Influence of Cultivar, Process, Maturity, and Planting Date on the Dimethyl Sulfide and Hydrogen Sulfide Levels in Sweet Corn

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Dimethyl sulfide (Me$_2$S), as measured by gas chromatography in cooked corn headspace, ranged between 0.43 and 17.0 ppm and varied significantly due to date of planting (or season) and cultivar. These factors also caused significant differences in levels of hydrogen sulfide (H$_2$S), which was measured colorimetrically and which ranged between 34 and 208 ppb. Me$_2$S decreased with increasing maturity while H$_2$S showed no significant change. Processing demonstrated the most notable changes, with mean Me$_2$S levels highest in canned corn (4.18 ppm), followed by fresh corn (1.08 ppm) and frozen corn (1.04 ppm). This trend was somewhat reversed with H$_2$S, canned corn containing 94.9 ppb, fresh corn 66.9 ppb, and frozen corn 138.1 ppb. Variations in canning and freezing processes also resulted in differences in Me$_2$S and H$_2$S. Panelists often did not respond to differences in Me$_2$S and H$_2$S levels with differences in aroma scores.

Dimethyl sulfide (Me$_2$S) and hydrogen sulfide (H$_2$S) were identified as major contributors to cooked sweet corn aroma (Flora, 1973; Flora and Wiley, 1974). Self et al. (1963) reported relatively high levels of these two volatiles in cooked sweet corn. Bills and Keenan (1968) measured Me$_2$S levels of 5.7-14.2 ppm in canned corn and levels of 0.3-6.8 ppm in frozen sweet corn. Williams et al. (1972) found concentrations of Me$_2$S in commercially processed yellow corn ranging from 10.1 to 16.0 ppm. Williams and Nelson (1973) found significant differences in the Me$_2$S potentials of 21 sweet corn hybrids. They also found that blanching reduced the Me$_2$S potential.

The work reported after Self et al. (1963) on sweet corn flavor volatiles has dealt almost exclusively with Me$_2$S (Bills and Keenan, 1968; Williams et al., 1972; Williams and Nelson, 1973). Research has shown that, although Me$_2$S lends the dominant character to sweet corn aroma, other compounds, notably H$_2$S, also contribute significantly to this response (Flora, 1973).

The purpose of this study was to determine the influence of cultivar, maturity of the corn at harvest, planting date, and commercial processing techniques on the levels of Me$_2$S and H$_2$S. The levels measured were thought to approximate concentrations of Me$_2$S and H$_2$S experienced by the consumer. Sensory panels were employed to determine if consumers responded to aroma differences when significant differences in the concentrations of Me$_2$S and H$_2$S were indicated.

EXPERIMENTAL SECTION

Planting, Harvesting, and Processing. The sweet corn was grown at the University of Maryland Plant Research Farm, Beltsville, Md. Five commercially grown cultivars were included in the study: Northrup King 199 (NK 199), Buttersweet, Code 556, Stylepak, and Silver Queen. The first four are processing cultivars while Silver Queen is a fresh market corn. Each cultivar was planted in blocks on each of two planting dates, May 27 and June 16, 1972.

Corn was harvested to cover a fancy to standard range of maturity. Objective evaluation of maturity was made by succulometer, alcohol insoluble solids (AIS), kernel size, and pericarp determinations. Two to six harvests were made from each cultivar in each planting.

The harvested corn was processed immediately after picking. The processing was essentially by standard commercial procedures. Blanched corn was frozen in plastic freezer bags and in evacuated heat sealed plastic bags to simulate "flavor tight" pouches. Two variations of canned corn were also used to simulate those being marketed. Whole kernel corn packed in brine and whole kernel corn packed under a 635 mm Hg vacuum in 1 oz of brine were processed in No. 303 total C-enamel cans. Some of the cans were packed with unblanched corn in order to study the effect of blanching on the volatiles.

It was not possible to analyze the fresh corn completely upon harvest. Thus 200 g of the freshly cut, unwashed, and unblanched corn was weighed into pint-size plastic freezer bags with 100 ml of 0.1 N HCl and frozen, so that corn of a semifresh, uncooked state would be available for later analysis of H$_2$S.

Gas Chromatographic Determination of Me$_2$S. The concentration of Me$_2$S in the samples was determined by gas chromatographic headspace analysis. This method was chosen largely because of its simplicity. However, the most important consideration was that the level of Me$_2$S in the headspace above cooked corn would be that level experienced by a consumer. The precision of the method also proved quite adequate, having a coefficient of variability of 6.0%.

The headspace analysis procedure used in this study was similar to those used by Buttery and Teranishi (1961) and Rasekh and Kramer (1971). Four hundred grams of corn drained on an 8 mesh screen was weighed into a 500-ml erlenmeyer flask. The total weight was brought up to 550 g with drain juice and distilled water. The flask was covered with a 132-cm^2 piece of Durafilm, onto which was bonded a 6.25-cm^2 rubber patch, and sealed tightly. The flask was then immersed in boiling water for 10 min to simulate a normal cooking procedure and then transferred to a 50° water bath. After equilibrating for 15 min, a Hamilton 1-ml gas-tight syringe was flushed in the confined headspace above the cooked corn and a 1-ml vapor sample was removed for gc analysis. Another 1-ml vapor aliquot was analyzed at 30 min. The procedure was gauged to meet the two conditions stated by Buttery and Teranishi (1961) as necessary to keep constant for reproducible analyses: the temperature of the food mixture at the time of sampling and the ratio of water to sample. In addition to the regular samples, special testing was carried out to determine the effect of atmospheric cooking on canned corn flavor volatiles and also to determine how the flavor volatiles were distributed between the liquid and solid phases in canned corn.

An Aerograph 204 gas chromatograph, in conjunction with a Hewlett-Packard Model 3370-B digital print-out integrator, was used for the headspace volatiles analysis. After screening several packed columns, including Carbowax 20M, Apiezon, various polyesters, and others, for

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Table I. Adjusted Means Showing Differences in Me₂S and H₂S Levels and Aroma Scores (Adjustment for AIS Covariate)

	Me_2S, ppm	H_2S, ppb	Aroma score[c]
Cultivar[a]			
NK 199	2.28b	102.43b	5.04ab
Code 556	1.89b	111.89b	4.99b
Buttersweet	3.27a	108.30b	5.46ab
Stylepak	2.25ab	131.71a	5.58a
Silver Queen	1.56b	90.30c	5.03ab
Planting			
1. May 27	1.92a	105.08a	5.21a
2. June 16	2.69b	112.78b	5.23a
Process[b]			
Fresh	1.08a	66.89a	5.85a
Brine can	4.26b	102.99b	4.76c
Vacuum can	4.09b	86.76c	4.51c
Frozen	1.02c	137.86d	5.43b
Vacuum frozen	1.05c	148.29e	5.03b

[a] Differences determined by Duncan multiple range test. [b] Differences determined by linear comparison. [c] Scale of 0–10, 10 being most acceptable.

Table II. Effects of Process and Maturity on Me₂S Levels (ppm) in Samples from NK 199 Cultivar

Maturity score and grade[a]	Canned	Frozen	Fresh
38.0, Fancy	10.6	1.6	1.7
30.9, Standard	1.8	0.67	0.43

[a] Twigg et al. (1956).

resolution of the volatiles, a 2.4 m × 3.12 mm stainless steel column packed with 10% SE-30 on 80–100 acid-washed Chromosorb W was used in the separations. The initial column temperature was 50°; programmed at 4°/min after 1 min up to 80°. Other conditions were: injector, 180°; detector (FID), 180°; and carrier gas, N_2 at 15 ml/min.

Colorimetric Determination of H₂S. H_2S was determined by a colorimetric procedure reported by Brenner et al. (1953) and later used by Miers (1966) for measuring H_2S in processed tomatoes.

After headspace analysis of canned and frozen corn, or neutralization with NaOH and subsequent cooking in the case of fresh corn frozen in HCl, each flask was placed overnight at 1° to minimize volatilization of the H_2S and was analyzed the next day.

Standard Curves. Standard curves were determined for Me_2S and H_2S to cover the ranges of concentrations present in the corn. The procedure followed for the Me_2S standard was a modification of the procedure described by Williams et al. (1972). One milliliter of previously cooled Me_2S was dissolved in 99 ml of cold ethylene glycol. One milliliter of this solution was diluted to 100 ml with distilled water. Appropriate aliquots of these solutions were used to make Me_2S concentrations of 0.1, 15, 1.0, 5.0, 10.0, and 20.0 ppm. The aliquots were brought up to 550 ml with distilled water in 500-ml erlenmeyer flasks and covered, cooked for 10 min in boiling water, and equilibrated in a 50° water bath, and 1-ml headspace samples were taken at 15 and 30 min for gc analysis. Detector response was linear between 1 and 10 ppm, and nonlinear below 1 and above 10 ppm.

The standard curve for H_2S was prepared using sodium sulfide according to the procedure of Brenner et al. (1953).

Sensory Panels. Most of the samples analyzed for Me_2S and H_2S were also submitted to sensory evaluation. Panelists were asked to score the aroma of each sample on a 0–10 preference scale, 10 being most acceptable. The sensory panels were conducted as reported by Flora and
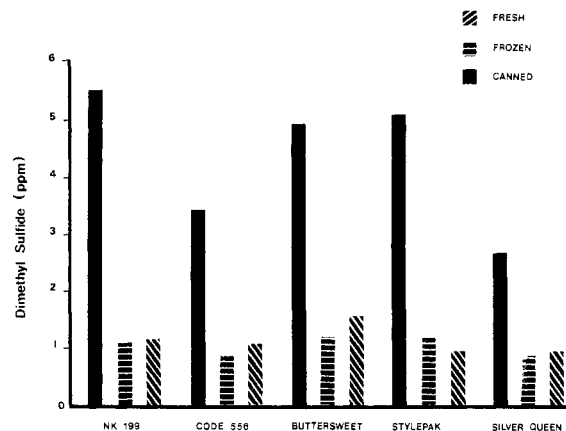
Figure 1.

Wiley (1974) and set up so that specific factors could be compared.

Statistical Analysis. Results were analyzed graphically and statistically. Statistical analyses were performed on a Univac 1108 computer at the University of Maryland Computer Science Center. Multiple analyses of variance were performed for Me_2S and H_2S to determine differences due to cultivar, maturity, process, and planting. Values were analyzed as a complete factorial design with plantings, cultivars, and processes as main effects. Alcohol insoluble solids (AIS) were entered in the design and analyzed as a covariate. Multiple analyses of variance were also performed using mean sensory panel scores to determine if panelists responded to aroma differences among cultivars, processes, and maturities, and between plantings.

RESULTS

Effects of Cultivar, Planting, Process, and Maturity on Me₂S. Me_2S concentrations ranged from 0.43 to 17.0 ppm in the 200 samples analyzed. Me_2S levels in the sweet corn were significantly different among maturities, among cultivars, between plantings, and among fresh, frozen, and canned corn. Apparently there was no significant effect of processing vacuum packed canned corn as opposed to canned corn packed in brine on Me_2S concentration. There was also no difference in Me_2S between frozen and vacuum pouch frozen corn. These results are summarized in Table I.

The most marked differences in Me_2S concentrations were among processes. Canned corn contained significantly higher levels of Me_2S than fresh or frozen corn. The fresh corn produced slightly more Me_2S than the frozen corn. These process differences are illustrated in Table II. For some reason, the corn harvested from the second planting contained more Me_2S than the corn harvested from the first planting. A Duncan multiple range test using adjusted cultivar means for Me_2S indicated that the Buttersweet cultivar contained significantly higher levels of Me_2S than NK 199, Code 556, and Silver Queen. Silver Queen had the lowest average Me_2S content with 1.56 ppm. These results are summarized in Table I. Figure 1 dramatically illustrates the higher levels of Me_2S in canned corn and the significantly lower level of Me_2S in the fresh market Silver Queen cultivar than in the processing cultivars. The effect of maturity on Me_2S in fresh cooked corn is illustrated in Figure 2. It is quite evident that the general trend sees a decline in the Me_2S concentration as maturity increases. A more dramatic difference may be seen in Table II. Frozen and canned corn had similar patterns of Me_2S decline with increasing maturity. Unblanched canned samples had 4.72 ppm of Me_2S as compared with 3.28 ppm in blanched samples, a significant difference.
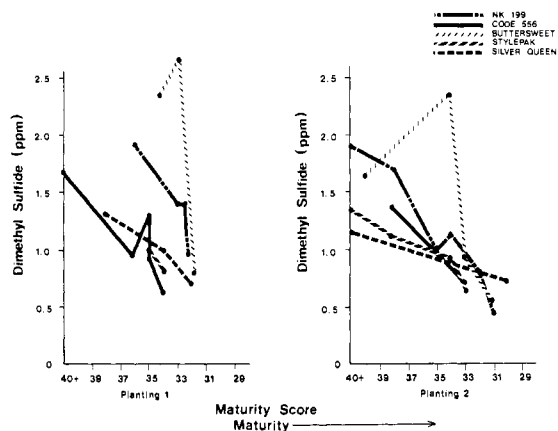
Figure 2.

There appeared to be a loss of Me₂S when the corn was cooked prior to analysis. A canned sample prepared for analysis under closed conditions contained about 2 ppm of Me₂S, while its replicate, which was atmospherically cooked prior to sealing of the flask, contained 1 ppm of Me₂S. Corn liquor apparently contains as much Me₂S as the equivalent amount of kernels plus liquor in the proportions found in canned corn.

Effects of Cultivar, Planting, Process, and Maturity on H₂S. The corn from the second planting produced significantly higher levels of H₂S than that from the first planting. As was the case for Me₂S, the most marked differences in H₂S levels were among processes. All processes contained significantly different levels of H₂S. The highest level of H₂S was found in the vacuum pouch frozen corn, followed by frozen, brine packed canned, vacuum pack canned, and fresh cooked corn. A Duncan multiple range test for cultivars indicated that Stylepak had a significantly higher concentration of H₂S than the others while the Silver Queen was significantly lower in H₂S than the others. Table I summarizes these findings. Hydrogen sulfide concentrations ranged from 34 to 208 ppb. Statistical analysis indicated that there was no significant adjustment for maturity.

There was no significant difference in H₂S levels between unblanched canned corn and blanched canned corn as determined by paired comparison and rank sum total techniques. Although tests showed nonsignificance between the 21 pairs tested, it appears that the H₂S levels may tend to be higher in the blanched samples. Fourteen of 21 blanched samples showed higher levels of H₂S than their unblanched counterparts. The average concentration of H₂S in all blanched samples was 98.4 ppb whereas in the unblanched samples it was 89.8 ppb.

Detectable Sensory Differences. Panelists responded to differences in aroma among maturities, among cultivars, and among fresh, frozen, and canned corn. They did not respond to differences in aroma between plantings, between brine pack canned corn and vacuum packed canned corn, or between frozen pack corn and vacuum pack frozen corn. These results are summarized in Table I.

They preferred the aroma of fresh corn over processed corn and frozen corn over canned corn (Table I). Cultivar differences indicate that the panelists could only really detect a difference in aroma between NK 199 and Code 556.

The effect of maturity on aroma scores of fresh, canned, and frozen sweet corn was not clear in the first planting. However, the second planting results indicated that the panel scores declined with increasing maturity. There were indications that these factors might reach an optimum in some cultivars before they begin to decline.

The panelists did not score the preblanched canned samples significantly differently from the unblanched canned samples from the aroma standpoint.

DISCUSSION

The results indicated that canned corn produced relatively high levels of Me₂S, and frozen corn contained relatively high levels of H₂S. These results were also borne out by smell. When the flasks of cold canned corn that had been cooked, analyzed by gc, and stored overnight in a cold room were opened for H₂S analysis, a strong Me₂S odor was quite noticeable. However, the Me₂S odor of cold frozen corn was drowned out by a much stronger odor like that of H₂S.

Attempts to correlate Me₂S and H₂S values with panel scores were generally unsuccessful. Although panel scores declined with maturity as did Me₂S, a trend could not be established by plotting panel scores against Me₂S levels of corn of different cultivars, processes, and maturities. This phenomenon would indicate that although Me₂S and H₂S have proven to be important aroma components in sweet corn, they are not sufficient in themselves to account for the differences in aroma responses. Consequently, more interest should be shown in the total complement of flavor volatiles of sweet corn. Volatile ratios and profiles might better explain the aroma response differences.

The panelists preferred the fresh and frozen corn aromas over that of the canned corn. The Me₂S levels in the fresh and frozen corn were significantly lower than in the canned corn. The strenuous heat treatment applied to the canned samples would account for the higher level of Me₂S due to exhaustion of its precursor as suggested by Bills and Keenan (1968). This corresponds to results reported by Heatherbell et al. (1971) in which they found canning produced an increase in the Me₂S level in carrots. Miers (1966) and Nelson and Hoff (1969) reported increases in H₂S and Me₂S in tomato juice due to canning. On the other hand, fresh and frozen corn, not being subjected to the strenuous heat process, can draw on their precursor reserves during cooking for flavor volatiles. It appears that frozen corn may lose some Me₂S during blanching or storage since it contains slightly less than its fresh analog. It is obvious that blanching does indeed cause a loss of Me₂S when one remembers the significantly higher levels of Me₂S produced in unblanched canned corn than in corn blanched prior to canning. However, panelists did not respond to these latter differences.

Panel scores showed the same pattern of decreasing with maturity as did Me₂S. Thus the Me₂S level in sweet corn may possibly be used as an indicator of maturity or quality, should there be a satisfactory correlation between Me₂S and maturity scores of AIS.

Differences in Me₂S levels were recorded for cultivars. It is of interest to note that the fresh market cultivar used in this study, Silver Queen, produced less Me₂S in canning than any of the processing cultivars. This difference is illustrated in Figure 1. Referring to Table I, one can see that Silver Queen had overall Me₂S and H₂S levels below those of the processing cultivars. These results are contradictory to results reported by Williams and Nelson (1973) in which they reported that fresh market cultivars had higher Me₂S potentials than processing cultivars.

The differences due to plantings are equally difficult to explain. Although panelists were not able to detect aroma differences between the two plantings, there were differences in Me₂S and H₂S levels. Whether the planting date itself or season caused this effect is open to question.

Thus, a number of factors influence the concentrations of Me₂S and H₂S in sweet corn. However, changes in Me₂S and H₂S levels are not often met by differences in panel aroma responses, suggesting that there is more involved in sweet corn flavor than just Me₂S and H₂S. Results have indicated that lower levels of Me₂S such as in

fresh and frozen corn are preferred. But is fresh corn aroma preferred over frozen because of the high concentration of H₂S in frozen corn? Obviously, more research needs to be done in this area to answer this and other questions.

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Nutritional Quality of Processed Milk Containing Carrageenan

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Nutritional studies were conducted on rats fed diets containing carrageenan that had been mixed into skim milk at a concentration equal to that of the protein and heat sterilized under conditions routinely used in the manufacture of canned liquid milk. When fed in a simulated milk powder diet, the processed carrageenan, at a dietary level of 4%, had no influence (compared to cerelose or cellulose) on growth rate, diet energy efficiency, absorption of protein, fat, or calcium, blood coagulability, utilization of protein for growth (PER), or the utilization of iron (anemic rats). Gross and microscopic examination of the cecum and colon, after 6 months feeding, revealed none of the abnormalities associated with the feeding of degraded (hydrolyzed) carrageenan to susceptible species. These results support the conclusion that food grade carrageenan, at its present or anticipated use level in food, does not constitute a hazard.

Carrageenan is the generic name for a group of sulfated polygalactans of high molecular weight present in a number of red seaweeds. The principle commercial source of carrageenan is the alga *Chondrus crispus,* also known as "Irish moss" or "carragheen moss," in reference to the Irish coastal town of Carragheen, where for centuries it was collected for use in the preparation of the milk pudding, blanc mange.

The reactivity of carrageenan with milk protein to form a stable gel has found wide application in the dairy and related industries. Typical applications include the prevention of separation and sedimentation in aseptically canned and sterilized milk products, the suspension of cocoa particles in chocolate drinks, the prevention of whey separation and ice crystal formation in ice cream, and the stabilization of whipped products.

The carrageenan products of predominant use in the food industry are mixtures of κ- and λ-carrageenan, with the proportions selected determined by the characteristics desired, κ-carrageenan for gel formation and λ for increased viscosity.

Chemically κ-carrageenan consists of alternating units of sulfated *d*-galactose and 3,6-anhydro-*d*-galactose in approximate equimolar amounts (O'Neil, 1955); λ-carrageenan consists almost entirely of sulfated *d*-galactose (Smith *et al.,* 1955). The molecular weight of κ-carrageenan is between 1.8 and 3.2 × 10⁵ and that of λ-carrageenan is between 4 and 7 × 10⁵ (Smith *et al.,* 1954).

The safety of carrageenan for use in food products has been recognized in the United States by its inclusion in the first GRAS list (FDA, 1959), and subsequently as a regulated additive (FDA, 1961). The joint FAO–WHO Expert Committee on Food Additives (World Health Organization, 1970) established an acceptable daily intake for man of 50 mg/kg, *i.e.* approximately 3.5 g per day for the average man.

Several years ago the safety of carrageenan as a food additive was questioned as a result of a series of studies in England (Marcus and Watt, 1969; Watt and Marcus, 1969, 1970; Watt *et al.,* 1970) in which ulcerations of the cecum and proximal colon were found in rabbits, guinea pigs, and rats drinking water containing a degraded carrageenan product. The product (Ebimar), which had been used in Europe for over 10 years in treatment of peptic ulcer, is produced by the degradation of ι-carrageenan, usually by acetolysis, to a molecular weight of less than 30,000 (Anderson and Soman, 1966). ι-Carrageenan, isolated from *Eucheuma spinosium,* differs from κ-carrageenan in that sulfate is present at the C₂ position of the 3,6-anhydrogalactose.

The implication of the Marcus and Watt studies, that degraded carrageenan was a hazardous therapeutic agent, was contested (Bonifils, 1970; Maillet *et al.,* 1970; Sharratt *et al.,* 1971), and further investigations on the biological properties of native and degraded carrageenan have been published (Beattie *et al.,* 1970; Dewar and Maddy,

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