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γ-HYDROXY-γ-METHYLGLUTAMIC ACID IN *POLYSTICHUM*ACROSTICHOIDES

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Key Word Index—*Polystichum acrostrichoides*; Aspidiaceae; Christmas fern; amino acid; γ -hydroxy- γ -methylglutamic acid.

Plant. Polystichum acrostichoides (Michaux) Schott (Christmas fern) (voucher specimen No. 72–64 of Appalachian State University Phytochemical Herbarium). Source. Watauga and Avery Counties, North Carolina. Use. Ornamental. Previous work. Other amino acids;¹ γ-hydroxy-γ-methylglutamic acid in sister species (P. tripteron).²

Present work. Dried and finely divided leaves were extracted $2 \times$ with 50% EtOH. The combined extract, reduced in volume under diminished pressure, was applied to an ion exchange column (CG-120 H⁺ form) and washed with H₂O, followed by 50% EtOH. The amino acid fraction was eluted with 2 N NH₃, the solvent evaporated under diminished pressure, the residue dissolved in 1·0 N HOAc and applied to an ion exchange column (CG-400 OAc' form). Using 1·0 HOAc, the last amino acid eluted from the column was γ -hydroxy- γ -methylglutamic acid, a major amino acid constituent. The identity was confirmed by electrophoretic and chromatographic comparison of the isolated amino acid and its derivatives with authentic samples.

Identical electrophoretic mobilities of the amino acid and of the standard were observed at pH 1·9, 3·6 and 6·5. $M_{\rm glutamic\ acid}$ at pH 1·9 = 0·88. Mobilities of the dansyl derivatives at pH 4·4 matched. Chromatographic data (R_f) observed which has not been previously reported: TLC on cellulose; PrOH-H₂O (7:3), 0·44; EtOH-H₂O (7:3), 0·74; n-BuOH-HOAc-H₂O (4:1:1), 0·19. TLC on silica gel; PrOH-H₂O (7:3), 0·18; EtOH-H₂O (7:3), 0·42; n-BuOH-HOAc-H₂O (4:1:1), 0·03; MeCOEt-pyridine-H₂O-HOAc (70:15:15:2), 0·02. Ascending PC; n-BuOH-HOAc-H₂O (4:1:1), 0·29*; PhOH-H₂O (4:1), 0·17; PhOH-H₂O-28% NH₃ (160:40:1), 0·15; EtOH-H₂O-28% NH₃ (18:1:1), 0·02. PC of DNP derivative using 0·75 M phosphate buffer⁴ yields R_f of 0·53.

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^{*} Peterson³ reports $R_{glu} = 0.99$ which is consistent with this datum.

¹ Panvisavas, R., Worthen, L. and Bohm, B. (1968) Lloydia 31, 63.

² TANAKA, M., NAKAMURA, S., NISIZAWA, K. and MIWA, T. (1971) Bot. Mag. (Tokyo) 84, 41.

³ Peterson, P. (1968) J. Chromatog. 38, 301.

⁴ SMITH, I. (1969) Chromatographic and Electrophoretic Techniques, 3rd Edn, Vol. I, p. 178, Wiley-Interscience, New York.