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Production of Volatile Flavor Compounds in Ultrahigh-Temperature Processed Milk during Aseptic Storage

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Volatile flavor compounds in ultrahigh-temperature (UHT) processed milk were investigated to determine their role in off-flavor development during aseptic storage. The milk samples were processed at 145 °C for 3 s with and without added ascorbic acid and stored at 3, 22, and 35 °C for 5 months. Flavor isolates were prepared through steam vacuum distillation and subsequent extraction of the distillate with dichloromethane. The isolates were analyzed using gas chromatography and mass spectrometry. The milk was regularly analyzed by various chemical methods and evaluated by a taste panel during storage. Twenty-six compounds were identified, seven of which were not previously reported in UHT milk. Gas chromatographic profiles indicated that 2-pentanone, 2-heptanone, 2-nonanone, and *n*-hexanal increased most in concentration during storage. The rate of increase in odd carbon-numbered methyl ketones (C_{3-13}) was dependent upon storage temperature, whereas the rate of increase in aldehydes was dependent upon both oxygen content and temperature of storage. Although methyl ketones were the most abundant class of compounds, aldehydes appeared to be most important in contributing to the off-flavor of stored UHT milk.

Development of stale and/or oxidized flavors during storage is a primary deterrent for acceptability of ultrahigh-temperature (UHT) processed milk. The flavor often appears within a month at room temperature and increases gradually as a function of time. Ashton (1965) reported that, at higher temperatures of storage (21–38 °C), the UHT milk packed aseptically in waxed paper–polyethylene laminates showed signs of developing an incipient oxidative rancidity or cardboardy flavor at about 19 days. Kirk et al. (1968) observed that in UHT milks the rate of staling was a function of storage temperature and paralleling the development of staleness were increases in carbonyl compounds as well as the disappearance of many unidentified components. Ashton et al. (1969) reported that the course of off-flavor development was not only associated with storage temperature but affected by light and oxygen as well. Zadow and Birtwistle (1973) reported that the major factors influencing flavor changes during storage of UHT milk were the level of dissolved oxygen present in the product after processing and the storage temperature. The influence of initial oxygen content on the flavor of samples stored at 2 °C was comparatively minor, most samples being considered "very good" independent of their oxygen content. For samples stored at 20 °C an initially low oxygen content resulted in a poor flavor performance during the first few weeks of storage while an initially high oxygen content resulted in the development of oxidized or rancid flavors at the early stages of storage. Samples with an intermedate initial oxygen content (P_{O_2} 60–100 mmHg) were preferred to achieve a balance between these extremes. Storage at 38 °C resulted in marked visual browning of samples and a rapid decrease in acceptability of flavor independent of oxygen concentration. Thomas et al. (1975) investigated the effect of dissolved oxygen content on flavor and chemical changes during storage of indirectly heated UHT milk. Flavor acceptability increased to a maximum after a few days of storage and then declined with storage. The increase was associated with less off-flavor described as cabbagy and the decrease with more "stale" off-flavor descriptions. Milks with higher initial oxygen contents were preferred up to 8–13 days, but thereafter acceptability was independent of initial oxygen content.

Several workers have studied the volatile compounds present in stored dairy products (Arnold and Lindsay, 1968; Arnold et al., 1966; Bassette, 1958; Bingam, 1964;

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Kirk et al., 1968; Muck et al., 1963; Nawar et al., 1963; Parks and Patton, 1961; Parks et al., 1964; Patel et al., 1962; Scanlan et al., 1968). Aldehydes (C_1–C_{10}, C_{12}, C_{14}), methyl ketones (C_3–C_{15} odd carbons), fatty acids (C_6–C_{16}, even carbons), furfuraldehyde, 2-methylheptanal, benzaldehyde, δ-decalactone, δ-dodecalactone, γ-dodecalactone, o-aminoacetophenone, dimethyl sulfide, benzothiazole, naphthalene, and dichlorobenzene have been identified. Arnold et al. (1966) considered the stale flavor of concentrated sterile milk to be due to longer chain methyl ketones, δ-decalactone, benzaldehyde, and o-aminoacetophenone.

This work was undertaken to isolate and identify volatile flavor compounds from UHT milk and to investigate effects of certain processing and storage parameters such as oxygen content, temperature, and ascorbic acid on changes in concentration of volatile flavor compounds during aseptic storage.

EXPERIMENTAL SECTION

Preparation of UHT Milk. A Cherry-Burrell No-Bac Unitherm Model 40 and a Dole Aseptic Canning System Model 60SA-2 located in the Borden Research Laboratories at Syracuse, N.Y., were used for processing all samples. Raw mixed herd milk which contained 3.5% butter fat was secured from Grade A milk producers in the vicinity of Central New York. The milk was preheated in two stages to 77 and 109 °C, sterilized at 145 °C for 3 s, and cooled to 74 °C before homogenization. It was homogenized at 211 kg/cm^2, cooled to 21 °C, and packed aseptically in 250-mL C-enamelled tin cans. Two kinds of samples were prepared: one lot was untreated control milk and the other lot had 200 mg of L-ascorbic acid added per liter of milk before processing (AA milk). The day after processing, the samples were transported to St. Paul, Minn., by air freight and stored in our laboratory at 3, 22, and 35 °C for various analyses during 5 months of storage.

Chemical Analysis. L-Ascorbic acid was determined by the 2,6-dichlorophenolindophenol titration method (Toothill et al., 1970).

Oxygen content of each sample was measured by an oxygen-permeable electrode with a YSI Oxygen Monitor Model 53 (Yellow Spring Instrument Co., Inc., Yellow Spring, Ohio) at a constant temperature of 23 °C.

Brown pigment formation was estimated by the trypsin digestion method (Choi et al., 1949). Milk samples with added trypsin were incubated at 45 °C for 2 h. Absorbance was measured in a Coleman Junior II Spectrophotometer Model 620 at 420 nm against distilled water. Time interval between filtration and absorbance reading were kept uniform to avoid an increase in reading (Patton, 1952).

Steam Vacuum Distillation. The apparatus used for the steam vacuum distillation consisted of a glass steam generator, vapor trap, 12-L sample flask, large condenser, distillate collection flask, Davies condenser, and three liquid nitrogen Dewar flasks. The steam generator was kept between 43 and 45 °C, and manometer readings were held between 8.5 and 9.0 mmHg during the distillation by manipulating a manostat for vacuum control. Ice–water was used to cool the condensers.

Isolation of Volatiles. Six liters of milk, which had been prewarmed to 29 °C, was placed in the 12-L round-bottomed flask of the distillation system with 0.3 g of purified tristearin added as an antifoaming agent (Jeon et al., 1976). One milliliter of 1-decanol–dichloromethane solution (5 μL/mL) was then added to provide an internal standard. The distillation was performed for 3.0 h and 318 ± 15 mL of the distillate collected in the ice–water trap. After completing the distillation, the distillate was transferred to a 1-L separatory funnel. The liquid nitrogen traps were rinsed with 100 mL of redistilled dichloromethane. The distillate was extracted five times with 20 mL of dichloromethane. A total of 200 mL of dichloromethane was used. The dichloromethane extract was dried with 0.5 g of anhydrous magnesium sulfate, filtered, and then evaporated to 0.20 mL under a gentle stream of nitrogen gas on a warm heating plate. Immediately after concentrating, 5.0 μL of this concentrate was injected into the gas chromatograph for analysis.

Gas Chromatography. A Hewlett-Packard Model 7620A Research Gas Chromatograph equipped with a hydrogen flame ionization detector was used in this study. Separation of volatiles was accomplished using a 3 m × 0.32 cm o.d. stainless steel column packed with 10% Carbowax 20M on 80–100 Chromosorb P (Applied Science Laboratories, Inc., State College, Pa.). Since aliphatic aldehydes cochromatographed with the same carbon numbered methyl ketones on Carbowax 20M, a short column (0.30 m × 0.32 cm) of 10% FFAP on 60–70 mesh Chromosorb W (Wilkens Instrument & Research, Inc., Walnut Creek, Calif.) was attached to the inlet of the Carbowax column. The FFAP column adsorbed the aliphatic aldehydes, permitting the quantification of both classes of carbonyls (Allen, 1966). The column oven was programmed from 50 to 200 °C at 4 °C/min with a 2-min postinjection hold and a 20-min hold at a final limit. The carrier gas (helium) flow rate was 35 mL/min and the injection port temperature was 230 °C. A Hewlett-Packard Model 3370B integrator was used to determine gas chromatographic peak areas.

Mass Spectrometry. A LKB Model 9000 combined gas chromatograph–mass spectrometer was used to identify the compounds utilizing the same columns and temperature programs described above. Mass spectra were obtained with a constant accelerating voltage of 3500 V with 20 and/or 70 eV and a scanning time of 5 s over a m/e range of 10 to 300.

Quantitative Estimation of Volatile Compounds. To estimate the amounts of volatile flavor compounds in UHT milk, known amounts of standard compounds (0.08–0.5 ppm) were added to pasteurized, homogenized milk and mixed well. The milk sample was then vacuum-distilled, extracted, and concentrated with the same procedures used for the UHT milk. A control sample of milk was also analyzed. The concentrations of volatile compounds in UHT milks were then estimated from the peak areas of known concentrations by taking ratios of the peak areas.

Sniffing Gas Chromatographic Effluents. The odor properties of gas chromatographic peaks were assessed by sniffing the gas chromatographic effluents by three experienced laboratory personnel. The carrier gas flow was split approximately 1:10, the smaller part going to the flame detector and the larger part to a sniffing port.

Sensory Evaluation of Milk. Milk samples were evaluated for flavor by four trained panel members. Coded samples were adjusted to 16 °C before testing and presented randomly to the panel. Acceptability rating scale was 1 to 8, with 1 indicating extreme dislike and 8 indicating extreme liking. Intensities of stale flavor were rated on a scale of 0 to 4, with 0 indicating none and 4 indicating pronounced.

RESULTS AND DISCUSSION

Retention of Ascorbic Acid. L-Ascorbic acid in the control milk was essentially depleted within a week when stored at 22 and 35 °C. The initial rate of loss of ascorbic acid was slightly lower at 3 °C storage but was depleted

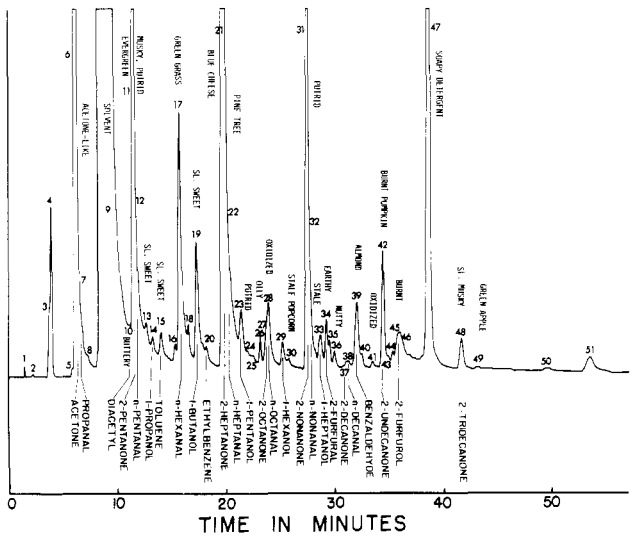

Figure 1. A typical gas chromatogram obtained from the flavor isolate of UHT milk stored 3 months at 35 °C.

after about 30 days. Ascorbic acid-added milk (AA milk) initially followed a pattern similar to that of the control milk but leveled off at about 170 mg/L of milk during storage. This "leveling-off" might be due to the depletion of dissolved oxygen in the system.

Dissolved Oxygen. Oxygen levels in the control milk decreased slowly after initially high rates of loss and showed temperature dependency. Dissolved oxygen decreased to about 1 ppm after about 60 days of storage at 35 °C, while it decreased slowly at 22 °C throughout storage with about 2 ppm still remaining after 5 months. At 3 °C storage, the dissolved oxygen essentially leveled off at about 4.5 ppm after 30 days. However, in the AA milk, dissolved oxygen was rapidly depleted until levels reached a constant value of 1 ppm after about 15 days of storage at 22 and 35 °C. At 3 °C, the rate of depletion was slower but still reached a level of 1 ppm after 5 months of storage.

Nonenzymatic Browning. Absorbance values indicated that little browning occurred in the control milk stored at 3 and 22 °C while the milk stored at 35 °C increased slightly in color throughout storage. Addition of ascorbic acid caused an increase in the rate of browning at all three storage temperatures. It has been suggested that ascorbic acid might be directly involved in the formation of pigments through the action of dehydroascorbic acid as a reductone (Hodge, 1953).

Identification of Volatile Compounds. A typical chromatogram obtained from the flavor isolate of 3-month-old UHT milk is shown in Figure 1. Odor properties of each peak are noted at the nearest possible space. At peak no. 11, for example, an evergreen odor was noted momentarily and then a strong musky and putrid odor prevailed over the entire peak region. A strong green grass or cut-grass odor was sniffed at peak no. 17. A very intense blue cheese (aged) odor was detected at peak no. 21 and then changed to a short note of pine tree-like odor at the region of peak no. 22. Names of compounds identified are also listed at the bottom of the chromatographic peaks. The identification was based on mass spectrometry, odor property, cochromatography of unknown and authentic compound, and a 10% FFAP aldehyde subtractor column. Peak no. 3, 4, 9, and 13 originated from the solvent (Jeon et al., 1976) and are an unknown, cyclohexane, dichloromethane, and chloroform, respectively. Aldehydes such as propanal (peak no. 7), *n*-pentanal (peak no. 12), *n*-heptanal (peak no. 22), and

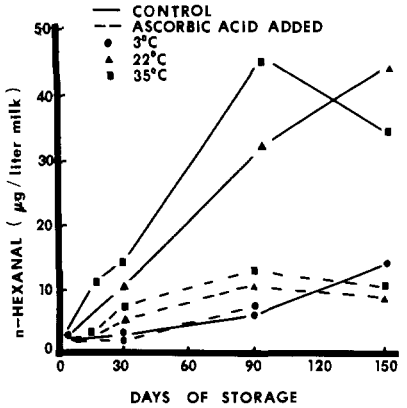

Figure 2. Changes in concentration of *n*-hexanal in control and ascorbic acid-added milk (200 mg/L) during storage.

n-nonanal (peak no. 32) overlapped with the same carbon-numbered methyl ketones. Due to the low concentration of aldehydes (relative to the methyl ketones), we were not able to obtain mass spectra for the aldehydes. However, we could observe the subtraction of the aldehyde peaks by the FFAP column. Poor subtraction for *n*-pentanal by the FFAP column (Withers, 1972) was improved by starting the temperature programming at 50 °C with a 2-min hold. The *n*-hexanal peak was used as an indicator for the deterioration of the FFAP column. Peak no. 17 was identified as *n*-hexanal. Peak no. 33 was tentatively identified as 1-heptanol by mass spectrometry and cochromatography. However, the mass spectrum was not clean enough for positive identification. The panel detected a strong popcorn-like odor at peak no. 30 and a fairly strong green apple or apple pie-like odor at peak no. 49 which could not be identified. Peak no. 47 is the internal standard (1-decanol) which was added to milk. Among the compounds identified, *n*-pentanal, *n*-octanal, *n*-nonanal, *n*-decanal, 1-pentanol, and 1-hexanol are reported for the first time in UHT milk.

Changes in Concentration of Volatile Compounds. Most compounds identified in Figure 1 appeared to be present in fresh UHT milk. However, aliphatic aldehydes other than *n*-hexanal may not be present initially. The presence of 1-butanol was confirmed in fresh UHT milk but no other alcohols were detectable.

Increases in concentration of volatiles during storage occurred primarily in carbonyl compounds. Those exhibiting distinct increases were aliphatic aldehydes, methyl ketones, and 1-butanol. Although Kirk et al. (1968) observed the disappearance of many of the less volatile components in UHT milk during storage, there were no measurable decreases in concentration of any components in this study.

Figure 2 shows the formation of *n*-hexanal during storage. In the control milk, the rate of *n*-hexanal formation was faster at 35 °C than at 22 °C. However, concentration reached a maximum after 90 days at 35 °C while concentration continued to increase at 22 °C storage. This might be attributable to oxygen availability. The dissolved oxygen at 35 °C storage was essentially depleted at 90 days but this was not the case at 22 °C storage. The small increases in hexanal during storage of AA milk might be due to limited oxygen availability. The added ascorbic acid did effectively scavenge oxygen in the AA milk. One of the functions of ascorbic acid as an antioxidant in food systems has been to scavenge oxygen and thereby prevent oxidation of oxygen-sensitive food constituents (Bauernfeid and Pinkert, 1970). However, it is also possible that ascorbic acid might be directly involved in the inhibition of

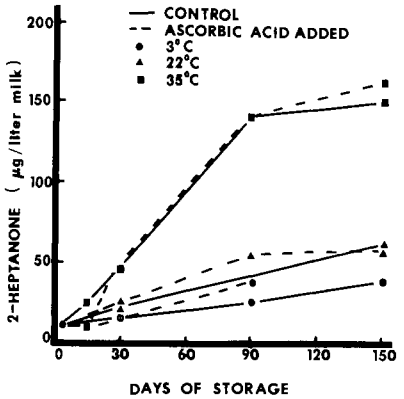

Figure 3. Changes in concentration of 2-heptanone in control and ascorbic acid-added milk (200 mg/L) during storage.

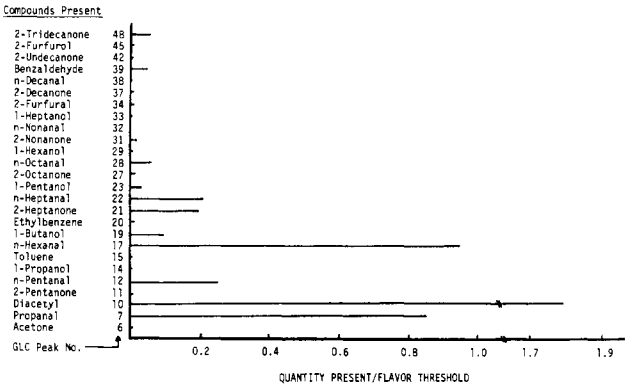

Figure 4. An aromagram obtained from UHT control milk stored at 35 °C for 3 months.

n-hexanal formation by lowering oxidation–reduction potential (Bell et al., 1962) or by inhibiting free radical chain reaction (Haase and Dunkley, 1969). Another possible source of antioxidants might be small amounts of SH groups remaining in the AA milk (Jeon, 1976).

The pattern of changes in concentration of *n*-pentanal, *n*-heptanal, and *n*-octanal during storage were similar to *n*-hexanal. The concentrations of *n*-nonanal in the control milk increased steadily during storage independent of storage temperature. The concentration of *n*-nonanal in the AA milk did not change significantly during storage except for a small increase in the 22 °C milk. *n*-Decanal might be present initially at low concentration (0.3 ppb) and did not increase in concentration during storage. The changes in concentration for propanal were not estimated because of the isolation techniques used.

The formation of odd carbon-numbered methyl ketones appeared to depend primarily upon storage temperature and was independent of oxygen content and ascorbic acid concentration. This is illustrated in Figure 3 with 2-heptanone. The concentrations of 2-heptanone at each storage temperature were similar in the control and AA milk. Similar patterns were found for 2-pentanone, 2-nonanone, 2-undecanone, and acetone, although changes in acetone concentration were difficult to measure due to the isolation techniques used. 2-Octanone and 2-tridecanone did not increase in concentration significantly during storage. 2-Decanone was initially present at a level of about 0.3 ppb and exhibited no change. The greatest increase in concentration occurred in 2-heptanone, followed by 2-pentanone, perhaps acetone, 2-nonanone, and 2-undecanone. The odd carbon-numbered methyl ketones are known to be generated from β-keto alkanoic acid esters of glycerides, which constitute about 0.04% of butterfat (Parks et al., 1964). The mechanism of formation involves hydrolysis and decarboxylation upon heating (Day, 1966). In addition, Schwartz et al. (1966) reported that methyl ketone formation followed a first-order reaction in butteroil with an activation energy of 26.5 kcal/mol. Kinsella (1969) therefore postulated that the formation of methyl ketones could occur spontaneously in stored dairy products because of the low activation energy.

Among the alcohols identified, only 1-butanol showed a significant increase in concentration during storage. Other aliphatic alcohols did not show measurable changes in concentration. The formation of 1-butanol during storage was not affected by the different levels of dissolved oxygen or ascorbic acid. It appeared to be dependent only upon the storage temperature. The source and mechanism involved in the formation of 1-butanol is not readily explicable. Stark and Forss (1966) suggested that alkanols (C_{1-18}) in oxidized butter were formed by the oxidation of unsaturated fatty acids from the primary alkoxy radicals resulting from the decomposition of lipid hydroperoxides. However, they observed that *n*-pentanol was the major *n*-alkanol formed by the oxidation process and that the amount of 1-butanol formed was very small. It is questionable whether 1-butanol is formed as an oxidation product since its concentration was not affected by oxygen content or the presence or absence of ascorbic acid. Reduction of methyl ketones results in the formation of 2-alkanols but not 1-alkanols (Day, 1966). It might be postulated that 1-butanol is a nonenzymatic browning product. However, the formation of 1-butanol steadily increased in the control milk during storage at 22 °C and little browning occurred.

Flavor Significance of Volatile Compounds. Methyl ketones were quanitatively the most abundant class of compounds in flavor isolates. For example, the flavor isolate from the control milk stored at 35 °C for 3 months showed that methyl ketones represented 79% of total peak area and 2-heptanone alone represented 24%. However, the aromagram shown in Figure 4 suggests that the flavor contributed by methyl ketones is not as significant as indicated by the quantity present. The aromagram was obtained from the UHT control milk stored at 35 °C for 3 months. Each bar was calculated by dividing the actual amount of a compound present in UHT milk by its average threshold in milk. Most flavor threshold values used were taken from data in the literature (Patton and Josephson, 1957; Day et al., 1963; Langler and Day, 1964; Scanlan et al., 1968; Kinsella, 1969). Threshold values for 1-propanol, 1-pentanol, 1-heptanol, toluene, ethylbenzene, 2-furfural, benzaldehyde, and 2-decanone in milk were estimated in our laboratory as 2.0, 0.4, 0.7, 0.5, 0.2, 1.5, 0.3, and 0.3 ppm, respectively. Figure 4 shows that diacetyl is the only compound present above its threshold value. Consequently, diacetyl is expected to contribute to the flavor of UHT milk. Scanlan et al. (1968) have suggested that diacetyl contributes to the rich or heated note in the flavor of heated milk. Diacetyl concentration was found to be about 16 ppb in our UHT milk while Scanlan et al. (1968) reported 38 ppb. This difference might be due to the differences in heat treatments. The milk samples prepared by Scanlan et al. (1968) were more severely heated. Their samples were preheated at 82 °C for 30 min and then sterilized at 146 °C for 4 s. Scanlan et al. (1968) suggested that diacetyl in heated milk was a fission product of the Maillard reaction (Hodge, 1953).

Although no off-flavor compounds were present above their threshold values, aldehydes appeared to be most important in contributing to the off-flavor of the milk. Following 3 months of storage, *n*-hexanal and propanal

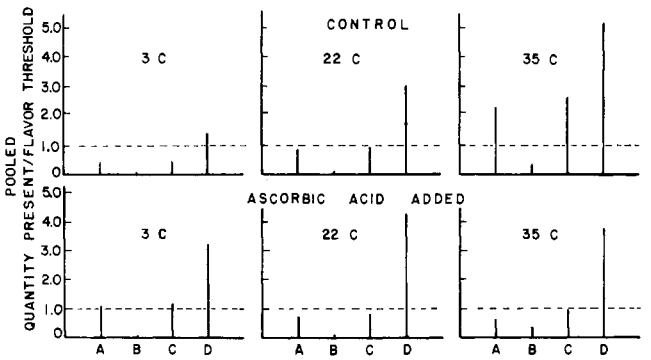

Figure 5. Pooled flavor values of quantity present/flavor threshold for carbonyl compounds at 3 months of storage: (A) *n*-alkanals, (B) methyl ketones, (C) *n*-alkanals plus methyl ketones, and (D) overall flavors.

were present at concentrations near their thresholds. *n*-Pentanal and *n*-heptanal were the next two most significant flavor compounds. As indicated earlier, flavor contribution by methyl ketones appeared to be less than anticipated based on their concentrations in stored milk. Although 2-heptanone was present in the largest quantity in stored milk, its concentration is far below threshold level. However, Langler and Day (1964) reported that a mixture of methyl ketones exhibited a synergistic interaction whereby a perceptible flavor was evident when the concentrations of all components in the mixture were far below their average thresholds. Similarly, a mixture of aliphatic aldehydes has been reported to compositely give rise to a characteristic oxidized flavor at their subthreshold concentrations (Lillard and Day, 1961; Day et al., 1963; Parks et al., 1963).

Although aldehydes or ketones may not interact to give a strictly additive function at their subthreshold concentrations, the values for quantity present/flavor threshold (flavor value) were totaled and are shown in Figure 5 to demonstrate the possible significance of carbonyl compounds to the flavor of stored UHT milk. In the control milk, the pooled values for aldehydes and ketones are distinctly different among storage temperatures. At 35 °C storage, the flavor value is far above the threshold and at 22 °C approximately threshold. However, the flavor value is far below the threshold at 3 °C storage. In the AA milk, the flavor values are all marginal at the thresholds. The value is slightly above the threshold at 3 °C, slightly below at 22 °C and very close at 35 °C.

Figure 6 shows mean scores of stale flavor intensities given by a four-membered taste panel. Stale flavor intensity increased throughout storage in all samples and tended to develop at a faster rate in control than in the AA samples. The time of appearance during storage in control samples was temperature dependent, whereas the effect of storage temperature on the development of stale flavor was inconsistent in the AA milk. Detection of stale flavor as early as 17 days of storage in the AA milk at 22 °C is not readily explicable. It is interesting that flavor values for carbonyl compounds in Figure 5 follow the same trend as stale flavor intensity ratings at 3-month storage shown in Figure 6.

Synthetic mixtures, made up of all the identified compounds at three different concentrations equivalent to those concentrations in 3-month-old UHT milk stored at 3 (low), 22 (medium), and 35 °C (high), were added to fresh pasteurized, homogenized milk and evaluated by a panel of seven judges for stale and other flavor defects. The synthetic compounds added were all, except diacetyl, below their threshold concentrations. Coded samples of

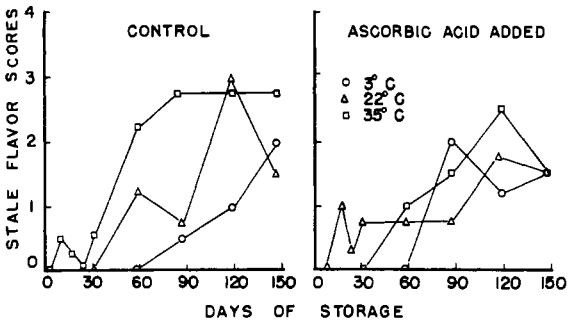

Figure 6. Mean scores of stale flavor intensities in control and ascorbic acid-added UHT milk during storage.

pasteurized homogenized milk and the UHT milk stored about 10 months at 22 °C were also provided for flavor evaluation. All judges except one responded to the stale flavor criticism from the milk containing synthetic mixtures. One judge indicated chemical odor at the high concentration of synthetic compounds and another indicated feedy odor. Two judges indicated oxidized flavor as well as stale flavor. Mean scores of stale flavor intensities (0 = none, 4 = pronounced) received were as follows:

fresh milk	fresh milk + synthetic mixtures			old UHT milk
	low	medium	high	
0.7	1.07	1.43	2.14	3.57

Analysis of variance indicated that the stale flavor scores obtained from fresh milk, fresh milks containing three levels of synthetic mixtures, and old UHT milk were significantly different at $p < 0.05$. Furthermore, Bonferroni's significant difference test showed that all the mean scores were significantly different from one another. These results indicated that the volatile flavor compounds identified were at least partly involved in the flavor deterioration of UHT milk.

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Isolation and Identification of Volatiles from Catawba Wine

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The volatile composition of three Catawba wines prepared from grapes grown in the vineyards of the New York State Agricultural Experiment Station during the 1976 vintage were analyzed by instrumental and sensory means. The three wines differed according to the enological technique employed for their production. Volatiles were isolated by solvent extraction, separated and quantified by gas chromatography, and identified by combined gas chromatography–mass spectrometry. Although some variation in volatile composition due to processing technique was observed, sensory analyses comparing the wines with corresponding model solutions indicate that the major identifiable components are of little importance in determining the aroma of Catawba wine as influenced by processing technique.

Catawba vines have been cultivated in the northeastern United States for over 150 years. Currently, in New York, over 10 000 tons are produced annually and over 90% of that is used for wine production (New York State Crop Reporting Service, 1976). Catawba grapes can be used in the production of either white or rosé wines depending upon enological technique, and much of the white wine produced is used in sparkling wine cuvées.

The literature dealing with the volatile composition of wines and wine grapes is extensive. Kahn (1969) and Webb and Muller (1972) have tabulated hundreds of compounds that have been identified in wines and other alcoholic beverages.

Many native American grape varieties including Concord and Catawba have characteristic aroma components that appear to be unique to some varieties with *labrusca* parentage. Although many workers (Holley et al., 1955; Neudoerffer et al., 1965; Stevens et al., 1965; and Stern et al., 1967) have studied the volatile composition of the Concord variety, no such investigations have been conducted with Catawba.

Methyl anthranilate, a compound long thought to be of major importance in the aroma of *labrusca* varieties (Sale and Wilson, 1926), now appears to be far less important than previously thought (Nelson et al., 1977a). Friedman (1976) believes that methyl anthranilate is of little importance in the aroma of Concord grapes, even though its concentration in that variety is relatively high. Amerine et al. (1959) noted that the distinctive Catawba aroma was apparently not due to methyl anthranilate and that other more important compounds must be present.

This report examines the volatile composition of Catawba wines prepared by three different enological techniques. In addition, it attempts to assess the effect of processing technique on the occurrence of these volatiles in Catawba wine.

MATERIALS AND METHODS

Wine Preparation. Catawba grapes were harvested at 16.6° Brix in October of 1976. The fruit was divided into three 20-kg lots for fermentation. From one lot a white Catawba wine was prepared by immediately pressing the crushed grapes while rosé wines were prepared from the other two. One of the rosé wines was prepared by fermenting the juice in contact with the skins for 5 days [rosé (FS)] while the other was thermally vinified [rosé (TV)]. Thermal vinification consists of heating the crushed grapes in a steam kettle to 60 °C for 15 min, followed by immediate pressing. Fermentations were conducted at 20 °C and other standard enological procedures as described by Nelson et al. (1977b) were followed in each case.

Volatile Isolation. The Catawba wine volatiles were isolated using organic solvent extraction with Freon 113 (1,1,2-trichloro-1,2,2-trifluoroethane, "Precision Cleaning Agent, Du Pont"). Equal volumes of wine and Freon (2700 mL) were stirred for 1 h. The Freon phase was then drawn off, dried over anhydrous magnesium sulfate, and con-

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