Relative Response Factors for Toxaphene Components Using Different Detectors

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1. Introduction

The composition of toxaphene residues in environmental samples and food is very complex. Quantification of this pesticide is very complicate because of the enormous number of toxaphene congeners. A validated method for quantification of the total toxaphene has not been developed yet. Using gas chromatography with electron capture detection (GC-ECD) estimated toxaphene results are influenced by interferences caused by other chlorinated compounds [1]. An overestimation of the true toxaphene level was observed, using the more selective electron capture negative ion mass spectrometry (MS-ECNI)[2]. An other point is the very low response of at least one important toxaphene congener under ECNI conditions in comparison to electron impact mass spectrometry (EI-MS) and GC-ECD[3].

The aim of this work was to investigate the response factors of eighteen toxaphene congeners using different types of detectors. The results of this study are provided the quantification of the total amount of toxaphene in environmentally relevant samples.

2. Experimental

Materials

The toxaphene congeners were isolated and purified at the department of chemistry of the university of St. Petersburg . Structure elucidation was performed by NMR (Bruker, Model AMX 500, 500 MHz)[4]. A solution in isooctane was prepared from all congeners usually containing 5 ng/ul. From these

solutions, two mixtures containing 13 toxaphene congeners each were prepared for this investigation. Table 1 gives an overview of the composition of either mixture. Mixture 2 contains three structurally not fully identified chlorobornanes. In both mixtures the concentration of most chlorobornanes was 500 pg/ul.

Table 1: Composition of chlorobornane mixtures 1 and 2

No	IUPAC name	Mixture 1	Mixture 2
1	2exo,5,5,9,9,10,10-heptachlorobornane		x
2	2endo,3exo,5endo,6exo,8,8,,10,10- octachlorobornane	x	x
3	2exo,3endo,5exo,8,9,10,10-heptachlorobornane	x	
4	2,2,5endo,6exo,8,9,10-heptachlorobornane	x	x
5	2,2,5,5,9,9,10,10-octachlorobornane	x	x
6	2endo,3exo,6exo,8,9,10,10-heptachlorobornane	x	x
7	2,2,3exo,5endo,6exo,8,9,10-octachlorobornane	x	
8	2endo,3exo,5endo,6exo,8,8,9,10,10-nonachlorobornane	x	
9	2exo,3,3,5exo,6endo,9,9,10,10-nonachlorobornane		x
10	2,2,3exo,5,5,9,9,10,10-nonachlorobornane	x	x
11	nonachlorobornane		x
12	nonachlorobornane		x
13	2,2,3exo,5,5,8,9,10,10-nonachlorobornane	x	
14	nonachlorobornane		x
15	2,2,5,5,8,9,9,10,10-nonachlorobornane	x	
16	2,2,3exo,5endo,6exo,8,9,9,10,10-decachlorobornane	x	x
17	2exo,3,3,5exo,6endo,8,9,9,10,10-decachlorobornane	x	
18	2,2,5,5,6exo,8,9,9,10,10-decachlorobornane	x	x

Instrumental

All GC measurements in this study were performed on 60m × 0.32mm × 025µm DB-5 columns in HP 5890 gas chromatographs. The study was carried out with following detection systems:

- a Ni^{63}-ECD operating with nitrogen at 300°C (ECD)
- a double focusing mass spectrometer Finnigan MAT 95 in selected ion monitoring mode using one mass group (MAT 95/1)
- the same double focusing mass spectrometer in selected ion monitoring mode using two mass groups (MAT 95/2)
- a quadrupol mass spectrometer Finnigan SSQ 700 in selected ion monitoring mode using one mass group (SSQ700).

Injections were performed in the split and splitless mode. The GC oven temperature programme used for the entire investigation was as follows: initial temperature 90°C held for 4 min, first ramp to 180°C at 40°/min , second ramp to 250°C at 2°/min and third ramp to 300°C at 5°/min. The splitless injector temperature was 230°C and the GC-MS interface temperature 300°C.
All MS measurements were performed in ECNI mode using ammonia as reactant gas. Temperature of the ion source was 140°C on both systems and the electron energy was 100 eV . The resolution on the quadrupol MS was set to nominal mass resolution. The magnetic sector MS was used in selected ion monitoring mode (SIM) under voltage scan conditions at a resolution of 1000. The masses selected for the SIM technique were on both MS systems: 341, 343, 345, 375, 377, 379, 409, 411, 413, 415, 443, 445, 447. The dwell time for the entire cycle was set to 1 sec.
The covered mass range of the MAT 95/1 SIM procedure (from 341 to 447 AMU) was very large. Due to the extreme reduction of the acceleration voltage in this procedure the high mass signals might have been discriminated. Therefore an additional acquisition series (MAT 95/2) was performed using a SIM system containing two time windows and splitting the mass range into two parts. Heptachlorobornanes and octachlorobornanes were recorded in the first time window containing the ions in range of 341-377 amu as well as nonachlorobornanes and decachlorobornanes in the second time window containing the ions in range of 409-447 amu. This could be performed easily since all hepta- and octachlorobornanes elute earlier from the GC column than the nona- and decachlorobornanes.
The relative response factors (RRFs) of the mass spectrometric results were determined using the total peak area of all selected ions.

3. Results

All solutions of individual chlorobornanes were analyzed by GC-MS in full scan (mass range 60-600 amu) on the MAT 95 under ECNI conditions. The aim of this investigation was to detect any other dominant ions of chlorobornanes apart from the known $[M\text{-}Cl]^-$ and $[M\text{-}HCl\text{-}Cl]^-$ ones which could be used as additional ions for the SIM techniques. No additional dominant ions were detectable. Usually, the base peaks were the mentioned $[M\text{-}Cl]^-$ signals accompanied by some $[M\text{-}HCl\text{-}Cl]^-$ and $[M\text{-}HCl\text{-}HCl\text{-}Cl]^-$ signals. The intensity of the fragment signals varied to some extent depending on the type of the congener.

Since it was reported that the injector type does affect the response of chlorobornanes [5], both mixtures were analysed by GC-ECD using the splitless and the split injection technique (split ratio 1:15). The results of this investigation are listed in Table 2.
No obvious differences were observed between both injection modes. Moreover, almost no differences in between of the response factors of all chlorobornanes examined were found. The range of RRF values in splitless and split injection mode was 0.66-1.61 and 0.81-1.48, respectively. We did not expect any major advantage from the preferred on-column injection technique [5], and performed the remaining injections in the splitless mode.

Table 2: Relative response factors of eighteen chlorobornanes determined with GC-ECD using splitless and split (ratio 1:15) injection technique

Comp.	Relative response factor		Comp.	Relative response factor	
	splitless	split		splitless	split
1	1.24 +/-0.02	1.07 +/-0.01	**10**	1.20 +/-0.02	1.24 +/-0.02
2	1.27 +/-0.03	1.25 +/-0.08	**11**	0.90 +/-0.04	1.33 +/-0.08
3	1.61 +/-0.03	1.28 +/-0.03	**12**	1.38 +/-0.02	1.56 +/-0.02
4	1 (reference)	1 (reference)	**13**	1.05 +/-0.03	1.16 +/-0.03
5	1.11 +/-0.01	1.07 +/-0.05	**14**	0.88 +/-0.01	0.93 +/-0.03
6	1.23 +/-0.03	1.02 +/-0.04	**15**	0.77 +/-0.02	0.87 +/-0.02
7	1.23 +/-0.05	1.48 +/-0.04	**16**	0.77 +/-0.01	0.90 +/-0.02
8	0.95 +/-0.06	1.05 +/-0.01	**17**	0.66 +/-0.01	0.81 +/-0.02
9	1.05 +/-0.03	1.23 +/-0.04	**18**	0.78 +/-0.04	0.86 +/-0.02

Mixtures 1 and 2 as well as a solution of technical toxaphene were injected three times each into the mentioned instruments GC-ECD; GC-SSQ 700; GC-MAT 95/1 and GC-MAT 95/2. The recorded peak areas were used for the calculation of the relative response factors. The calculation was performed on the basis of the reconstructed ion current of compound **4** (2,2,5endo,6exo,8,9,10-heptachlorobornane = Toxicant B). Both mixtures 1 and 2 contained this compound. The ECD and ECNI-MS response of this „reference" chlorobornane **4** was comparable to the total response of the technical toxaphene (using corrections for the injected amounts).

As mentioned above, the ECD response factors of the chlorobornanes investigated do not notably differ. Additionally, the mean response of all chlorobornanes and of technical toxaphene is almost the same. In contrast to the findings of the EC detection, the response of the investigated chlorobornanes shows large differences when using ECNI-MS detection (Figure 1). Results obtained from measurements with the SSQ 700 varied between 2,25 and <0.1. Similar results were obtained from measurements with the magnetic sector MS, whereby the variation of the RRF values was between 5 and 0.001 using one mass range and between 4 and 0.02 using two mass ranges. The compound **15** (2,2,5,5,8,9,9,10,10-nonachlorobornane, one of our proposed indicator compounds [2]) gave a fairly low response. As observed by other authors [3], most of the time $[M\text{-}Cl]^-$ ions of this chlorobornane were hardly detected and $[M\text{-}HCl\text{-}Cl]^-$ signals were recorded as base peaks. Similar results were obtained by Lau [3] for two other chlorobornanes with 2,2,5,5 chlorine substitution in the ring. One of these compounds was the chlorobornane **5** (2,2,5,5,9,9,10,10-octachlorobornane), which did not show the predicted low response in our investigations. Most of the decachlorobornanes tend to give lower responses with the MAT 95 mass spectrometer compared to those toxaphene components containing a lower number of Cl-atoms. As observed in full scan, decachlorobornanes do not form other dominant ions apart from $[M\text{-}HCl\text{-}Cl]^-$ (m/z 375, 377) and $[M\text{-}HCl\text{-}HCl\text{-}Cl]^-$ (m/z 343, 345).

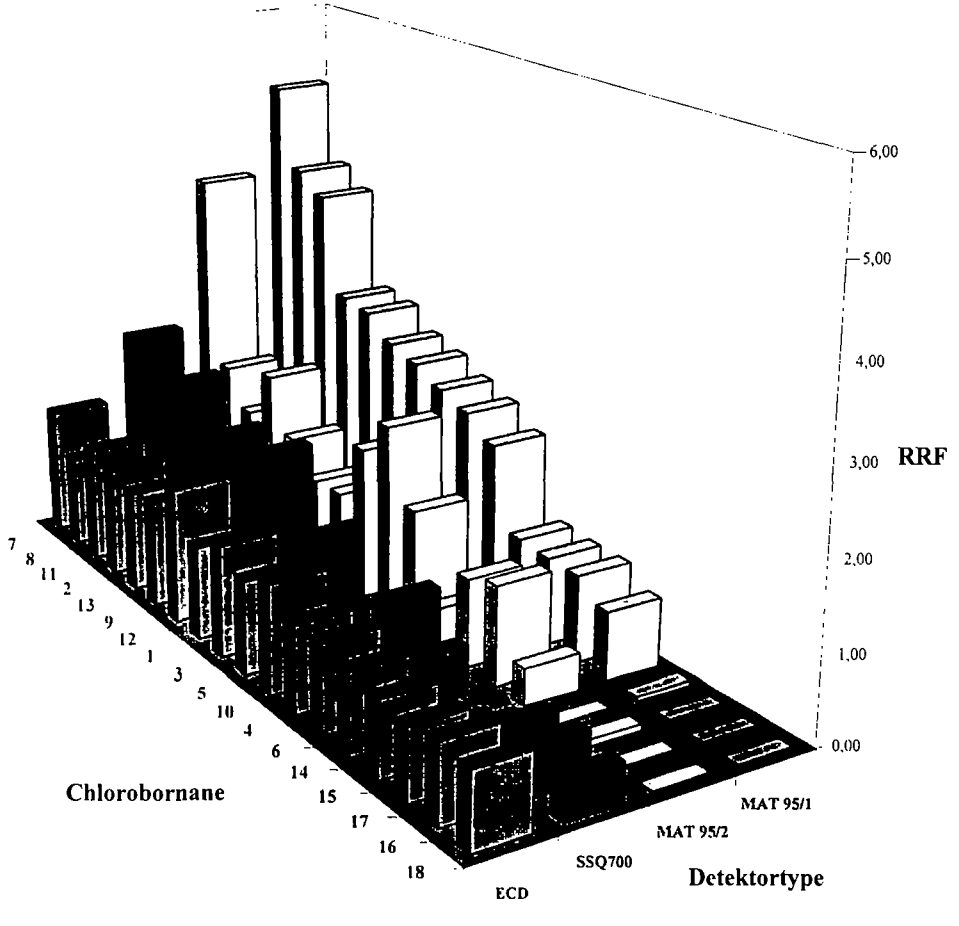

Figure 1: Relative response factors of eighteen chlorobornanes determined with GC/ECD and three GC/ECNI-MS systems.

4. Conclusions

Response factors of chlorobornane acquired under ECNI conditions with different mass spectrometers and SIM procedures differ to a large extend. This shows that the ECNI reaction of certain chlorobornanes depends strongly from source conditions in general and the number of ions recorded in particular.

Since response factors of most chlorobornanes in environmental samples are unknown, the total response of all chlorobornanes in a sample was compared so far to the total response of a toxaphene standard. In this case, the result of the quantification of total toxaphene using GC/ECNI-MS will be strongly affected by chlorobornanes producing high response. Other chlorobornanes with very low response factors will hardly influence the quantitation result, even if they are present in high concentrations. Quantitative results acquired under ECNI-MS conditions will be neither correct nor comparable if not recorded under totally identical conditions.

The best estimation of total toxaphene in environmental samples will achieved by ECD measurements because of the balanced response for all chlorobornanes. Prior to the determination by ECD it is necessary to identify all chlorobornanes by GC-ECNI-MS in the adequate sample. Since the peak sequence is influenced by the vacuum of a direct GC-MS interface an open split interface is required for comparability of the chromatograms [6]. The total amount of residual toxaphene can be calculated on the basis of the total chlorobornane peak area in samples compared to the ECD response of a known concentration of technical toxaphene.

5. References

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