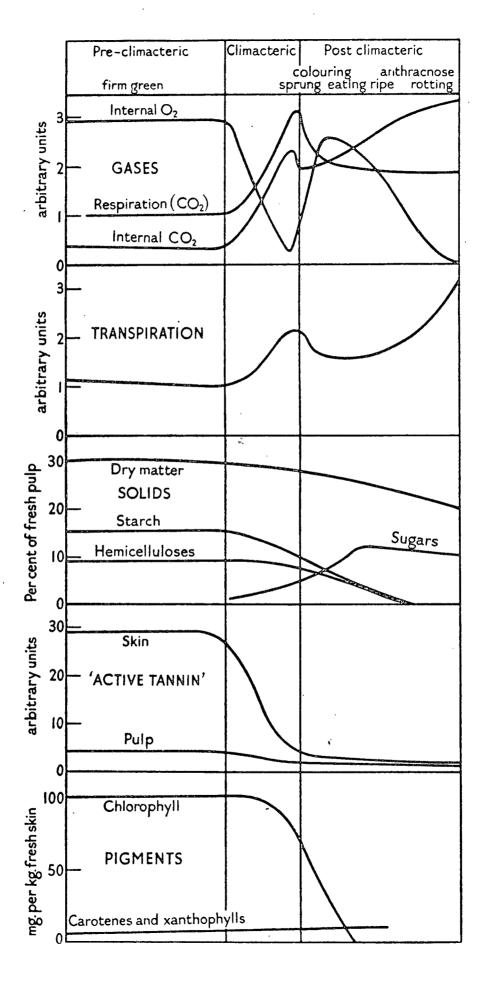
fresh peel to near zero, carotene remains approximately constant at 1.5 - 3.5 µg/g fresh peel and xanthophyll also remains constant at 5 - 7 µg/g fresh peel. Looney and Patterson (1967) reported that chlorophyllase activity in Gros Michel peel reached a maximum at the climacteric peak. It was suggested that chlorophyll destruction was due to the loss of structural integrity in the chloroplasts. The ability of ethylene to stimulate ripening indicates that it may have a direct effect on chlorophyll destruction. Ethylene-treated bananas ripen with uniformly colored peels (vonLoesecke 1950; United Fruit Sales Corp. 1964).

Color measurement in bananas has received little attention. Gottreich et al. (1969) reported that early attempts to determine state of ripeness on the basis of pulp color failed because of difficulties in preparing stable standard colors. Finney et al. (1967) used reflectance spectrophotometry to evaluate the relationship between peel color and firmness in Valery bananas. They reported that loss of chlorophyll associated with the change in color from green to yellow was related to the change in reflectance at 675 nm.

Texture

Softening in fruits can be atributed to the interconversions of several structural polysaccharides. McCready and McComb (1954) demonstrated that pectic substances play a major role in the loss of firmness in fruit tissue.





vonLoesecke (1950) indicated that during ripening, soluble pectic substances in the pulp of Gros Michel bananas increased from 0.3 to 4.0% while protopectin decreased from 0.5 to 0.2% on a fresh weight basis.

The role of cellulese and hemicellulose in textural change in the ripening banana was investigated by Barnell (1943). He reported that cellulose remained constant while hemicellulose decreased from 8 - 10% in the pulp of green fruits to about 1 - 2% in ripe fruits. Hemicellulose content fluctuated during ripening and this suggested to him that it acted as a form of reserve carbohydrate. It was concluded that hemicellulose was hydrolyzed to give substances which may serve as substrates for respiration.

Sarkissian (1965) used a modified Chatillon Fruit and Vegetable Tester to determine texture-firmness in ripe banana fruit tissue. This device is a pressure tester used in testing soft fruits such as strawberries. Pressure was applied at right angles to the cut surface of 2 inch unpeeled cross sections of fruit. He reported that at advanced peel colors the pulp of Valery bananas was much firmer than that of Gros Michel.

Finney et al. (1967) used a sonic technique to measure changes in firmness of Valery bananas as they ripened. Cylindrical sections of pulp were vibrated longitudinally and the resonant frequencies were used to calculate

Young's modulus of elasticity which is defined as the ratio of stress to strain. It is a measure of resistance to force and therefore, of firmness. They found that softening of the banana during ripening was associated with a decrease in Young's modulus of elasticity from 272 X 10^5 dynes/cm² in light green fruit to 85 X 10^5 dynes/cm² in the full yellow stage.

Carbohydrates

Starch is the predominant carbohydrate in green bananas. During ripening it is hydrolyzed to sugars.

vonLoesecke (1950) reported the starch content of green banana pulp as 20 - 25% and that of ripe fruit as 1 - 2%.

The peel contains 3% starch which is also hydrolyzed during ripening. Sugars normally increase from 1 - 2% in green fruit to 15 - 20% in the pulp of ripe fruit.

There is some disagreement in the literature with regard to the form of the different sugars in ripe bananas. Eheart and Mason (1966) analyzed bananas bought on the wholesale market in Washington D.C. and found that reducing sugars were present at 10.34% while sucrose content was 8.54%. United Fruit Sales Corp. (1964) lists the sugar content of a fully ripe banana as follows: sucrose 12.7%, levulose (fructose) 3.7% and dextrose (glucose) 4.8%.

Poland et al. (1938) reported that during ripening, glucose, fructose and sucrose maintain nearly constant proportions.

Acidity

Acidity of banana pulp rises to a maximum at or soon after the climacteric and may show a slight decrease as ripening progresses (vonLoesecke 1950). The pH ranges from 5.0 - 5.8 for the pulp of green fruit to 4.2 - 4.8 in post-climacteric fruit (Simmonds 1966). Titrable acidity of Gros Michel pulp changes from 2.96 in the preclimacteric state to 4.95 following the climacteric then to 3.66 m.equiv/100g fresh pulp at full ripeness. Eheart and Mason (1966) reported the acidity of fully ripe banana pulp as 0.27% malic acid [4.03 m.equiv/100g].

Studies by Stewart $\underline{\text{et}}$ $\underline{\text{al}}$. (1960) and Miller and Ross (1963) indicate that L-malic and citric acids are the predominant acids in banana pulp at all stages of ripeness.

Moisture

In spite of transpiration losses, the moisture content of banana pulp increases during ripening. vonLoesecke (1950) gave values for five clones ranging from 63 - 74% to 68 - 77%. Palmer (1971) suggested that the net increase in pulp moisture is the combined effect of respiration and osmosis. Sugar increases more rapidly in the pulp than in the peel and this creates an osmotic gradient causing water to move from peel to pulp. Stratton and vonLoesecke (1931) found that during ripening the osmotic pressure of Gros Michel peel increases from 6 to 11.5 atmospheres and that of the

pulp increases from 6 to 25 - 27 atmospheres.

Osmotic transfer of water results in changes of pulp-to-peel ratio on a fresh weight basis. Simmonds (1966) observed that the ratio is about 1.2 - 1.6 in green fruit and rises with normal ripening to 2.2 - 2.4 at advanced ripening. Microbial invasion and dehydration give rise to further increases in the pulp-to-peel ratio. vonLoesecke (1950) suggested that pulp-to-peel ratio could be useful as an index of ripeness.

Anatomy and Histology

Bananas are arranged in nodal clusters or "hands" which are borne on a nodal base or "cushion" attached to a stalk. This entire structure is roughly cylindrical in shape and is known commercially as a bunch or "stem".

The fruit is vegetatively parthenocarpic; i.e. it develops a mass of edible tissue without pollination. The pulp consists of relatively undifferentiated tissue divided into 3 segments by the carpellary margins. The degenerated ovules are arranged centripetally in each segment. Wolfson (1928) has studied the anatomy of the fruit, and his work forms the basis for subsequent observations by vonLoesecke (1950). The peel consists of an epidermal layer which is underlayed by parenchyma cells interspersed with fibrovascular bundles. Chloroplasts are found in the outer layer of parenchyma known as the chlorenchyma.

The epidermal cells are slightly convex on the

upper surface and slight grooves appear where the edges of adjoining cells meet, giving a striated appearance to the peel surface. Transpiration and gaseous exchange take place through stomata and a thin layer of cutin protects the epidermis from desiccation and other forms of injury.

Parenchyma cells make up the bulk of the peel tissue. They are large and thin walled, increasing in size away from the epidermis.

A thin layer of cytoplasm containing numerous plastids, line the inside of the cell wall. In the innermost cells of the peel the plastids become centers of starch accumulation. A sap-filled vacuole occupies the center of the cell. The cell sap is a clear fluid which consists mainly of dissolved sugars, organic acids, phenolic compounds and water.

The fibrovascular bundles are scattered throughout the parenchyma and run parallel to the longitudinal axis of the fruit. In addition to imparting strength and rigidity to the peel they serve as conveyors of water and metabolites. Individual fiber cells are long, narrow and thick walled. Inner bundles are less fibrous but more complex. They are surrounded by a ring of laticiferous tissue interspersed with parenchyma cells (Ram et al. 1962). The vascular elements of the fruit all converge and anastomose in the region of the pedicel.

The latex system is found in both the peel and

the pulp, and consists of large thin-walled, barrel shaped cells which are joined end to end in single lines. In the peel they are generally associated with vascular bundles, although they frequently occur by themselves. Barnell and Barnell (1945) noted that latex cells in the peel are of two well-defined types. Most of the tannin in the fruit is found in the latex (Barnell and Barnell 1945).

The pulp-peel boundary consists of a few layers of parenchyma cells with large intercellular spaces.

During ripening this region becomes more porous with the result that peeling of the fruit is made easier. Upon removal of the peel prominent longitudinal bundles are seen adhering to the peel. These bundles give rise to the characteristic depressions on the surface of the pulp.

Pulp cells are thin walled and may be long or isodiametric. The inner cells are arranged in rows radiating from the septae. These consist largely of parenchyma cells and vascular bundles which pass into the placental axis. The placental axis consists chiefly of spongy parenchymatous tissues. In green as well as ripe bananas there are few or no intercellular spaces.

During ripening, cell size remains constant while starch granules decrease in size and number (vonLoesecke 1950). Latex tubes are present in the pulp of ripe fruit, but they contain no latex. This is attributed to tannin condensation (Barnell 1943). The ripening behavior of the cell wall and its components has not been studied.

EXPERIMENTAL METHODS

Sampling Procedures

Materials

Hard green Valery bananas (Chiquita brand) were purchased from a local wholesale establishment. The fruits had been brought in by truck from the docks at Seattle, Washington and were treated with ethylene (approx. 1 ft^3 per 1000 ft^3 storage space) for 24 hr before being used in the experiments.

Ripening

A closed-cycle heated air dryer equipped with a cooling coil attached to a refrigeration unit was used as a ripening chamber. This apparatus is described in detail by Bhargava (1970). Fruits were ripened at $16 \pm 1^{\circ}$ C and $25 \pm 1^{\circ}$ C. A relative humidity of 85 - 95% was maintained during ripening, by injecting steam into the unit. Temperature and relative humidity in the ripening compartment were recorded by a hygrometer (Hydrodynamics Inc. Model 15-4050 E) and a hygrothermograph.

A 40 lb carton of fruit was used for each ripening trial. Upon receiving the fruits the "hands" were broken up and individual fruits were replaced at random in the carton, which was then placed in the ripening chamber.

Each temperature treatment was replicated three times. Groups 1, 2 and 3 were ripened at 16 \pm 1° C and groups 4, 5 and 6 at 25 \pm 1° C. Experiments were terminated

when the fruits acquired a peel color index of 8.

Sampling

Eight fruits were sampled every 2-3 days during ripening at $16\pm1^{\circ}\text{C}$, and every day during ripening at $25\pm1^{\circ}\text{C}$. Five fruits were used individually in the determination of length, pulp-to-peel ratio, color and force-deformation behavior. The pulp from these fruits was pooled and used to study flow behavior. The other three fruits were collectively used in the determination of alcohol insoluble solids (AIS), sugars, moisture, pH and titrable acidity. Physical Properties

Length

A vinyl metric tape was used to measure the distance from the trimmed stem end to the distal end of the fruit. This was done along both the convex and concave sides. The mean of these two measurements was taken as the length of the fruit.

Pulp-to-peel ratio

Whole and peeled fruits were weighed to obtain gross and pulp weight. Peel weight was then derived by difference and the pulp-to-peel ratio was calculated as the quotient of pulp and peel weights.

Color

A banana ripening chart with color plates was used to evaluate the color index (CI) of fruits, (United Fruit Sales Corp. 1964). The CI for a given day represented

the mean for the eight fruits sampled.

Spectrophotometric measurement of peel color was carried out on two 35 mm discs of peel removed from the fruit at the distal and stem ends. Each disc was placed in a plastic tissue culture dish and flattened with a black backing. The dish was covered and clamped to the sample holder of the reflectance unit of a Unicam SP 800 recording spectrophotometer. Diffuse reflectance of the peel within the visible spectrum (450 to 800 nm) was recorded on a logarithmic scale at a scan rate of 200 nm/min. Fresh magnesium oxide was used to calibrate the instrument at 100% reflectance.

Reflectance values at 470, 672 and 730 nm were read from the spectra and the mean reflectance obtained for the two sections constituted the reflectance data for each fruit. Reflectance ratios were then computed for 470 and 672, 672 and 470 as well as 730 and 672 nm respectively. The Index of Variance Reflectance (IVR) proposed by Powers et al. (1953) as a criterion for color measurement in fruits, was calculated using the following formula:

$$IVR = \frac{R_{730} - R_{672}}{R_{672}}$$

Rheological Properties

Force-deformation behavior

A cylindrical specimen of tissue was prepared by

cutting a section of the peeled fruit 2.5 cm long, using a parallel string slicing device. Cross sectional area at each cylinder end was calculated from the average diameter measured with vernier calipers. The mean of these measurements was designated as the cross sectional area of the specimen.

The section was subjected to parallel plate compression along the longitudinal axis using an Instron Model TMM universal testing machine. Loading rate was 0.5 cm/min and chart speeds of 2 and 5 cm/min were used. Force-deformation curves of the type shown in Figure 2 were obtained for five fruits on each sampling day. From these curves values were obtained for maximum force, linear limit, deformation at one kg of force and the energy absorbed by the sample from initial loading to the linear limit (linear limit energy). To compensate for differences in cross sectional area of the cylindrical samples, maximum force and linear limit were divided by the cross sectional area and the one kg deformation was multiplied by that area.

Flow behavior

Pulp from the fruits used in the study of force-deformation characteristics was sliced and blended with 25% (W/W) water in a high speed Waring blendor for 8 min. Preliminary trials indicated this amount of water was optimum for slurry preparation since green pulp could be macerated to a smooth consistency while the particulate

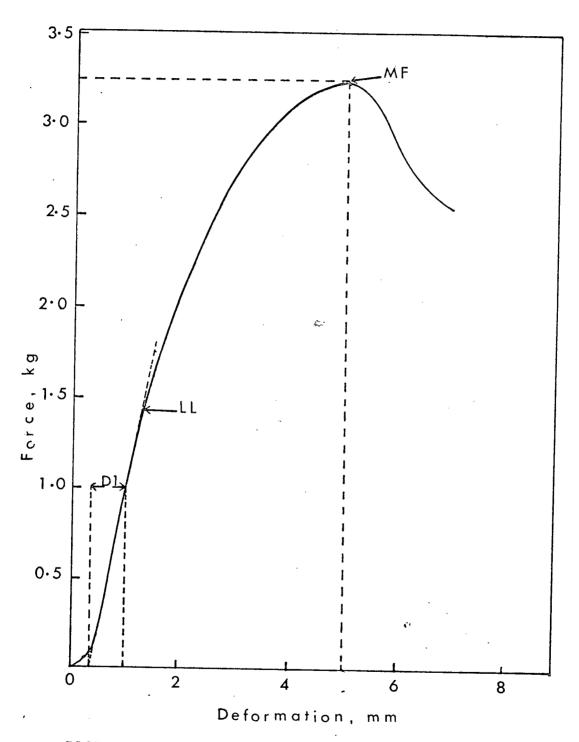


FIGURE 2. Typical force-deformation curve for banana pulp tissue. MF, maximum force; LL, linear limit; D1, deformation due to 1 kg force.

matter in over-ripe puree did not settle out of the suspension. After blending, the puree was allowed to stand at room temperature for 30 min.

About 100 ml of puree was used for each determination. All measurements were obtained with a Haake Rotovisko concentric cylinder viscometer equipped with an MVl spindle (gap width 0.96 mm). The sample was kept at 20° C using a Kryomat constant temperature bath connected to the water jacket that surrounded the sample holder.

During each determination the spindle rotation speed was varied stepwise from maximum to minimum. The viscometer transmission was then disengaged and the shear stress relaxation was recorded until a constant value was attained. This measurement was used as a yield stress for the sample. The sample was then tested from low to high shear rates to complete the viscometric measurements. Shear rates ranged from 8.5 to 1370 sec⁻¹. The torque due to viscous drag in the fluid at known shear rates was sensed by a Moseley Autograf strip chart recorder.

Flow behavior curves were constructed using shear rate, shear stress and yield stress data derived from the viscometric tests. Two forms of the widely used power-law flow model were fitted to these data.

and $\tau = m\dot{\gamma}^n + \tau_y$ [3]

where $\tau = \text{shear stress (dynes cm}^{-2})$

 τ_{y} = yield stress (dynes cm⁻²)

 $\dot{\gamma}$ = shear rate (sec⁻¹)

m = a parameter, the consistency coefficient

n = a parameter, the flow-behavior index.

The flow parameters \underline{m} and \underline{n} were evaluated with a computer using the method of least squares and a non-linear curve fitting technique. This procedure included evaluation of statistical parameters that would indicate the accuracy with which the flow models fitted the data.

Chemical Properties

Moisture

A modified AOAC (1965) method was used in which duplicate 5 g samples were dried in a hot air oven at 100 - 103°C for 18 hr. After cooling in a desiccator for 30 min, the dried samples were weighed and percent moisture computed on a fresh weight basis.

рН

A 40% slurry was made by mixing an appropriate amount of puree with distilled water and the pH was measured with an Instrument Laboratory pH meter.

Alcohol Insoluble Solids

Determination of alcohol insoluble solids (AIS) was based on an AOAC (1965) method. Duplicate 5 g samples of puree were extracted in 250 ml boiling 80% ethyl alcohol. Extraction was carried out for 30 min in a water bath at

about 85°C. The hot solution was vacuum-filtered through Whatman No.2 filter paper in a Buchner funnel. The residue was washed with an equal volume of hot 80% ethyl alcohol, then dried in a hot air oven at 100 - 103°C for 2 hr. After cooling in a desiccator for 15 min, the samples were weighed and AIS calculated as a percentage of fresh weight. Dried AIS material was placed in sample bottles and stored in a desiccator to be used later for determination of starch.

Reducing sugars

These were measured by the method of Ting (1956) with some modifications by Furuholmen et al. (1964). The method is based on the reduction of alkaline ferricyanide, which is then converted to a blue-green arsenomolybdate complex. The absorbance of this complex at 515 nm is then measured with a spectrophotometer. All reagents used in this method were as described by Ting (1956).

The filtrate obtained during AIS determination was cooled to room temperature and made up to 1000 ml with distilled water. At this concentration ethanol did not interfere with the test. A one ml aliquot of this dilute extract was transferred by pipette to a 100 ml volumetric flask and 5 ml ferricyanide reagent added. The flask was swirled then heated in a boiling water bath for 10 min. After heating the flask was quickly cooled in a running water bath. The contents were then partially neutralized with 10 ml 1M $_2$ SO $_4$ and shaken until gas evolution ceased.

Four ml of arsenomolybdate reagent were added, the mixture was mixed thoroughly and made up to volume. The flask was allowed to stand for 15 min.

Absorbance of the ferrocyanide-arsenomolybdate complex was measured at 515 nm and a slit width of 0.016 mm with a Hitachi-Perkin Elmer spectrophotometer. A reagent blank with water was used to standardize the spectrophotometer and glucose and fructose solutions were used to construct a standard curve. The reducing sugar content of each alcoholic extract was determined in duplicate.

Total sugars

A 50 ml sample of the dilute extract was placed in a 200 ml beaker with 10 ml 6M HCl. The beaker was swirled and allowed to stand at room temperature for 18 hr. Following inversion the mixture was partially neutralized with 5 ml 10M NaOH and the pH adjusted between 5 and 7 with 1M NaOH. The solution was then transferred to a 200 or 250 ml volumetric flask and made up to volume. One ml of this solution was transferred to a 100 ml volumetric flask and total sugars were determined using the procedure described for reducing sugars. Two determinations were carried out on each inverted sample.

Starch

A modified AOAC (1965) method was used to measure starch. A sample of dried AIS ($\sim 0.5 \rm g$) was added to a mixture of 200 ml water and 20 ml 6M HCl. This was then

refluxed for 2.5 hr and after cooling at 20°C in an ice bath, treated with 10 ml 10M NaOH. The pH was adjusted between 5 - 7 with 1M NaOH and one ml of saturated lead acetate solution was added for clarification. The mixture was then left overnight in a refrigerator to allow settling of the suspended particles. The supernatant was filtered through Whatman No.2 filter paper using a light vacuum, then transferred to a 500 or 1000 ml volumetric flask and made up to volume. A one ml aliquot of this solution was used to determine reducing sugars as outlined before. Starch was calculated in duplicate as 0.90 times the reducing sugar equivalent.

Titrable acidity

The AOAC (1965) method was used to determine titrable acidity in samples of frozen puree. The puree had been sealed in plastic bags and kept frozen at -37° C. Twenty g of puree was used to make a slurry with 100 ml distilled water. The slurry was titrated with 0.10M NaOH to pH 8.1 using a pH meter and magnetic stirrer. Titrable acidity was determined in duplicate and expressed in m.equiv/100g.

Histochemical Properties

Fruits used in these studies were ripened at 15 - 17°C and 85% relative humidity. During ripening a sample of five fruits were removed every two days for histochemical and histological studies. Free-hand

transverse sections of fresh pulp tissue were cut with a razor blade and tested for starch and esterified pectins. Cross sections of pulp 5 mm thick from the middle of the fruit were used to study "tannin" distribution.

Starch

The method of Jensen (1962) was used in this determination. An IKI solution consisting of 0.3g iodine and 1.5g potassium iodide dissolved in 100 ml distilled water was the reagent used. The tissue section was placed on a glass slide and 3 drops of reagent added.

After 2 min excess reagent was washed off with distilled water and the section examined under a microscope. Starch granules were stained dark blue.

Esterified pectins

The hydroxylamine-ferric chloride reaction (Gee et al. 1959) was the test used. Five drops of an alkaline hydroxylamine reagent was placed on a slide and a section lowered on to it. After 5 min an equal volume of solution consisting of one part concentrated HCl and 2 parts 95% ethyl alcohol was added. Excess solution was drained off and the slide flooded with 10% FeCl₃ in 60% ethyl alcohol containing 0.1M HCl. The presence of esterified pectin was indicated by a red color when the section was examined under a microscope.

Tannins (polyphenols)

This test was adapted from the method of Jensen

(1962). It is not specific for tannin since other polyphenols react with the reagent; however, in fruits such as bananas where tannins constitute the largest group of polyphenols this test is useful. Cross sections of pulp cut from the middle of the fruit were placed in a petri dish and covered with a 10% solution of ferric chloride in 60% ethyl alcohol containing 0.1M HCl. After 5 min the sections were thoroughly washed with distilled water and examined for "tannin" location. This was indicated by a dark blue precipitate.

Histological Properties

Sample preparation

A 2.5 cm cross section of pulp from the middle of the fruit was cut into 4 mm slices. Cylindrical cores of tissue 5 mm in diameter were then cut out within the septae close to the degenerated ovules.

Tissue cores were fixed in Navashin's solution (Jensen 1962) at 0° C for 24 hr. A 15 min vacuum infiltration was necessary at the outset of fixation to remove air from the tissue. Navashin's solution is made from two solutions mixed (1:1) before use. Solution A consists of 5g chromium trioxide, 50 ml glacial acetic acid and 320 ml distilled water. Solution B is a maxture of 200 ml formalin and 175 ml distilled water. After fixation the tissue was washed for 30 min in cold water and then dehydrated at 0° C using the schedule of Feder and O'Brien (1968).

The tissue was infiltrated with Tissuemat paraffin (melting point 56.5°C) in a vacuum oven at 62°C. The same material was used for embedding the tissue. The molten paraffin was poured into paper molds and tissue cores were properly oriented using hot needles. The molds were then lowered into a water bath containing crushed ice. When the surface and solidified the mold was submerged and allowed to harden completely. After the block had completely cooled, it was removed from the water, wrapped in aluminum foil and stored in a refrigerator.

Sectioning and staining

Pieces of paraffin containing tissue were attached to wooden blocks using a hot spatula. Excess paraffin was removed from around the tissue and the wooden blocks were placed in the jaws of a Spencer rotary microtome which was used to obtain ribbons $10 - 12 \mu$ thick. The upper surface of a precleaned slide was coated with a thin layer of Haupt's adhesive (Jensen 1962). A few drops of 4% formalin were added and segments of the ribbons were floated on the slide which was then placed on a hot plate at 35° C to allow the sections to expand. Excess formalin was drained and the slide was left to dry overnight on the hot plate.

Tissuemat was removed from the slides by soaking in xylene. After partial hydration in an ethyl alcohol series (100, 95, 70 and 50%), the slides were stained with

safranin-toluidine blue. The staining schedule was adapted from the safranin-fast green method outlined by Jensen (1962). Stained slides were passed through an alcohol series (70, 95 and 100%) and finally through 3 changes of xylene. They were then dried for 24 hours before mounting in Permount with 22 x 60 mm coverslips (No.1). A further drying period of 24 hr was necessary before examination.

A Wild M20 light microscope fitted with a 35 mm Asahi Pentax camera was used to observe and photograph the sections at magnifications of 100 and 400 diameters.

RESULTS AND DISCUSSION

. Changes During Ripening

Physical properties

Fruit size: All variables examined were closely related to each other (Table 1). Generally there was not much difference among groups although fruits in groups 1 and 5 were longer and heavier than the others (Table 2). Length and gross weight showed the most variation in all experimental groups. Variation in size was not considered excessive because the fruits were derived from different "hands" and bunches and were subject to seasonal and geographical effects.

TABLE 1. SIMPLE CORRELATIONS AMONG SIZE CHARACTERISTICS OF BANANAS (pooled data, n = 195)

	Length	Gross wt.	Pulp wt.
Gross wt.	0.847		
Pulp wt.	0.805	0.962	
Cross section area	0.489	0.762	0.831

Pulp-to-peel ratio: All groups of fruit showed a steady increase in pulp-to-peel ratio (Figure 3). Ratios ranged from 1.35 in green fruits to 2.14 in over-ripe fruits. The results of this study are in agreement with the observations of vonLoesecke (1950) and Simmonds (1966).

TABLE 2. MEANS, STANDARD DEVIATIONS AND t-TEST RESULTS FOR SIZE CHARACTERISTICS OF BANANAS.

Group	Length (cm)	Gross wt. (g)	Pulp wt. (g)	Area (cm ²)
1	23.22 ^{ab} 1	181.0 ^{ab}	114.73 ^a	6.97 ^a
	1.64	17.63	14.41	0.48
2	21.76 ^{cd}	172.84 ^c	111.05 ^a	7.18 ^a
	1.91	25.36	16.67	0.64
3	21.26 ^c	171.89 ^{ac}	110.87 ^a	7.24 ^a
	1.75	31.07	31.07	0.80
4	22.03 ^{ce}	173.72 ^{acd}	111.67 ^a	7.06 ^a
	2.33	42.57	31.2	1.11
5	23.90 ^a	191.46 ^{bd}	119.93 ^a	7.04ª
	2.00	35.13	24.89	0.99
6	22.66 ^{bde}	178.29 ^{bc}	109.58 ^a	6.90 ^a
	2.46	33.02	22.18	0.70

¹ Means in a column sharing the same letter do not differ at P = 0.05.

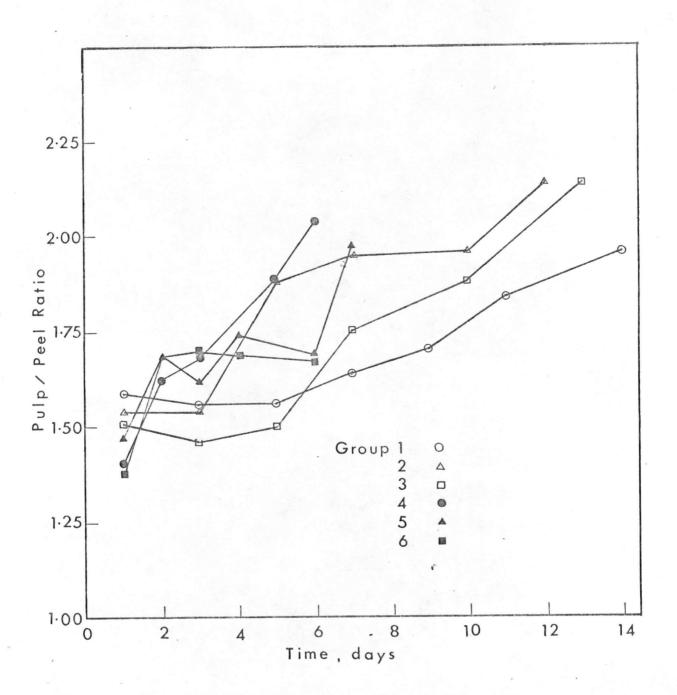


FIGURE 3. Changes in pulp-to-peel ratio of bananas during ripening.

Color: The change in color index during ripening is shown in Figure 4. In most groups a change in color was observed after one day of ripening. Rate of change was somewhat variable although that of groups 4, 5 and 6 tended to be less so.

Typical reflectance curves of banana peel during ripening are shown in Figures 5 and 6. There was an overall increase in reflectance during ripening with the greatest changes occurring at 672 nm. Reflectance changes at that wavelength were associated with the reduction in chlorophyll content of the peel. Finney et al. (1967) found that chlorophyll in banana peel was associated with a change in reflectance at 675 nm while Powers et al. (1953) reported that reflectance at 678 nm was related to changes in surface concentration of chlorophyll in lemons.

Index of Variance Reflectance (IVR) decreased during ripening, but ripe fruits in groups 4, 5 and 6 tended to have higher values than groups 1, 2 and 3.

Rheological properties

Force-deformation behavior: Green fruit with a color index of 2 were hard and brittle with cylindrical sections able to withstand up to 65 kg of force. After one or two days the ripening of all groups showed a marked increase in softening. This was reflected by sudden changes in maximum force, linear limit and linear limit energy.

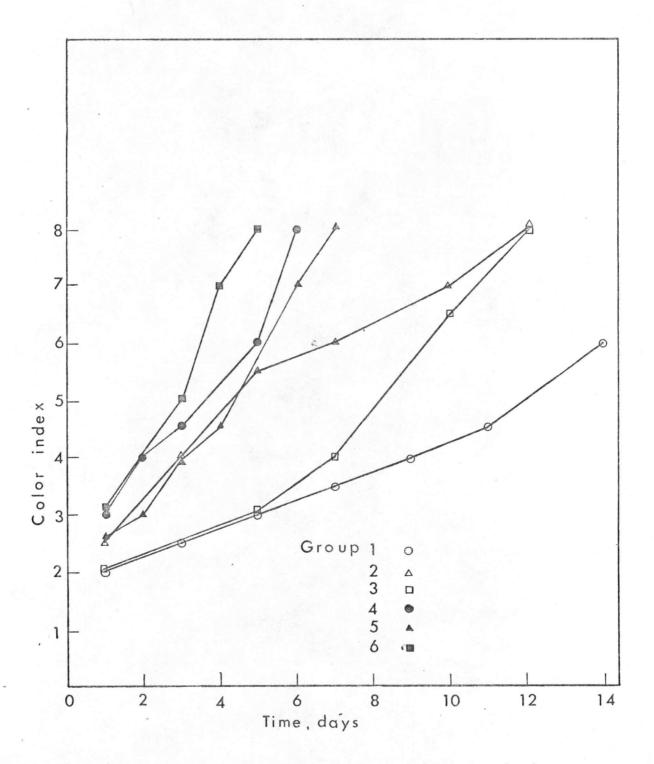


FIGURE 4. Changes in color index of bananas during ripening.

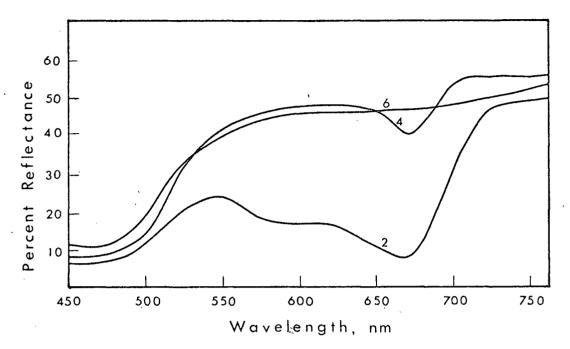


FIGURE 5. Reflectance curves for the peel of bananas ripened at 16 \pm 1 °C. Numbers on the curves indicate peel color index.

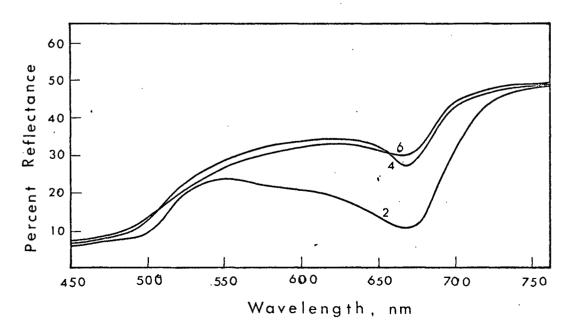


FIGURE 6. Reflectance curves for the peel of bananas ripened at 25 \pm 1° C. Numbers on the curves indicate peel color index.

Changes in linear limit during ripening are illustrated in Figure 7. When the fruits from all groups attained full ripeness (full yellow), linear limit remained relatively constant and showed less variation among groups.

Deformation increased steadily during early ripening. In groups 1, 2 and 3 the ripe fruits were not as readily deformed as in the other groups. Linear limit energy decreased with ripening in all groups with the rate of change being greatest in groups 4, 5 and 6. There was much variation in green fruit and, to a lesser extent, in ripe fruit.

Flow behavior: Data from all groups fitted the two power-law flow models well. The mean coefficient of determination for the power-law with yield stress (Equation [3]) was 0.97 and that for power-law (Equation [2]) was 0.96. Thus both models accurately describe the flow behavior of banana puree, however the power-law with yield stress was selected for discussion in this presentation.

During ripening the fruit puree decreased in consistency. The power-law consistency coefficient (\underline{m}) decreased and flow behavior index (\underline{n}) increased, i.e. the puree approached Newtonian flow. The rate of change in \underline{m} was greatest in groups 4, 5 and 6 (Figure 8), while values for n tended to be larger in the same groups.

The parameter \underline{m} correlated well (Table 3) with force deformation variables as well as with starch and sugar



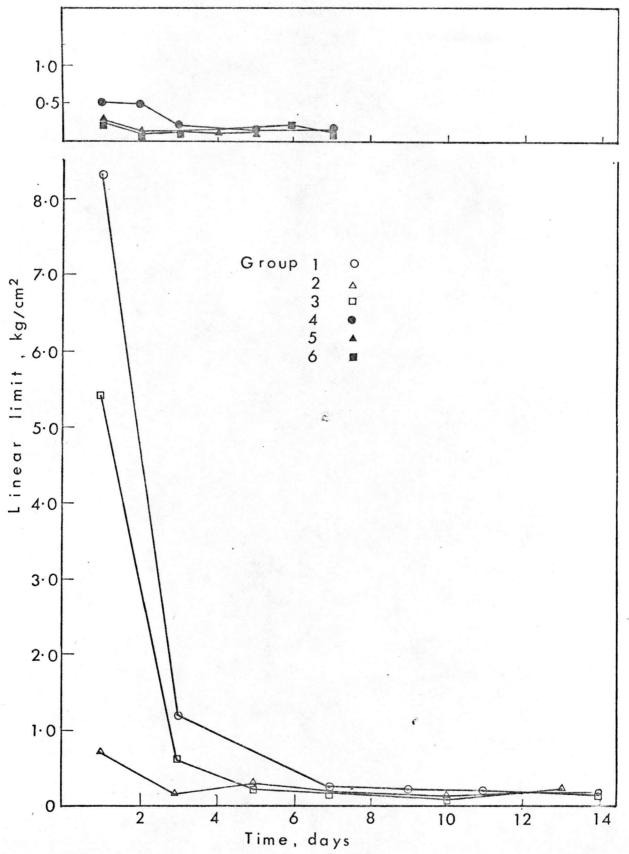


FIGURE 7. Changes in linear limit of banana pulp tissue during ripening.

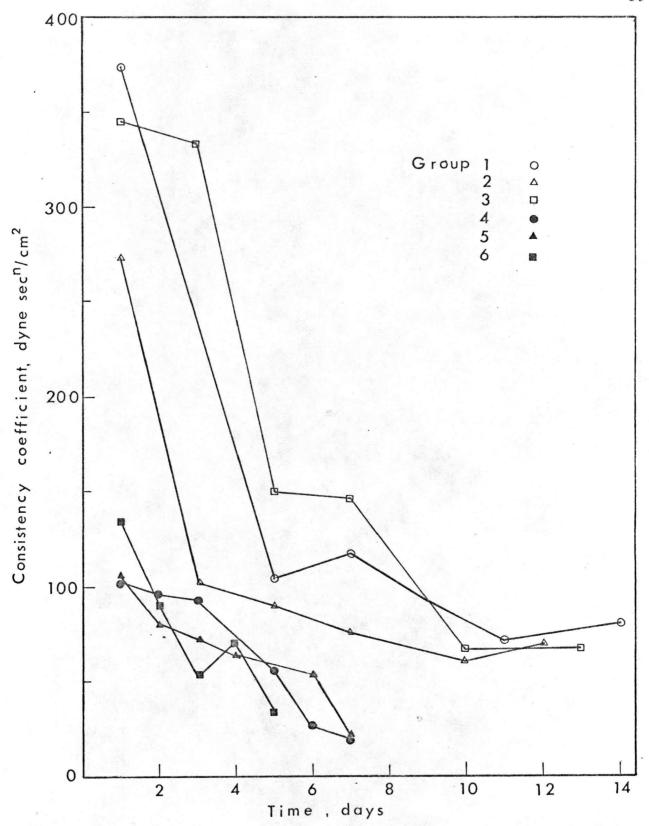


FIGURE 8. Changes in consistency coefficient of banana puree during ripening.

content. Yield stress values decreased during ripening from 270 - 350 dynes/cm² in green fruits to 10 - 25 dynes/cm² in over-ripe fruits. Decrease in yield stress followed a pattern similar to that of the parameter m.

Chemical properties

Moisture: Moisture content increased from 71.3 to 76.0% in groups, 1, 2 and 3 and from 72.6 to 78.7% in groups 4, 5 and 6 (Figure 9). Increases were within the range of tabulated values (vonLoesecke 1950) and were steady throughout ripening in all groups.

Acidity: Peak acidity occurred in fully ripe fruits although levels were variable among groups. pH decreased from 5.0 - 5.4 in green fruits to 4.2 - 4.8 in fully ripe fruits, then increased with advanced ripening. Titrable acidity increased with initial ripening but decreased with continued ripening. Values ranged from 3.3 - 5.0 m.equiv/100g in green fruits to 4.85 - 6.0 m.equiv/100g in fully ripe fruits and 4.0 - 5.5 m.equiv/100g in over-ripe (heavily spotted) fruits. This suggests that acids are used as respiratory substrates.

Maximum titrable acidity did not generally coincide with minimum pH. This is probably due to differences in buffering capacity of the organic acids present at different stages of ripeness.

Alcohol insoluble solids: There was a steady decrease in AIS during ripening. Green fruits contained

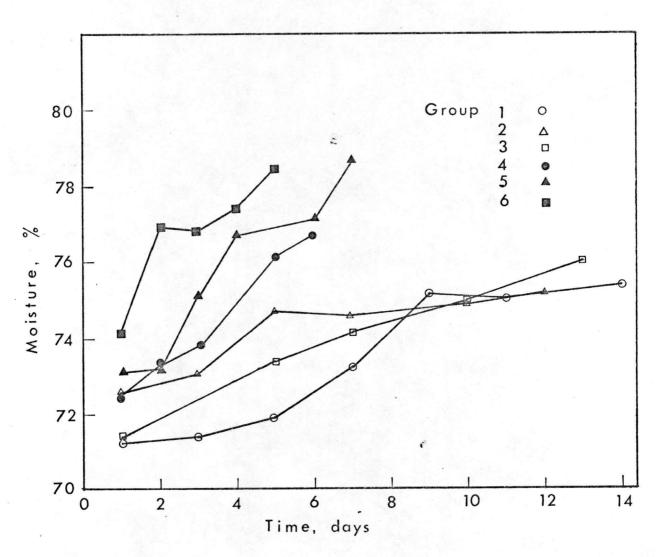


FIGURE 9. Changes in moisture content of banana pulp during ripening.

20 - 25% while in over-ripe fruits the level was generally less than 5%. The rate of decrease was faster and more uniform in groups 4, 5 and 6 (Figure 10). In addition these groups tended to have less AIS than the others.

Changes in AIS during ripening represents transformation of starch and structural materials such as
cellulose, hemicellulose and pectic substances and are closely
related to rheological properties.

Sugars: Total sugar increased during ripening in all groups (Figure 11). Groups 4, 5 and 6 appeared to have slightly large amounts of sugar in over-ripe fruits. With advanced ripening, the sugar content appeared to reach a plateau.

Reducing sugars increased steadily during ripening (Figure 12). The rate of increase as well as the final amount in over-ripe fruit was greatest in groups 4, 5 and 6. Reducing sugars did not decrease in over-ripe fruits, indicating that non-reducing sugars are probably utilized in respiration before reducing sugars.

In this study non-reducing sugars constitute a larger percentage of total sugar. This is in agreement with compositional data given by United Fruit Sales Corp. (1964). In general reducing sugars accounted for about 25% of total sugar content up to full ripeness.

Starch: Changes in starch were generally parallel to those of AIS (Figures 10 and 13). Starch content varied

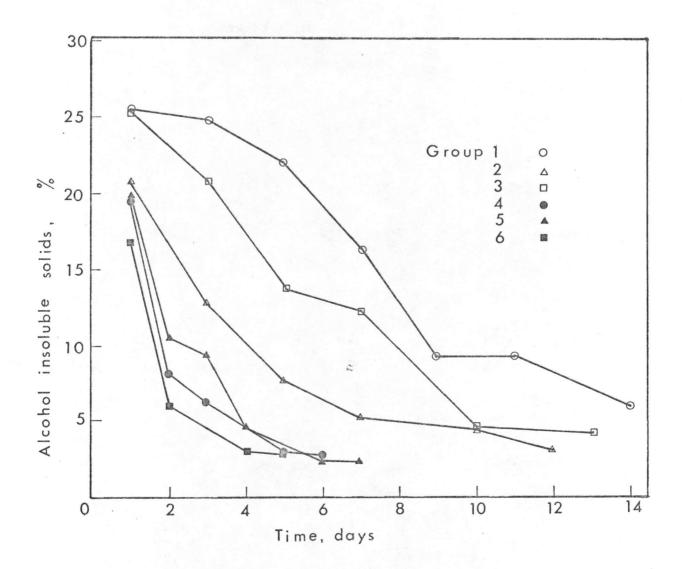


FIGURE 10. Changes in AIS content of banana pulp during ripening.

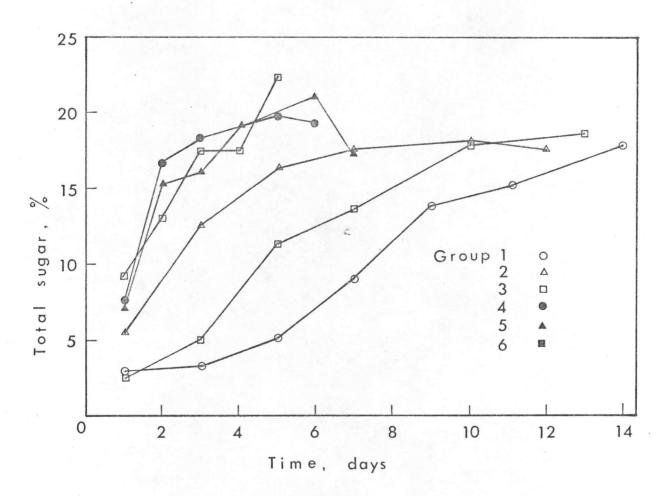


FIGURE 11. Changes in total sugar content of banana pulp during ripening.

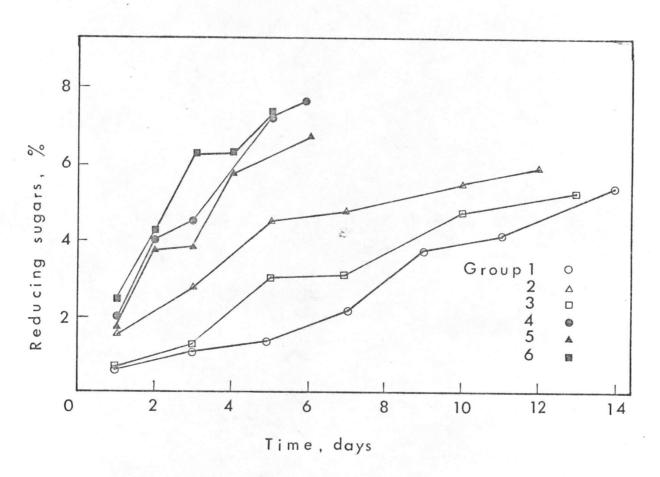


FIGURE 12. Changes in reducing sugar content of banana pulp during ripening.

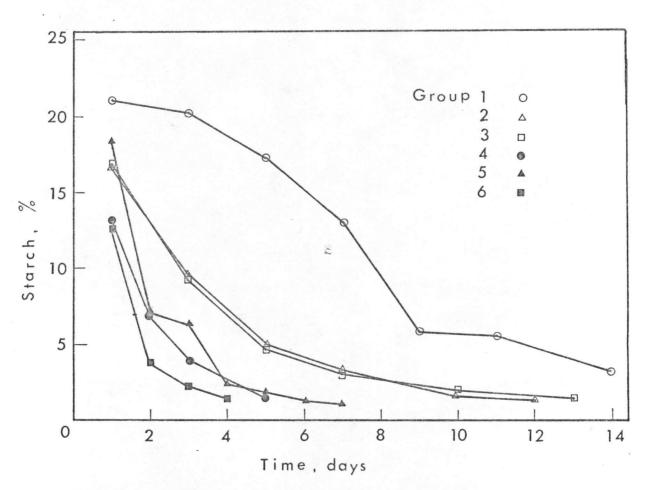


FIGURE 13. Changes in starch content of banana pulp during ripening.

from 17 - 22% in green fruits to less than 2% in ripe fruits.

Histochemical properties

Figure 14 shows that tannins are associated primarily with the carpellary margins. A longitudinal section through one of the margins reveals that latex cells are present in large numbers. Barnell and Barnell (1945) also found that banana fruit tannins occur mainly in latex cells.

During ripening there was a decline in the level of tannins (polyphenols) in the pulp (Figure 14). Goldstein and Swain (1963) have stated that loss of astringency in ripe bananas results from a decrease in "active" tannins due to polymerization.

Starch disappearance began in the placental region at color index 3 and progressed towards the peel. At color index 7, starch was confined largely to the cells at the periphery of the pulp with some starch-laden cells scattered at random throughout the pulp tissues. Starch granules in the central part of part of ripe fruits appeared to decrease in size, however the persistence of starch at the periphery made it difficult to observe whether a similar trend occurred.

Pulp tissue from green fruit (color index 2) did not give a positive reaction with hydroxylamine reagent. At the onset of yellowing (color index 3) an intense red color, indicative of esterified pectins, was obtained with the

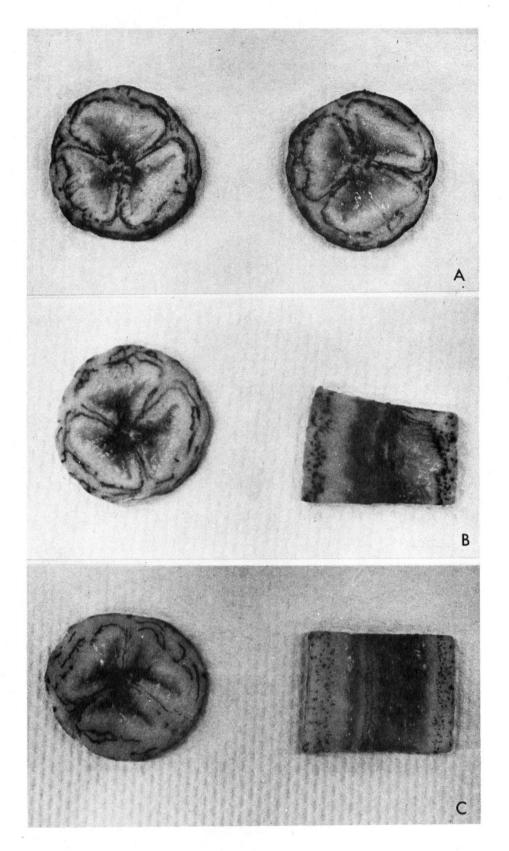


FIGURE 14. Sections of banana pulp tissue showing tannin distribution. A = color index 2; B = color index 4; C = color index 6.5.

reagent. The first appearance of esterified pectins coincided with the decrease in starch at the placental region.

As ripening progressed the red color decreased in intensity and at color index 7 esterified pectins were present only in vascular tissue.

Changes in pectic constituents are considered a major factor in the textural qualities of fruits (McCready and McComb 1954; Pilnik and Voragen 1970). These changes are characterized by an increase in the proportion of soluble to insoluble fractions resulting from depolymerization and de-esterification (Pilnik and Voragen 1970). Thus softening is associated with a decrease in esterified pectins. In this study esterified pectins were present in larger amounts at color index 3 than in hard green bananas. Similar observations in peaches were reported by Reeve (1959). It is apparent that the ratio of soluble to insoluble pectic substances does not explain adequately the role of these substances in banana fruit texture.

Histological properties

This study revealed that during ripening the cell wall in pulp tissue loses its rigidity. Figure 15A shows that the cell wall in pulp tissue at color index 2 is rigid and well defined, but as ripening progresses the cell wall softens such that cells lose their characteristic shapes.

At color index 6.5 to 7.0 there was an apparent thickening of the cell wall (Figure 15C). In contrast to

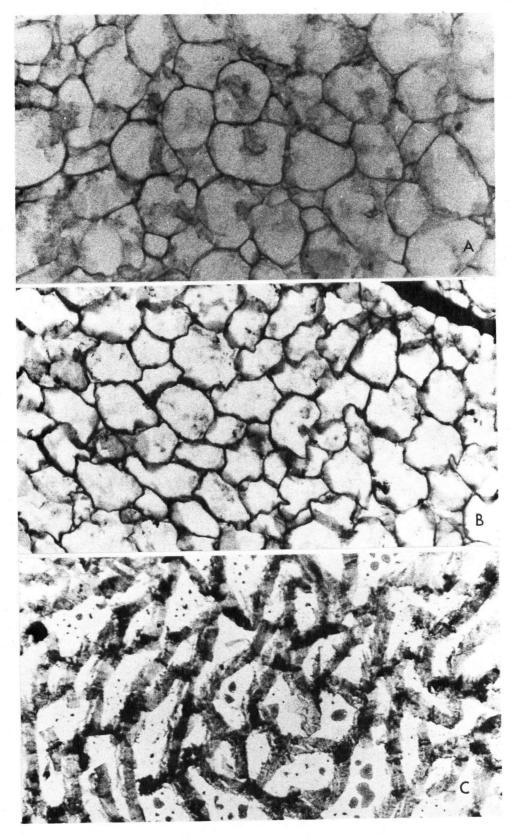


FIGURE 15. Photomicrographs of banana pulp tissue during ripening at $16 \pm 1^{\circ}C$ (X 320).

A = color index 2; B = color index 4; C = color index 7.

loss of rigidity, cell wall thickening did not occur gradually. This phenomenon has not been previously reported in banana tissue, but has been observed in peaches at early stages of ripeness by Reeve (1959). Cell wall hydration can result in thickening, but hydration in banana pulp is associated with the climacteric (Bauer and Workman 1964).

It is possible that wall thickening may have been induced by sample preparation, but if this is so, there occurs at advanced ripening a sudden structural modification which predisposes the cell wall to such thickening. At advanced ripening pulp tissue disintegrates during handling and sections from such tissue show cellular debris within the cells and multiple fractures in the cell wall (Figure 15C).

Effect of Ripening Temperature

Examination of changes during ripening showed that rates of change in groups receiving similar temperature treatments tended to be similar, however the groups were not necessarily at the same stage of ripeness when the experiments began. Since peel color is the most widely used index of ripeness, changes in physical, rheological and chemical properties were studied as a function of color index. This method would enable comparison of these properties at similar stages of ripeness based on peel color. The effect of ripening temperature on physical, rheological and chemical properties was studied by comparing changes in the pooled

data of low and high temperature groups as a function of color index.

Physical properties

During low temperature (16 \pm 1°C) ripening pulpto-peel ratio increased linearly. High temperature (25 \pm 1°C) resulted in slightly lower values which did not increase steadily throughout ripening (Figure 16).

The rate of color change in high temperature groups was roughly twice that in low temperature groups. Change in color index per day was 0.76, 1.18 and 0.89 for low temperature groups and 1.8k, 1.44 and 2.00 for high temperature groups. Persistence of a chlorophyll peak in ripe fruits (Figure 6) was associated with the lack of development of full yellow in high temperature-ripened fruits and resulted in higher IVR values. It is possible that chlorophyll retention may be an initial sign of "boiling"-- a commercial condition which is experienced during ripening above 30°C (Wilkinson 1970). Intense spotting and considerable rotting of the peel at advanced ripening in high temperature groups was probably due to fungal growth (Hall 1967).

Rheological properties

Fruits ripened at low temperature were much firmer than those ripened at high temperature. Linear limit of ripe fruit tissue in both temperature groups were not appreciably different, however deformation due to 1 kg

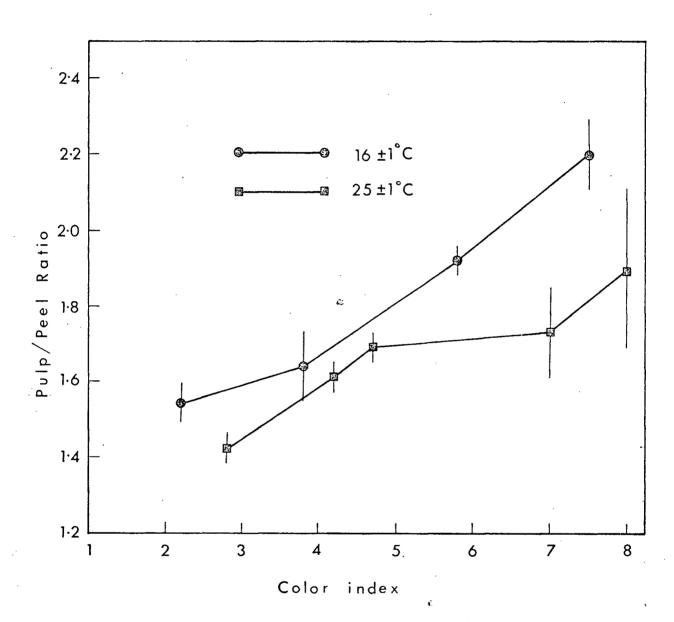


FIGURE 16. Effect of ripening temperature on pulp-topeel ratio of bananas. Vertical bars represent ± one standard deviation.

force showed very different patterns in the two treatments (Figure 17). This variable remained more or less constant in ripe fruits (beyond color index 4) at low temperature but increased linearly with high temperature ripening.

The effects of ripening temperature on flow behavior was reflected in all power-law parameters. Consistency coefficient and yield stress were generally lower while flow behavior index was higher in high temperature groups.

Chemical properties

High temperature resulted in increased moisture content in pulp tissue. This was in contrast to lower pulp-to-peel-ratios (Figure 16). Apparently weight increases due to moisture are nullified by increased structural breakdown and hydrolysis.

Acidity was not affected by temperature treatment.

There was considerable overlapping of pH and titrable acidity in both treatments.

Alcohol insoluble solids (AIS) and starch were affected in a similar pattern by temperature. High temperature resulted in lower levels of both variables throughout ripening; while the AIS content of over-ripe fruits remained constant in low temperature groups, it continued to decline in high temperature groups. Starch content of over-ripe fruits in both treatments decreased slightly on continued ripening.

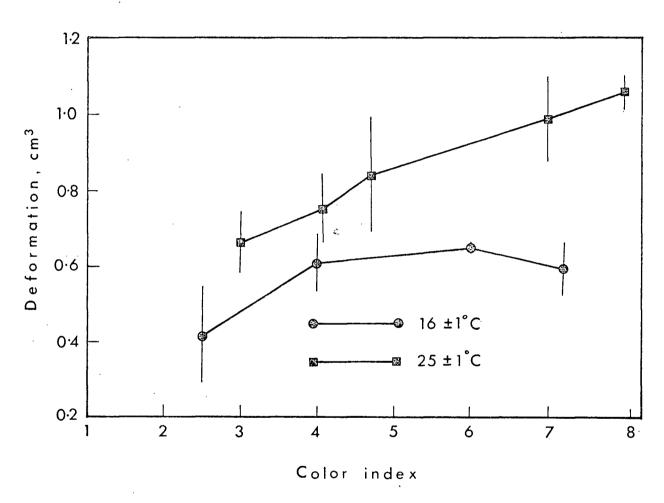


FIGURE 17. Effect of ripening temperature on deformation of banana pulp tissue under 1 kg force. Vertical bars represent ± one standard deviation.

Total sugar content of banana pulp was increased by high temperature, although there was more variation in high temperature groups (Figure 18). The 16°C treatment was characterized by a more or less constant level of reducing sugars in ripe fruits, while the 25°C temperature resulted in a linear increase in these sugars throughout ripening. This accounted for the overall increase in sugar content of high temperature groups.

In this study the ripening treatment was applied during the entire experimental period. Under commercial conditions the fruit is considered ripe at color index 4, thus the effect of ripening temperature should be considered up to that stage of ripeness. Subsequent changes are generally referred to as storage changes and are treated separately. Furthermore, since the study was limited to two temperature treatments, it is not possible to discuss fully the effects of temperature on ripening behavior.

The results support the observations of Sanchez Nieva et al. (1969) and Dalal et al. (1969) that bananas ripened at high temperature are more susceptible to mechanical injury. Reduction of shelf life by high temperature (United Fruit Sales Corp. 1964) is also illustrated by the rates of ripening in high temperature groups. The lack of complete yellowing in high temperature-ripened fruit is probably the initial symptom of a physiological disorder. Haard and

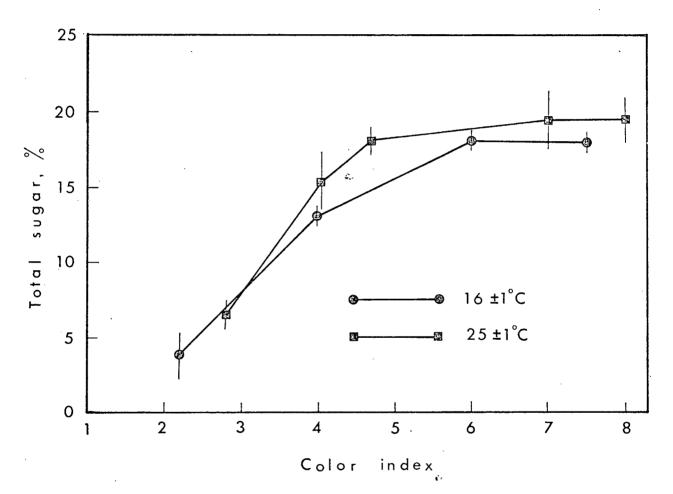


FIGURE 18. Effect of ripening temperature on total sugar content of banana pulp tissue.

Vertical bars represent ± one standard deviation.

Hultin (1969) reported that a similar effect on yellowing was brought about by low relative humidity.

Relationships Among Properties

It has been demonstrated elsewhere in this study that ripening is accompanied by several changes and that ripening temperatures exert a considerable effect on the properties of bananas. The relationships among representative parameters of physical, rheological and chemical properties were studied by pooling data from the three groups in each temperature treatment and calculating simple correlations. This would indicate whether relationships between different variables are the same at both ripening temperatures. In addition, it would be possible to assess the value of peel color as an index of ripeness under different ripening conditions.

Simple correlations among selected variables during ripening at $16 \pm 1^{\circ} C$ and $25 \pm 1^{\circ} C$ are shown in Tables 3 and 4 respectively. In general, the correlations were highly significant (P \leq 0.01) during both treatments. When data from both treatments were pooled (Table 5) correlation coefficients were generally intermediate between those obtained fro the separate treatments. However, correlations between pulp-to-peel ratio and other variables decreased when the data were pooled. There is little indication that ripening temperature exerts a major influence on the

TABLE 3. SIMPLE CORRELATIONS AMONG SELECTED PHYSICAL, RHEOLOGICAL AND CHEMICAL PROPERTIES OF BANANAS RIPENED AT 16 \pm 1 $^{\circ}$ C. (n = 110).

	Color index	Pulp/ peel	IVR	Linear limit	Maxi- mum force	Defor- mation	m *	Total sugar	Reduc- ing sugar	AIS	Starch
Pulp/peel	0.810										7 1 7
IVR	-0.754	-0.672									
Linear limit	-0.452	-0.311	0.606								
Maximum force	-0.474	-0.332	0.628	0.996							
Deformation	0.645	0.532	-0.716	-0.645	-0.654						
m*	-0.670	-0.540	0.704	0.709	0.715	-0.531					
Total sugar	0.881	0.746	-0.904	-0.587	-0.607	0.781	-0.702				
Reducing sugar	0.947	0.814	-0.814	-0.516	-0.535	0.729	-0.696	0.939			
AIS	-0.903	-0.751	0.896	0.597	0.617	-0.781	0.670	-0.994	-0.999		
Starch	-0.896	-0.744	0.904	0.632	0.632	-0.778	0.719	-0.993	-0.943	0.999	
Moisture	0.952	0.791	-0.798	-0.533	-0.552	0.734	-0.634	0.921	0.966	-0.949	-0.940

^{*} Power-law consistency coefficient.

TABLE 4. SIMPLE CORRELATIONS AMONG SELECTED PHYSICAL, RHEOLOGICAL AND CHEMICAL PROPERTIES OF BANANAS RIPENED AT 25 \pm 1° C. (n = 85).

	Colorindex	Pulp/ peel	IVR	Linear limit	Maxi- mum force	Defor- mation	m *	Total	Reduc ing sugar	AIS	Starch
Pulp/peel	0.661										
IVR	-0.734	-0.629									
Linear limit	-0.427	-0.483	0.645	•	`						
Maximum force	-0.432	-0.520	0.630	0.982							
Deformation	0.726	0.690	-0.583	-0.464	-0.47,7					•	
m* ,	-0.885	-0.702	0.703	0.413	0.427	-0.680					
Total sugar	0.761	0.538	-0.814	-0.549	-0.510	0.492	-0.760				
Reducing sugar	0.934	0.674	-0.818	-0.521	-0.507	0.725	-0.905	0.838			
AIS	-0.816	-0.599	0.867	0.583	0,551	-0.609	0.802	-0.933	-0.927		
Starch	-0.826	-0.599	0.849	0.533	0.503	-0.604	0.811	-0.919	-0.925	0.981	
Moisture	0.805	0.477	-0.694	-0.503	-0.473	0.716	-0.795	0.683	0.884	-0.828	-0.829

^{*} Power-law consistency coefficient.

TABLE 5. SIMPLE CORRELATIONS AMONG SELECTED PHYSICAL, RHEOLOGICAL AND CHEMICAL PROPERTIES OF RIPENING BANANAS. (Pooled data, n=195).

	Colorindex	Pulp/ peel	IVR	Linear limit	Maxi- mum force	Defor- mation	m *	Total sugar	Reduc- ing sugar	AIS	Starch
Pulp/peel	0.718										
IVR	-0.613	-0.613									
Linear limit	-0.380	-0.212	0.556								
Maximum force	-0.402	-0.230	0.579	0.996	•						
Deformation	0.598	0.356	-0.583	-0.505	-0.566	p.					
m* .	-0.640	-0.420	0.677	0.710	0.719	-0.566				•	
Total sugar	0.820	0.570	-0.866	-0.544	-0.566	0.681	-0.709				
Reducing sugar	0.909	0.625	-0.785	-0.457	-0.481	0.757	-0.702	0.905			
AIS	-0.853	-0.597	0.877	0.555	0.578	-0.711	0.713	-0.936	-0.936		
Starch	-0.853	-0.594	0.878	0.564	0.587	-0.706	0.727	-0.933	-0.933	0.994	
Moisture	0.873	0.561	-0.749	-0.474	-0.496	0.740	-0.648	0.850	0.939	-0.911	-0.906

^{*} Power-law consistency coefficient.

relationship between selected variables, although correlation coefficients tended to be higher at the lower temperature (Table 3).

Variations in color index account for 63.7 and 82.6% of the variation in moisture and reducing sugar respectively, when all the data were pooled. At both ripening temperatures, Index of Variance Reflectance (IVR) was closely related to color index with correlation coefficients of -0.754 and -0.734 for low and high temperature respectively. This variable, on the average, surpassed color index in correlations with rheological properties.

Correlations among rheological and chemical properties were on the average higher during ripening at low temperature. An exception to this trend was found in the relationship between reducing sugars and consistency coefficient where the correlation coefficients were -0.905 and -0.696 during high and low temperatures respectively. Some rheological properties, such as deformation and consistency coefficients, were closely related to chemical properties.

Correlations among chemical constituents were very high during both treatments. Moisture correlated well with all variables except pulp-to-peel ratio during ripening at high temperature. This is probably due to the high degree of variability in pulp-to-peel ratio during this

treatment.

In spite of its relatively low correlation with rheological properties, color index was the best overall index of fruit quality. Yet the demonstrated effect of ripening temperature on total sugar, moisture and deformation indicates that it should be given careful consideration if external appearance is used to estimate internal quality of bananas. Pulp-to-peel ratio may be a good index of maturity in green fruits (Simmonds 1966) but does not appear to be a good index of stage of ripeness. It is influenced to a large extent by fruit size and ripening temperature. Among the rheological parameters, deformation and consistency coefficient had the best correlation with chemical properties but their usefulness as indices of ripeness are limited by the time and equipment needed for determination.

CONCLUSIONS

The ripening rate of bananas at $25 \pm 1^{\circ}$ C was roughly twice that at $16 \pm 1^{\circ}$ C. High temperature-ripened fruits were characterized by chlorophyll retention which prevented the development of full yellow color. Pulp-to-peel ratio increased during ripening in both treatments but was somewhat lower in fruits ripened at the higher temperature.

Deformation due to 1 kg force increased linearly during ripening at high temperature, while it remained fairly stable beyond color index 6 during ripening at low temperature. Maximum force and linear limit as well as consistency coefficient were generally lower during ripening at high temperature.

Total sugar and moisture content of pulp tissue were higher in fruits ripened at high temperature. Reducing sugars increased linearly throughout ripening at high temperature while at low temperature they remained essentially constant beyond color index 6. Starch and AIS levels were somewhat higher in low temperature-ripened fruits. This indicates that starch hydrolysis was enhanced by high temperature treatment.

Ripening was characterized by a gradual loss of rigidity as well as an apparent thickening of the cell wall

in the pulp of over-ripe fruits. Tannins decreased but did not completely disappear during ripening. Esterified pectins were present in the largest amount at color index 3 and decreased during ripening. Starch granule disappearance began in the central region of the pulp tissue and progressed towards the peel as ripening continued.

Peel color was evaluated by color index and IVR was the best index of stage of ripeness in fruits ripened at both low and high temperature. However, in view of the effects of ripening temperature on some chemical and rheological properties, the relations between these properties and color index would be influenced by ripening temperature. This suggested that ripening temperature may affect eating quality of the ripe fruit, as well as the accuracy of peel color as an index of ripeness.

Correlations among rheological and chemical properties were highly significant although correlations among the chemical variables were higher than those among the rheological variables. Variations in tissue strength (maximum force) could be explained largely by variations in starch and AIS while variations in softening (deformation) could be accounted for mainly by variations in total and reducing sugars. In general, ripening temperatures did not appear to influence greatly the correlation coefficients among the different properties examined.

LITERATURE CITED

- Arthur, H.B., Houck, J.P. and Beckford, G.L. 1968. Tropical Agribusiness Structures and Adjustments Bananas. Graduate School of Business Administration, Harvard University, Boston.
- Assoc. Offic. Agric. Chem. 1965. Official methods of analysis, 10th ed., Washington, D.C.
- Barnell, H.R. 1941. Studies in tropical fruits XIII. Carbohydrate metabolism of the banana fruit during storage at 53°F and ripening at 69°F. Ann. Bot. 5: 607-646.
- Barnell, H.R. 1943. Studies on tropical fruits XV. Hemicellulose metabolism of the banana fruit during ripening. Ann. Bot. 7: 297-323.
- Barnell, H.R. and Barnell, E. 1945. Studies in tropical fruits XVI. The distribution of tannins within the banana and changes in their condition and amount during ripening. Ann. Bot. 9: 77-99.
- Bauer, J.R. and Workman, M. 1964. Relationships between cell permeability and respiration in ripening banana fruit tissue. Plant Physiol. 39: 540-543.
- Bhargava, V.K. 1970. Drying of wheat grain in thin layers.
 M.A.Sc. Thesis. University of British Columbia,
 Vancouver, Canada.
- Biale, J.B., Young, R.E. and Olmstead, A.J. 1954. Fruit respiration and ethylene production. Plant Physiol. 29: 168-174.
- B.C. Dept. Industrial Development Trade and Commerce. 1972. External trade 1970. Economic Statistics Branch, Victoria.
- Burg, S.P. and Burg, E.A. 1965. Relationship between ethylene production and ripening in bananas. Bot. Gaz. 126: 200-204.
- Dalal, V.B., Nagaraja, N., Thomas, P. and Amla, B.L. 1969. Some aspects of the storage of dwarf Cavendish bananas at refrigerated temperature for export. Indian Food Packer 23: 1-6.

- Eheart, J.S. and Mason, B.S. 1966. Sugar and acid in the edible portion of fruits. J. Am. Diet. Assoc. 50:130-132.
- Feder, N. and O'Brien, T.P. 1968. Plant microtechnique: some principles and new methods. Am. J. Bot. <u>55</u>: 123-142.
- Finney, E.E., Ben-Gera, I. and Massie, D.R. 1967. An objective evaluation of changes in firmness of ripening bananas using a sonic technique. J. Food Sci. 32: 642-646.
- Food and Agriculture Organization of the United Nations.
 1971. FAO Commodity Review and Outlook 1970-71.
 Commodities Division, Rome.
- Furuholmen, A.M., Winefordner, J.D., Knapp, F.W. and Dennison, R.A. 1964. Potato sugars. J. Agr. Fd. Chem. 12: 109-112.
- Gee, M., Reeve, R.M. and McCready, R.M. 1959. Reaction of hydroxylamine with pectinic acids. Chemical studies and histochemical estimation of the degree of esterificiation of pectic substances in fruit. J. Agr. Fd. Chem. 7: 34-38.
- Goldstein, J.L. and Swain, T.S. 1963. Changes in tannins in ripening fruits. Phytochem. 2: 371-383.
- Gottreich, M., Temkin-Gorodeiski, N., Peled, A., Spodheim, R. and Aharoni, Y. 1969. The determination of the stage of ripeness of bananas by colorimetry. Tropical Agriculture 46: 239-245.
- Haard, N.F. and Hultin, H.O. 1969. Abnormalities in ripening and mitochondrial succinoxidase resulting from storage of preclimacteric fruit at low relative humidity. Phytochem. 8: 2149-2152.
- Haarer, A.E. 1964. Modern Banana Production. Leonard Hill, London.
- Hall, E.G. 1967. Technology of banana marketing.CSIRO Food Preserv. Q. 27: 36-42.
- Jensen, W.A. 1962. Botanical Histochemistry. W.H. Freeman and Co.

- Looney, N.E. and Patterson, M.E. 1967. Chlorophyllase activity in apples and bananas during the climacteric phase. Nature 214: 1245-1246.
- McCready, R.M. and McComb, E.A. 1954. Pectic constituents in ripe and unripe fruits. Food Res. 19: 530-535.
- McGlasson, W.B. 1970. The ethylene factor, p.475-519. In Hulme, A.C. (ed.) Biochemistry of Fruits and Their Products. Vol. I. Academic Press.
- Miller, C.L. and Ross, E. 1963. Non-volatile organic acids of the dwarf Cavendish (Chinese) variety of bananas. J. Food Sci. 28: 193-194.
- Murata, T. 1970. Studies on post-harvest ripening and storage of banana fruits VIII. Effect of ripening methods on the fruit qualities. J. Food Sci. Technol. (Japan) 17: 462-466 (In Japanese).
- Murata, T. and Ku, H.S. 1966. Studies on post-harvest ripening and storage of banana fruits. Part 5. Physiological studies of chilling injuries in Bananas II. J. Food Sci. Technol (Japan) 13: 466-471 (In Japanese).
- Palmer, J.K. 1971. The banana, p. 65-105. <u>In Hulme</u>, A.C. (ed.) Biochemistry of Fruits and Their Products Vol. II. Academic Press.
- Pilnik, W. and Voragen, A.G.J. 1970. Pectic substances and other uronides, p. 53-87. <u>In Hulme</u>, A.C. (ed.) Biochemistry of Fruits and their Products. Vol. I. Academic Press.
- Poland, G.L., Manion, J.T., Brenner, M.W. and Harris, P.L. 1938. Sugar changes in the banana during ripening. Ind. Eng. Chem. 30: 340-342.
- Powers, J.B., Gunn, J.T. and Jacob, F.C. 1953. Electronic color sorting of fruits and vegetables. Agric. Eng. 34: 149-154 and 158.
- Pratt, H.K. and Goeschl, J.D. 1969. Physiological roles of ethylene in plants. Ann. Rev. Plant Physiol. 20: 541-576.

- Ram, H.Y.M., Ram, M. and Steward, F.C. 1962. Growth and development of the banana plant. 3B. The structure and development of the fruit. Ann. Bot. 26: 657-673.
- Sanchez Nieva, F., Hernandez, I. and Buesco de Vinas, C.
 1969. Ripening of Montecristo bananas. J. Agric.
 Univ. Puerto Rico 53: 274-283.
- Sarkissian, I.K. 1965. Texture-firmness differences in ripe Valery versus Gros Michel fruit. United Fruit Research Dept., New York (mimeograph).
- Seelig, R.A. 1969. Fruit and Vegetable Facts and Pointers Bananas. United Fresh Fruit and Vegetable Association, Washington, D.C.
- Simmonds, N.W. 1966. Bananas. 2nd ed. Longmans.
- Standard Fruit and Steamship Co. 1964. Ripening and warehousing of Cabana bananas. Technical Service Department, New Orleans.
- Statistics Canada. 1972. Summary of Imports. Vol. 26, No.1 Information Canada, Ottawa.
- Steward, F.C., Hulme, A.C., Freiberg, S.R., Hegarty, M.P., Pollard, J.K., Rabson, R. and Barr, R.A. 1960. Physiological investigations of the banana plant.

 1. Biochemical constituents detected in the banana plant. Ann. Bot. 24: 83-116.
- Stratton, F.C. and vonLoesecke, H. 1931. Changes in osmotic pressure of bananas during ripening. Plant Physiol. 6: 361-365.
- Ting, S.V. 1956. Rapid colorimetric methods for simultaneous determination of total reducing sugars and fructose in citrus juices. J. Agr. Fd. Chem. 4: 263-266.
- United Fruit Sales Corp. 1964. Banana ripening guide. Boston. 32 pp.
- United Fruit Sales Corp. 1970. Your guide to greater profits.
 Boston. 24 pp.
- U.S. Department of Agriculture. 1954. The commercial storage of fruits and vegetables and florist and nursery stocks. Agricultural Handbook No.66. Agricultural Marketing Service, Washington, D.C.

- U.S. Department of Agriculture. 1971. World demand prospects for bananas in 1980. Foreign Agricultural Report No.69. Economic Research Service, Washington, D.C.
- vonLoesecke, H.W. 1950. Bananas. 2nd ed. Interscience Publishers.
- Wilkinson, B.G. 1970. Physiological disorders of fruit after harvesting, p. 537-554. <u>In Hulme A.C.</u> (ed.) Biochemistry of Fruits and their Products. Vol.1. Academic Press.

£.,.

Wolfson, A.M. 1928. Banana morphology. United Fruit Co., Research Dept. Bull. No.6.