Bagley, R. W., Bauernfeind, J. C., in "Insect Juvenile Hormones", Menn, J. J., Beroza, M., Ed., Academic Press, New York, N.Y., 1972, p 113.
Bowers, W. S., *Science* **164**, 323 (1969).
Bowers, W. S., *Bull. Soc. Entomol. Suisse* **44**, 115 (1971).
Dorn, S., Oesterhelt, G., Suchý, M., Trautmann, K. H., Wipf, H.-K., submitted to *J. Agric. Food Chem.* for publication (1976).
Frischknecht, M., manuscript in preparation (1975).
Gerig, L., Federal Research Station, Liebefeld Bern, personal communication, 1975.
Gill, S. S., Hammock, B. D., Casida, J. E., *J. Agric. Food Chem.* **22**, 386 (1974).
Hammock, B. D., Gill, S. S., Casida, J. E., *J. Agric. Food Chem.* **22**, 379 (1974).
Hangartner, W., Peyer, B., Meier, W., *Meded. Fac. Landbouurvet. Rijksuniv. Gent* **36**, 866 (1971).
Harrison, R. B., Holmes, D. C., Roburn, J., Tatton, J. O'G., *J. Sci. Food Agric.* **18**, 10 (1967).
Henrick, C. A., Staal, G. B., Siddall, J. B., *J. Agric. Food Chem.* **21**, 354 (1973).
Hindley, N. C., Andrews, D. A. (to F. Hoffmann-La Roche Co.), Belgian Patent 806775 (1974).
Hoffmann, L. J., Ross, J. H., Menn, J. J., *J. Agric. Food Chem.* **21**, 156 (1973).
Hoffmann, W., Pasedach, H., Pommer, H., *Justus Liebigs Ann. Chem.* **729**, 52 (1969).
Hoppe, T., Suchý, M., *Bull. OEPP* **5**, 193 (1975).
Hoppe, T., personal communication, 1975.
Meier, W., Kolar, O., Ramses, E., *Mitt. Schweiz. Landwirtsch.* **23**, 1 (1975).
Menn, J. J., Beroza, M., Ed., "Insect Juvenile Hormones", Academic Press, New York, N.Y., 1972.
Mori, K., *Bull. Soc. Entomol. Suisse* **44**, 17 (1971).
Pallos, F. M., Menn, J. J., Letchworth, P. E., Miaullis, J. B., *Nature (London)* **232**, 486 (1971).
Pauling, H. (to F. Hoffmann-La Roche Co.), Belgian Patent 783055 (1972).
Pfiffner, A., in "Aspects of Terpenoid Chemistry and Biochemistry", Goodwin, T. W., Ed., Academic Press, London, 1971, p 95.
Quistad, G. B., Staiger, L. E., Schooley, D. A., *J. Agric. Food Chem.* **23**, 299 (1975).
Sarmiento, R., Mc Govern, T. P., Beroza, M., Mills, G. D., Redfern, R. E., *Science* **179**, 1342 (1973).
Scheurer, R., Ruzette, M. A., *Z. Angew. Entomol.* **77**, 218 (1974).
Schooley, D. A., Bergot, B. J., Dunham, L. L., Siddall, J. B., *J. Agric. Food Chem.* **23**, 293 (1975).
Sláma, K., Romaňuk, M., Šorm, F., Ed., "Insect Hormones and Bioanalogues", Springer-Verlag, Wien, New York, N.Y., 1974.
Staal, G. B., *Annu. Rev. Entomol.* **20**, 417 (1975).
Suchý, M., Sláma, K., Šorm, F., *Science* **162**, 582 (1968).
Vogel, W., Masner, P., Frischknecht, M., manuscript in preparation (1975).
WHO, personal communication; monthly report of the Filariasis Research Unit, Rangoon, Burma, Feb 11, 1964.
Wright, J. E., Mc Govern, T. P., Sarmiento, R., Beroza, M., *J. Insect Physiol.* **20**, 423 (1974).

Received for review April 10, 1975. Accepted September 18, 1975.

Volatile Components of Pecan Leaves and Nuts, *Carya illinoensis* Koch

Naresh V. Mody, Paul A. Hedin,* and William W. Neel

The volatile constituents of leaves and immature pecan nuts (including the shucks), *Carya illinoensis* Koch (Juglandaceae), were analyzed by GLC–MS and found to contain 38 compounds including 7 monoterpene hydrocarbons, 7 sesquiterpene hydrocarbons, 11 terpene alcohols, 1 terpene aldehyde, 1 terpene ketone, and 11 other aldehydes, ketones, alcohols, and esters. These studies were initiated to identify those constituents that could conceivably attract the pecan weevil, *Curculio caryae* Horn, to the leaves and nuts, their primary food source. The constituents could also be precursors of the pecan weevil sex pheromone.

Pecans, *Carya illinoensis* Koch (Juglandaceae), are commercially grown in a "Belt" across the southern United States that includes North Carolina, South Carolina, Georgia, Florida, Alabama, Mississippi, Louisiana, Arkansas, Oklahoma, Texas, New Mexico, and Arizona. The pecan trees are attacked by several insects including the pecan weevil, *Curculio caryae* Horn. Indeed, in one central Mississippi orchard (Neel, 1970), 30–40% of the pecans were infested. Likewise, the average weevil population per tree in an Oklahoma orchard in 1968 and 1969 was estimated to be 1962 and 6130, respectively (Raney et al., 1970).

The work reported herein was a survey of the volatile components from pecan leaves and nuts made in anticipation of a further effort to determine any role in plant attraction and nutrition of the pecan weevil. The preliminary work on the pecan weevil sex attractant was reported recently by Mody et al. (1973).

A search of the literature revealed that mature pecan leaves and nuts have been analyzed for various constituents and properties including sterols and choline, carbohydrates, fatty acids in the oil, trace elements, growth regulators, amino acids and proteins, minerals, and vitamins. However, the only study of volatile constituents was made with roasted pecans by Wang and Odell (1972). They found 19 carbonyl compounds, pyridine, 8 pyrazines, 7 acids, 5 alcohols, and 1 lactone. The presence of most of these components is due to roasting, because except for 2-furfuraldehyde they are not found in unroasted pecans.

EXPERIMENTAL SECTION

Preparation of Essential Oil. Fresh, mature pecan leaves of the cultivar Stuart were gathered in July. Green nuts with shucks (involucre) and shells that had not hardened and with kernels that were in the water stage were gathered in August. Both the leaves and the nuts

Boll Weevil Research Laboratory, Agricultural Research Service, U.S. Department of Agriculture, Mississippi State, Mississippi 39762 (N.V.M., P.A.H.) and the Department of Entomology, Mississippi State University, Mississippi State, Mississippi 39762 (W.W.N.).

Table I. Volatile Compounds in Pecan Leaves and Nuts

Leaves	Nuts	I_k OV-17[a]	Mol wt.	Compd	Ref or MS fragmentation[b]
+	0	1215	96	2-Furaldehyde[c]	Stenhagen et al. (1969)
+	0	940	98	$C_6H_{10}O$	55, 43, 83, 41, 98
+	0	910	98	C_7H_{14}	55, 83, 41, 43, 98
+	0	1105	100	(*E*)-3-Hexen-1-ol[c]	Stenhagen et al. (1969)
+	0	1110	100	(*Z*)-3-Hexen-1-ol[c]	Stenhagen et al. (1969)
+	0	1185	106	Benzaldehyde[c]	Stenhagen et al. (1969)
+	0	1305	108	Benzyl alcohol[c]	Stenhagen et al. (1969)
+	0	1225	120	Acetophenone[c]	Stenhagen et al. (1969)
+	0	1300	120	Phenylacetaldehyde[c]	Stenhagen et al. (1969)
0	+	1135	134	*p*-Cymene[c]	Ryhage and von Sydow (1963)
0	+	1100	136	α-Phellandrene	Ryhage and von Sydow (1963)
0	+	1110	136	α-Pinene[c]	Ryhage and von Sydow (1963)
0	+	1115	136	Camphene	Ryhage and von Sydow (1963)
0	+	1130	136	Sabinene	Ryhage and von Sydow (1963)
0	+	1140	136	α-Terpinene	Ryhage and von Sydow (1963)
+	+	1150	136	Limonene[c]	Ryhage and von Sydow (1963)
+	0	1390	152	1-*p*-Menthen-9-al	Stenhagen et al. (1969)
+	0	1380	152	Methyl salicylate[c]	Stenhagen et al. (1969)
+	0	1270	154	Linalool[c]	von Sydow (1963)
+	+	1325	154	Borneol	von Sydow (1963)
+	+	1335	154	α-Terpineol[c]	von Sydow (1963)
0	+	1170	154	1,8-Cineole[c]	Stenhagen et al. (1969)
+	0	1210	154	$C_{10}H_{18}O$ alcohol	43, 71, 55, 93, 121
+	0	1320	154	Menthone	von Sydow (1963)
+	+	1330	154	Terpin-4-ol[c]	von Sydow (1963)
+	0	1530	154	$C_{10}H_{18}O$ alcohol	57, 95, 93, 69, 67
+	0	1325	156	Menthol[c]	von Sydow (1963)
+	0	1405	166	Ethyl salicylate[c]	Stenhagen et al. (1969)
+	+	1470	204	$C_{15}H_{24}$	81, 80, 123, 55, 161
+	+	1495	204	α-Santalene	Hirose (1967)
+	+	1540	204	β-Caryophyllene[c]	Stenhagen et al. (1969)
+	+	1630	204	Humulene[c]	Stenhagen et al. (1969)
+	+	1665	204	β_2-Bisabolene	Hirose (1967)
+	+	1680	204	α-Cubebene	Hirose (1967)
+	+	1750	204	α-Ferulene	Hirose (1967)
+	+	1845	220	$C_{15}H_{24}O$ alcohol	69, 55, 93, 71, 107
+	+	1915	220	$C_{15}H_{24}O$ alcohol	67, 82, 105, 123, 55
+	+	1965	222	Cubenol	Hirose (1967)

[a] Kováts indices (1961). [b] Five most abundant ion values are inferred. [c] Compared with an authentic sample.

were gathered from an orchard near Columbus, Miss. All samples were stored at −10°C until the tissue was chopped, ground, and steam distilled for isolation of the essential oil. The distillate was extracted with redistilled CH_2Cl_2. The CH_2Cl_2 was dried with anhydrous Na_2SO_4, and the solvent was removed in vacuo at 35°C to give an essential oil.

Column Chromatography. To obtain fractions that could be more conveniently analyzed by GLC–MS, the essential oil was chromatographed on a 2 × 25 cm jacketed Florisil column that was cooled with ice to prevent column cracking. The column was eluted with 200-ml portions of pentane, 10% Et_2O in pentane, 25% Et_2O in pentane, and 100% Et_2O. Progress of the elution and recombination of all fractions into four reconstructed fractions were monitored by TLC. The developed TLC plate was visualized by spraying with 3% vanillin in 0.5% concentrated H_2SO_4 in MeOH and then heated to 100°C.

Analytical GLC-MS. The four fractions obtained by preparative TLC as described above were introduced separately into a Hewlett Packard 5930 Quadrupole mass spectrometer interfaced with a 250 ft × 0.03 in. stainless steel capillary gas chromatographic column coated with OV-17. Carrier gas flow was 8.0 ml/min of He. The GLC unit was programmed from 80 to 180°C at 2°C/min. The final temperature was maintained for 20 min. Mass spectra were obtained at 70 eV. Fragment ion values were compared with those of Stenhagen et al. (1969), von Sydow (1963, 1964), and Hirose (1967). GLC retention times are presented as Kováts indices (I_k) (1961). Comparisons were made with authentic samples when available.

RESULTS AND DISCUSSION

Structures for 31 components and elemental formulas for 7 others were proposed as present in leaves and/or nuts (Table I). Nuts contained 7 monoterpene hydrocarbons, but leaves contained only limonene. Leaves contained at least 9 oxygenated monoterpenoids; nuts contained only 4. Leaves contained 2-furaldehyde, (*E*)- and (*Z*)-3-hexen-1-ol, benzaldehyde, benzyl alcohol, acetophenone, phenylacetaldehyde, and both methyl and ethyl salicylate, but none of these were found in nuts. Both the leaves and nuts contained 7 sesquiterpene hydrocarbons and 3 sesquiterpene alcohols. Because the relative concentrations of the constituents varied widely between samples, the percentage contents of each are not included. However, the terpenoids accounted for approximately 75% of the total oil that could be analyzed by GLC, and no individual component comprised more than 10%. Of all the constituents found in green leaves and nuts, only 2-furaldehyde was also found in roasted pecans (Wang and Odell, 1972). Since roasting would not be expected to remove all the terpenoids by volatilization or degrade them by temperature effects, the terpenoids must be present chiefly in the green shucks and shells. By comparison, roasted pecans contained mostly pyrazines and carbonyl compounds.

Since nearly all sex pheromones identified in insects of the order Coleoptera, which includes the pecan weevil, are terpenes, it is interesting that the food sources of pecan weevil larvae and adults do contain terpenoids. However, there is not specific information as yet that has been gathered from studies with other Coleoptera that a closely

related precursor is utilized for pheromone biosynthesis. In fact, studies of the male boll weevil, *Anthonomus grandis* Boheman, show that ^{14}C is readily incorporated into all four of the pheromones from common metabolic precursors such as mevalonate, glucose, and acetate (Mitlin and Hedin, 1974). Nevertheless, pecan weevils may either require terpenoids or be attracted to them.

LITERATURE CITED

Hirose, Y., *Shitsuryo Bunseki* **15**, 162 (1967).
Kovats, E., *Anal. Chem.* **181**, 351 (1961).
Mitlin, N., Hedin, P. A., *J. Insect Physiol.* **20**, 1825 (1974).
Mody, N. V., Miles, D. H., Neel, W. W., Hedin, P. A., Thompson, A. C., Gueldner, R. C., *J. Insect Physiol.* **19**, 2063 (1973).
Neel, W. W., *Miss. Farm Res.* **33**, 3 (1970).
Raney, H. G., Eikenbary, R. D., Flora, N. W., *J. Econ. Entomol.* **63**, 697 (1970).
Ryhage, R., von Sydow, E., *Acta Chem. Scand.* **17**, 2025 (1963).
Stenhagen, E., Abrahamsson, S., McLafferty, F. W., Ed., "Atlas of Mass Spectral Data", Vol. 1, 2, and 3, Interscience, New York, N.Y., 1969.
von Sydow, E., *Acta Chem. Scand.* **17**, 2504 (1963).
von Sydow, E., *Acta Chem. Scand.* **18**, 1099 (1964).
Wang, P. S., Odell, G. V., *J. Agric. Food Chem.* **20**, 206 (1972).

Received for review June 30, 1975. Accepted September 17, 1975. This work was partially financed by the Environmental Protection Agency through Grant No. R-802659-02-0. Mention of a proprietary product in this paper does not constitute endorsement of the product by the U.S. Department of Agriculture.

Pyrolysis of Chitin, a Potential Tobacco Extender

William S. Schlotzhauer, Orestes T. Chortyk,* and Paul R. Austin[1]

Chitin and tobacco were pyrolyzed separately and in admixture under identical conditions. The pyrolyzates were fractionated and analyzed for neutrals, bases, phenols, and carboxylic acids. Except for acidic products, chitin produces much smaller quantities of pyrolytic products than tobacco and in different ratios. Chitosan was also pyrolyzed to determine the effect of deacetylation on the carboxylic acid profile of the chitin pyrolyzate. It was concluded that both chitin and chitosan could become suitable tobacco extenders.

Current increases in tobacco leaf prices coupled with the smoking and health controversy have led numerous investigators and tobacco companies in search of tobacco extenders or "synthetic tobaccos". These extenders are to be mixed into cigarettes, replacing some tobacco and reducing the harmful tar and nicotine contents. Difficulties encountered in developing acceptable modified smoking materials have included unsatisfactory rate and continuity of burn, smoke temperature, ash retention, and more subtle problems of aroma, flavor, and biological response. Natural cellulose or modified cellulose products have been of recent interest as tobacco fillers or substitutes, accounting for 10–50% of cigarette composition ("Tobacco Situation", 1973; *Tab.-J. Int.*, 1974; *World Tob.*, 1974; *Tob. Reporter*, 1975). In addition to the substitute materials currently in use, two natural carbohydrate polymers, chitin and chitosan, are also of interest as potential tobacco extenders ("Pacific Northwest Sea", 1973; Pariser and Bock, 1972).

Chitin, a mucopolysaccharide, poly(*N*-acetyl-D-glucosamine) $[(C_8H_{12}O_5N)_n]$, occurs widely in nature, for example, in the cell walls of fungi and shells of insects and crustaceans. Chitin is recovered from "crab meal" after alternate hydrolyses with mild alkali and acid which remove proteineous and calcereous constituents. Chitosan (2-deoxy-2-aminoglucose polymer), a deacetylated chitin, is normally prepared by strong alkaline hydrolysis of chitin. The product is then subsequently neutralized and isolated ("Pacific Northwest Sea", 1973; Tracey, 1957). Deacetylated chitin has been utilized as an adhesive for tobacco particles in reconstituted tobacco sheet formulation (Moshy and Germino, 1966). A low tar yield in smoke of chitin led to its trial as a tobacco extender (Austin, 1975) and prompted this study of chitin pyrolysis.

In the initial trials, it was determined that chitin and chitosan could be added in substantial amounts to tobacco blends without altering significantly such physical properties as packing ability, burning rate, and ash retention, or grossly affecting organoleptic attributes such as aroma, taste, or mildness. Since identification of the major pyrolytic products of chitin would provide additional evaluation of this material as a tobacco extender, chitin and tobacco were pyrolyzed separately and in admixture under identical conditions; the pyrolyzates were analyzed for gas–liquid chromatographically volatile, ether-soluble neutrals, bases, phenols, and carboxylic acids. Chitosan was also pyrolyzed to determine the effect of deacetylation on the carboxylic acid profile of the chitin pyrolyzate.

EXPERIMENTAL SECTION

Preparation of Materials for Analysis. Chitin and chitosan, obtained from the Food, Chemical, and Research Laboratories (Seattle, Wash.), were used without further chemical purification. The tobacco was North Carolina, flue-cured, whole leaf (provided by the Tobacco Research Laboratory, Agricultural Research Service, USDA, Oxford, N.C.). It was ground to 32 mesh in a ball mill to promote homogeneity upon admixture with chitin. Prior to pyrolysis, all samples were equilibrated over 76% glycerol in water for 48 hr to standardize moisture at 12.0% in

Tobacco Laboratory, Agricultural Research Service, U.S. Department of Agriculture, Athens, Georgia 30604.
[1]Present address: College of Marine Studies, University of Delaware, Newark, Delaware.