contents were those of fruit.

On the basis of the data of Table III, together with similar data obtained for vegetables by Herrero-Latorre et al. (1986), the contributions of these kinds of food to the mean intake of selenium in Galicia/person per day have been estimated by multiplying the mean Se content of each group by the mean consumption of that kind of food in Galicia/person per day (OERGA, 1980). The results are shown in Table IV togeter with those of similar calculations carried out using the mean consumptions recommended in the balanced diet tables of Randoin et al. (1969). It may be noted that in spite of their high Se levels, mushrooms make a relatively modest contribution to total Se intake because their consumption is not widespread in Galicia. The total Se intake in food of vegetable origin, 11.20 μg/person per day, is very similar to the figure of 13.01 μg/person per day obtained using the tables of Randoin et al. (1969). The fact that both figures are considerably lower than those for other parts of the world [in Canada, for example, cereals alone provide 62–112 μg of Se/person per day (Thompson et al., 1975)] may be attributed to the low selenium content of soils.

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LITERATURE CITED

Analytical Methods Committee, Royal Society of Chemistry, *Analyst* **1979**, *104*, 778.
Hartley, W. J. *N.Z. J. Agric.* **1959**, *100*, 259.
Herrero-Latorre, C.; Mejuto-Martí, M. C.; Bollaín-Rodríguez, M. H.; Bermejo-Martínez, F. *Anales Bromatologia* **1987**, *39*(1), 133.
Ihnat, M. *J. Assoc. Off. Anal. Chem.* **1976**, *59*, 911.
Instituto Nacional de Estadistica, Ministerio de Economía, Observatorio Estadístico Regional de Galicia (OERGA) *La Alimentación en Galicia*, Santiago de Compostela, 1980.
IUPAC *Pure Appl. Chem.* **1984**, *56*, 645.
Kubota, J.; Allaway, W. H.; Carter, D. L.; Cary, E. E.; Lazar, V. A. *J. Agric. Food Chem.* **1967**, *15*, 448.
Levander, O. A. *J. Am. Diet. Assoc.* **1975**, *66*, 338.
Levander, O. A. *Proc. Symp. Selenium–Tellurium Environ.* **1976**, 26.
Mondragon, M. C.; Jaffe, W. G. *Arch. Latinoam. Nutr.* **1976**, *26*, 341.
Morris, W. C.; Levander, O. A. *J. Nutr.* **1970**, *100*, 1383.
National Research Council, Agricultural Board, Committee on Animal Nutrition, Subcommittee on Selenium (J. E. Oldfield, Chairman) *Selenium in Nutrition*; National Academy of Sciences: Washington, DC, 1971.
Olson, O. E.; Palmer, I. S. *J. Food Sci.* **1984**, *49*, 446.
Pipponen, S.; Liukkonen-Lilja, E.; Kuusi, T. *Z. Lebensm. Unter. Forsch.* **1983**, *177*, 257.
Pipponen, S. *Kem. Kemi.* **1984**, *11*, 180.
Randoin; Legalli; Duchene In *Rations Equilibrees*, 8th ed.; Foucher, E. Ed.; Hygiene: Paris, 1969; Vol. I, p 35.
Rosenfield, I.; Beath, O. A. *Selenium, Geobotany, Biochemistry, Toxicity and Nutrition*; Academic: New York, 1964.
Schwarz, K.; Foltz, C. M. *J. Am. Chem. Soc.* **1957**, *79*, 3292.
Thompson, J. N.; Erdoby, P.; Smith, C. *J. Nutr.* **1975**, *105*, 274.

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Green Leaf Headspace Volatiles from *Nicotiana tabacum* Lines of Different Trichome Morphology

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Volatile compounds from fully expanded whole leaves of four greenhouse-grown vegetative (green) tobacco lines were isolated by entrainment in purified air followed by Tenax trapping, characterized by capillary GC and GC–MS, and quantified by capillary GC. Identified headspace compounds, which comprised about 50% of the number of GC peaks and weight of the total estimated volatiles, were (*E*)-β-ocimene, (*Z*)-3-hexenyl acetate, (*Z*)-3-hexen-1-ol, linalool, β-caryophyllene, (*E*)-β-farnesene, solanone, methyl salicylate, nicotine, and neophytadiene. Yields of total estimated volatile compounds among the replicated leaf samples of the tobacco lines ranged from 6.6 to 38.5 ng/g wet weight. The means of amounts of volatile components that were believed to be emitted as a result of wound-induced lipoxygenase-hydroperoxide lyase activities were higher for analyses performed with 5-L entrainment flasks than with 12-L flasks. The leaf surface emissions for individual volatile compounds are discussed in terms of genetic differences including leaf trichome morphologies among Tobacco Introduction (TI) 1112, TI 1406, and TI 1068 and KY 14 burley. The results obtained do not support the view that the majority of headspace volatiles are emitted from exudate-secreting glandular heads of leaf trichomes. However, there were significant differences in yields of some compounds among genetic lines of tobacco.

There have been relatively few reports on the occurrence, quantities, and biogenesis of volatile compounds in green vegetative tobacco as compared with cured tobacco. It is recognized, however, that knowledge about the composition of volatiles in tobacco prior to harvest could be useful in the understanding of the biogenesis of compounds in postharvest tobacco that affect the quality of leaf and may play a role in plant–pest interactions. The occurrence of at least 25 compounds in steam distillates and five compounds in the headspace of burley tobacco stalk and their quantitative analyses were recently reported (Andersen et al., 1986). Burton and Kasperbauer (1985) showed that the composition of some volatile components in green burley leaf at harvest changed during air-curing. Several reports have appeared on volatiles in terms of the effects of cultural practices on cured tobacco (Weeks and Seltmann, 1986), composition in flue-cured tobacco (Lloyd

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et al., 1976; Rix et al., 1977; Enzell and Wahlberg, 1980), and changes during curing (Enzell and Wahlberg, 1980) and during aging (Sakaki et al., 1985).

Volatile aldehydes, alcohols, and esters containing primarily six and nine carbon atoms are believed to be derived from lipoxygenase-hydroperoxide lyase mediated oxidations of unsaturated fatty acids such as linoleic and linolenic acids (Galliard and Phillips, 1976; Galliard, et al., 1976; Hatanaka et al., 1978; Wu and Liou, 1986). Several volatile compounds of this type were present in hexane extracts of steam distillates (SDE) of burley tobacco stalk, but none were detected in the headspace volatiles of the same tissue (Andersen et al., 1986). In contrast, volatile sesquiterpenoids, which presumably accumulate in specialized secretory structures analogous to tobacco trichomes (Loomis and Croteau, 1980) or in specialized internal accumulatory cells present in some plants (Henderson et al., 1970) were not detected in SDE, but they were present in the headspace of green tobacco stalk (Andersen et al. , 1986). These compositional differences in volatiles obtained by these methodologies can be rationalized from the following inherent features of the techniques used for their isolations: (1) a high degree of damage to all leaf cells (including those of leaf trichomes) and presumed resultant activity of the lipoxygenase enzyme system during the preparation (chopping) of tobacco tissue for steam distillation; (2) a low degree of wounding, resulting in reduced activity of lipoxygenase and damage to leaf epidermal cells and trichome structures during the gas entrainment and trapping of headspace volatiles.

The purpose of this investigation was to isolate, identify, and quantify headspace volatiles of vegetative (green) burley tobacco leaf. A comparison was made of the quantities of volatile compounds obtained from genetic lines of burley tobacco that varied in trichome morphology and density to determine whether the majority of leaf volatiles are emitted from the glandular heads of secreting trichomes.

EXPERIMENTAL SECTION

Plant Materials. Tobacco plant seedlings differing in trichome morphology (*Nicotiana tabacum* L. cv. KY 14, TI 1068, TI 1112, and TI 1406) (Nielsen et al., 1982; Severson et al., 1985) were transplanted to the greenhouse soil floor in mid-October 1986. No chemical agents were applied after transplanting, but other recommended cultural and fertilization practices were followed (Andersen et al., 1969). The plot design was a randomized complete block and included 3 replicates of each tobacco line as subplots with 12 plants each. Whole leaves that were approximately 20 in. long were harvested at 3 p.m. each day from November 24 to December 5 for immediate weighing and headspace analyses. Harvests were completed before the floral bloom stage.

Isolation of Volatiles. Headspace compounds were isolated from 200 g of tobacco leaves with minor modifications of the method previously described for tobacco stalk (Andersen et al., 1986). The trapping agent was 0.5 g of Tenax packed in a glass tube attached to an entrainment vessel. Two isolations of volatiles from samples of one tobacco line/replicate were carried out on a given day with a 45/50 standard taper (Ⓢ) 5-L (run A) and a 71/60 Ⓢ 12-L (run B) round-bottomed flask as the entrainment vessels at 30 °C for leaves swept with activated charcoal/molecular seive filtered purified air at a flow rate of 500 mL/min for 20 h. Freshly picked whole leaves were loosely rolled and placed in the flasks with as much care as possible to minimize leaf injury. Isolations were then carried out immediately. After a run, each Tenax trap was eluted with 15 mL of *n*-hexane (distilled in glass). The hexane was concentrated to 100–1000 μL in a microstill fitted with a Vigreux column. The volume was measured, and an internal standard of cumene in hexane was added to the volatiles solution, yielding a known concentration of about 20 ng/μL. This solution was used directly for capillary GC or GC–MS analysis.

In addition to tobacco headspace isolations, a single isolate of 200 g of freshly harvested field-grown red clover leaves including top portions of stems and some immature seed pods was prepared in a 5-L entrainment flask in the same manner that was used for tobacco leaves. This isolate provided a mixture that contained (*E*)-β-ocimene and (*E*)-β-farnesene that were required as standards (Buttery et al., 1984).

Capillary GC and GC–MS Analyses. A Hewlett-Packard 5880A GC was used with a 60 m × 0.32 mm Supelcowax 10 column (Supelco, Inc.) for capillary GC analyses as follows: inlet temperature, 240 °C; operation, splitless mode; He carrier linear velocity, 31 cm/s; column temperature, held 1 min isothermal and then programmed from 60 to 220 °C at 2 °C/min and held at 220 °C for 30 min; FID detector, 240 °C. Kovats' indices were calculated for separated components relative to hydrocarbon standards (Perry, 1981).

Yields of total volatile oils and individual volatiles in Tenax-trapped headspace eluates were estimated from FID peak areas of components and the internal standard used in the GC analyses. Quantitative results were subjected to a statistical analysis of variance. Duncan's multiple-range test was used to make comparisons among treatment/line means.

Aliquots of the solutions of the volatile isolates used for capillary GC were analyzed by GC–MS using a Hewlett-Packard Model 5985A instrument as previously described with the capillary GC column conditions described in this subsection. Electron impact MS and chemical ionization MS with methane were obtained. Identifications of compounds were confirmed by matching EI- and CI-MS data, comparison with published spectra, and cochromatography of components with authentic compounds.

Reagents. Chemical compounds for cochromatography and comparisons of mass spectra and GC retention times were obtained from commercial or other sources. Reagent-grade hexane was redistilled on a Vigreux column. Tenax GC adsorbent (60–80 mesh) was obtained from Alltech Associates Inc.; it was conditioned before use by washing each 0.5-g portion with 100 mL of hexane and then preheating it 2 h in the glass column used in the headspace apparatus at 250 °C in a stream of purified nitrogen.

RESULTS AND DISCUSSION

Scanning Electron Photomicrographs of Tobacco Lines. Variations in leaf trichome morphologies and leaf surface (underside) areas among the tobacco lines are shown in Figure 1. Leaves of Tobacco Introduction (TI) 1406, TI 1068, and KY 14 that were selected for full expansion all had glandular headed trichomes. Trichomes of TI 1112 were headless and presumably nonsecreting. There were fewer glandular trichomes per unit surface area on TI 1406 than on TI 1068 and KY 14. Glandular trichome densities on the leaves of these lines were previously reported; Tobacco Introduction 1406 had glandular trichomes that were nonsecreting, whereas TI 1068 and KY 14 burley tobacco had glandular secreting trichomes (Nielsen et al., 1982).

Quantitative Estimates of Total Headspace Volatiles. The means of amounts (wet-weight basis) of total volatile compounds (identified and unknown) in the 5-L

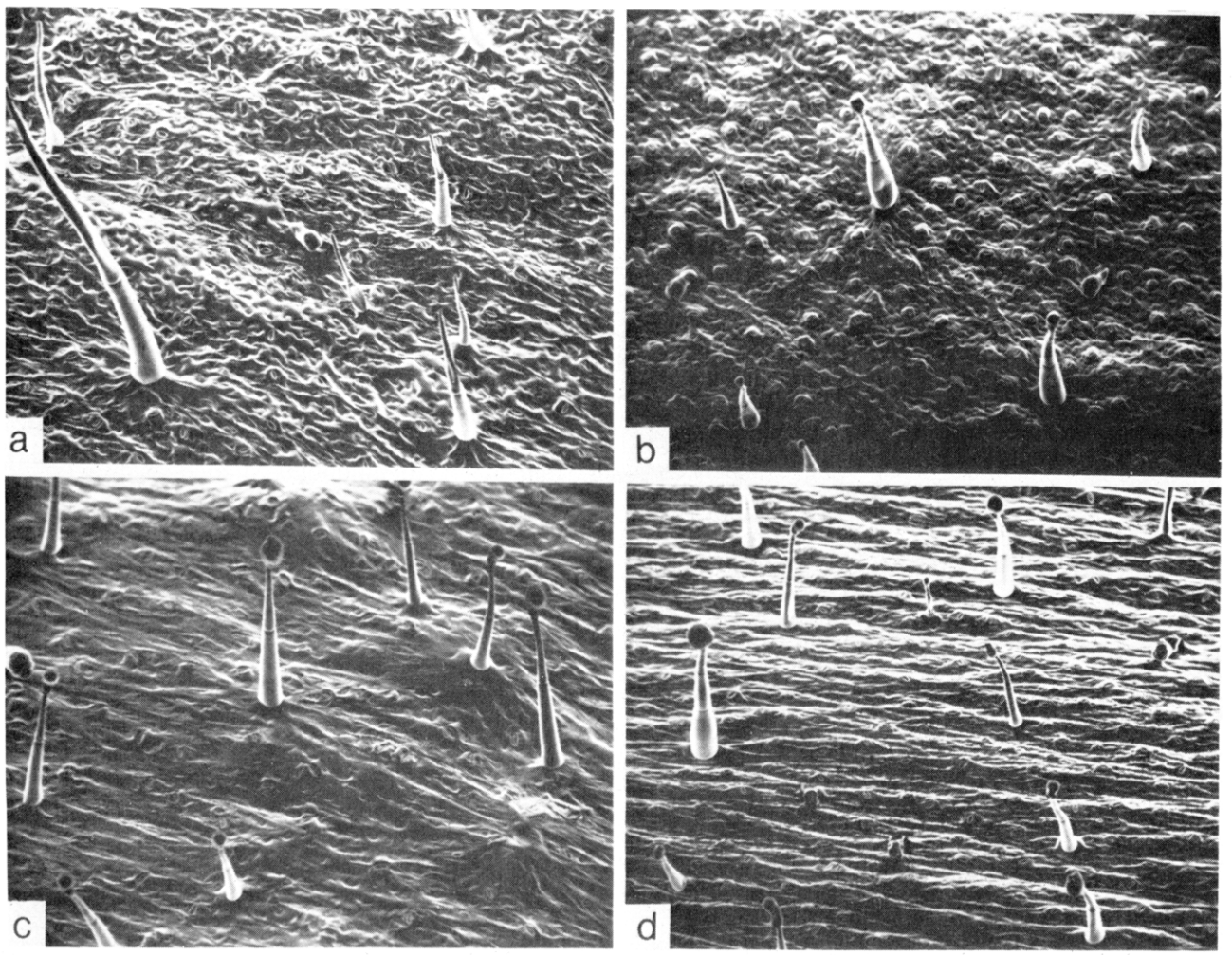

Figure 1. (a) Nonglandular headless trichomes of TI 1112 leaf. (b) Glandular-headed trichomes without exudate of TI 1406 leaf. (c) Glandular-headed trichomes with exudate of TI 1068 leaf. (d) Glandular-headed trichomes with exudate of KY 14 burley leaf. Underside leaf surfaces ×65 are shown.

run A trapped headspace preparations from fully expanded leaves for each of the four tobacco lines were as follows: TI 1112, 38.5 ng/g; TI 1406, 25.0 ng/g; TI 1068, 16.6 ng/g; KY 14, 15.7 ng/g. Corresponding amounts in the 12-L run B preparations were as follows: TI 1112, 30.0 ng/g; TI 1406, 12.6 ng/g; TI 1068, 8.7 ng/g; KY 14, 6.6 ng/g. The purpose for using two different sized entrainment vessels for headspace analyses was twofold: (1) to demonstrate the effect of small partially controlled amounts of leaf wounding on the yields of total and individual headspace volatiles; (2) to determine the compositional nature of lipoxygenase-generated volatiles.

The mean yields of total volatiles among the four tobacco lines obtained with 5-L entrainment flasks were 1.3–2.4-fold higher than the corresponding yields obtained with 12-L flasks. One probable explanation for this increase is the increased injury of leaves that occurred when they were placed in the narrower opening of the smaller flask (45/50 ⅋) as compared to the larger flask (71/60 ⅋). Such injury of plant tissue as, for example, detachment of leaves, leaf tearing, fruit disruption, etc., is known to activate a group of enzymes including lipoxygenase and hydroperoxide lyase required for the generation of volatile compounds such as aldehydes, alcohols, and esters containing six and nine carbon atoms (Galliard and Phillips, 1976; Galliard et al., 1976; Hatanaka et al., 1978; Wu and Liou, 1986). A second and perhaps less tenable explanation for more total volatiles obtained with smaller flask size is that there was a greater linear air flow per unit area of leaf surface and less chance of binding to active sites on the glass in the smaller flask. Thus, more volatiles may have been entrained in the smaller flask even though a uniform flow rate was maintained in the Tenax traps connected to both flasks. Comparison of our observed yields of total volatile compounds in whole leaves of *Nicotiana tabacum* lines (6.6–38.5 ng/g) with reported yields of volatiles in leaves (and stems) of other species such as clover (100 ng/g) (Buttery et al., 1984) and wheat (10–50 ng/g) (Buttery et al., 1985) indicates that tobacco leaves grown under our conditions emitted gross quantities of volatiles similar to the other compared plant species.

The results indicate that the leaves from tobacco lines with nonsecreting and fewer trichomes per unit area (TI 1406) and with headless nonsecreting trichomes (TI 1112) emitted higher total yields of headspace volatiles than leaves from lines with glandular-headed secreting trichomes of higher density (TI 1068, KY 14). The data are consistent with emissions of large fractions of the total volatile compounds from the epidermal cells or stomata of the leaf surfaces, rather than from glandular secreting trichomes, per se.

Identification of Tobacco Leaf Headspace Volatiles. The identification of 10 volatile compounds from tobacco leaves estimated to comprise about 50% of the weight of total estimated volatiles emitted from the 4 tobacco lines are summarized in Table I. Each compound represented a single GC peak, and their sums accounted for about half the total number of GC peaks obtained for each tobacco

Table I. Headspace Volatiles of Vegetative Green Tobacco Leaf[a]

compound	Kovats' GLC index[b]	major MS (EI) ions[c]	MW	other evidence	class/probable precursor(s)
(*E*)-β-ocimene	1268	93, 80, 53, 105, 121, 67	136	MS (CI); cochromatography	monoterpenoid/geranyl pyrophosphate (Goodwin et al., 1983)
(*Z*)-3-hexenyl acetate	1337	43, 67, 82	142	MS (CI); cochromatography	aliphatic ester/lipoxygenase action on unsaturated fatty acids (Sekiya et al., 1984)
(*Z*)-3-hexen-1-ol	1401	57, 67, 82	100	MS (CI); cochromatography	aliphalic alcohol/lipoxygenase action on unsaturated fatty acids (Sekiya et al., 1984)
linalool	1560	93, 71, 55, 121	154	cochromatography	monoterpenoid/geranyl pyrophosphate (Goodwin et al., 1983)
β-caryophyllene	1633	41, 93, 133, 69, 79, 105	204	MS (CI); cochromatography	sesquiterpenoid/farnesyl pyrophosphate (Goodwin et al., 1983)
(*E*)-β-farnesene	1681	69, 55, 93, 120, 133, 79, 161	204	MS (CI); cochromatography	sesquiterpenoid/farnesyl pyrophosphate (Goodwin et al., 1983)
solanone	1756	93, 121, 136, 79, 107, 55	194	MS (CI); cochromatography	diterpenoid degradation product of thunberga-2,7,11-triene-4,6-diol (Demole and Dietrich, 1977)
methyl salicylate	1825	120, 92, 152, 39, 65, 53	152	MS (CI); cochromatography	phenylpropanoid derivative/shikimic acid pathway (Goodwin et al., 1983)
nicotine	1925	84, 133, 162, 42	162	MS (CI); cochromatography	alkaloid/nicotinic acid and ornithine (Enzell et al., 1977)
neophytadiene	1931	43, 55, 68, 82, 95, 123	278	MS (CI); cochromatography	diterpenoid/geranyl geranyl pyrophosphate (Goodwin et al., 1983)

[a] All identifications except for linalool were based on KY 14 leaves; linalool was identified from TI 1068 leaves. [b] Kovats' GLC index for the Supelcowax 10 capillary column. [c] One each 14 mass units listed in order of decreasing intensity.

Table II. Effect of Entrainment Flask Size on Yields of Tobacco Leaf Headspace Volatiles

compound	yield,[a] ng/10 g leaves	
	5-L flask	12-L flask
(*E*)-β-ocimene	18 a	21 a
(*Z*)-3-hexenyl acetate	14 a	0.9 b
(*Z*)-3-hexen-1-ol	122 a	25 b
linalool	2.3 a	4.3 a
β-caryophyllene	3.9 a	3.4 a
(*E*)-β-farnesene	11 a	7.9 a
solanone	3.9 a	3.5 a
methyl salicylate	5.2 a	14.7 a
neophytadiene	51 a	45 a

[a] Yields are expressed as means of results for three replicates each of four genetic lines, namely TI 1112, TI 1406, TI 1068, and KY 14. Values in each horizontal row not followed by the same letter are significantly different at $P = 0.05$.

Table III. Yields of Leaf Headspace Volatiles Emitted from Tobacco Lines with Different Trichome Morphologies

compound	yield,[a-d] ng compound/10 g leaf			
	TI 1112	TI 1406	TI 1068	KY 14
(*E*)-β-ocimene	80 a	6.0 b	<1.0 b	<1.0 b
(*Z*)-3-hexenyl acetate	1.6 a	2.2 a	<1.0 a	<1.0 a
(*Z*)-3-hexen-1-ol	26 a	20 a	23 a	30 a
linalool	<1.0 a	<1.0 a	17 b	<1.0 a
β-caryophyllene	<1.0 a	<1.0 a	14 b	<1.0 a
(*E*)-β-farnesene	3.2 a	<1.0 b	<1.0 b	<1.0 b
solanone	14 a	<1.0 b	<1.0 b	<1.0 b
methyl salicylate	30 a	29 ab	<1.0 b	<1.0 b
neophytadiene	61 ab	90 a	8.7 b	21 ab
total identifiables	216	147	63	51

[a] Values are analytical means of three replicate samples. [b] Values in each horizontal row not followed by the same letter are significantly different at $P = 0.05$. [c] Analyses carried out in a 12-L round-bottomed entrainment flask (run B). [d] Yields of nicotine were <1.0 ng/10 g in all cases.

line. All the identifications except for that of linalool were based on their presence in a headspace isolate of KY 14 tobacco leaves that was more concentrated (×4) than that used for volatile quantifications. Linalool was identified in a headspace isolate of TI 1068 leaves. Pure standards of the monoterpenoid (*E*)-β-ocimene and the sesquiterpenoid (*E*)-β-farnesene were not available. In lieu of pure standards, the identificiations of these terpenoids were aided by cochromatographic verification of the appropriate peaks in the red clover headspace isolate with those in tobacco headspace isolates. The Kovats' indices and mass spectra of these terpenoids in red clover were in good agreement with those reported by Buttery et al. (1984) and matched those found in the KY 14 leaf headspace isolate.

Quantitative Estimates of Individual Headspace Volatiles. The effect of leaf wounding during isolations of volatiles on yields of individual headspace components was demonstrated in Table II. Comparisons of amounts of a compound (means on a wet-weight basis) obtained from headspace isolates of TI 1112, TI 1406, TI 1068, and KY 14 obtained with a 5-L entrainment vessel (run A) with those of a 12-L vessel (run B) indicated that significant differences were obtained between runs A and B only for (*Z*)-3-hexenyl acetate and (*Z*)-3-hexen-1-ol, the only two components identified that are considered to be products of lipoxygenase-lyase activity. The elevated yields of these two known products of lipoxygenase-lyase activity in run A compared to run B account for part of the higher yields of total volatile compounds in run A.

Amounts of individual components of the headspace volatiles from leaves of each tobacco line obtained during analyses performed with the 12-L entrainment flask are given in Table III. The results obtained do not support the view that the exudate-secreting glandular heads of leaf trichomes emit the majority of headspace volatiles. However, there were some significant differences in yields of some compounds among the genetic lines of tobacco. It is interesting to note that leaves of the TI 1112 and TI 1406 lines with headless and nonsecreting glandular-headed trichomes, respectively, emitted greater quantities of identifiable volatiles than leaves of the TI 1068 and KY 14 cultivars that had secreting glandular-headed trichomes.

Quantities of the monoterpenoid (*E*)-β-ocimene were larger from the tobacco line with headless nonsecreting trichomes (TI 1112) than they were from tobacco lines with nonsecreting glandular headed trichomes (TI 1406) and "normal" glandular headed secreting trichomes (TI 1068, KY 14). The presence of one hydrocarbon monoterpenoid isomeric to (*E*)-β-ocimene was reported in cured tobacco (Enzell et al., 1984).

In contrast to the monoterpenoid (*E*)-β-ocimene, significant quantities of the sesquiterpenoid β-caryophyllene accumulated only in the leaf headspace of the "normal" glandular-headed trichome tobacco line, namely TI 1068. β-Caryophyllene was reported present in tobacco (Enzell et al., 1984) and in the headspace of green tobacco stalk (Andersen et al., 1986).

The acyclic diterpenoid neophytadiene was generally the most abundant compound in the leaf headspace of the tobacco lines. The compound has been found in most tobacco types (Enzell et al., 1984; Burton and Kasperbauer, 1985) and was present in the headspace of green tobacco stalk (Andersen et al., 1986). However, there was significantly less neophytadiene from TI 1068 leaves than from TI 1406 leaves.

Solanone was present in the headspace of TI 1112 leaf but was not detectable in the headspace of green leaves of the other tobacco lines at the concentrations used for quantifications. Solanone was previously reported in the steam distillate of green tobacco stalk (Andersen et al., 1986) and in the essence and essential oil of flue-cured tobacco (Lloyd et al., 1976).

Methyl salicylate was quantified in the headspace of TI 1112 and TI 1406 tobacco lines, but it was not detectable at concentrations used for quantifications in TI 1068 or KY 14 headspaces. The presence of this compound was recently reported in the headspace of green tobacco stalk (Andersen et al., 1986).

Quantities of (*Z*)-3-hexenyl acetate were present at detectable concentrations in the headspace of TI 1112 and TI 1406 leaves. (*Z*)-3-Hexen-1-ol commonly referred to as leaf alcohol was the second most abundant volatile compound, and it was emitted by all of the tobacco leaf samples. (*Z*)-3-Hexenyl acetate and (*Z*)-3-hexen-1-ol were not detected in the headspace of green tobacco stalk (Andersen et al., 1986). Lipoxygenase-lyase-mediated products in the headspace of plant leaves such as tobacco can be regarded as inducible, naturally occurring, volatile compounds that may have a physiological or defense role that has not been elucidated. The possible role of tobacco volatiles and trichomes in resistance to insects has been reviewed (Stipanovic, 1983).

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Registry No. (*Z*)-AcO$(CH_2)_2$CH=CHEt, 3681-71-8; (*Z*)-HO$(CH_2)_2$CH=CHEt, 928-96-1; *o*-$HOC_6H_4CO_2Me$, 119-36-8; (*E*)-β-ocimene, 3779-61-1; linalool, 78-70-6; β-caryophyllene, 87-44-5; (*E*)-β-farnesene, 18794-84-8; solanone, 1937-54-8; nicotine, 54-11-5; neophytadiene, 504-96-1.

LITERATURE CITED

Andersen, R. A.; Lowe, R.; Vaughn, T. H. "Plant Phenols and Polyphenoloxidase in *Nicotiana tabacum* during Greenhouse Growth, Field Growth and Air Curing". *Phytochemistry* **1969**, *8*, 2139–2147.

Andersen, R. A.; Hamilton-Kemp, T. R.; Fleming, P. D.; Hildebrand, D. F. *Biogeneration of Aromas*; Parliment, T. H., Croteau, R., Eds.; ACS Symposium Series 317; American Chemical Society: Washington, DC, 1986; pp 99–111.

Burton, H. R.; Kasperbauer, M. J. "Changes in Chemical Composition of Tobacco Lamina during Senescence and Air Curing. 1. Plastid Pigments". *J. Agric. Food Chem.* **1985**, *33*, 879–883.

Buttery, R. G.; Kamm, J. A.; Ling, L. C. "Volatile Components of Red Clover Leaves, Flowers, and Seed Pods: Possible Insect Attractants". *J. Agric. Food Chem.* **1984**, *32*, 254–256.

Buttery, R. G.; Xu, C.; Ling, L. C. "Volatile Components of Wheat Leaves (and Stems): Possible Insect Attractants". *J. Agric. Food Chem.* **1985**, *33*, 115–117.

Demole, E.; Dietrich, P. *Recent Advances in the Chemical Composition of Tobacco and Tobacco Smoke*; McKenzie, J. L., Ed.; Proceedings of the American Chemical Society Symposium; Pierce Printing: Ashokie, NC, 1977; pp 3–20.

Enzell, C. R.; Wahlberg, I. "Leaf Composition in Relation to Smoking Quality and Aroma". *Rec. Adv. Tobacco Sci.* **1980**, *6*, 64–122.

Enzell, C. R.; Wahlberg, I.; Aasen, A. J. *Progress in the Chemistry of Organic Natural Products*; Herz, W., Griseback, H., Kirby, G. W., Eds.; Springer-Verlag: New York, 1977; Vol. 34, p 48.

Enzell, C. R.; Wahlberg, I.; Rybage, R. "Mass Spectra of Tobacco Isoprenoids". *Mass Spectrom. Rev.* **1984**, *3*, 395–438.

Galliard, T.; Phillips, D. R. "The Enzymatic Cleavage of Linoleic Acid to C_9 Carbonyl Fragments in Extracts of Cucumber (*Cucumis sativus*) Fruit and the Possible Role of Lipoxygenase". *Biochim. Biophys. Acta* **1976**, *431*, 278–287.

Galliard, T.; Phillips, D. R.; Reynolds J. "The Formation of *cis*-3-Nonenal, *trans*-2-Nonenal and Hexanal from Linoleic Acid Hydroperoxide Isomers by a Hydroperoxide Cleavage Enzyme System in Cucumber (*Cucumis sativus*) Fruits". *Biochim. Biophys. Acta* **1976**, *441*, 181–192.

Goodwin, T. W.; Mercer, E. I. *Introduction to Plant Biochemistry*, 3rd ed.; Pergamon: New York, 1983.

Hatanaka, A.; Sekiya, J.; Kajiwara, T. "Distribution of an Enzyme System Producing *cis*-3-Hexanal and Hexanal from Linolenic and Linoleic Acids in Some Plants". *Phytochemistry* **1978**, *17*, 869–872.

Henderson, W.; Hart, J. W.; How, P.; Judge, J. "Chemical and Morphological Studies on Sites of Sesquiterpene Accumulation in *Pogostemon cablin* (Patchouli)". *Phytochemistry* **1970**, *9*, 1219–1228.

Lloyd, R. A.; Miller, C. W.; Roberts, D. L.; Giles, G. A.; Dickerson, J. P.; Nelson, N. H.; Rix, C. E.; Ayers, P. H. "Flue-Cured Tobacco Flavor. I. Essence and Essential Oil Components". *Tobacco Sci.* **1976**, *20*, 40–48.

Loomis, W. D.; Croteau, R. In *The Biochemistry of Plants; Lipids*; Stumpf, P. K., Ed.; Academic: New York, 1980; Vol. 4, p 363.

Nielsen, M. T.; Jones, G. A.; Collins, G. B. "Inheritance Pattern for Secreting and Nonsecreting Glandular Trichomes in Tobacco". *Crop Sci.* **1982**, *22*, 1051–1053.

Perry, J. A. *Introduction to Analytical Gas Chromatography*; Marcel Dekker: New York, 1981; p 246.

Rix, C. E.; Lloyd, R. A.; Miller, C. W. "Headspace Analysis of Tobacco with Tenax Traps". *Tobacco Sci.* **1977**, *21*, 93–96.

Sakaki, T.; Fukuhara, K.; Niino, K.; Sakuma, H.; Sugawara, S. "Changes in the Composition of Headspace Volatiles of Flue-Cured Tobacco by Aging". *Agric. Biol. Chem.* **1985**, *49*, 1785–1791.

Severson, R. F.; Johnson, A. W.; Jackson, D. M. "Cuticular Constituents of Tobacco: Factors Affecting Their Production and Their Role in Insect and Disease Resistance and Smoke Quality". *Rec. Adv. Tobacco Sci.* **1985**, *11*, 105–174.

Stipanovic, R. D. *Plant Resistance to Insects*; Hedin, P. A., Ed.; ACS Symposium Series 208; American Chemical Society: Washington, DC, 1983; p 69.

Weeks, W. W.; Seltmann, H. J. "Effect of Sucker Control on the Volatile Compounds in Flue-Cured Tobacco". *J. Agric. Food Chem.* **1986**, *34*, 899–904.

Wu, C.-M.; Liou, S.-E. "Effect of Tissue Disruption on Volatile Constituents of Bell Peppers". *J. Agric. Food Chem.* **1986**, *34*, 770–772.

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