

## A Comparison of the Chemical Composition of Boiled and Roasted Aromas of Heated Beef

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Using a modified Likens and Nickerson extraction procedure followed by low temperature/high vacuum distillation, representative samples of aroma volatiles were obtained from beef while cooking by microwave radiation and by conventional means. Separation of the components of the isolates was achieved by gas chromatography and the majority of the components were identified by combined gas chromatography/mass spectrometry. A comparative study was undertaken of the effect of cooking variations on the volatile components liberated. Such variations included heating in the presence and in the absence of water and for different periods of time for both conventional and microwave heating methods. Results indicated that certain carbonyl compounds, sulfides, pyrroles, and pyridines are probably important contributors to roasted notes as opposed to boiled or brothy qualities of heated beef aroma.

Desirable beef flavor is developed during heating and the nature of the flavor depends on the way in which the meat is cooked. Differences in flavor due to cooking are probably a direct function of temperature and degree of moisture in the meat (Dwivedi, 1975). Hornstein and Crowe (1960b) showed that the precursors of cooked meat flavor are water soluble and when lyophilized, the dry powder developed on heating an aroma reminiscent of roast meat; in contrast, boiling an aqueous solution of the powder developed a brothy, boiled meat aroma. Characteristically, boiled beef flavor is produced in the presence of water whereby the temperature during boiling does not exceed 100 °C; roasted aromas are liberated under relatively dryer heating conditions where the temperature rises above 100 °C (Wasserman, 1972). An acceptably cooked roast beef topside, for example, shows a temperature gradient from about 60 °C (rare) or 80 °C (well-done) in the interior to about 160 °C or more at the surface. The flavor of roast meat therefore is believed to derive essentially from the surface layers.

Some of the earliest attempts at analyzing *boiled* beef aroma reported the presence of simple carbonyls, thiols, sulfides, and ammonia (Bender and Ballance, 1961; Hornstein and Crowe, 1960a, b; Kramlich and Pearson, 1960; Sanderson et al., 1966; Self et al., 1963; Yueh and Strong, 1960). In 1968, Chang et al. proposed that these did not represent the complete spectrum of volatile components, nor indeed were they the important ones. In their search for the crucial constituents, they isolated 2,4,5-trimethyl-3-oxazoline and 3,5-dimethyl-1,2,4-trithiolane from two fractions possessing boiled beef aroma (Chang et al., 1968). Later work at the same laboratories however showed that when synthesized, these compounds did not possess the odor of boiled beef (Hirai et al., 1973). Persson and von Sydow (1973) reaffirmed that the trithiolane was probably not important, while Brinkman et al. (1972) provided evidence that it may be formed during the isolation procedure. The latter also identified 5,6-dihydro-2,4,6-trimethyl-1,3,5-dithiazine (thialdine) which may have been an artefact from the interreaction of ammonia, hydrogen sulfide, and acetaldehyde. However, due to the great ease of its formation, it was proposed as a genuine component. Further attempts at searching for the character impact compounds of boiled beef aroma resulted in the identification of some furanones (Hirai et al., 1973; Tonsbeek et al., 1968), 1-(methylthio)ethanethiol (Brinkman et al., 1972), and thiophene-2-carboxaldehyde (Hirai et al., 1973). None of the compounds mentioned

has been proved to be chiefly responsible for the aroma, which therefore remains as a concept of a delicate yet complex mixture of many components (Hornstein, 1967).

Studies involving procedures wherein flavor volatiles have been isolated from beef heated to temperatures exceeding 100 °C have involved canned beef (Brennan and Bernhard, 1964; Persson and von Sydow, 1973; von Sydow, 1971), pressure cooking (Mussinan et al., 1973; Wilson et al., 1973), frying (Watanabe and Sato, 1971a,b; 1972a,b; Yamato et al., 1970), and conventional roasting (Flament and Ohloff, 1971; Liebich et al., 1972). The volatiles identified have shown considerable similarities and roasted notes characteristically predominate in the aromas; these have frequently been attributed to the many alkylpyrazines identified, since they are typical Maillard browning reaction products frequently identified in roasted foods (Bondarovich et al., 1967; Flament and Ohloff, 1971; Johnson et al., 1971; Marion et al., 1967; Watanabe and Sato, 1971b, 1972a,b). Thus, roasted aroma qualities are not peculiar to conventional oven roasted beef.

Despite the great progress recently made in the study of cooked beef flavor, the search for specific character impact compounds has not proved particularly successful. It is likely that the majority of the aroma constituents liberated during cooking play a significant role in the overall flavor character, which is probably largely determined by the subtle quantitative balance of numerous volatile components. It is not unreasonable therefore to expect that precise differences between the cooked beef flavors described as boiled (or brothy) and roasted may be similarly explained. A study of these differences was the purpose of the work here reported.

Table I summarizes the literature results to date and compares the classes of volatile compounds identified in boiled vs. roasted aromas of heated beef (with no regard to concentrations).

### EXPERIMENTAL SECTION

Aroma volatiles were extracted from 1 kg of lean minced beef (*Longissimus dorsi* muscle from Friesian steers of similar histories) during boiling—both conventionally and by microwave radiation—using a modified Likens and Nickerson extraction apparatus with twice distilled 2-methylbutane solvent as previously described (MacLeod and Coppock, 1976). The meat sample was brought to the boil (30 min conventional; 5 min microwave) and the volatiles extracted for varying periods of time. Reasons for selecting the aroma isolation procedure adopted for the boiled beef samples have been given (MacLeod and Coppock, 1976). In order to perform direct and valid comparisons of two sets of samples possessing characteristic boiled and roasted aroma qualities, respectively,

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Table I. Summarized Comparison of Volatile Components of Boiled (B) and Roasted (R) Aroma of Heated Beef (1960 to Date)

Chemical class	Boiled <sup>a</sup> aroma		Roasted <sup>b</sup> aroma	
	No. identified	Comment	No. identified	Comment
Alkanes	6 <sup>c</sup>	Mostly high mol wt	22	High and low mol wt R > B
Alkenes	2 <sup>c</sup>	Mostly high mol wt	4	
Benzenoids	9 <sup>c,d</sup>		36	R > B
Alcohols	11	Mostly low mol wt	28	R > B
Aldehydes				
Saturated	10	Mostly low mol wt	22	High and low mol wt R > B
Unsaturated	3 <sup>c</sup>	Mostly high mol wt	14	
Ketones	7	Mostly low mol wt	24	High and low mol wt
Acids	10	B > R	1	
Lactones	2	15 identified in uncooked beef, however (Watanabe and Sato, 1968)	16	
Esters	1 <sup>c</sup>		6	R > B
Ethers	2 <sup>c</sup>		1	
Amines	2	B only		
Thiols	5	Mostly low mol wt B > R	3	
Sulfides	8		17	R > B
Thiophens and derivatives	2 <sup>c</sup>		24	R > B
Furans and derivatives	6 <sup>c</sup>		16	R > B
Pyrroles	1 <sup>d</sup>		2	
Pyridines			2	R only
Pyrazines	3 <sup>d</sup>		35	R > B
Quinoxalines			2	R only
Thiazoles	2 <sup>d</sup>		10	R > B
Trithianes			2	R only
3,5-Dimethyl-1,2,4-trithiolane	Present <sup>c</sup>		Present	
Thialdine	Present		Present	
2,4,5-Trimethyl-3-oxazoline	Present	B only		

<sup>a</sup> Boiled aroma used in this context for procedures where in general the temperature during isolation did not exceed 100 °C—with two exceptions (Brinkman et al., 1972; Hirai et al., 1973) (Bender, 1961; Chang, 1972; Hornstein, 1967; Hornstein and Crowe, 1960a,b; Kramlich and Pearson, 1958; Sanderson et al., 1966; Self et al., 1963; Tonsbeek et al., 1968, 1971; Yueh and Strong, 1960). <sup>b</sup> Roasted aroma used in this context for procedures wherein the temperature during isolation exceeds 100 °C and includes fried, canned, pressure-cooked, and roasted beef (Brennan and Bernhard, 1964; Flament and Ohloff, 1971; Liebich et al., 1972; Mussinan et al., 1973; Persson and von Sydow, 1973; von Sydow, 1971; Watanabe and Sato, 1971a,b; 1972a,b; Wilson et al., 1973; Yamato et al., 1970). <sup>c</sup> Isolation involves some heating at 105–110 °C. <sup>d</sup> Some identified in boiled beef (Liebich et al., 1972) but conditions unspecified.

it was necessary to develop the roasted qualities by means which simulated as closely as possible (and therefore standardized on) the isolation procedure used for the boiled beef samples in all respects other than the one experimental variable being studied. This was achieved by a preliminary standardized open heating period (1 h conventional; 20 min microwave) to volatilize much of the meat endogenous water, after which extraction commenced immediately and heating was continued, in the case of the conventionally heated series by means of an oil bath at 160 °C. For both heating methods, the volatiles were extracted for varying periods of time: conventional extraction times—15 min, 30 min, 1 h, 4 h; microwave extraction times—5 min, 15 min, 30 min, 1 h.

Concentration of the extracts to a constant volume of 400  $\mu$ l was achieved by low temperature/high vacuum distillation and the isolates were analyzed by routine gas chromatography and combined gas chromatography/mass spectrometry (MacLeod and Coppock, 1976). Peak areas were calculated and normalized to a standard injection volume and attenuation for all chromatograms. No area was recorded where the peak was too small to measure accurately. Total peak areas were calculated and for samples extracted up to 1 h, each peak area was expressed as a percentage of the total area, i.e. relative percentage abundance (RPA).

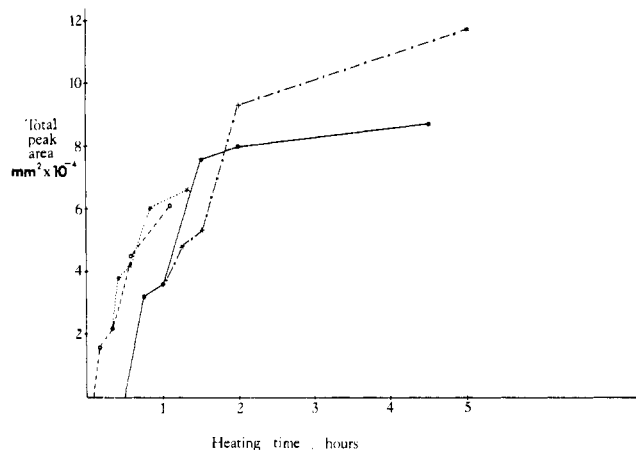
## RESULTS AND DISCUSSION

Sensory evaluations of the aromas of both sets of samples confirmed that they possessed the required boiled

and roasted qualities. All samples produced by the dry heating conditions were described by both trained and untrained panelists as possessing strong roasted aroma qualities, characteristic of roast beef. Using paired comparison directional difference tests and grouping tests, both trained and untrained judges differentiated correctly a significant number of times between the aroma of these samples and that of the boiled beef samples.

Table II (Supplementary Material; see paragraph at end of paper) presents values for absolute area and RPA of each peak in each of the isolates obtained under dry heating conditions, together with peak identifications. Similar values for boiled beef isolates have been reported (MacLeod and Coppock, 1976).

Figure 1 shows the effect of variation in heating time on the total organic volatiles extracted for both sets of samples. For samples possessing the strong roast aroma qualities, corrections have been made for the initial standardized open heating periods (1 h conventional; 20 min microwave) and also for the total volatiles liberated during this period ( $3.6 \times 10^4$  mm<sup>2</sup> conventional;  $2.2 \times 10^4$  mm<sup>2</sup> microwave). It can be seen that after a conventional heating time of 1.5 h there is a sharp increase in total volatiles extracted to values surpassing the figures for the boiled beef heated for the same periods of time. Thus, loss of endogenous water accompanied by higher cooking temperatures increases the rate of chemical reactions occurring during boiling, thus boosting the amount of volatile products; thermal degradations not normally associated with boiling may also be initiated resulting in



**Figure 1.** Effect of heating time on total organic volatiles extracted for boiled and roasted aromas: (—) conventional boiled aroma; (---) conventional roasted aroma; (- · -) microwave boiled aroma; (· · ·) microwave roasted aroma.

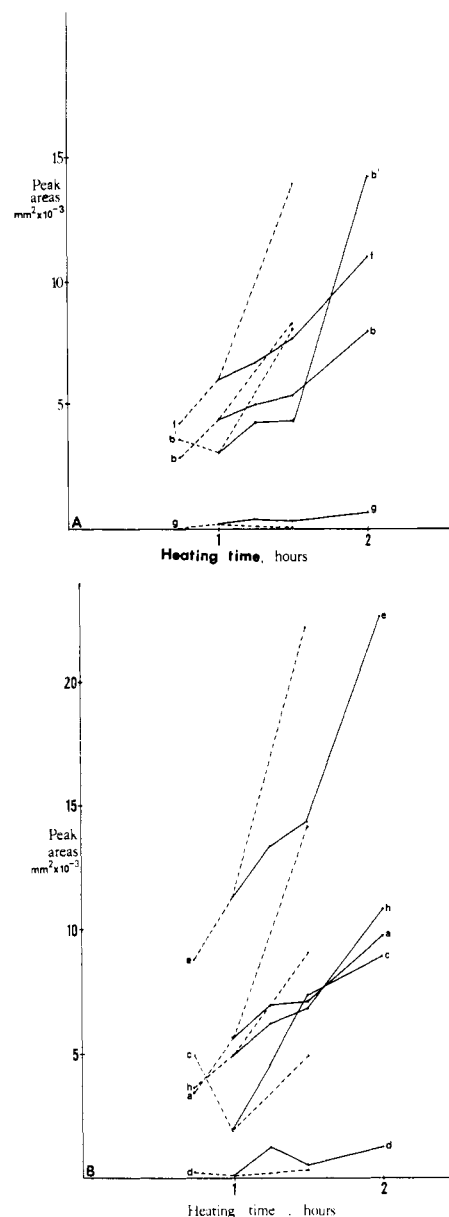
the formation of different volatile components which may contribute to characteristic roasted notes in heated beef.

Rates of production of the various classes of compounds represented in the total organic volatiles of Figure 1 under both wet and dry conventional heating conditions are compared in Figures 2A and 2B. Once more the figures plotted for the latter have been corrected for the quantities of each class of compounds liberated during the pre-extraction heating process. From the slopes of the graphs it can be concluded that conventional dry heating, compared with moist heating, causes decreased rates of production for the alkanes, alkenes ( $<C_{20}$ ), and furans and increased production rates for the aldehydes, ketones, and pyrroles. For the  $C_{20}$  alkenes, the benzenoid compounds, and the pyrazines, rates of production are rather less initially under dry heating conditions but later their production increases at approximately the same rate for both sets of samples.

Similar trends are shown for the microwave isolates but additionally the sulfides are produced more rapidly under dry rather than wet conditions; the pyrazines show decreased production under dry heating conditions initially but later their rate of production considerably surpasses their rate for the boiled samples.

Since rates of production of the various components of the isolates vary, their relative percentage abundance (RPA) values will also vary (Table II of supplementary material; see paragraph at end of paper) and this variation will account for sensory differences in the aromas. For this reason, Figure 3 compares variations in proportions of the classes of components identified in the conventional isolates possessing boiled aroma and roasted aroma qualities, respectively. The increased production rate under conventional dry heating conditions earlier described for the aldehydes, ketones, and pyrroles produces higher RPA values for these classes in the conventional isolates obtained under dry heating conditions than in the boiled beef samples and are therefore likely to be important in the characteristic roast aroma, particularly since several members of these classes showed little if any variation within the boiled beef samples.

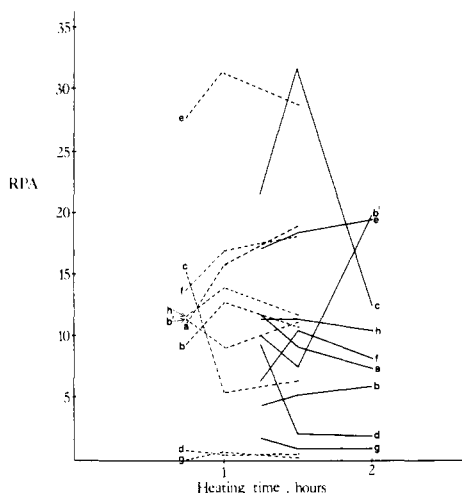
The very high aldehyde RPA for the samples possessing roasted aromas is particularly due to 3-methylbutanal. This agrees with the findings of Liebich et al. (1972), who found that carbonyl compounds, including 3-methylbutanal, were probably responsible for roasted beef flavor. This aldehyde is obtained by the Strecker degradation of



**Figure 2.** Comparison of the effect of heating time on the production of various classes of volatile components in conventional boiled and roasted aroma: (- · -) conventional boiled aroma; (—) conventional roasted aroma; (a) alkanes; (b) alkenes  $< C_{20}$ ; (b')  $C_{20}$  alkenes; (c) aldehydes; (d) ketones; (e) benzenoids; (f) furans; (g) pyrroles; (h) pyrazines.

leucine. Self et al. (1963) and Hodge et al. (1972) report that many Strecker aldehydes contribute to "browned", roasted, or toasted aromas.

For the microwave samples too, the aldehydes and ketones represent greater proportions of the isolates obtained by dry heating, the 3-methylbutanal reaching up to 40% of the total isolates (as measured by GC response). Increased concentration of sulfides is also shown. Persson and von Sydow (1973) demonstrated that carbonyl and sulfur compounds are responsible for the high heat off-flavor ("retort" flavor) of canned beef. Several sulfides have been identified by Mussinan and Katz (1973) in the strong roast aroma emanating on heating a model meat system containing hydrolyzed vegetable protein, L-cysteine, D-xylose, and water. Similarly, Wilson et al. (1973) identified many sulfur compounds in the aroma of pressure-cooked beef, particularly at high temperature. Sulfides and 3-methylbutanal were among certain com-



**Figure 3.** Comparison of the effect of heating time on the percentage relative abundances of classes of volatile components in conventional boiled and roasted aroma: (---) conventional boiled aroma; (—) conventional roasted aroma; (a) alkanes; (b) alkenes < C<sub>20</sub>; (b') C<sub>20</sub> alkenes; (c) aldehydes; (d) ketones; (e) benzenoids; (f) furans; (g) pyrroles; (h) pyrazines.

pounds implicated in burnt aroma of canned beef by Persson and von Sydow (1974). Further studies at our laboratories have indicated a positive correlation between the odor descriptions roast meat and burnt.

For both the conventional and microwave samples possessing roasted aromas, the alkanes, alkenes (<C<sub>20</sub>), benzenoid compounds, and furans show lower RPA values than for the boiled beef in general.

As a group, the pyrazines are present in approximately equal concentrations in conventional boiled and roast aroma, with higher amounts in the microwave samples. This enhanced microwave effect has been previously discussed (MacLeod and Coppock, 1976). Maga and Sizer (1973) report that optimum concentrations of pyrazines probably exist and that a high pyrazine level can give off-odors. As shown in Table I, pyrazines have not previously been identified in boiled beef aroma, except by Liebich et al. (1972) who did not specify their experimental conditions for boiled beef aroma isolation. However, in the present work, the paired comparison sensory tests showed a significantly correct differentiation between samples possessing boiled and roast aroma. Reineccius et al. (1972) have shown that pyrazine formation can occur at time-temperature relationships as low as 30 min at 70 °C. Production is quite rapid and linear during the first 30 min of roasting at 150 °C and above this temperature, pyrazine yields are highly variable—possibly due to pyrazine destruction after their formation (Koehler and Odell, 1970). The mechanisms for the formation of alkylpyrazines at lower, as opposed to higher, temperatures are discussed by Koehler and Odell (1970) and by Hough et al. (1952). Thus, although certain alkylpyrazines more than likely account for some roasted notes in cooked foods, they are clearly not the sole contributors to the spectrum of rather more desirable odor qualities associated with the roasted aroma of beef compared with that of boiled beef.

Only two pyrroles have previously been identified in heated beef aroma (Liebich et al., 1972; Watanabe and Sato, 1972a,b). Further evidence to support the present suggested implication of pyrroles in roasted aroma was revealed from an isolate obtained from relatively strongly heated beef (extraction time 4 h and dry heating conditions). This sample possessed a very strong roast beef odor and contained pyrrole at an RPA level as high as 6%.

Additional pyrrole compounds not listed in Table II were also identified in this particular sample, these being an ethylpyrrole, a trimethylpyrrole, a dimethylethylpyrrole, and a C<sub>5</sub> saturated substituted pyrrole, probably *N-n*-pentylpyrrole. In this sample, 3,4-dimethylpyridine was also additionally identified. Although proline and myoglobin may act as precursors for the pyrroles, they are also, together with the pyridines, products of Maillard browning reactions (Watanabe and Sato, 1972a,b). Pyridines have a burnt and heavy fruity odor while pyrroles have burnt odors in concentration, but when diluted the odors have been variously described as nutty, corny, and bready (Hodge et al., 1972; Reynolds, 1970).

Thus, there is some evidence that carbonyl compounds (particularly 3-methylbutanal), sulfides, pyrroles, and pyridines are probably important in roast aroma of heated beef. Further work at these laboratories has suggested that the benzenoids and furans may be associated with the desirable qualities associated with well-cooked boiled beef. Odor descriptions of all samples have been achieved and detailed comparisons of chemical and sensory data are currently in progress.

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**Supplementary Material Available:** Table II showing absolute peak areas and relative percentage abundances of beef aroma isolates obtained under dry heating conditions (7 pages). Ordering information is given on any current masthead page.

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## Comparison of Nutrient Composition and of Enzyme Activity in Purple, Green, and White Eggplants

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Tissues of purple, green, and white varieties of eggplant, *Solanum melongena*, grown under identical conditions in the same area, were analyzed for percent moisture and total solids. The total solids were further analyzed for crude protein, amino acid composition, ether-extractable lipids, crude fiber, ash, and nitrogen-free extract. Trace elements in the dried matter were determined by neutron activation analysis. Activities of three enzymes (polyphenol oxidase, alcohol dehydrogenase, and catalase) were compared in the three varieties. Composition of the total solids and activities of these enzymes in the three varieties are discussed with respect to their effects on keeping quality of fresh or processed eggplants. Based upon these data, the green variety appears to have better properties for processing than the more popular purple variety.

The growing interest in nutrient composition of foods can be attributed in part to the new FDA nutritional labeling regulations. In the past there was little concern for variations in nutrient contents of many established vegetables because the development of new varieties emphasized higher yields or better processing and storage characteristics, rather than nutrient composition. Recent research on the requirements of several essential trace elements has stimulated investigations on the sources and availabilities of these minerals in plant foods (Leveille et al., 1974).

To ensure a year-round supply of fresh and processed fruits and vegetables we need a better understanding of the initial composition and of certain enzymes that catalyze biochemical changes in fresh vegetables during extended storage, or before and during processing. Adverse changes due to overripening or deterioration of the tissue can lower final quality of the product.

Eggplants are grown only to a limited extent in northern climates because they need a long, warm, frost-free growing season of 14 to 16 weeks for good yields (Combs et al., 1973). Although the purple fruit is the most widely used, other varieties are known that differ in size, shape, and color. A white variety has been grown in Europe for many years, but apparently for ornamental purposes only (Dumonthay, 1936). A round, light green eggplant has

been grown in India (Viraktamath et al., 1963) and is currently appearing in home gardens in the southern United States, but it has not been found to a significant degree in fresh produce markets. A pink and a black eggplant has been cultivated in India (Singh et al., 1974), but their acceptance has been based only on external characteristics such as color, shape, and size of the fruit.

Polyphenol oxidase (EC 1.10.3.1) is probably a major cause of discoloration in eggplants that have been subjected to rough handling, storage, peeling, and processing. This enzyme utilizes the tannins and phenolic compounds in the tissue as substrates, causing brown products. Lipoxygenase (EC 1.13.1.13), which can also produce off-flavors from polyunsaturated fatty acids, was found in purple eggplants by Pinsky et al. (1971). The first comparison of lipoxygenase activity and its inhibition by KCN in purple, green, and white eggplants was recently reported (Flick et al., 1975). Two other enzymes in fresh vegetables which may be related to keeping quality are catalase (EC 1.11.1.6), which catalyzes decomposition of the hydrogen peroxide that is toxic to plant cells, and alcohol dehydrogenase (ADH) (EC 1.1.1.1), which converts the resulting aldehyde products of lipoxygenase activity to alcohols in tea seeds (Hatanaka and Harada, 1972). These studies were undertaken to compare the nutrient composition and three enzymes that can affect flavor and quality of purple eggplants with the two lesser known green and white varieties.

### MATERIALS AND METHODS

Since many factors (i.e., soil conditions, irrigation, fertilization, growing areas, and maturity at harvest) can influence the levels of specific nutrients or components in

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