hydrochalcones are at least slightly better antioxidants than the corresponding chalcones.

Dihydrookanin (VII) and its isomer (X) are particularly good antioxidants, the latter being the better of the two.

CONCLUSIONS

The present study establishes that polyhydroxydihydrochalcones are more effective than the corresponding chalcones as antioxidants for lard at 120 °C. In this respect, therefore, they are analogous to the polyhydroxydihydrocinnamic acids, which are superior to the corresponding cinnamic acids. This is presumably due to the fact that the dihydro derivatives form more stable free radicals than the unsaturated compounds. Why this should be so, however, is not at this stage obvious.

Registry No. I, 480-41-1; II, 60-82-2; III, 520-33-2; IV, 35400-60-3; V, 484-76-4; VI, 489-73-6; VII, 72666-14-9; VIII, 73692-51-0; IX, 552-58-9; X, 57765-66-9; XI, 72704-76-8; XII, 94324-67-1; XIII, 88191-22-4; XIV, 94324-68-2.

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Mass Spectral Identification of a Metabolite of Chlorpropham in Potatoes

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Chlorpropham (CIPC), isopropyl *N*-(3-chlorophenyl)carbamate, is a selective herbicide that also is used as a sprout suppressant on stored potatoes. Residues of CIPC have been encountered in table-ready potatoes examined under the Total Diet Program conducted by the Food and Drug Administration. A second compound, detected by GC, was found to be associated with many of these residues. This suspected metabolite of chlorpropham was identified by GC–MS, with comparison to synthesized reference standards, as isopropyl *N*-(3-chloro-4-methoxyphenyl)carbamate. Home-grown potatoes, previously free of all detectable residues, were found to contain isopropyl *N*-(3-chloro-4-methoxyphenyl)carbamate 21 days after chlorpropham postharvest application, thus confirming this methoxy derivative as a formerly unreported metabolite of CIPC. Levels of this metabolite as high as 0.063 ppm (French fries) have been encountered in various table-ready potatoes.

The Food and Drug Administration (FDA) monitors residues of pesticides, industrial chemicals, metals and nutrients in the nations food supply, through several programs. One of these, the Total Diet Program, is one of FDA's oldest residue surveillance studies, having been in operation since 1964. Results of this study are published periodically (Johnson et al., 1984a,b; Podrebarac, 1984a,b).

Each sample (market basket) consists of 232 retail grocery items from each of three cities in one of four geographic region of the country. Samples reflect dietary preferences for eight age–sex groups represented in the United States and are based on data obtianed from the Nationwide Food Consumption Survey (U.S. Department of Agriculture) and the Second National Health and Nutrition Examination Survey (National Center for Health Statistics) (Pennington, 1982, 1983).

Each food is prepared for consumption (table ready), just as it might be in the average home. For example, oranges are peeled, meats are roasted, baked, or fried, and potatoes are baked, boiled, scalloped, etc. Foods requiring this processing are prepared by dieticians in institutional kitchens. After preparation, the items are examined individually or as recipe items (e.g., meatloaf, lasagna, soups, etc.). Thus, 234 table-ready foods (including 13 recipe items) are screened for pesticides, herbicides, industrial chemicals, toxic metals, and selected nutrients.

Extraction and cleanup procedures used routinely for pesticides and industrial chemicals are described elsewhere (AOAC, 1980; FDA, 1981; Storrherr et al., 1971; Carson, 1981; Krause, 1980; Hopper, 1982).

With the exception of *N*-methylcarbamates, which are determined by liquid chromatography, organic chemical contaminants are determined by gas chromatography (GC). Routinely, sample eluates are initially examined by GC with a relatively nonpolar stationary phase and electron-capture (EC) or element-specific detectors (halogen, P, S, N). Confirmation of residues detected is attempted by using one or more stationary phases of different polarity.

Retention data, element-specific detector responses, cleanup column elution patterns, and expert interpretation by the residue chemist combine to identify all but the most unusual chemical contaminants. Those residues that defy identification are classified as unidentified analytical responses (UAR's) and become candidates for mass spectrometric (MS) analysis.

One such UAR has occasionally been detected in some of the several potato items (i.e., mashed, boiled, baked, scalloped, French fries, and potato chips). The occurrence of this compound has always been associated with residue

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levels of chlorpropham (CIPC) and was, therefore, suspected to be a metabolite or impurity of this organic carbamate. Chlorpropham, isopropyl *N*-(3-chlorophenyl)carbamate, when used as a sprout suppressant on potatoes is applied postharvest at a rate of nearly 17 ppm (Thomson, 1981).

This investigation was initiated with the intent of establishing the identity and determining the source of this UAR and thereby aid the toxicologist in evaluating its significance in our nations food supply.

EXPERIMENTAL SECTION

Mass Spectrometry. Identification of the metabolite of chlorpropham was achieved through mass spectrometric (GC–MS) analysis performed on a VG 7070E GC–MS interfaced to an 11-250 data system (VG Analytical, Manchester, England) and a Varian 3700 gas chromatograph. Ion source operating temperature was maintained at 250 °C with an ionizing voltage of 70 eV for electron impact ionization. Methane-positive ion chemical ionization was performed at 200 °C and 45 eV. Capillary column analyses were accomplished with split mode injection on OV-1 (30 m × 0.25 mm i.d.) fused silica column (J & W Scientific, Inc., Rancho Cordova, CA) eluting directly into the MS source and by using helium carrier gas (head pressure 30 psi, split 50 mL/min) and a column oven temperature of 180 °C. Low-resolution, EI, full-scan mass spectra collected at a 1 s/decade scan rate were normalized after background subtraction.

Gas Chromatography. Quantitation was accomplished with a 1.8 m × 2 mm i.d. glass column packed with 5% OV-101 on 60–80-mesh Chromosorb WHP and fitted in a Tracor 560 gas chromatograph with a Hall electrolytic conductivity detector (HECD). The column flow rate of helium was 50 mL/min with a temperature of 200 °C.

Assay of Sprout Nip. Sprout Nip EC (complimentary sample from PPG Industries, Inc., Pittsburgh, PA) was sequentially diluted with ethyl acetate to a concentration consistent with Hall electrolytic conductivity detection (HECD) and determined by GC with comparison to a reference standard of chlorpropham.

Treatment of Potatoes. Red Pontiac potatoes from a home garden were washed at harvest and stored for 2 weeks. Twelve tubers, weighing approximately 100 g each, were dipped for 15 s in a freshly prepared 5% (commercial strength; Thomson, 1981) aqueous solution of Sprout Nip EC, allowed to dry, and stored in dark at room temperature in beakers covered with aluminum foil. Potatoes were analyzed in duplicate at 1-week intervals following procedures described in the "Pesticide Analytical Manual of the Food and Drug Administration" (FDA, 1981). Two undipped 100-g tubers were used as the control.

Synthesis of Isopropyl *N*-(Chloromethoxyphenyl)carbamates. Isopropyl *N*-(3-chloro-4-methoxyphenyl)carbamate was synthesized by adaptation of a procedure by Strain (1956) for preparation of *N*-(chlorophenyl)carbamates.

To 0.1 mol (15.8 g) of 3-chloro-*p*-anisidine (Aldrich Chemical Co., Milwaukee, WI) dissolved in 100 mL of benzene was added 30 mL of water and 8.44 g (0.11 mol) of sodium bicarbonate and mixed vigorously. After being cooled to 5 °C, 0.1 mol (12.2 g) of isopropyl chloroformate (complimentary sample from PPG Industries, Pittsburg, PA) was added dropwise over a period of 2 h and the temperature kept at 5–10 °C during reaction. Dilute hydrochloric acid was added to the reaction mixture, and the organic layer was separated, washed consecutively with dilute hydrochloric acid and water, and dried over anhydrous sodium sulfate. After evaporation of the benzene on a steam bath with a stream of nitrogen, the residue was recrystallized twice from ethyl alcohol. After purification through sublimation, white crystals with melting range of 95.0–96.0 °C were obtained.

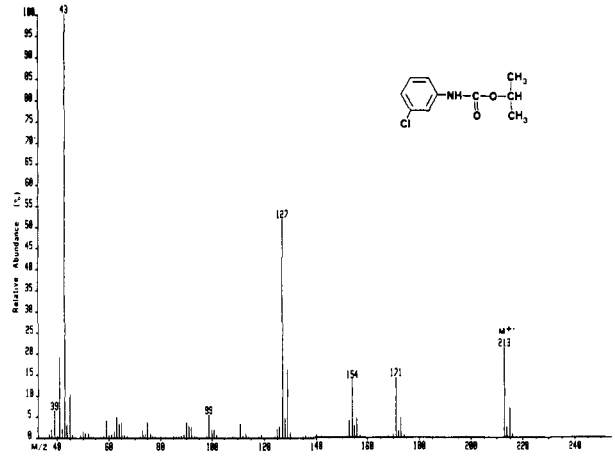

Figure 1. Electron impact mass spectrum of chlorpropham [isopropyl *N*-(3-chlorophenyl)carbamate].

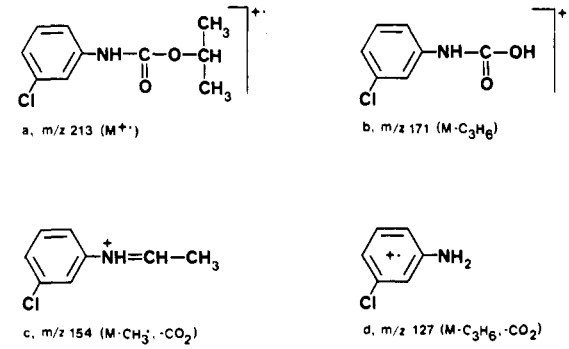

Figure 2. Suggested structures of major electron impact fragments of chlorpropham.

Similarly, isopropyl *N*-(2-methoxy-5-chlorophenyl)carbamate prepared from 5-chloro-*o*-anisidine (Aldrich Chemical Co., Milwaukee, WI) yielded white crystals with a melting range of 53.0–55.0 °C.

RESULTS AND DISCUSSION

When the gas chromatography system described for the analysis of various potato products was used, the UAR was found to elute at a retention time relative to that of chloropyrifos (R_c) of 0.75, whereas the relative retention time (R_c) of chlorpropham corresponded with the literature value of 0.32. All of the UAR and a small portion of chlorpropham eluted in the 50% ethyl ether–petroleum ether eluate from a Florisil column (FDA, 1981); the largest fraction of chlorpropham, however, was present in the 15% mixed ether eluate. No evidence of this UAR or any other impurities was observed in the chromatograms of Sprout Nip EC.

The mass spectrum of the parent compound, chlorpropham (Figure 1), was investigated with the intent that it might provide clues for the elucidation of the spectrum of its metabolite. A fairly intense (30%) molecular ion, which is characteristic of carbamates with N-aromatic substituents, is seen at m/z 213 (Figure 2a). Elimination of propylene from the isopropyl ester moiety results in a peak at m/z 171 (Figure 2b). Loss of a methyl radical followed by elimination of carbon dioxide yields the even-electron ion at m/z 154 (Figure 2c) (Lewis, 1964a,b). A McLafferty rearrangement involving the elimination of both carbon dioxide and propylene, result in the production of the diagnostically significant ion at m/z 127 (Figure

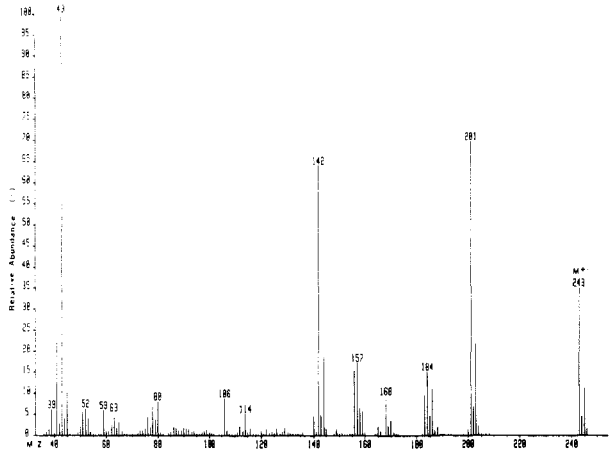

Figure 3. Electron impact mass spectrum of metabolite of chlorpropham.

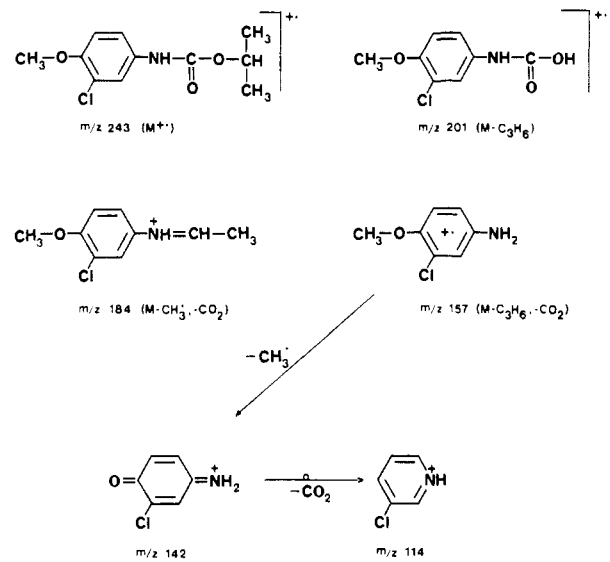

Figure 4. Suggested structures of major electron impact fragments of metabolite of chlorpropham.

2d) (Lewis 1964a,b; Thomson et al., 1966). Cleavage of the carbon–oxygen bond with charge retention by the isopropyl group gives rise to an abundant peak at m/z 43 (Budzikiewicz et al., 1967; Safe and Hutzinger, 1973).

The mass spectrum of the metabolite of chlorpropham (Figure 3) reveals an apparent molecular ion at 243, which was confirmed by methane-positive ion chemical ionization. Although somewhat more complex this spectrum bears a similarity to that of chlorpropham itself. Elimination of propylene (m/z 201); losses of methyl and subsequent explusion of carbon dioxide (m/z 184); the McLafferty rearrangement with losses of both carbon dioxide and propylene (m/z 157) all appear to be evident, indicating the ester portion of the molecule is unchanged. Consequently, the observed increase in molecular weight of 30 is probably the result of substitution on the phenyl ring. It has been demonstrated that potato tubers in storage will attach a methoxy group (probably through the hydroxyl structure) on a compound containing a substituted phenyl ring (Heikes et al., 1979). Additionally, it has been shown that a loss of 15 daltons and the formation of the relatively stable quinoid structure, followed by the rearrangement loss of carbon monoxide and the resultant pyridinium cation, are characteristic of o- and p-methoxyanilines. (This pattern is evident as ions at m/z 157, m/z 142, and m/z 114 for the chlorinated species in this spectrum.) These ions are displayed as Figure 4. By

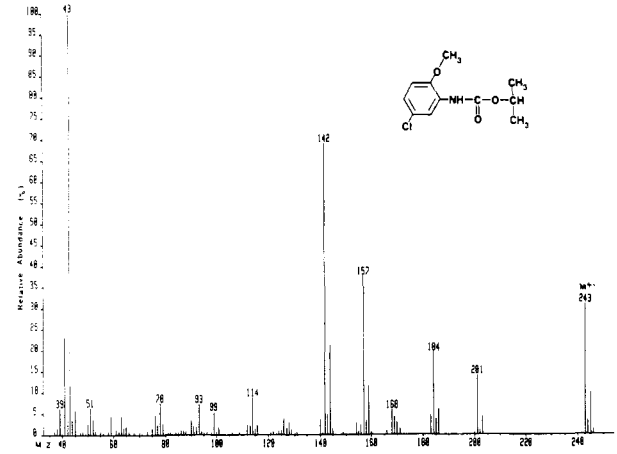

Figure 5. Electron impact mass spectrum of isopropyl N-(2-methoxy-5-chlorophenyl)carbamate (o-methoxychlorpropham).

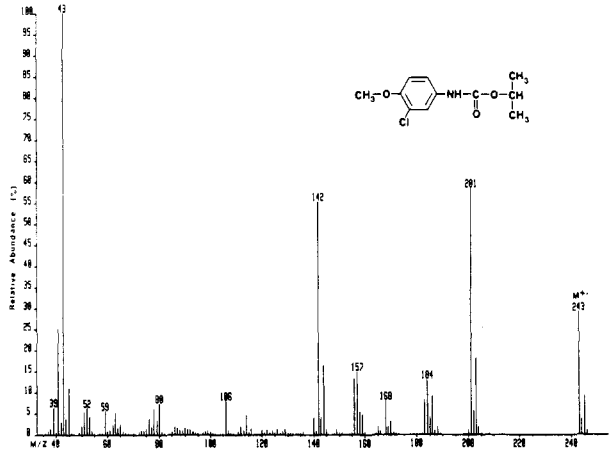

Figure 6. Electron impact mass spectrum of isopropyl N-(3-chloro-4-methoxyphenyl)carbamate (p-methoxychlorpropham).

contrast, the meta isomer was found to preferentially eliminate formaldehyde (Spiteller and Spiteller-Friedmann, 1962). The above evidence supported the hypothesis that the metabolite (UAR) accompanying residues of chorpropham in potatoes was indeed the ortho or para methoxy derivative of chlorpropham.

Synthesized reference standards, o-methoxychlorpropham [isopropyl N-(2-methoxy-5-chlorophenyl)carbamate] and p-methoxychlorpropham [isopropyl N-(3-chloro-4-methoxyphenyl)carbamate], give quite similar mass spectra (Figures 5 and 6, respectively), but only the retention data of the para derivative are identical with those of the metabolite found in potatoes. When the gas chromatographic conditions described under Experimental Section were used, both the synthesized p-methoxychlorpropham and the metabolite (UAR) exhibited retention times of 1.7 min. In contrast, the synthesized o-methoxychlorpropham showed a retention time of 1.4 min.

Home-grown potatoes dipped in a 5% aqueous solution of Sprout Nip EC and analyzed at weekly intervals showed no detectable levels of isopropyl N-(3-chloro-4-methoxyphenyl)carbamate (p-methoxy chlorpropham) until trace levels (0.0015 ppm) appeared at 21 days. Subsequent analysis showed increasing concentrations of this metabolite to a level of 0.17 ppm for the final determination at the end of 6 weeks.

In recent market basket samples, this metabolite has occurred in baked potatoes (0.0040 ppm), potato chips (0.0080 ppm), and French fried potatoes (0.063 ppm) accompanied by corresponding residues of chlorpropham of 0.25, 2.4, and 1.6 ppm, respectively.

Registry No. CIPC, 101-21-3; isopropyl *N*-(3-chloro-4-methoxyphenyl)carbamate, 94483-57-5.

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Volatile Components of Rooibos Tea (*Aspalathus linearis*)

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Rooibos tea (*Aspalathus linearis*) components have been identified by using capillary gas chromatography and gas chromatography/mass spectrometry. Samples were vacuum steam distilled/solvent extracted to yield a volatile oil for analysis. Among the 99 components positively or tentatively identified in the vacuum steam volatile oil are 26 ketones, 19 aldehydes, 16 alcohols, 12 esters, 9 hydrocarbons, 7 phenols, 4 acids, 3 ethers, and 3 miscellaneous components. The major components of the extract were shown to be guaiacol, 6-methyl-3,5-heptadien-2-one, damascenone, geranylacetone, β-phenylethyl alcohol, and 6-methyl-5-hepten-2-one. Headspace analysis of dry leaves yielded 218 positive or tentative identifications: 47 alcohols, 41 ketones, 39 aldehydes, 27 hydrocarbons, 24 esters, 13 ethers, 7 phenols, 6 acids, and 14 miscellaneous components.

Aspalathus linearis is a shrub native to the mountains of western South Africa. The stems are slender, and the leaves are linear and needlelike, 2–6 cm long. The leafy stems, when finely cut, fermented, and dried, are used as a substitute for tea from *Camellia sinensis* (Morton, 1983). Preliminary observations by one of the authors (J.F.M.) suggested that the plant might possess some repellent or antifeedant activity against cockroaches, so a study of Rooibos tea volatiles was initiated. Previous work had identified glycosylflavonoids and other high molecular weight components (Koeppen, 1962a,b, 1963, 1964; Koeppen and Roux, 1965), but no reference to more volatile components appears in the literature.

EXPERIMENTAL SECTION

Volatile Component Concentrate Preparation. Cured Rooibos tea was obtained from a South African producer. The material (450 g) was placed in a 5-L round-bottomed flask, and distilled water (2.5 L) was added. A modified Likens-Nickerson steam distillation/continuous extracton head was attached. Purified heptane (Burdick and Jackson; 110 mL) was used as the extracting solvent. The isolation was carried out under reduced pressure (40 mmHg) for 3-h intervals. The extraction head condenser was cooled with water/ethylene glycol at 0 °C, and a Dewar condenser filled with isopropyl alcohol/solid carbon dioxide was placed at the outlet of the system. After 3 h the heptane solution was replaced with fresh heptane. The process was then continued for a second 3-h period. This operation was repeated twice, for a total extraction period of 9 h. Each heptane extract was dried, filtered, and concentrated by careful vacuum distillation to remove the solvent. Capillary gas chromatography (GC) indicated all three extracts to be quite similar, so they were combined (4.3 mg, 9.6-ppm yield from the sample material; corrected for residual solvent).

Headspace Analysis. In a typical sequence, Rooibos tea (1 g) was placed in a sample tube (15 cm^3; 1.8 cm i.d. × 13.0 cm long) upstream from a Tenax-GC packed trap (0.635 cm o.d. × 7.6 cm long) and purified helium (25 cm^3/min) was passed through the sample, sweeping volatiles into the Tenax trap. A sampling of 1.5 h (at room temperature) was employed (2.25 L total). The trap was

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