

CHROM. 16,514

## Note

### A highly sensitive electron-capturing standard for application in high-temperature gas chromatography

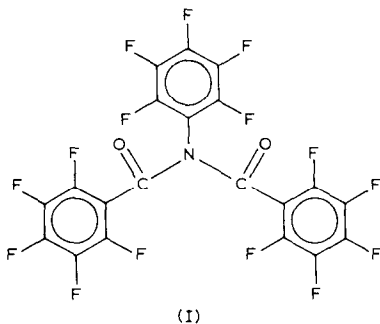
PETR HUŠEK

Research Institute of Endocrinology, Národní třída 8, CS-116 94 Prague 1 (Czechoslovakia)

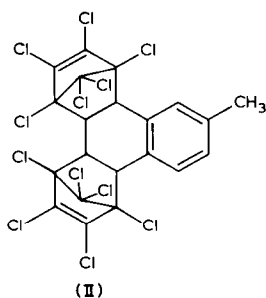
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For the analysis of thyroid hormones at low biological levels by electron-capture gas chromatography (GC-ECD)<sup>1-7</sup>, a convenient internal standard is required. The compound should be a good electrophore (absorbent of thermal electrons), with as many halogen atoms as possible and with a high molecular weight in the range of those of very heavy thyroidal substances.

Several compounds have been tested for this purpose. Sodium lasalocid was used in the determination of thyroxine as its persilylated derivative<sup>1</sup>, but it could be used only with a flame-ionization detector. Efforts have been devoted to producing halogenated thyronines, *e.g.*, 5'-chloro-3,5,3'-triiodothyronine (Cl-T<sub>3</sub>), which unfortunately has a retention time identical with that of reverse T<sub>3</sub> (rT<sub>3</sub>)<sup>2</sup>, and 3,5-diiodo-3',5'-dibromothyronine (Br<sub>2</sub>T<sub>2</sub>), which is eluted between T<sub>3</sub> and rT<sub>3</sub> in a packed column<sup>3</sup> and probably is also coeluted with rT<sub>3</sub> in a capillary column<sup>4-6</sup> (the standard was omitted in those investigations). Corkill *et al.*<sup>7</sup>, in studies of the GC-ECD analysis of thyroid hormones in the form of N,O-di(perfluoroacyl)methyl esters, synthesized a perfluorinated standard, N,N-dipentafluorobenzoylpentafluoroaniline (I). Its relative molar response (RMR) was found to be approximately five times higher than that of lindane<sup>7</sup>. The drawback of using of the three last-mentioned compounds is the necessity to synthesize them in the laboratory.



From the above reasons, our attention was drawn to some pesticide standards offered by Foxboro/Analabs (North Haven, CT, U.S.A.). However, none of the chlorinated naphthalenes tested gave satisfactory results for our purposes, *i.e.*, to function



as possible internal standards in the determination of thyroidal substances treated according to our procedure<sup>8</sup>. Finally, we obtained from Aldrich Europe (Beerse, Belgium) the 2-methylnaphthalene-bis(hexachlorocyclopentadiene) adduct II [MN(HCCP)<sub>2</sub>, Cat. No. 18.489-6]. This compound, serving as a precursor to substituted naphthalenes<sup>9</sup>, is relatively cheap and proved to be also stable under the conditions of high-temperature gas chromatographic (GC) analysis. Its sensitivity towards ECD was investigated by co-injection of known ECD standards (all compounds dissolved in heptane) into a short fused-silica capillary under conditions of temperature-programmed operation (Fig. 1). The molar responses are given in Table I, and it is apparent that the RMR of MN(HCCP)<sub>2</sub>, being approximately five times higher than that of lindane, is comparable to that of compound I<sup>7</sup>. However, the most important finding is that it can be used as an internal standard for the GC determination of thyroid hormones because it fits into the analytical range perfectly<sup>8</sup>.

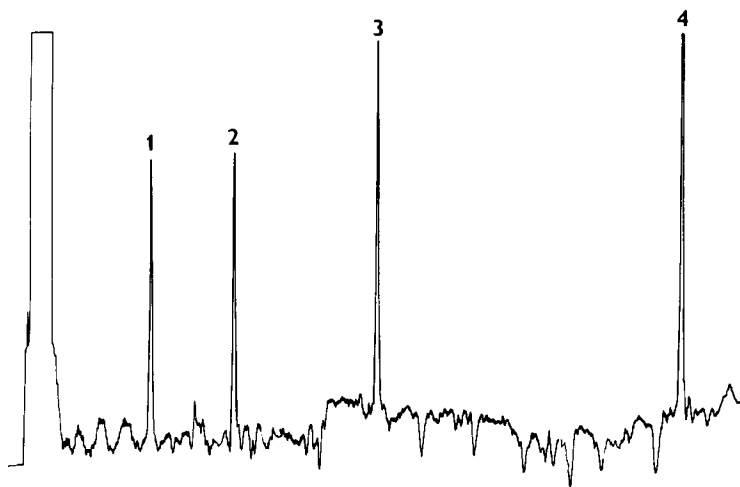


Fig. 1. GC-ECD analysis of 15 pg of each of (1) lindane, (2) aldrin, (3) *p,p'*-DDT and (4) MN(HCCP)<sub>2</sub> on a fused-silica column (5 m × 0.31 mm I.D.) with cross-linked OV-1 stationary phase in the temperature range 80–230°C (8°C/min). The flow-rate of the carrier gas (hydrogen) was held constant by a continual increase in the pressure (20–32 kPa) in order to maintain a constant ratio between the flow-rates of hydrogen and make-up nitrogen (1:5, 24 ml/min in total). Splitless injection (35 sec delay) of 2 μl of the compounds in heptane was performed at 250°C (glass insert of 4 mm I.D.) while maintaining the linear <sup>63</sup>Ni ECD (Hewlett-Packard 5736A gas chromatograph, attenuation × 2) temperature at 300°C.

TABLE I

## GC-ECD RELATIVE WEIGHT AND MOLAR RESPONSES

Reference solute: lindane.

<i>Compound</i>	<i>Molecular weight</i>	<i>Relative weight response</i>	<i>Relative molar response (RMR)</i>
Lindane	291	1.00	1.00
Aldrin	365	1.06	1.33
<i>p,p'</i> -DDT	355	1.45	1.77
MN(HCCP) <sub>2</sub>	688	2.18	5.14

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