

14. Ramadevi, B. and Suryanarayana Rao, V., *The East. Pharm.*, 1994, 1075.
15. Sakaguchi, T., Taguchi, K., *Pharm. Bull. (Japan)*, 1955, 3, 166.
16. Chatten, L.G., Krauz, S.I., *J. Pharm. Sci.*, 1971, 60, 107.
17. Sakaguchi, T., Taguchi, K., *Japan Analyst*, 1957, 6, 782.
18. Pradeary, D., *Am. Pharm. Fr.*, 1979, 37, 369.

Comparison of Individual Calibration Curve Method and Single Point Internal Standard Method in the Determination of Carbazole Alkaloids by GLC

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Quantitative determination of Carbazole alkaloids (from Crude plant extracts) was carried out by GLC using individual calibration curve method and a new single point internal standard method and the results compared.

GLYCOSMIS PENTAPHYLLA (family: **G**rataceae), an Indian medicinal plant, has been reported to elaborate carbazole alkaloids^{1,2}. This paper reports comparison of the two GLC methods reported earlier³ to determine the concentration of the alkaloids.

A stock solution of 1mg.ml⁻¹ in chloroform of each of the following carbazole alkaloids was prepared:

1. Carbazole
2. 3-Methylcarbazole
3. Glycozoline
4. Glycozolidine (internal standard)

Appropriate working concentrations were prepared by suitable dilutions of the stock solutions. Heptazolidine (1mg.ml⁻¹) in chloroform was used as internal standard. The plant extract (unknown solution) was prepared by first drawing a drop (23 mg)

from the light petroleum extract with a clean glass rod into a sample bottle. This was then extracted with 5 ml of chloroform. The internal standard was added to the solutions prior to analysis (500 µg.ml⁻¹). All samples were chromatographed on a Pye Unicam Series 204 Gas Chromatograph equipped with Flame Ionization Detector (FID) and a temperature programmer. Calibration curve was prepared by spiking a range of concentration (0-50 µg.ml⁻¹) in increasing order of each of the standard solution with the internal standard and the peak-height ratios were used against concentrations to plot the curves.

In the case of single point internal standard, the following formula 4 [$C_u = (R_u \times C_s) / R_s$] was used.

C_u = Concentration of unknown (µg.ml⁻¹)

C_s = Concentration of standard (µg.ml⁻¹)

R_u = Peak-height ratio of substance in unknown solution

R_s = Peak-height ratio of substance in standard solution

* For correspondence

Statistical analysis was performed using a paired student's t- test between the two methods. Table 1 gives the estimated concentrations of carbazole alkaloids using the two methods.

Table 1 : Estimated Concentrations of Carbazole alkaloids

Sample	Concentrations ($\mu\text{g.ml}^{-1}$)	
	Single Point	Individual Method
1. Carbazole	17.33*	18.00*
2. 3-Methyl Carbazole	186.67*	190.00*
3. Glycozolidine	189.70*	180.00*

* Represents $P < 0.5$ (Student's t-test)

The minimum detectable limit was found to be $5 \mu\text{g.ml}^{-1}$. The Calibration Curves were linear up to $850 \mu\text{g.ml}^{-1}$ with $r > 0.9$.

The concentrations of Carbazole alkaloids determined by the two methods (Table 1) were found to be similar and there was no significant difference between the two methods ($P < 0.5$). However, the single point international standard method was found to be faster and more convenient to use than the individual calibration curve method.

REFERENCES

1. Choudhury, B.K., Mustapha, A., and Bhattacharya, P., *J. Chrom.*, 1985, 42, 555.
2. Choudhury, B.K., Mustapha, A., Garba, M., and Bhattacharyya, P., *Phytochem.*, 1987, 26(7), 2138.
3. Garba, M., Mustapha, A., and Choudhury, B.K., *Ghana J. Chem.*, 1993, 1(7), 349.
4. Supelco In. Bellefonte, P.A., U.S.A., *Bulletin.*, 1977, 768A, 1.