Bull. Environ. Contam. Toxicol. (1993) 51:647–650

Environmental Contamination and Toxicology

Hexachlorobenzene and Hexachlorocyclohexane Residues in Pork as Affected by Weight and Sex

A. Ariño, A. Herrera, P. Conchello, and R. Lazaro

Department of Animal Production & Food Science (Veterinary Faculty), University of Zaragoza, c/Miguel Servet, 177, 50013 Zaragoza, Spain

Recent surveys showed a broadly distributed contamination of hexachlorobenzene (HCB) and hexachlorocyclohexane (HCH) residues in human milk and fat tissue in Northeast Spain, ranging from 2 to 4 ppm (mg/kg) on a fat basis (Camps et al., 1989; Ferrer et al., 1992). This area is host to an industry that has been producing the insecticide lindane (γ-HCH) until the middle of 1991. The four main isomers of HCH (α, β, γ, and δ) were detected in all samples of water, soil, vegetation and invertebrates collected in five nearby locations (Hernandez et al., 1991). Additionally, contamination of meat and meat products was also reported by Conchello (1991) and Lazaro (1991).

It has been demonstrated that exposure of animals to excessive amounts of environmental toxicants may decrease the wholesomeness of edible food products due to the presence of hazardous residues (Shull and Cheeke, 1983). Ruiter (1985) reported the accumulation ratios (tissue level to feed level) of some organochlorine pesticides observed in adipose tissues of swine: 8-11 for HCB, and 1-3 for HCH. Studies on the pattern of accumulation of organochlorine residues as affected by sex, age and weight were reported in bovine (Montes et al., 1988) and ovine carcasses (Conchello, 1991). In the present study, in addition to reporting the current levels of contamination in pork, the effect of weight and pig's sex on the chlorinated residual contamination is investigated.

MATERIALS AND METHODS

Thirty pork hind legs (19 from male-carcasses, 11 female) were randomly selected in a food processing plant. The whole pieces were weighed to establish three categories: light (< 9 kg), medium (9 to 11 kg) and heavy (> 11 kg). All pieces came from animals of the same age grown in a farm located in Zaragoza (Spain). Samples were taken by removing about 100 g of meat with a knife, and were passed 3 times through a food chopper, being mixed thoroughly after each grinding. Then, 10 g were taken for residue analysis. The analytical method used was originally described by Telling et al. (1977) and validated for animal fats by the British Ministry of Agriculture, Fisheries and Food (MAFF, 1979).

Briefly, the method involves extraction of organochlorine residues using acetone/hexane (1:4), partitioning into 2% aqueous Na_2SO_4, and drying and concentration of the hexane. Then, a sample extract containing 0.5 g fat in 2 ml of hexane is subjected to cleanup on a single 22-g column of neutral aluminium oxide. Residues are eluted with 150 ml hexane and the eluate concentrated to 2 ml and examined by GLC-ECD.

Send reprint requests to Dr. Herrera at the above address

The gas chromatograph Hewlett-Packard HP 5890 was equipped with an electron capture, Ni^{63} detector, and an HP 3396 A integrator, and was fitted with a glass chromatographic column (6', 1/4'', 2-mm i.d.) packed with 1.5% SP-2250/1.95% SP-2401 on 100/120 Supelcoport. The identity of the residues was confirmed on two more packed columns (3% SP-2100 and 4% SE-30/6% QF1). Operating conditions were: injector 250°C, column 200°C, detector 300°C, carrier gas argon-methane 45 ml/min. The peaks were identified by comparison of retention times with those of the standards on the several columns with different polarities. Residue concentrations in the samples were obtained by comparing peak areas from samples with those recorded for the standards. Recovery of pesticides at 10-ppb level was above 85%. Results are expressed as ppb (μg/kg on a fat basis), being 4 ppb on a fat basis the detection limit.

Mann-Whitney's and Kruskall-Wallis's nonparametric tests used for statistical analysis were designed according to Sachs (1978).

RESULTS AND DISCUSSION

Table 1 summarises the results by weight. Both HCB and lindane were detected in all samples, whereas α-HCH was present in 60-70% of pieces. The mean levels of HCB and α-HCH weakly increased with the weight of the pig's leg unlike the insecticide lindane which mean levels decreased in heavy pieces. One piece showed a level of α-HCH (206 ppb) above the MRL set up by the EEC for that isomer (200 ppb), and another sample reached 197 ppb. Maximum levels of HCB and lindane were below the MRL, being about 5-10 times smaller than the respective limits of 200 ppb for HCB and 1000 ppb of lindane.

Statistical analysis by Kruskall-Wallis's nonparametric test did not show significant differences ($p>0.05$) among pieces due to weight, so it may be concluded that this factor did not influence the level of HCB and HCH contamination. Conchello (1991) reported similar results in a study on ovine carcasses where HCB and α-HCH levels were not significantly different among pieces belonging to three weight categories. However, lindane concentration was significantly higher ($p<0.05$) in light ovine carcasses. Investigations carried out on bovine carcasses by Montes et al. (1988) showed no significant differences for the pattern of residue accumulation among different categories, being lactating cows less contaminated since they excrete a considerable amount of these compounds by way of the milk.

The effect of sex was more homogeneous since the three pesticides under study were less concentrated in female than in male pieces (Table 2). The effect is more pronounced with the α-HCH which mean levels averaged 54±16.7 ppb in male pieces over 8±2.2 ppb in female ones.The maximum levels of HCB, α-HCH, and lindane were also detected in male pieces. However, that differences were not statistically significant by Mann-Whitney's nonparametric test even though the level of significance for α-HCH is almost 0.05. In general, the fat content of porcine female pieces is higher than that of male ones (Bout and Girard, 1988). Therefore, the organochlorine contamination could be more diluted in female pieces due to the higher amount of fat.

Similar results were reported by Conchello (1991) in ovine where HCB and α-HCH levels in female carcasses were much smaller than those detected in male ones, while lindane showed quite the reverse. Again, there was not significant difference in either case.

Montes et al. (1988) also reported no significant differences considering the number of samples above the MRL between male and female bovine carcasses.

Table 1. Organochlorine residues in pork (μg/kg fat basis) related to the weight of pig's hind leg.

Pesticide	Weight	n	% Detected	Mean±SE	Maximum	"p" value
HCB	Light	10	100%	11 ± 1.7	23 ppb	p > 0.05
	Medium	10	100%	14 ± 2.1	25 ppb	
	Heavy	10	100%	15 ± 1.8	23 ppb	
α-HCH	Light	10	70%	33 ± 16.5	132 ppb	p > 0.05
	Medium	10	60%	25 ± 19.2	197 ppb	
	Heavy	10	70%	53 ± 23.3	206 ppb	
Lindane	Light	10	100%	47 ± 26.9	284 ppb	p > 0.05
	Medium	10	100%	58 ± 28.7	231 ppb	
	Heavy	10	100%	27 ± 6.8	80 ppb	

Kruskall-Wallis's nonparametric test is applied to seek differences among weight categories.

On the other hand, several studies done in Europe and USA on human fat contamination by organochlorine residues do not indicate significant differences between man and woman, even though some disparities remain to be fully explained (Ferrer et al., 1992).

Overall, no significant differences owing to sex and weight in the amount of organochlorine residues were discovered, so pieces from male and female carcasses belonging to three weight categories showed a similar degree of contamination. Although the organochlorine pesticides accumulate in the body lipids, this does not always mean that levels in animals are strictly related to the fat content. Nevertheless, the fat content of meat determines to an important extent the amount of these compounds ingested by the consumer.

Table 2. Organochlorine residues in pork (μg/kg fat basis) related to pig's sex.

Pesticide	Sex	n	% Detected	Mean±SE	Maximum	"p" value
HCB	Male	19	100%	15±1.4	25 ppb	p > 0.05
	Female	11	100%	11±1.8	23 ppb	
α-HCH	Male	19	68.4%	54±16.7	206 ppb	p > 0.05
	Female	11	63.6%	8±2.2	20 ppb	
Lindane	Male	19	100%	57±19.9	284 ppb	p > 0.05
	Female	11	100%	21±5.5	65 ppb	

Mann-Whitney's nonparametric test is applied to seek differences between sexes.

Acknowledgments. Dr. Ariño was supported by a Research Scholarship from the Spanish Ministerio de Educación y Ciencia. This research project was financed in part by the University of Zaragoza (Spain).

REFERENCES

Bout J, and Girard JP. (1988). Lipides et qualités du tissu musculaire: facteurs de variation. XX Journees de la Recherche Porcine en France, p. 88.

Camps M, Planas J, Gómez-Catalán J, Sabroso M, To-Figueras J, and Corbella J. (1989). Organochlorine residues in human adipose tissue in Spain: study of an agrarian area. Bull Environ Contam Toxicol 42: 195-201.

Conchello P. (1991). Organochlorine residues in ovine carcasses and meat, and effect of several culinary processes on the residual contamination. Doctoral Thesis. University of Zaragoza, Spain.

Ferrer A, Bona MA, Castellano M, To-Figueras J, and Brunet M. (1992). Organochlorine residues in human adipose tissue of the population of Zaragoza (Spain). Bull Environ Contam Toxicol 48: 561-566.

Hernandez LM, Fernandez MA, and Gonzalez MJ. (1991). Lindane pollution near an industrial source in Northeast Spain. Bull Environ Contam Toxicol 46: 9-13.

Lazaro R, Herrera A, Conchello MP, and Ariño A. (1991). Organochlorine pesticide residues in fresh sausages. Anal Bromatol 43: 373-382.

MAFF. (1979). Ministry of Agriculture, Fisheries, and Food. Determination of residues of organochlorine pesticides in animal fats and eggs. Analyst 5: 425-433.

Montes L, Lic SP, Tamayo R, Pinto M, and Cristi R. (1988). Chlorinated pesticide residues in slaughtered cattle of the IXth and Xth regions classified according to categories. Arch Med Vet 20: 126-134.

Ruiter A. (1985). Contaminants in meat and meat products. In: "Developments in meat science - 3". Ralston Lawrie (Ed.). Elsevier Applied Science Pub.

Sachs L. (1978). Estadistica Aplicada, 1st ed., Labor, Barcelona.

Shull LR, and Cheeke PR. (1983). Effects of synthetic and natural toxicants on livestock. J Ani Sci (Suppl. 2) 57: 330-354.

Telling GM, Sissons DJ, and Brinkman HW. (1977). Determination of organochlorine insecticide residue in fatty foodstuffs using a clean-up technique based on a single column of activated alumina. J Chromatog 137: 405-423.

Received October 1, 1992; accepted April 20, 1993.