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Note

High-performance liquid chromatographic separation of potato glycoalkaloids using a radially compressed amino column

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The majority of glycoalkaloids present in commercial potato varieties or tubers in breeding programs are of the solanidine series which includes α-chaconine, α-solanine and their metabolites. Because of their known acute toxicity[1–3], their possible chronic toxicity[4–6] and their characteristic bitter taste[7,8]; glycoalkaloids must be analyzed in all new potato varieties before they can be released commercially.

Of the possible methods for the analysis of potato glycoalkaloids[9], high-performance liquid chromatography (HPLC) is becoming the most widely used because it is fast, accurate, reproducible and can be used to determine both individual and total glycoalkaloids. Recently there have been several HPLC methods developed for glycoalkaloid analysis[9–14], one of which employed a reversed-phase radially compressed column[9] which prevents channeling, enhances reproducibility and permits a faster analysis. In this paper, a radial compression separation of potato glycoalkaloids using an amino Radial-Pak column is described.

EXPERIMENTAL

All solvents used were HPLC grade (Fisher Scientific, Fair Lawn, NJ, U.S.A.). α-Chaconine and α-solanine were obtained from the procedure of Bushway and Storch[15] while β_2-chaconine was a gift from Eugene A. Talley, Eastern Regional Research Center, USDA, Philadelphia, PA, U.S.A. The other glycosides were prepared from acid hydrolysis of α-chaconine and α-solanine employing the procedure of Filadelfi[8].

The HPLC system used consisted of a Model 6000A pump and a U6K injector (Waters Assoc., Milford, MA, U.S.A.), a Schoeffel 450 variable-wavelength detector (Westwood, NJ, U.S.A.) and an Omni-Scribe recorder (Houston Instruments, Austin, TX, U.S.A.).

HPLC separations were performed at ambient temperature with a Radial-Pak amino column (Waters Assoc.) 10 cm × 8 mm I.D. with 10-μm packing. Tetrahydrofuran–acetonitrile–water–methanol (55:30:10:5) was employed for the separation of glycoalkaloid metabolites while tetrahydrofuran–acetonitrile–water–methanol (50:25:15.5:9.5) was used for the analysis of α-chaconine and α-solanine. Flow-rates were 3.0 ml/min and 2.5 ml/min, respectively. The effluent was monitored at 215 nm and 0.04 a.u.f.s.

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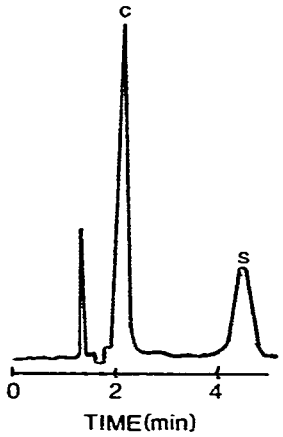
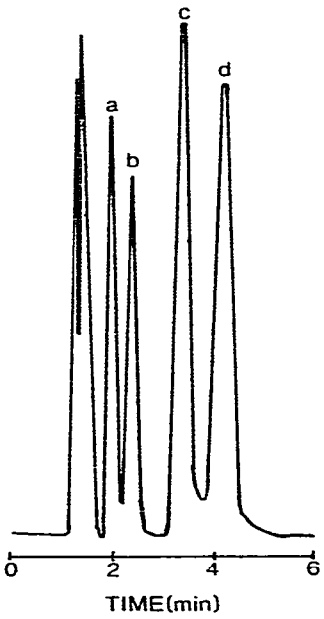


Fig. 1. Chromatogram of glycoalkaloids from an extract of potato tubers. Solvent system, tetrahydrofuran–acetonitrile–water–methanol (50:25:15.5:9.5); flow-rate, 2.5 ml/min; detector sensitivity, 0.04 a.u.f.s.; wavelength, 215 nm; chart speed, 1 cm/min. Peaks: c = α-chaconine; s = α-solanine.



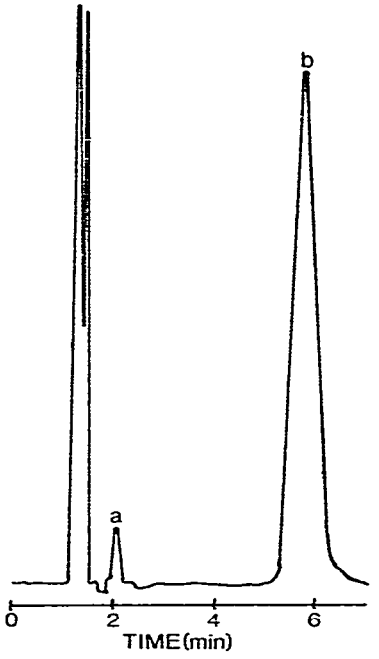


Fig. 2. Chromatogram of α-chaconine metabolites. Solvent system, tetrahydrofuran–acetonitrile–water–methanol (55:30:10:5); flow-rate, 3.0 ml/min; detector sensitivity, 0.04 a.u.f.s.; wavelength, 215 nm; chart speed, 1 cm/min. Peaks: a = γ-chaconine; b = β_2-chaconine; c = β_1-chaconine; d = α-chaconine.

Fig. 3. Chromatogram of α-solanine metabolites. Solvent system, tetrahydrofuran–acetonitrile–water–methanol (55:30:10:5); flow-rate, 3.0 ml/min; detector sensitivity, 0.04 a.u.f.s.; wavelength, 215 nm; chart speed, 1 cm/min. Peaks: a = γ-solanine; b = β-solanine.

RESULTS AND DISCUSSION

Fig. 1 shows the separation of α-chaconine and α-solanine in a potato extract. Analysis time was 4.5 min, a decrease in time of 40% over a method previously developed in our laboratory[11]. In addition analysis time was 25% less than the reversed-phase compression procedure of Morris and Lee[9] when they achieved baseline resolution. A comparison of the amount of α-chaconine and α-solanine present in tuber extracts was made between the existing μBondapak carbohydrate[11] and the amino Radial-Pak methods. The results were in good agreement.

Separations of the metabolites of α-chaconine and α-solanine were also developed (Figs. 2 and 3). Analysis time of the glycoalkaloid series γ-chaconine, β_1-chaconine, β_2-chaconine and α-chaconine was approximately 4 min. This reduced the chromatography time by 50% from a previous method[14] although no baseline resolution was achieved with the new method. However, compared to the method of Morris and Lee[9], the analysis time was shorter and the resolution better.

The metabolic products of α-solanine are separated within 6 min (Fig. 3) excluding α-solanine. A previously developed non-radial compression method[14] took 15 min while the reversed-phase compression procedure of Morris and Lee[9] took 3.5 min. Upon comparison of this new HPLC method to a previously developed procedure for quantifying potato glycoalkaloid metabolites[14], good agreement was shown between the two methods.

Furthermore, to demonstrate the applicability of the metabolite separation to the analysis of actual samples, a potato meal product containing metabolites was

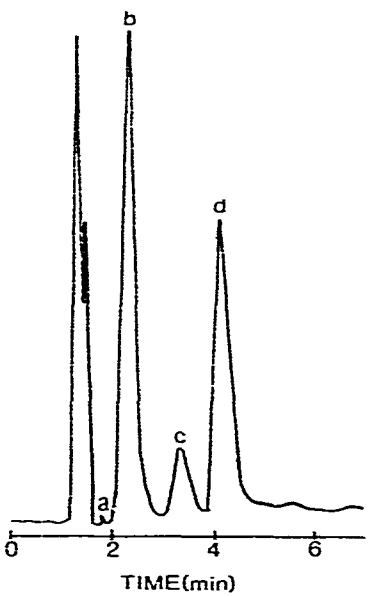


Fig. 4. Chromatogram of glycoalkaloids from a potato meal extract. Solvent system, tetrahydrofuran–acetonitrile–water–methanol (55:30:10:5); flow-rate, 3.0 ml/min; detector sensitivity, 0.04 a.u.f.s.; wavelength, 215 nm; chart speed, 1 cm/min. Peaks: a = γ-chaconine; b = β_2-chaconine; c = β_1-chaconine; d = α-chaconine.

extracted and analyzed. The separation is shown in Fig. 4. As shown in the figure, there were no compounds in the extract that appeared to interfere with the resolution of these metabolites. There were no metabolites of α-solanine present at the detection limit, stated in a previous paper[11]. However, it is not uncommon to observe metabolites of α-chaconine only since α-solanine is not as readily hydrolyzed.

Radial compression HPLC using an amino Radial-Pak column provides a quick and accurate means of analyzing the potato glycoalkaloids, α-chaconine, α-solanine and their metabolites in potato tubers and products. Also it could be used in studying the reaction kinetics of the enzyme(s) involved in the metabolism of α-chaconine and α-solanine.

REFERENCES

1 R. Jellema, E. T. Elema and Th. M. Malingré, *J. Chromatogr.*, 210 (1981) 121.
2 S. G. Willimott, *Analyst (London)*, 58 (1931) 431.
3 M. McMillan and J. C. Thompson, *Quart. J. Med.*, 48 (1979) 227.
4 A. M. Mun, E. S. Barden, J. M. Wilson and J. M. Hogan, *Teratology*, 11 (1975) 73.
5 R. F. Keeler, D. Brown, D. R. Douglas, G. F. Stallknecht and S. Young, *Bull. Environ. Contam. Toxicol.*, 15 (1976) 522.
6 R. F. Keeler, D. R. Douglas and G. F. Stallknecht, *Amer. Potato J.*, 52 (1975) 125.
7 S. L. Sinden, K. L. Deahl and B. B. Aulenbach, *J. Food Science*, 41 (1976) 520.
8 M. Filadelfi, *Ph.D. Thesis*, University of Guelph, Guelph, Ontario (1980).
9 S. C. Morris and T. H. Lee, *J. Chromatogr.*, 219 (1981) 403.
10 I. R. Hunter, M. K. Walden, J. R. Wagner and E. Heftmann, *J. Chromatogr.*, 119 (1976) 223.
11 R. J. Bushway, E. S. Barden, A. W. Bushway and A. A. Bushway, *J. Chromatogr.*, 178 (1979) 533.
12 I. R. Hunter, M. K. Walden and E. Heftmann, *J. Chromatogr.*, 198 (1980) 363.
13 P. G. Crabbe and C. Fryer, *J. Chromatogr.*, 187 (1980) 87.
14 R. J. Bushway, *J. Liquid Chromatogr.*, 5 (1982) 1313.
15 R. J. Bushway and R. H. Storch, *J. Liquid Chromatogr.*, 5 (1982) 731.