of dielectric distribution. A voltage of 3,000 v was applied. The spore cells were preferentially witheld in the column between the two plates while the vegetative cells and debris passed through the electric field. The material which was retained was refractionated under the same conditions. The particles retained between the plates the second time were 90% spores.

The particles fell freely in the solvent until they reached the upper edge of the condenser. Then there was a tendency for the particles to be attracted to the ungrounded side of the condenser. There was a strong tendency for the particles to form chains across the tube in the direction of the field. Chain formation is to be expected (1), but we have no explanation for the tendency of the particles to move to the ungrounded side of the apparatus. There was no evidence that the particles accumulated net charges and repelled each other as Pohl and Schwar (7) postulated.

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Dihydroxy Fatty Acids in *Cardamine impatiens* Seed Oil[1]

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Abstract

The oil of *Cardamine impatiens* L. (Cruciferae) seed includes glycerides of a series of saturated long-chain vicinal dihydroxy fatty acids, which make up 25% by weight of the mixed fatty acids. The mixture of diols, after transesterification of the oil with methanol, can be crystallized from an ether solution of the mixed methyl esters and has the following composition: methyl 13,14-dihydroxydocosanoate, 66%; methyl 15,16-dihydroxytetracosanoate, 24%; methyl 9,10-dihydroxyoctadecanoate and methyl 11,12-dihydroxyeicosanoate, about 5% each. Chemical proof is presented showing that essentially all the diols have the *erythro* configuration.

Introduction

NUMEROUS PLANT SOURCES have been shown to contain various monohydroxy acids in substantial quantities. Among these are seeds oils from the genus *Lesquerella,* family Cruciferae, which contain large amounts of either 12-hydroxy-*cis*-9,*cis*-15-octadecadienoic acid (12) or 14-hydroxy-*cis*-11-eicosenic acid (13).

The presence of 25% of saturated long-chain vicinal dihydroxy acids in the seed oil of *Cardamine impatiens,* a crucifer, is reported here. This seed oil is the first known that contains a large amount of vicinal dihydroxy acids. Castor oil (4,14), *Strophanthus* seed oil (5) and *Cephalocroton* seed oil (3) have been reported to contain vicinal dihydroxy acids in amounts of 4% or less. Some other examples of fatty acids with vicinal hydroxyl groups have been found in extracts of plant parts other than seeds (6,7), in wheat stem rust uredospores (15) and in *Lycopodium clavatum* L. spores (10).

The unusual infrared (IR) band at 8.1 μ in the spectrum of *Cardamine* oil has not been assigned to any structural feature. We feel that it is due to a deviation from the usual pattern of glyceride structure found in seed oils. This possibility is being investigated, and the results will be reported later.

Experimental

Gas-Liquid Chromatographic Conditions

Analyses by gas-liquid chromatography (GLC) were done on either a nonpolar Apiezon L column or a polar LAC-2-R 446 column, or both. The operating parameters of the polar column were the same as described previously (8). The nonpolar column (275 × 0.6 cm) was packed with 10% Apiezon L on 60–80 mesh Celite and was operated under the following conditions: Injection port, 287C; column bath, 257C; detector bath, 268C; detector current, 200 ma; fraction collector, 277C; helium flow rate, 102 ml/min at an inlet pressure of 40 psi. Identification of components was based on their equivalent chain lengths (9) as compared to equivalent chain lengths of similar, known materials. All percentages reported are area percent.

Oil Preparation and Analysis

Oil was obtained from ground seeds (36.0 g) of *Cardamine impatiens* L. by Soxhlet extraction with petroleum ether (bp 30–60C). The solvent was removed *in vacuo* at ca. 40C, yielding 11.9 g or 33.2% of oil. The ultraviolet (UV) spectrum showed a maximum at 230 mμ, $E^{1\%}_{1cm} = 14$ (in absolute ethanol) and weaker maxima at 270 and 276 mμ. IR analysis of the oil as a film on sodium chloride plates showed hydroxyl absorption at 2.88 μ and a rather strong unidentified band at 8.1 μ. Analysis of *Cardamine* oil by thin-layer chromatography (TLC) on silica gel G plates, with petroleum ether-ethyl ether (70–30) as the developing solvent, showed a large unknown spot with an R_f intermediate between those of the spots obtained for triglycerides containing one and two hydroxyl groups per molecule.

Preparation of Methyl Esters and Crystallization of Diols

A 1.958 g sample of *Cardamine* oil was refluxed 3 hr under nitrogen, with 75 ml of 1% hydrochloric acid in methanol; the solution was diluted with water; and the esters were recovered by ether extraction; yield, 1.876 g. The mixed methyl esters were dissolved in 10 ml of ethyl ether and crystallized overnight at −18C. The crystals (0.459 g) were filtered off, washed with cold ether and dried. A second crystallization yielded only 0.005 g of material. The liquor remaining from the crystallization was concentrated *in vacuo* and GLC gave the following composition: $C_{16:0}$, 4.4%; $C_{18:0}$, 0.4%; $C_{18:1}$, 14.6%; $C_{18:2}$, 26.6%; $C_{18:3}$, 4.1%; $C_{20:0}$, 0.8%; $C_{20:1}$, 7.0%; $C_{20:2}$,

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0.8%; $C_{22:0}$, 1.3%; $C_{22:1}$, 33.4%; $C_{22:2}$, 0.7%; $C_{24:0}$, 0.3%; and $C_{24:1}$, 5.6%. Analysis by IR and TLC showed that no hydroxyl was present.

Analysis of Diol Methyl Esters

The crystalline product had no measurable optical activity (5% in $CHCl_3$, 1 dm tube, sodium D-line), and it melted at 107–108C without recrystallization. The IR spectrum of the crystallized methyl esters (2.3% in chloroform, 1 mm sodium chloride cells) was nearly identical to that obtained for methyl-*threo*-9,10-dihydroxystearate. TLC indicated that only a trace of normal methyl esters was present.

The crystallized product consumed no hydrogen at 25C and 1 atm over platinum oxide catalyst.

An isopropylidene derivative was formed by warming the crystals briefly in acetone containing a trace of mineral acid. This was shown by the disappearance of IR hydroxyl absorption and the appearance of new bands near 7.3 and 9.0 μ.

Cleavage of Diol Methyl Esters

A 0.026-g sample of the crystallized methyl esters was oxidized with permanganate-periodate essentially as described by von Rudloff (11). The reaction mixture was reduced with bisulfite, made strongly alkaline and evaporated to dryness on a rotary evaporator. After the solid residue was cooled in ice and acidified with concenterated hydrochloric acid, the small volume of saturated salt solution obtained was extracted 10 times with 20-ml portions of ether. The extract was washed twice with small volumes of saturated sodium chloride solution and dried over anhydrous sodium sulfate. The ether solution was concentrated at much below room temperature *in vacuo* to ca. 0.1 ml. Analysis of the concentrate by GLC showed that nonanoic acid was the only monobasic acid present in the oxidation products. The remainder of the solvent was removed, the residue was esterified with 1% sulfuric acid in methanol and the recovered esters were analyzed by GLC. The following percentages of dibasic components were obtained: Nonanedioate, 5.1%; undecanedioate, 4.4%; tridecanedioate, 66.0%; and pentadecanedioate, 24.5%.

Determination of Diol Configuration

A 0.238-g sample of the dihydroxy acids (obtained by saponification of the crystallized methyl esters) was treated with hydrogen bromide in glacial acetic acid according to the method of Ames and Bowman (2). Twelve milliliters of ca. 25% hydrogen bromide in acetic acid was added to the sample. Concentrated sulfuric acid (1.2 ml) was then added dropwise with cooling and swirling. After the mixture was allowed to remain at room temperature with occasional swirling for 19.5 hr, it was heated in an oil bath at 95–100C for 6.5 hr. An additional 2 ml of the hydrogen bromide/acetic acid solution was added after 3 hr of heating. The reaction mixture was cooled, diluted with water while being swirled and extracted with hexane until the extract was colorless. The extract was washed to neutrality with water, and the solvent was removed on a rotary evaporator. The recovered products were converted to ethyl esters by refluxing with 75 ml of 2% sulfuric acid in absolute ethanol; yield, 0.376 g. IR analysis showed no hydroxyl absorption remained.

These ethyl esters were debrominated as described by Ames and Bowman (2). Granulated zinc (3 g, 30 mesh) was boiled 5 min with 13 ml of absolute ethanol containing 0.3 ml of 50% aqueous hydrogen bromide. The brominated esters (dissolved in 2 ml of ethanol) were added to the mixture, which was then refluxed for 1 hr. The zinc was filtered out of the cooled reaction mixture and washed well with ethyl ether. These washings were combined with the original filtrate. The resulting solution was washed with dilute sulfuric acid and finally with water until neutral. Solvent was removed to yield 0.224 g of product.

Quantitative IR analysis indicated only 5% isolated *trans* unsaturation (1) was present. Analysis of the monoenoic ethyl esters by GLC gave the following composition: $C_{16:1}$, 0.5%; $C_{18:1}$, 2.5%; $C_{20:1}$, 5.5%; $C_{22:1}$, 64.0%; $C_{24:1}$, 25.6%; and an unknown, 1.9%.

Similar treatment of a pure sample of *threo*-9,10-dihydroxystearic acid yielded a product containing 90% isolated *trans* unsaturation.

Results and Discussion

The seed oil of *Cardamine impatiens* was a rather viscous light-colored oil which remained fluid at −18C. The IR spectrum showed hydroxyl absorption and an unusual band at 8.1 μ. UV analysis indicated there was no *trans* and no conjugated unsaturation.

Methyl esters obtained by transesterification of the oil were a mixture of solid and liquid phases at room temperature, and became completely liquefied only on heating to about 80C. Nonoxygenated fatty acids present in the oil in largest quantity are a C_{18} monoene, 11%; a C_{18} diene, 20%; and a C_{22} monoene presumed to be erucic acid, 26%. In this respect *Cardamine* oil is quite similar to a number of other crucifer oils.

The solid diol esters were not very soluble in ethyl ether, and could readily be crystallized from a 10% ether solution of the mixed methyl esters. This concentration appeared to be near the solubility limit of the diols, and nearly quantitative recovery was obtained by one crystallization at −18C. A very clean separation was obtained in this manner, since the nonoxygenated fatty acid esters remained in solution at this temperature. TLC of the diol esters indicated they contained only a trace of the normal methyl esters. Removal of solvent from the crystallization filtrate yielded the soluble methyl esters which contained no hydroxyl as shown by IR analysis and TLC. The complete composition according to GLC is given in the experimental section.

An attempted crystallization from acetone resulted in nearly complete conversion of the diols to isopropylidene derivatives, apparently because a trace of mineral acid had been carried over from the esterification reaction. A pure sample of methyl *threo*-13,14-dihydroxydocosanoate was heated in acetone with a trace of sulfuric acid, and the IR spectrum of the isopropylidene derivative was essentially the same as the spectrum obtained for the corresponding derivative of the unknown diols.

The IR spectrum of the diol methyl esters was nearly identical to the spectrum of methyl *threo*-9,10-dihydroxystearate, and both were somewhat different from the spectrum of a monohydroxy ester. The apparent absence of optical activity of the dihydroxy acids (or esters) is surprising, since they contain two asymmetric carbon atoms. This nonactivity may be a result of nearly complete internal compensation because the *erythro*-diol groupings, flanked as they are by long chains of methylene groups, resemble *meso*-forms. A similar case of a naturally occurring hydroxy fatty acid that appears to be optically inactive has previously been reported (12).

The absence of monobasic acids other than nonanoic acid in the oxidative cleavage products shows that the hydroxyl groups are located on carbons 9 and 10 from the methyl end of each acid. Percentages of the four dibasic acids in the oxidation mixture have been interpreted as representing the composition of the original diol mixture. These values agree with the composition of the monoene mixture obtained from the diols by the Ames-Bowman procedure (2).

The conclusion that the vicinal dihydroxy acids of *Cardamine* oil have the *erythro* configuration is based on the observation that they are converted almost exclusively to *cis*-olefins by the procedure of Ames and Bowman (2). This procedure involves replacement of both hydroxyls with bromines by hydrogen bromide in acetic acid, and subsequent debromination with zinc. It is assumed that the bromination step inverts the configuration of one of the hydroxyl-bearing carbons, but not the other (16,17). *trans*-Elimination of the bromines by an E2-type mechanism is then effected by zinc. The net result is transformation of an *erythro*-diol to a *cis*-olefin. A small amount of isolated *trans*-unsaturation in these monoenes may indicate that some *threo*-diol is present in the mixture or that the conversion of diol to olefin is not completely stereospecific.

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N,N-Dialkylamides of Long Chain Fatty Acids as Plasticizers[1]

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Abstract

A number of N,N-dialkylamides have been prepared, characterized and evaluated as plasticizers for poly(vinyl chloride-vinyl acetate) copolymer. Among these are the N-oleoyl derivatives of diisopropyl, dibutyl, diisobutyl, diamyl, dihexyl, diheptyl, dioctyl, di-2-ethylhexyl and didecylamines. Also included are the N,N-dibutylamides of 2-ethylhexanoic, neodecanoic, neotridecanoic, palmitic, stearic, linoleic, erucic, ricinoleic, naphthenic, dimer, pinic, epoxystearic, animal, cottonseed, hydrogenated cottonseed, rapeseed, *Limnanthes douglasii* seed and parsley seed acids. Optimum low-temperature plasticizing properties are achieved for the N-oleoyl derivatives of dibutyl, diamyl and dihexyl amines. These low-temperature properties are comparable to those of the adipate and sebacate plasticizers without the adverse volatility characteristics of the adipates. Compatibility of the N,N-dialkyloleamides extends through the dihexyl derivative. A brief heat stability study of some selected plasticized polylvinyl chloride copolymer compositions indicates that the thermal stabilization of amide plasticizers is not an insurmountable problem.

Introduction

IT HAS BEEN SHOWN in previous publications that many N,N-disubstituted long chain fatty acid amides such as the morpholides (1), N,N-bis(2-acyloxyethyl)amides (2), piperidides (3) and N,N-bis(2-alkoxyethyl)amides (4), are acceptable primary plasticizers for polyvinyl chloride resins (PVC). Recent work has shown that still another type, the dialkylamides, characterized by excellent sebacate-like low-temperature performance, must be added to the group. This report deals with the preparation, characterization and plasticizer evaluation of a number of N-fatty acyl derivatives of various C_3 to C_{10} symmetrical dialkylamines and also includes a brief heat stability study of selected plasticized PVC compositions.

Experimental

The oleic acid (Emersol 233LL Elaine) and dimer acid (Empol 1014) were products of Emery Industries. Oleoyl chloride was prepared from the oleic acid. Neodecanoyl and neotridecanoyl chloride were obtained from Euron Laboratories. The "animal acids" mixture was an Armour & Co. product, Neofat 65, having the following compositional specifications: 2% myristic, 26% palmitic, 16% stearic, 48% oleic and 8% linoleic acids. Hydrogenated cottonseed acids were derived from a selectively hydrogenated oil (5) which had an iodine value of 73.0 and a thiocyanogen value of 68.0. Rapeseed, cottonseed, *Limnanthes douglasii* seed and parsley seed fatty acids were obtained by saponification of the respective oils. Ethyl-2,2-dimethyl-3-chlorocarbonylcyclobutaneacetate, used in the preparation of the pinic acid derivative, was furnished by the Naval Stores Laboratory of this Division. Linoleic acid, 95% purity, was obtained from the Northern Regional Research Laboratory. Di-*n*-hexylamine was a Sapon Laboratories product. All the other dialkylamines and acids were Eastman Kodak Co. products, "white label" when available. The naphthenic acid had a neutralization equivalent of 217.

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