lycoricidine must be expressed by the formulae **1** and **2**, respectively. The stereochemical problems are being investigated. Narciclasine, isolated from the fresh bulbs of daffodils, for which Piozzi, *et al.*[5] have recently proposed the structure (**13**), seems to be iden tical with lycoricidinol.

Lycoricidinol and lycoricidine have growth-inhibiting activity on *Avena coleoptile* sections and rice seedling test[1] (Fig. 3). They also have marked inhibitory action on cell division in tobacco tissue culture[1] (Fig. 4), and they have carcinostatic activity[6] (Fig. 5).

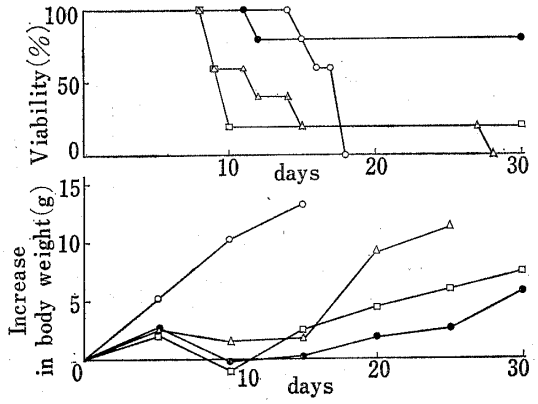

Fig. 5. Antitumor Activity of Lycoricidinol and Lycoricidine using Ehrlich Carcinoma

○ control □ lycoricidinol (100 μg)
● mitomycin C (10 μg) △ lycoricidin (100 μg)

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5) F. Piozzi, C. Fuganti, R. Mondelli, and G. Geriotti, *Tetrahedron*, **24**, 1119 (1968).

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Gas Chromatography of Principal Hexosamines

Recently, gas chromatographic tequniques have been developed for the detection and quantitative estimation of hexosamines by converting them to acetyl[1], trimethylsilyl[2] and N-acetyl-[3] or N-carboethoxy-trimethylsilyl[4] derivatives, although multiplicity of peaks caused by anomeric isomers or decomposition of the derivatives during gas chromatographic separation are disadvantages of these methods.

We have overcome the difficulties by converting hexosamines to corresponding amino alcohols with sodium borohydride followed by trifluoroacetylation[5] of them with trifluoroacetic anhydride (TFAA) in tetrahydrofuran (THF). The procedure was as follows:

1) H.G. Jones, J.K.N. Jones and M.B. Perry, *Can. J. Chem.*, **40**, 1559 (1962).
2) J. Kärkkäinen, A. Lehtonen and T. Nikkari, *J. Chromatog.*, **20**, 457 (1956).
3) C.C. Sweeley and B. Walker, *Anal. Chem.*, **36**, 1461 (1964).
4) M.D.G. Oates and J. Schrager, *J. Chromatog.*, **28**, 232 (1967).
5) Z. Tamura and T. Imanari, *Chem. Pharm. Bull.* (Tokyo), **15**, 246 (1967).

To 0.5 ml of aqueous sample solution (containing 100—500 μg of a mixture of hexosamines) 0.5 ml of 1% $NaBH_4$ in water was added. The mixture was allowed to stand for 30 min at room temperature and the excess of $NaBH_4$ was destroyed by the addition of drops of 0.5N HCl–MeOH. The solution was evaporated to dryness *in vacuo*, 2 ml of MeOH was added and evaporated to remove boric acid as methyl borate. To the residue, 1 ml of MeOH was added, the precipitate of NaCl was removed and again the solution was evaporated to dryness. The residue was dissolved in 0.5 ml of water and passed through a DEAE–Sephadex column (borate form) (0.8 mm×30 mm) and the column was washed with 10 ml of water. The amino alcohols were eluted with 2 ml of 0.5 N HCl–MeOH and the effluent was evaporated to dryness. The boric acid was removed as described above, and to the residue 0.2 ml of THF and 0.1 ml of TFAA was added at 0° and the solution allowed to stand for 10 min. One to three μl of the reaction mixture was injected to a gas chromatograph.

Gas chromatography was performed on a Shimadzu Model GC–1C gas chromatograph equipped with a hydrogen flame ionization detector. The separation of trifluoroacetates of three hexosaminols was investigated (Table I) and the complete separation was achieved on 2% XF–1105 column (Fig. 1).

Moreover linear calibration curves for galactosamine and mannosamine were obtained by the internal standard method (Fig. 2).

Table I. Retention Times of Trifluoroacetates of Hexosaminols

Stationary phase Column temperature	2% XF–1105 180°	2% QF–1 180°	2% OV–1 105°
Glucosaminol	7.30 min	4.23 min	5.50 min
Galactosaminol	8.28	4.90	6.62
Mannosaminol	9.22	4.46	5.85

column: Glass tube (1.8 m×4 mm *i.d.*) was packed with aech stationary phase on Gas-Chrom P (80—100 mesh).
carrier gas: N_2 80 ml/min sens. 100 range 0.4 V

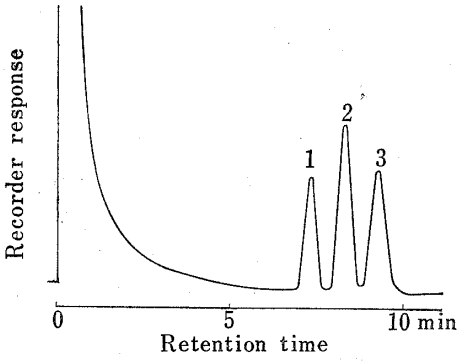

Fig. 1. Separation of Three Hexosaminols as the Trifluoroacetates

peak: 1. glucosaminol 2. galactosaminol 3. mannosaminol
conditions: the same as 2% XF–1105 column in Table I

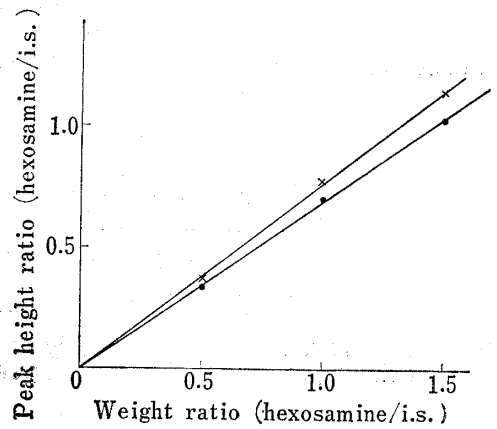

Fig. 2. Calibration Curves for Galactosamine (—×—) and Mannosamine (—•—) Using Glucosaminol as an Internal Standard (i.s.)

This procedure has proved useful in our laboratory for the analysis of hexosamines in mucopolysaccharides and glycoproteins. A complete report will be published in the near future.

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