
SHORT COMMUNICATIONS

A SIMPLE AND SENSITIVE METHOD TO MONITOR THE PRESENCE OF DI (2-ETHYL HEXYL) PHTHALATE (DEHP) IN I.V. FLUIDS
A. C. FERNANDEZ, P. V. VEDANARAYANAN and MARY VASANTHA BAI
BMT Wing, Sree Chitra Tirunal Institute for Medical Sciences and Technology, Trivandrum 695 012, India.

Di(2-ethyl hexyl) phthalate (DEHP) is used extensively as a plasticizer in the manufacture of flexible polyvinyl chloride (PVC) plastics. Flexible PVC enjoys wide popularity for the fabrication of containers to store blood, blood products, dextrose and saline solutions used in hospitals. Three million bags were used in USA, alone in 1982¹. These bags are reported to contain 30–40% of DEHP by weight². It is also true that a significant amount of DEHP enters into the patient through transfused blood or saline stored in PVC bags^{3,4}. This amount is reported to vary from 5 to 50 mg per bag in the case of blood bags². Recently toxic potential of DEHP has attracted wide attention¹, evidence for its carcinogenic potential in animals has become available⁵ and proof that it can modulate the immune system of the experimental animal is at hand⁶. These facts indicate an alarming situation when one considers the massive use of fluids stored in PVC bags irrespective of the age and health status of the patient in day-to-day clinical practice.

The above facts prompted us to look for the occurrence of phthalic acid esters in various samples of commercially available sodium chloride injections (IP) and concentrations of DEHP as high as 11 mg/500 ml of saline were detected⁷. Of the two methods used for the estimation of DEHP, the ultraviolet spectroscopic method (UV method) was simple and cheap. But the UV absorbing impurities known to be present in the I.V. fluids often may lead to erroneous data. The separation of DEHP from other impurities prior to the estimation therefore became necessary. Thin layer chromatography could successfully be used as a very sensitive method to identify even the microgram presence of phthalates in a given sample.

DEHP was obtained from Indo-Nippon Co. (Bombay). All other reagents (BDH, India) used were either of analytical or spectroscopic grade and used as received.

One mg and 5 mg of DEHP were dissolved separately in 1 ml each of carbon tetrachloride.

Samples, in varying quantities (5, 10, 15 and 20 μ l) were applied on silica G coated glass plates using a microsyringe. The plate was kept in a closed glass tank containing *n*-hexane and ethyl acetate (9:1 v/v) as carrier. The spots were visualized by keeping the developed and dried plates in iodine chamber for 10 min.

Plates spotted with 1 mg in 1 ml sample showed a faint yellow colour at the 5 μ l (5 μ g) site and strong yellow-yellowish brown at all other sample sites including those applied with 5 mg in 1 ml sample. The minimum quantity (detection limit) for observing the colour was found to be 10 μ g.

Resorcinol-sulphuric acid, *p*-nitroaniline, thymol sulphuric acid⁸, 2% phosphomolybdic acid in ethanol⁹ and other agents have been used for visualization of phthalate. All these techniques involve the use of more than a single chemical, acids and require heat treatment. Though iodine is known to give spots in TLC with practically all organic compounds and especially aromatic ones^{10,11}, this is the first attempt to use it for visualization of DEHP.

16 July 1987; Revised 27 August 1987

-
1. Third annual report on carcinogenes, September 1983, Public Health Service, USA.
 2. Pierre Blais, *Can. Res.*, 1981, 13.
 3. Needham, T. E. and Corlly, *New Eng. J. Med.*, 1973, **94**, 398.
 4. Lawrence, W. H., *Clin. Toxicol.*, 1979, **15**, 447.
 5. Douglas, J. F. and Hartwell, W. V., *Toxicologist*, 1981, **4**, 129.
 6. Dogra, R. K. S., Khanna, S., Megale, S. L. and Shukla, *Indian J. Exp. Biol.*, 1985, **2**, 315.
 7. Rathinam, K., Fernandez, A. C., Vedanarayanan, P. V., Bhujle, V. V. and Sreenivasan, K., *Toxicol. Lett.*, 1983, **15**, 329.
 8. Practice of thin layer chromatography, (ed.) J. C. Touchstone, John Wiley, New York, 1978, p. 204.
 9. Guess, W. H., Jacob, J. and Autian, J., *Drug Intell.*, 1967, **1**, 120.
 10. Brante, *Nature (London)*, 1949, **163**, 651.
 11. Marini Bettoto and Guarins, *Experientia*, 1950, **6**, 309.
-