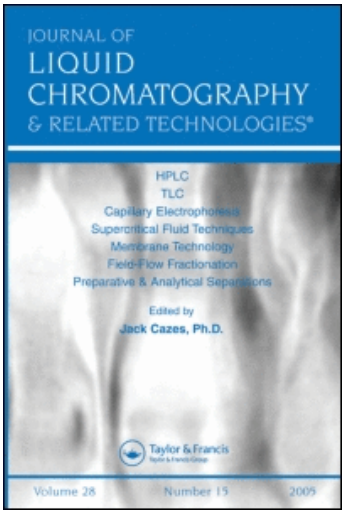

Journal of Liquid Chromatography & Related Technologies
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597273

Analysis of Dehydroabietic Acid in Kraft Mill Effluents by High-Performance Liquid Chromatography

R. K. Symons[a]
[a] Environment Protection Authority Laboratory, East Melbourne, Victoria

To cite this Article Symons, R. K.(1981) 'Analysis of Dehydroabietic Acid in Kraft Mill Effluents by High-Performance Liquid Chromatography', Journal of Liquid Chromatography & Related Technologies, 4: 10, 1807 — 1815
To link to this Article: DOI: 10.1080/01483918108064848
URL: http://dx.doi.org/10.1080/01483918108064848

JOURNAL OF LIQUID CHROMATOGRAPHY, 4(10), 1807-1815 (1981)

ANALYSIS OF DEHYDROABIETIC ACID IN KRAFT MILL EFFLUENTS BY HIGH-PERFORMANCE LIQUID CHROMATOGRAPHY

R.K. Symons
Environment Protection Authority
Laboratory Services Branch
P.O. Box 41, East Melbourne, Victoria, 3002

ABSTRACT

A rapid high-performance liquid chromatographic method for the determination of dehydroabietic acid in kraft mill effluent is described. Selective solvent extraction of the samples with dichloromethane was performed. The samples were isocratically analysed on a Rad-Pak C18 column using 75% acetonitrile in water (0.1% acetic acid added). Detection was carried out at 220 nm and 267 nm on a variable wavelength detector with a detection limit of 0.01 mg/L.

INTRODUCTION

Toxic components of pulp and paper effluents are complex mixtures of organic and inorganic moieties either naturally occurring, added or formed during the pulping process.(1) Many of those components have been tested for toxicity.(2,3) Dehydroabietic acid (DHAA) is one of several naturally occurring resin acids extracted from softwood trees during the kraft process and is a major contributor of toxicity in non-bleached pulp effluent.(4-7) Its 96-h LC 50 (static bioassay) for

juvenile freshwater coho salmon is 0.75 to 1.8 mg/L.(1,2,5,6,8) Concentration of DHAA in mill effluents is highly variable ranging from 0.4 to 51.8 mg/L.(1)

Colorimetric (9) and gas chromatographic (GC) (10-13) procedures for the analysis of resin acids in mill effluents have been widely used. However, each suffers certain inadequacies with respect to separation, detection and identification of DHAA from other resin acids. The non-specificity and insensitivity of the colorimetric procedure to DHAA has already been noted,(2) while incomplete derivatization due to the bulk of involatile materials present, interference from decomposition of labile substances, and lengthy analysis times has hindered quantitative GC analyses. (15)

High-performance liquid chromatography (HPLC) has been developed for the analysis of a wide range of trace organic compounds in waters and waste waters, (14,16) and is free of many of the problems associated with GC.

By using a selective solvent extraction procedure and HPLC, a rapid and quantitative method has been developed for the determination of trace levels of DHAA in mill effluents at a detection limit of 0.01 mg/L.

MATERIAL AND METHODS

Glassware

The glassware used was a Kuderna-Danish evaporator with 3 mL concentrator tubes; 50 and 100 mL volumetric flasks; and 2000 mL separatory funnels. All glassware was soaked in a synthetic detergent, rinsed and dried. Prior to use all glassware was rinsed with dichloromethane.

Chemicals

All solvents used were specially purified for HPLC (Ajax Chemicals, Melb., Aust.).

Dehydroabietic acid was obtained from Dr. A.A. Sioumis (CSIRO, Div. Chem. Tech., Melb., Aust.) and a purity of 95% was determined by GC/MS of the TMS derivatives.

Apparatus

The chromatography was conducted using a Laboratory Data Control (Riveria Beach, Florida, USA) liquid chromatograph with dual Constametric III solvent delivery pumps, a Gradient Master, Rheodyne 7125 loop injector, and a Waters Assoc. (Bedford, Mass., USA) Radial Compression Separation System, Rad-PAK A (10 cm x 5 mm, C_{18}) column. Detection was accomplished via a Spectromonitor III variable-wavelength absorbance detector.

Retention times and peak areas were electronically acquired on a Hewlett-Packard (Avondale, PA, USA) HP3388A reporting integrator.

Chromatographic Conditions

The separations were achieved isocratically with a mobile phase composition of 75% acetonitrile in water (0.1% acetic acid) at a flow rate of 2.0 mL/min, and detection was carried out at 220 nm and 267 nm. The radial compression module was at ambient temperature in its fully compressed mode. The injections were made with a 20 µL sample loop.

Preparation of Standards and Samples

The standard solution of DHAA was prepared by dissolving 10 mg in a small amount of dichloromethane and diluting to 100 mL

with acetonitrile. This stock solution and subsequent working standards were stored at 4°C.

Samples of 2-L volumes were collected in plastic bottles, delivered to the laboratory on ice, and were stored at 4°C prior to extraction.

The sample, 500 mL, was adjusted to pH 12 with 10M NaOH and was extracted with three successive 60 mL volumes of dichloromethane to produce a base-neutral fraction. The dichloromethane extracts containing neutral compounds, were discarded.

The pH of the aqueous fraction was adjusted to 2 with conc. HCl and extracted with dichloromethane (3 x 60 mL). The acid-extractable fraction containing the DHAA and other resin acids were combined in a Kuderna-Danish evaporator which was connected to a rotary evaporator and the volume reduced under vacuum to approximately 2 mL. The concentrate was evaporated to dryness under a stream nitrogen and the residue dissolved in 1.0 mL acetonitrile before injection into the HPLC.

Identification and Quantitation of Peaks

Peaks observed in the sample chromatographic profiles were identified by (i) retention times in comparison with standards; (ii) coinjection with standards; (iii) peak area ratios at two different wavelengths (220 and 267 nm); and, (iv) GC/MS of the TMS derivatives of fractions collected from the HPLC. Both peak height and area measurements were used for the quantitation of DHAA in the samples.

RESULTS AND DISCUSSION

The HPLC profiles of DHAA obtained with UV detection at 220 nm and 267 nm shown respectively in Figures 1 and 2

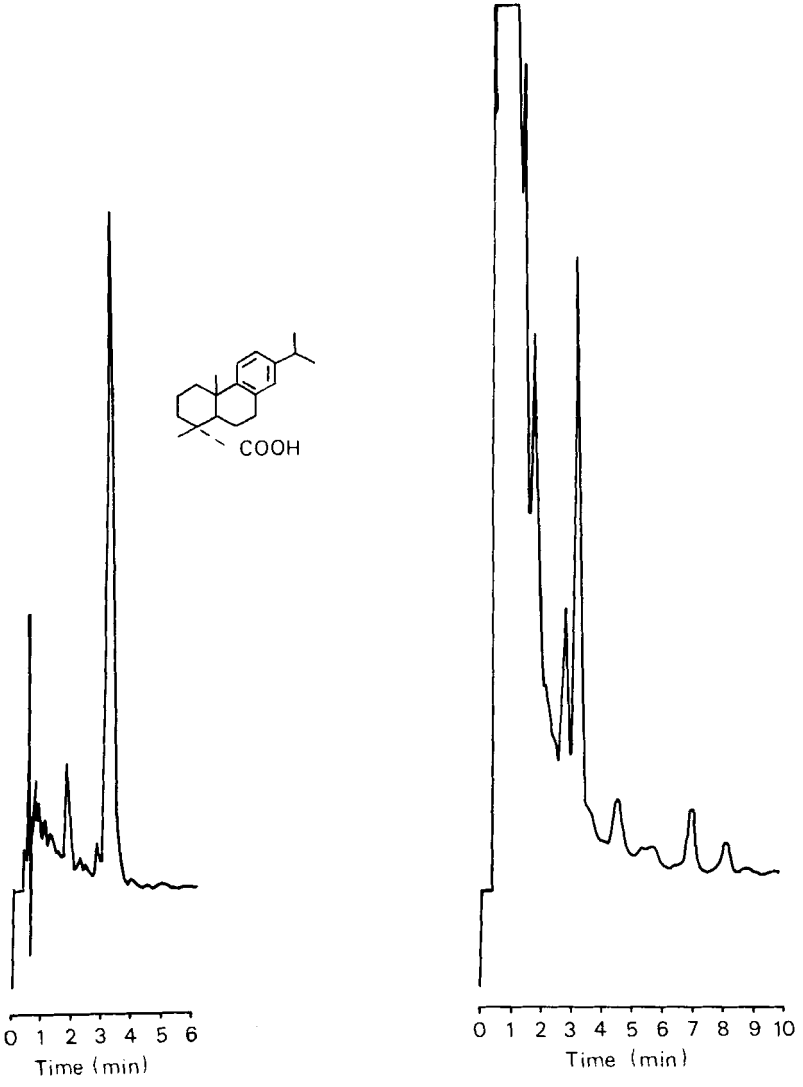

FIGURE 1

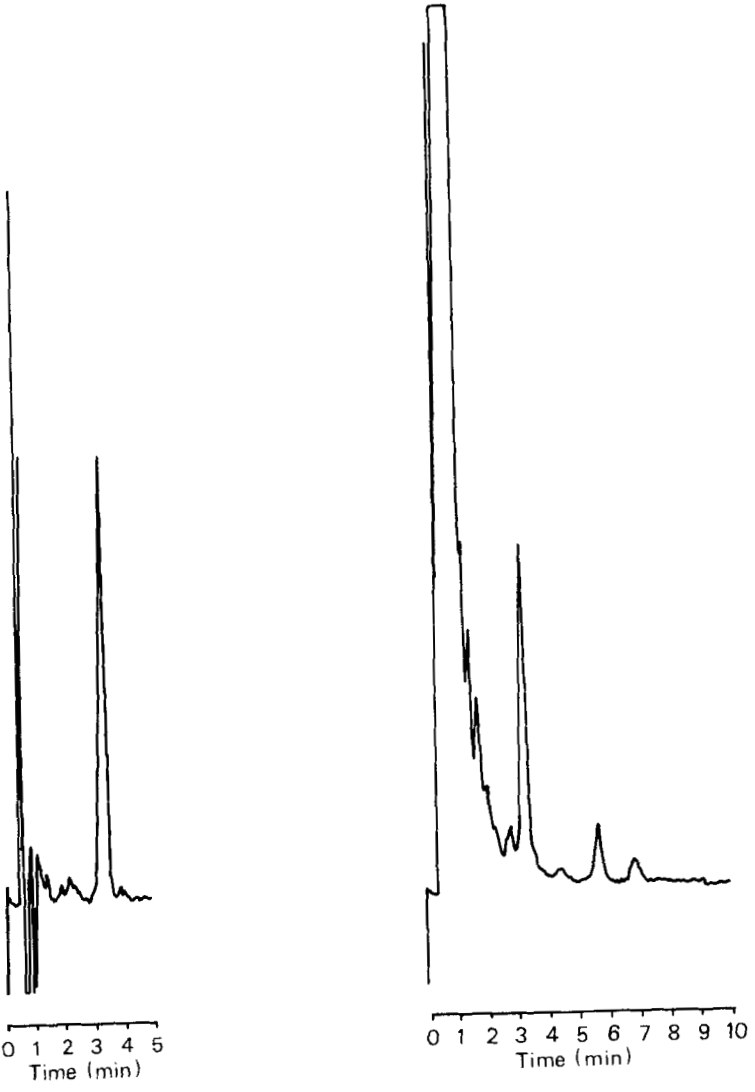

FIGURE 2

show good separation from other extractives with a retention time for DHAA of 3.25 minutes. It was also observed that DHAA exhibits UV absorption at both 220 and 267 nm wavelength, the main difference being in their relative intensities, with the 220 nm wavelength exhibiting a five-fold increase in sensitivity. The use of absorbance ratios to identify compounds in samples has been used, (17) but cannot be relied upon if interferences are present.

Any tendency for sample components to dissociate while being chromatographed, as in the case of resin acids, frequently leads to excessive peak broadening or a tailing peak. The addition of 0.1% acetic acid to the mobile phase decreases the pH sufficiently to suppress dissociation making the sample component less polar and more amenable to analysis, i.e., a much improved peak shape results. (18)

No attempt was made to quantitate any of the other compounds present however, results of GC/MS analysis suggested the presence of several resin acids of the abietic and pimaric-acid type. Further investigations relating to the identification of these resin acids by HPLC and their extraction from kraft mill effluent by Sep-Pak C18 cartridges (Waters Assoc.) is being carried out.

ACKNOWLEDGEMENTS

I would like to thank Dr. D. Burke for performing and interpreting the GC/MS data and Dr. A. Sioumis for supplying the resin acid standards.

REFERENCES

1. Hutchins, F.E., Toxicity of Pulp and Paper Effluents - A Literature Review, US EPA - 600/3-79-013, (1979).

2. Leach, J.M. and Thakore, A.N., Isolation of Toxic Constituents of Kraft Pulp Mill Effluents, CPAR, Can. For. Serv., Proj. Rept., 11-5, (1974).

3. Rogers, I.H., Isolation and Chemical Identification of Toxic Components of Kraft Mill Wastes, Pulp Paper Mag. Can. 74, 9, 111. (1973).

4. Leach, J.M. and Thakore, A.N., Toxic Constituents in Mechanical Pulping Effluents, Tappi, 59, 2, 129, (1976).

5. Leach, J.M. and Thakore, A.N., Identification of the Toxic Materials in Sulphite Pulp Mill Effluents, CPAR, Can. For. Serv., Proj. Rept., 407-3, (1978).

6. Iwama, G.K., Greer, G.L., and Larkin, P.A., Changes in Some Hematological Characteristics of Coho Salmon in Response to Acute Exposure to Dehydroabietic Acid at Different Exercise Levels, J. Fish. Res. Board Can., 33, 2, 285, (1976).

7. Rogers, I.H., Mahood, H.W., James, B.C., Servizi, J.A. and Gordon, R.W., Identifying Extractives Toxic to Aquatic Life, Pulp Paper Mag. Can., 80, 9, 94, (1979).

8. Davis, J.C. and Hoos, R.A.W., Use of Sodium Pentachlorophenate and Dehydroabietic Acid as Reference Toxicants for Salmonoid Bioassays, J. Fish. Res. Board Can., 32, 411, (1973).

9. Carpenter, W.L., "Colourimetric Measurement of Resin Acid Soaps in Pulp and Paper Mill Wastes", Tappi, 48, 11, 669 (1965).

10. Nestler, M.F.H. and Zinkel, D.F., "Quantitative Gas-Liquid Chromatography of Fatty and Resin Acid Methylesters", Anal. Chem., 39, 10, 1118, (1967).

11. Jaye, N.M., Proveaur, A.T., and Lawrence, R.V., "Improved Procedure for the Analysis (GLC) of Resin Acids", J. Am. Oil Chem. Soc., 51, 5, 195, (1974).

12. Holmpom, B., Avela, E., Pekkala, S., "Capillary Gas Chromatography - Mass Spectrometry of Resin Acids in Tall Oils Resin", J. Am. Oil Chem. Soc., 51, 9, 397, (1974).

13. Hasson, J.C. and Kulkarni, M.V., "Open-Tube-Column Gas Ghromatography of Resin Fluxes", Anal. Chem., 44, 9, 1586 (1972).

14. Horvath, C., and Melander, W., "Liquid Chromatography with Hydrocarbonaceous Bonded Phases; Theory and Practice of Reversed Phase Chromatography", J. Chromat. Sci., 15, 393, (1977).

15. Keith, L.H., "Identification of Organic Compounds in Unbleached Treated Kraft Paper Mill Wastewaters", Environm. Sci., Techol., 10, 6, 555, (1976).

16. Crathorne, B., Watts, C.D., and Fielding, M., "Analysis of Non-volatile Organic Compounds in Water by High-performance Liquid Chromatography", J. Chromat., 185, 671, (1979).

17. Krstulovic, A.M., Rosie, D.M. and Brown, P.R., "Selective Monitoring of Polynuclear Aromatic Hydrocarbons by High-Pressure Liquid Chromatography with a Variable Wavelength Detector", Anal. Chem., 48, 1383 (1976).

18. Majors, R.E., "Recent Advances in High-Performance Liquid Chromatography Packings and Columns, J. Chromat. Sci., 15, 334, (1977).