ELSEVIER

Journal of Chromatography A, 705 (1995) 313–317

JOURNAL OF CHROMATOGRAPHY A

Gas chromatographic determination of 5-chloro-2-(2,4-dichlorophenoxy)-phenol in the waste water of a slaughterhouse

Milica Graovac, Miša Todorović, Milan I. Trtanj, Miroslav M. Kopečni, Jožef J. Čomor*

Vinča Institute of Nuclear Sciences, Chemical Dynamics Laboratory, P.O. Box 522, YU-11001 Belgrade, Yugoslavia

First received 22 November 1994; revised manuscript received 7 February 1995; accepted 3 March 1995

Abstract

A straightforward analytical method for the determination of 5-chloro-2-(2,4-dichlorophenoxy)-phenol (commercially known as Irgasan DP 300) in the waste water of a slaughterhouse was developed. The waste water, first cleaned from fats and oils by petroleum ether (b.p. 40–80°C) at pH 11, was extracted by benzene at pH 1 and this Irgasan-containing extract, was further purified by sodium sulfate/silica gel adsorption. After preconcentration and diazomethane derivatization of the extract, capillary GC–ECD determination was applied. The procedure yields a routine recovery of 88.1% regardless of the sample concentration. The lower detection limit for Irgasan in the waste water was found to be 0.2 ng l^{-1}.

1. Introduction

Due to its excellent bactericide properties and very high chemical stability, 5-chloro-2-(2,4-dichlorophenoxy)-phenol (commercially known as Irgasan DP 300) has been widely used in various cosmetic products and as an active component in many commercial disinfectants. One of such disinfectants, coded Albin® DP (product of Albus, Novi Sad, Yugoslavia) has been extensively used in numerous Yugoslav slaughterhouses for the disinfection of working tools.

As pointed out, Irgasan is very stable; it was detected in an industrial waste-water stream following waste water treatment [1], in river water and sediments [2,3], and even in fish tissue [4]. Further, it was confirmed that Irgasan reacts with chlorine in water, producing dichloro- and trichloro-2-(2,4-dichlorophenoxy)-phenols, followed by the decomposition of these intermediates to chlorophenols [5]. As a consequence, the commonly used chlorination of waste water, in order to deodorize and disinfect effluents prior to discharge, will convert Irgasan to no less harmful chlorophenols. Next, incineration of the municipal sewage sludge contaminated with Irgasan represents a potential environmentally hazardous process, because of the ability of chlorinated 2-phenoxyphenols (named predioxines) to undergo ring closure at elevated temperatures, producing extremely toxic polychlorinated dibenzo-*p*-dioxins [6–8].

Since Irgasan DP 300 is a recognized environmental pollutant, we decided to develop a pro-

* Corresponding author.

0021-9673/95/$09.50
SSDI 0021-9673(95)00313-4

cedure for its determination in a very special waste-water stream of a slaughterhouse, being a known source of contamination. Previously reported methods were found to be inadequate for precise quantitative determinations of Irgasan in the slaughterhouse waste waters. A reported GC–MS method [1] for Irgasan identification in waste water was stated to be valuable only for qualitative analysis [1]. Renberg [4] published three methods for Irgasan determination in (a) water, (b) fish tissue and (c) soil, based on anion-exchange preconcentration. Method (a), developed for river water, applied to the more complex slaughterhouse waste water gave far lower values than expected. We believe that the reason is the very high Cl^- content in this waste water (several orders of magnitude higher than the Irgasan concentration) and that the ion exchanger is saturated by Cl^- anions instead of by Irgasan. Methods (b) and (c), assuming a rather complex matrix, could have been adopted to our waste water, but the resulting method would be too complicated and time consuming. Therefore, our strategy for the determination of Irgasan was based on simple and fast extraction procedures.

2. Experimental

2.1. Reagents and reference compounds

All solvents used throughout this work were Merck products, of pro analysi grade. The reference sample of Irgasan DP 300 was granted from Albus (Novi Sad, Yugoslavia). Silica gel, particle size of 0.2–0.5 mm, was a product of Kemika (Zagreb, Croatia) and was dried overnight at 105°C. Diazomethane for derivatization was freshly prepared every week.

2.2. Instrumentation and operating conditions

Gas chromatographic measurements were carried out on a Spectra Physics Model SP7100 equipped with an electron-capture detector operating at 350°C. The chromatographic column was a SPB™-5 (Supelco) fused-silica capillary column: 30 m × 0.32 mm I.D., 1.0 μm film thickness. Nitrogen was used as a carrier gas, the injections were made in the splitless mode and the injector was operated at 250°C. The temperature program was started at 200°C and immediately ramped to 250°C at 20°C/min; then without hold-time ramped further to 300°C at 2.5°C/min and held for 5 min.

2.3. Sample preparation and GC–ECD analysis

The pure organic samples (20–250 ml reference organic solutions or organic extracts after clean-up procedure) were preconcentrated in a Kuderna–Danisch apparatus, then evaporated to dryness applying a slow stream of purified dry air to the liquid surface. The residue was redissolved in 0.1 ml of dry diethyl ether containing 10 ng of hexachlorobenzene as an internal standard, and finally 0.1 ml of diethyl ether saturated with diazomethane was added. Injections were made 5 min after the derivatization and the samples prepared in this way were stable during several days.

3. Results and discussion

3.1. Extraction procedure

Selection of the organic extractants for trace analysis in waste-water streams is critical and, therefore it must be performed carefully. As our goal was to develop a method for determination of Irgasan at a concentration as low as possible, the extractants were not tested for selectivity.

The best conditions for extraction were selected on the basis of the distribution coefficient of Irgasan between water and *n*-hexane, carbon tetrachloride, benzene, cyclohexane and dichloromethane. The distribution coefficients were determined as follows: 25 ml of organic solutions containing 5 $\mu g\ ml^{-1}$ of Irgasan were vigorously mixed for 15 min with the same volume of distilled water the pH of which was previously adjusted to a certain value. The lower pH values were adjusted by adding an 8 *M* H_3PO_3 solution to water, while the higher pH

values were adjusted with 1 *M* NaOH. The use of phosphoric acid for pH adjustments during extraction is recommended in the case of phenols, in order to avoid possible oxidation processes which can be initiated when using other, oxidising, acids [9]. After phase separation, an aliquot of 20 ml of organic phase was analysed as described earlier.

The obtained distribution coefficients are shown in Fig. 1. As expected, the distribution coefficient strongly depends on the pH of the aqueous phase: the higher the acidity, the higher the distribution coefficient (a result of the suppression of the phenolic group dissociation). Among the tested extractants, benzene shows the highest distribution coefficient, allowing the extraction of 90.9% of Irgasan in a single step when the water-phase pH is adjusted to 1.0 and the volume ratio used for extraction is 1:4 (benzene–water). This volume ratio was selected as an optimal one, taking into account the Irgasan recovery and benzene consumption. On the other hand, the low distribution coefficient at high pH values of the water phase allows us to remove efficiently fats, oils and tissue residues by extraction with suitable extractants, thereby reducing the amount of unwanted matrix, without significant loss of Irgasan.

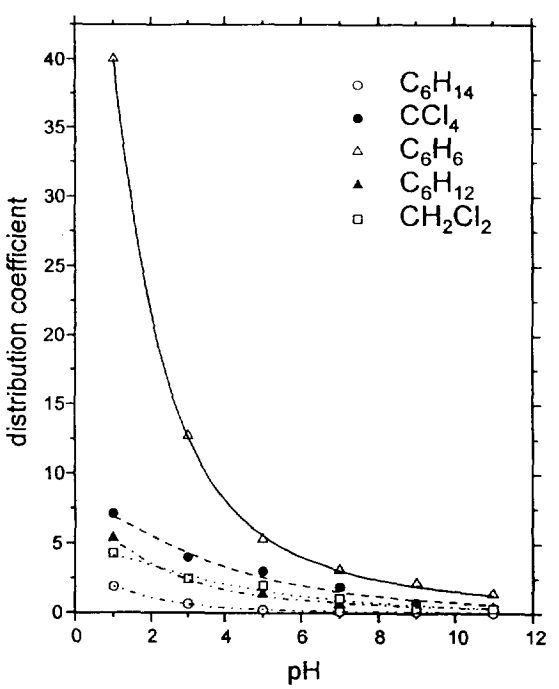

Fig. 1. Experimentally obtained distribution coefficients of Irgasan DP 300 between various organic extractants and water of different pH values.

Petroleum ether is the solvent of choice for cleaning water and solid samples from oils and being a loosely defined mixture of low boiling hydrocarbons its extraction capabilities are well aproximated by hexane. The negligible losses of Irgasan by petroleum ether extraction at pH 11 are corrected by the recovery factor.

3.2. Sample clean-up

Though the concentration of fats and oils can be significantly reduced in the samples by extraction (with petroleum ether at pH 11), this is usually not sufficient, since the fat content in the slaughterhouse waste water very often exceeds several $g\,l^{-1}$. In order to extend the life of the chromatographic columns, another sample clean-up procedure is necessary. A short chromatographic column (1 cm I.D. made from glass), filled with 2 g of anhydrous Na_2SO_4 and 4 g of freshly dried silica gel (0.2–0.5 mm particle size) was used for this purpose. After all the organic extract had passed the column, it was rinsed with 25 ml of benzene and all the eluates were joined. The columns were freshly filled for every sample without recycling the adsorbents to avoid possible sample contamination.

3.3. The optimised procedure and recovery

Based upon the dependence of the distribution coefficient on pH, the following procedure was selected for determination of Irgasan in the waste water: Take 1000 ml of the waste water, adjust its pH to 11.0 by adding 1 *M* NaOH, transfer the sample to a mixing funnel, add 250 ml of petroleum ether and shake vigorously for 15 min. After the phases have been separated, adjust the water-phase pH to 1.0 by adding 8 *M* H_3PO_4, transfer the sample to a mixing funnel, add 250 ml of benzene and shake vigorously for 30 min. Separate the phases and allow the

organic phase to pass through the clean-up column, wash the column with 25 ml of benzene and join the eluates. Transfer the cleaned benzene extract to the Kuderna–Danish apparatus and concentrate it to less than 1 ml, rinse the apparatus with 1 ml of benzene, join the concentrate and the benzene rinse in a 2-ml receiving vessel and evaporate the sample to dryness applying a slow stream of cleaned dry air to the liquid surface. Add 0.1 ml of diethyl ether solution with the internal standard (100 ng ml^{-1} of hexachlorobenzene) and 0.1 ml of diazomethane in ether. After 5 min or more inject 5 μl of the sample into the GC injector.

The recovery of Irgasan was determined by analysing the spiked waste-water samples taken from slaughterhouses not using disinfectants based on Irgasan DP 300. Six groups of spiked samples (three samples each) were prepared resulting in an initial Irgasan concentration of 1, 10, 25, 50, 75 and 100 ng l^{-1}. The concentration of Irgasan in water was adjusted by addition of an adequate amount of acetone containing 50 ng ml^{-1} of Irgasan. The spiked samples were homogenised for 1 h by stirring and then analysed according to the described procedure. Based on the known initial concentration, the recovery of all the samples was calculated and the results are shown in Fig. 2. It is obvious that the Irgasan concentration has no significant effect on the recovery (the calculated slope of the data is 0.003% ng^{-1} l) and an average value of 88.1% (with an S.D. of 1.007) was used for all measurements.

The detection limit of the method described above was determined as the standard deviation of three consecutive analyses of the same spiked sample containing 0.5 ng l^{-1} of Irgasan. It was found that the detection limit of the described procedure for determination of Irgasan DP 300 in the slaughterhouse waste water amounts to 0.2 ng l^{-1}.

Fig. 3 shows two typical chromatograms of a spiked (a) and a real (b) sample from two different slaughterhouses. The chromatographic conditions were optimized to give the best resolution around the Irgasan peak to avoid any possible misinterpretation, other parts of the chromatogram were of no interest to our investigation. A small peak next to the Irgasan peak was observed in each chromatogram of spiked samples and of samples where Irgasan was detected. Identification of this compound would require a GC–MS analysis, which we could not afford. However, we believe that this

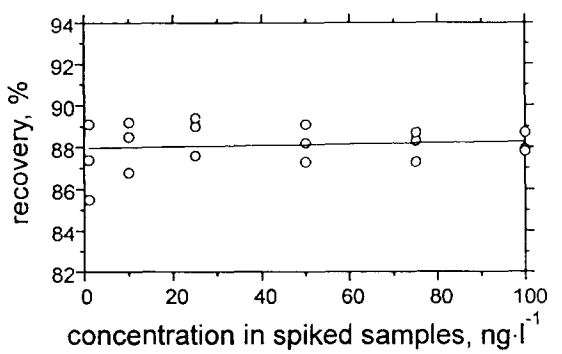

Fig. 2. Recovery of Irgasan DP 300 as a function of its total concentration in spiked samples.

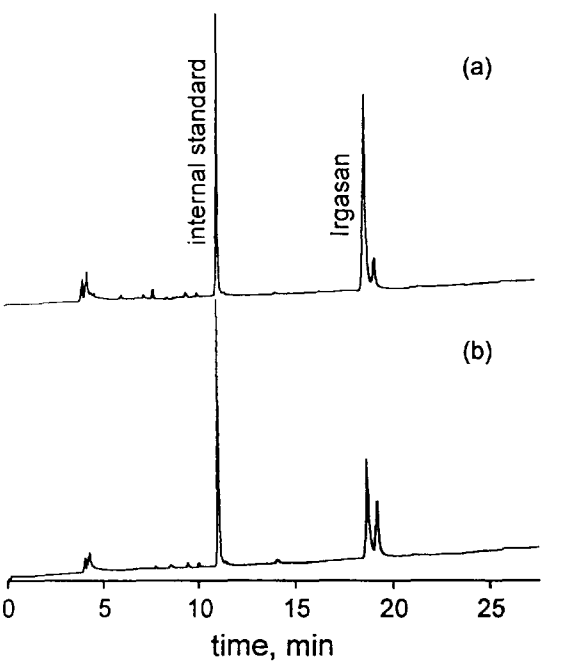

Fig. 3. Typical chromatograms of a spiked (a) and a real sample (b) from different slaughterhouses.

peak comes from an isomer of Irgasan, which is present as an impurity right from its synthesis.

Since the reproducibility of the method (tested by analysing 25 different samples three times) was better than 5%, it was decided to use the developed method in a large scale monitoring study of the waste waters of slaughterhouses in Belgrade and surroundings (these results will be available upon request till the end of 1995).

4. Conclusions

The procedure for the determination of Irgasan DP 300 in the slaughterhouse waste-water stream presented in this paper is carefully developed and optimized in order to get an Irgasan recovery as high as possible, giving very low detection limits, yet being fast and consuming chemicals as little as possible. As has been shown, the Irgasan recovery practically does not depend on the sample concentration in the range from 1 to 100 ng l^{-1}, offering us a reliable method for determination of Irgasan DP 300.

Acknowledgement

The authors are indebted to the Ministry of Science and Technology of the Republic of Serbia for partial financial support.

References

[1] R.A. Hites, V. Lopez-Avila, Anal. Chem., 51 (1979) 1452A.
[2] G.A. Jungclaus, V. Lopez-Avila and R.A. Hites, Environ. Sci. Technol., 12 (1978) 88.
[3] V. Lopez-Avila, R.A. Hites, Environ. Sci. Technol., 14 (1980) 1382.
[4] L. Renberg, Anal Chem., 46 (1974) 459.
[5] S. Onodera, M. Ogawa, S. Suzuki, J. Chromatogr., 392 (1987) 267.
[6] H.G. Langer, T.P. Brady, P.R. Briggs, Environ. Health Perspect., 5 (1973) 3.
[7] C.-A. Nilsson, L. Renberg, J. Chromatogr., 89 (1974) 325.
[8] C.-A. Nilsson, K. Andersson, C. Rappe, S.-O. Westermark, J. Chromatogr., 96 (1974) 137.
[9] M.M. Kopečni, M.V. Tarana, S.D. Čupić and J.J. Čomor, J. Chromatogr., 462 (1989) 392.