berries averaged 87.9% and of the leaves 66.4%.

As shown in Table I, the experimental plots that were sprayed only once at day 1 had 0.25 and 0.34 ppm BNOA residues on the berries when sprayed with 50 or 100 ppm AI, respectively. By day 5 to 6 the residues had dropped to <0.05 ppm. The experimental plots that were resprayed on day 5 showed residues of BNOA on the berries of 0.17 and 0.33 ppm and then dropped to <0.05 ppm by days 10 or 11. All mature berries at harvest (day 15) had residues <0.05 ppm.

The BNOA residues on the leaves showed similar trends of residue dissipation as on the berries except that the initial residues were much larger (5.05 and 8.40 ppm) when sprayed on day 1 with 50 or 100 ppm, respectively. On day 15 the residues had declined to 0.16 and 0.25 ppm. The residues on the resprayed leaves were proportionally higher but showed similar trends in residue dissipation as did the plots sprayed only one. No β-naphthol residues were found in either the berry or leaf samples.

The strawberry blossoms that were sprayed, sampled, and analyzed had residues of 4.7 and 7.6 ppm when sprayed with 50 or 100 ppm AI BNOA, respectively. Due to the limited number of plants available, no berries were sampled from sprayed blossoms. However, since the directly sprayed berries only gave residues of 0.25 and 0.34 ppm when sprayed with 50 or 100 ppm AI BNOA, respectively, it is probably reasonable to assume that by harvest time 15 days later, the residues would have been <0.05 ppm due to dissipation and dilution of the plant residues by growth and environmental factors.

β-Naphthoxyacetic acid residues dissipated very rapidly on berries and leaves of strawberry plants that were sprayed in the field. Apparently BNOA does not degrade in the field to its postulated mebabolite, β-naphthol, since it was not detected by high-pressure liquid chromatographic analysis at 0.05 ppm or higher.

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Effect of Maturity, Cultivar, Field History, and the Operations of Peeling and Coring on the Geosmin Content of *Beta vulgaris*

The purpose of this study was to obtain information about the origin and distribution of geosmin (*trans*-1,10-dimethyl-*trans*-(9)-decalol), the earthy odor component in beets. The geosmin content of *Beta vulgaris* L. cv. Ruby Queen was monitored throughout growth and storage. The data indicate that geosmin content is directly related to the surface area of the beet. In addition the geosmin content of eight different cultivars of *B. vulgaris* showed little variation except for two cultivars which had a large number of secondary roots (i.e., more surface area). An analysis of the distribution of geosmin in the root of Ruby Queen beets showed that the peels of the beets had the highest concentration. The high concentration of geosmin in the surface of the beet implies that the geosmin may be derived from the soil in which it is grown. However, the geosmin content of beets grown in two fields with different histories of cultivation was the same.

Geosmin (*trans*-1,10-dimethyl-*trans*-(9)-decalol) has been identified as an odorous metabolite of many soil microorganisms of the family Streptomyces (Gerber and Lechevalier, 1965; Collins et al., 1970; Kikuchi et al., 1972), and as a metabolite of several species of blue-green algae (Safferman et al., 1967; Medsker et al., 1968; Kikuchi et al., 1973). The strong earthy odor of this compound led scientists to the suspicion that it was responsible for musty off-odors sometimes present in water supplies and in fish (Gerber and Lechevalier, 1965; Safferman et al., 1967; Lovell, 1972). Geosmin was subsequently identified as an earthy odor contaminant of water, catfish, rainbow trout, and dry beans (Rosen et al., 1970; Lovell, 1972; Yurkowski and Tabachek, 1974; Buttery et al., 1976). In each case, earthiness was recognized as an abnormal odor produced by microorganisms. In contrast, geosmin, isolated from red table beets, was perceived as part of the characteristic aroma of beets (Murray et al., 1975; Acree et al., 1976).

It is not known whether the geosmin in table beets is an endogenous metabolite of the beet itself or a contaminant produced by microorganisms. In an attempt to answer this question, the changes in the geosmin content of one cultivar of *Beta vulgaris* (Ruby Queen) during 5 months of growth and 2 months of storage were measured by gas chromatography. In addition, the differences in the geosmin content of Ruby Queen beets grown in two different fields and the variations in the geosmin content of eight cultivars of *Beta vulgaris* were determined. Since the origin of geosmin may be reflected in its distribution within the root, differences in the geosmin content of various parts of Ruby Queen beets are presented in this paper.

EXPERIMENTAL SECTION

Beet Samples. An experimental field of lima silt loam soil at the New York State Agricultural Experiment Station was fertilized with 250 kg/acre of 10-20-20 fertilizer on May 2, 1977. The following day the field was treated with 2 kg/acre of Ro-Neet herbicide (Stauffer Chemical Co., Westport, Conn.). On May 4, 1977 eight cultivars of *Beta vulgaris* seed were planted about 0.75 in. deep at a rate of about 9 kg/acre with a mechanical planter. This field was fertilized again on June 22, 1977 with 25 kg/acre of ammonium nitrate. A commercial field of the same soil type was fertilized on May 2, 1977 with about 360 kg/acre of 10-20-20 fertilizer. The same herbicide was used on the

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commercial field as was used on the experimental field at the same rate of application. The commercial field was planted (also on May 4, 1977) with *Beta vulgaris* L. cv. Ruby Queen in the same manner at a rate of about 11 kg of seed/acre. The commercial field was fertilized again the last week of June with 70 kg/acre of ammonium nitrate.

After 6 weeks of growth, 3 kg of Ruby Queen beets was harvested from the experimental plot. This sample size was sufficient for one analysis of geosmin. Subsequent samples weighed at least 6 kg to allow for duplicate analyses. Ruby Queen beets growing in the experimental plot were sampled at approximate 3-week intervals thereafter. Twice during the season the Ruby Queen beets in the commercial plot and the experimental plot were sampled simultaneously. In the 24th week after planting 18 kg of Ruby Queen beets was harvested, washed, and stored at 0 °C and 90% RH. The beets were sampled after 1 month and after 2 months in storage. Sixteen kilograms of Ruby Queen beets were harvested from the experimental plot 20 weeks after planting to be used in measuring the geosmin content of three parts of the beet. All eight cultivars growing in the experimental plot were sampled at maturity in October of 1977.

Beet Juice. Each batch of beets was washed immediately after harvest, ground, digested with 10% (w/w) EtOH 95%, and pressed into juice (Tyler et al., 1978). Approximately 30 beets selected at random from each sample were weighed to determine the average weight of a beet in that sample.

The first sample of Ruby Queen beets harvested yielded only 1 L of juice. Duplicate 1-L juice samples were obtained for all other Ruby Queen beets harvested at different times and for each variety of *Beta vulgaris*. The °Brix of the juice of each variety was measured using a refractometer.

To determine the distribution of geosmin within the beet, approximately 16 kg of beets (cv. Ruby Queen, ca. 8 cm in diameter) was washed and an average 2 mm of the outer surface was removed by a four-cup peeler/corer (Pease Corp., Rochester, N.Y.). An average 16 mm of the axial, cylindrical core was simultaneously removed from the beet by the peeler/corer leaving the main body of the beet. The resulting peels, cores, and bodies were each ground into pulp. Samples of the resulting pulp (peels, 1550 g; cores, 1160 g; bodies, 2000 g) were made into juice as described previously. One liter of juice was obtained from the peels and bodies and 0.96 L from the cores. Five beets were used to estimate the average weight of the peel, core, and body of a beet. The range of weights for the peels, bodies, and cores was 25 to 29 g, 169 to 265 g, and 12 to 16 g, respectively.

Concentrated Extract. Each sample of beet juice was extracted with Freon 113 (1,1,2-trichloro-1,2,2-trifluoroethane) (Precision Cleaning Agent, Buffalo Solvent Corp., Buffalo, N.Y.), 40% by volume. Each extract was concentrated with a Buchi Rotovapor (Rinco Instruments, Greenville, Ill.) in a 20 °C water bath using Concentratubes (Laboratory Research Co., Los Angeles, Calif.). The extracts were purified by adsorption chromatography and analyzed for geosmin by gas chromatography. Recovery of geosmin from model solutions (10% w/w ethanol in water) and beet juice used in the development of this determination procedure averaged 109% with a standard deviation of 11.7% over the range of 0.31–10.00 ng/g (Tyler et al., 1978).

Gas Chromatography. A 4 m × 3 mm stainless steel column packed with 5% SP1000 on Chromosorb W was used in a Varian Model 1200 gas chromatograph equipped with a flame ionization detector. The column was operated isothermally at 190 °C with a carrier gas (He) flow rate of 25 mL/min. The geosmin content of each sample was calculated by measuring its peak height relative to the peak height of an internal standard (1-undecanol) (Tyler et al., 1978).

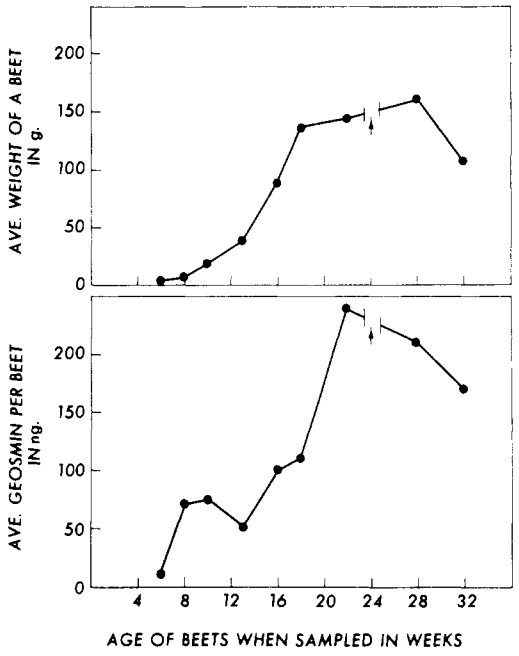

Figure 1. A plot of both the average geosmin content of a beet (ng) and the average weight of a beet (g) versus time in weeks after planting. At the 24th week (arrow) the beets were placed in storage.

RESULTS AND DISCUSSION

Figure 1 is a plot of both the average weight of a beet and the average geosmin content per beet versus time. The figure shows that the geosmin content of the beet increases approximately as the beet increases in weight up to the 24th week, at which time the beets were harvested and placed in storage. Figure 1 also illustrates that both the geosmin content and weight of the beets decreased during storage. A regression line obtained for the average geosmin per beet versus the average weight of a beet had a correlation coefficient of 0.87. The data showed no increased or decreased deviation from the regression line at the extremes of the range studied. These data indicate that the average geosmin content per beet increases and decreases as the average weight of the beet increases and decreases.

The beet cultivar Ruby Queen is almost spherical in shape at maturity and its volume is proportional to its weight. The surface area of a Ruby Queen beet can be approximated by the two-thirds root of its weight since the surface area of a sphere is proportional to the two-thirds root of its volume. A regression line of the average geosmin per beet versus the two-thirds root of the average weight of a beet has a correlation coefficient of 0.86 which is essentially the same as the correlation coefficient obtained for the average geosmin per beet versus the average weight of a beet. Therefore, these data do not indicate a strong relationship between the geosmin content per beet and the surface area of a beet.

Table I reports the change in the concentration of geosmin in beets (ng/g) with time. The concentration of geosmin in beets was at its highest level on the first sampling date and it decreased from that point to the level

Table I. Changes in the Geosmin Content of Beets (cv. Ruby Queen) during Growth and Storage

date sampled	concn of geosmin in beets, ng/g	av, ng/g
6/14 (6 weeks after planting)	13	13
6/28	16	
	8.3	12
7/12	4.2	
	3.6	3.9
8/03	1.4	
	1.4	1.4
8/22	1.1	
	1.2	1.2
9/08	0.8	
	0.8	0.8
10/03	1.3	
	2.1	1.7
11/17 (stored from 10/21)	1.2	
	1.4	1.3
12/14 (stored from 10/21)	1.8	
	1.4	1.6

Table II. Differences in the Geosmin Content of the Peels, Cores, and Bodies of Beets (cv. Ruby Queen)

location	av wt,[a] g	concn[b] of geosmin, ng/g	% of total geosmin in an av beet
peel	26	4.9	26
body	205	1.7	70
core	14	1.5	4

[a] The average from five beets (approximately 8 cm in diameter) of appropriate size for the peeler. [b] Approximately 3 kg each of the three beet parts were used in determining the geosmin concentration.

Table III. Comparison of the Geosmin Content of Beets Grown in Two Different Fields

date sampled	field type	av wt. of a beet, g	concn of geosmin in beets, ng/g	av ng/g
8/01	commercial	30	1.8	
			2.1	1.9
8/03	experimental	38	1.4	
			1.4	1.4
8/22	experimental	38	1.1	
			1.2	1.2
8/25	commercial	48	0.6	
			1.7	1.2

attained on the fourth sampling date at which point it remained approximately constant. A plausible explanation for these data is that the geosmin concentration decreases as the surface-to-volume ratio of the beet decreases. The surface-to-volume ratios of the cone and sphere which approximate the shape of a beet at different stages of growth decrease as the radii increase. Therefore, the surface-to-volume ratio of a beet decreases as the beet enlarges and later on in the growing season it becomes static as the growth rate slackens. This interpretation of the data is supported by the fact (presented in Table II) that the surface or peel of the beet has a higher geosmin concentration than any other part. Therefore, the surface area of the beet relative to the volume of the bulk of the beet is an important factor in determining the average concentration of geosmin throughout the beet.

Table II gives the average concentration of geosmin in the cores, peels, and bodies of beets. The concentration of geosmin in the peels is about three times that found in the bodies and cores of the beets. The peels of the beets contained 25% of the total geosmin in only 11% of the weight of the beets. The concentration of geosmin in the cores and bodies of the beets was nearly the same, indicating that the distribution of geosmin throughout most of the beet flesh is fairly uniform except for the outer 2 mm of peel.

The facts presented thus far show that the geosmin content increases as the weight and surface area of the beets increase. One would expect that an enlarging surface area would provide an increasing medium for the growth of actinomycetes which are known to exist in the rhizosphere of *Beta vulgaris* (Rouatt et al., 1951). Several of these actinomycetes are known to produce geosmin, especially *Streptomyces* (Gerber and Lechevalier, 1965). These actinomycetes may produce geosmin on the surface of the beet root in an incidental or symbiotic relationship. This may explain the increase of geosmin with beet size and surface area and is also consistent with the data indicating that the highest geosmin concentration is in the peel of the beet. Actinomycetes have been reported to grow in the middle lamellae of the beet root although in fewer numbers than at the beet surface (Lutman, 1945). Therefore, these microorganisms could be producing geosmin throughout the beet root with the highest production at the surface of the beet where the number of organisms has been reported to be highest. However, the data do not rule out the possibility that geosmin is produced endogenously by the growing cells. The most actively dividing cells of a growing beet are the periderm parenchyma cells and the cambial cells adjacent to the

Table IV. Comparison of the Geosmin Content of Eight Cultivars of *Beta vulgaris*

cultivar	date sampled	av wt of a beet, g	concn of geosmin in beets, ng/g	av, ng/g	av ng geosmin per beet	°Brix
Ruby Queen	10/03	143	1.3	1.7	240	11
			2.1			
Burpee's Golden	10/06	136	1.3	1.1	150	9
			0.9			
Detroit Dark Red	10/06	192	0.8	0.8	150	10
			0.8			
Nero	10/10	117	0.7	0.6	70	11
			0.5			
Sugar beet	10/10	270	2.8	3.1	840	20
			3.4			
Cylindra	10/12	363	0.7	0.8	270	9
			0.8			
Swiss chard	10/12	130	2.8	3.7	470	15
			4.5			
Crosby's Egyptian	10/13	140	0.9	1.0	140	9
			1.1			

periderm (Hayward, 1938). The outer 2 mm of the beet is partially composed of these cells and therefore could account for the higher geosmin concentration in this region. The fact that geosmin has never been identified in other root crops such as carrots (Buttery et al., 1968; Seifert and Buttery; 1978), even though actinomycetes have been reported to grow between the cells of these crops (Lutman, 1945), gives some credence to the idea that geosmin is an endogenous metabolite.

The soil composition may have an effect on the geosmin content of beets grown in it because geosmin has been isolated from soil (Buttery and Garibaldi, 1976) and the rhizosphere of *Beta vulgaris* has been reported to contain a higher concentration of actinomycetes than control soil samples (Rouatt et al., 1951). Spores of actinomycetes and geosmin may accumulate in the soil over several seasons and thus increase the geosmin content of beets grown on this soil. Table III shows the difference in geosmin content of beets harvested at similar times from two different fields. The major difference between the two fields was that the commercial field had a history of beet cultivation while the experimental one did not. According to the data in Table III there is no significant difference in geosmin content per gram on both sampling dates for the two fields studied, even though the beets in the experimental plot grew more between the two sampling dates.

The geosmin content of a beet may depend on its genotype. Table IV shows the geosmin content of eight cultivars of *Beta vulgaris*. Sugar beets and Swiss chard were significantly higher in geosmin content per gram and per beet than the other varieties. Sugar beet roots and Swiss chard roots may be higher in geosmin content than table beet roots because they have more secondary roots which gives them a larger surface-to-volume ratio (Hayward, 1938). Neither of these cultivars is used as a table beet, so consequently excessive earthy flavor is not a quality concern. It is interesting to note that sugar beets and Swiss chard were highest in sugar content also. It is possible that some relation exists between sugar production and geosmin synthesis. The most striking result in Table IV is that the geosmin content of all the red table beets fall within a fairly narrow range (0.6–1.7 ng/g). This narrow range suggests either that the concentration of this chemical is not genetically controlled or that a gene controlling geosmin production is buried deeply within the genotype along with vital functions of the beet plant. One would expect more variability in the range of geosmin contents for the widely differing cultivars studied if the geosmin content were genetically controlled.

This paper shows that geosmin was present at some level in every sample analyzed and that its amount was approximately proportional to the surface area of the root. Therefore, we can say that geosmin is a typical component of beets even though it is not yet known how it is produced.

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Spectrophotometric Determination of Copper in Alcoholic Beverages

By using 6-phenyl-2,3-dihydro-*as*-triazine-3-thione (PDTT), the copper content of different alcoholic beverages was determined spectrophotometrically, after dry ashing. Trace elements commonly present in these beverages had no interference in the determination. Recovery of added copper was 98%, and the method showed excellent agreement with the atomic absorption method of the AOAC.

Determination of copper concentration in foods and beverages has long been of significant interest. Several trace metals, including copper, have deleterious effects on color, aroma, and taste of alcoholic beverages. Besides the known toxic actions of copper, the action of copper in several important diseases was recently investigated (Klevay and Forbush, 1976; Raitses and Pityk, 1976; Zyka, 1971). The permissible maximum copper content in these beverages is defined by health regulations. It is therefore desirable to use a suitable method for its analysis.

A number of methods have been described for the determination of copper in alcoholic liquors. In view of the low concentration of copper in these beverages, the spectrophotometric (Szobolotzky, 1970; Maneva et al.,

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