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POLLUTION AND ITS EFFECTS ON OTTER POPULATIONS IN SOUTH-WESTERN EUROPE

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1 INTRODUCTION

The case of the otter (*Lutra lutra*) has been one of the clearest examples of effort to research and conserve an animal species in the 20th century. This resulted from the dramatic decline in numbers and restriction of range that occurred during the second half of the 20th century (Figure 1). During the 1970s and 1980s the disappearance of the species was confirmed in most of Western Europe (REUTHER, 1980; FOSTER-TURLEY, MACDONALD and MASON, 1990; MACDONALD and MASON, 1994).

The causes of this decline in most of these territories were: (1) direct persecution (MASON and MACDONALD, 1986), although the otter also disappeared in places where there was little persecution, (2) the transformation of its habitats (resting sites, breeding and sheltering places), and (3) also changes in the availability of the species upon which the otter fed (see summary in MASON and MACDONALD, 1986 and FOSTER-TURLEY, MACDONALD and MASON, 1990). However, this did not explain why this mustelid had also disappeared from catchments where the habitats had apparently not been modified and, in most cases, it neither included prey populations that had been affected such as those of our study areas. These include, for example, regions of both sides of a large area of the Pyrenean mountains, the Alps, the Sierra Nevada and coastal wetlands such as the Ebro and Po Deltas, the Camargue or Albufera of Valencia.

The otter was not the only animal to be drastically affected in those years. In Western Europe, some other predators also showed sudden declines and fell to their lowest distribution levels. The first indications of decline were found in species like the peregrine falcon (*Falco peregrinus*) (NEWTON, 1979). The most affected animals were, however, insectivorous and fish-eating species such as the osprey (*Pandion haliaetus*), and various species of *Ardeidae* (*Ardea purpurea,* for example). The otter's decline, as well as that of these other species, was considered to be the result of a group of cumulative or synergistically-acting causes.

Pollution and epizootic occurrences were suggested as possible causes of decline (OLSSON and SANDEGREN, 1983; MASON, 1989), as they were the latest and the most consistent with the simultaneous disappearance of otter populations and other species.

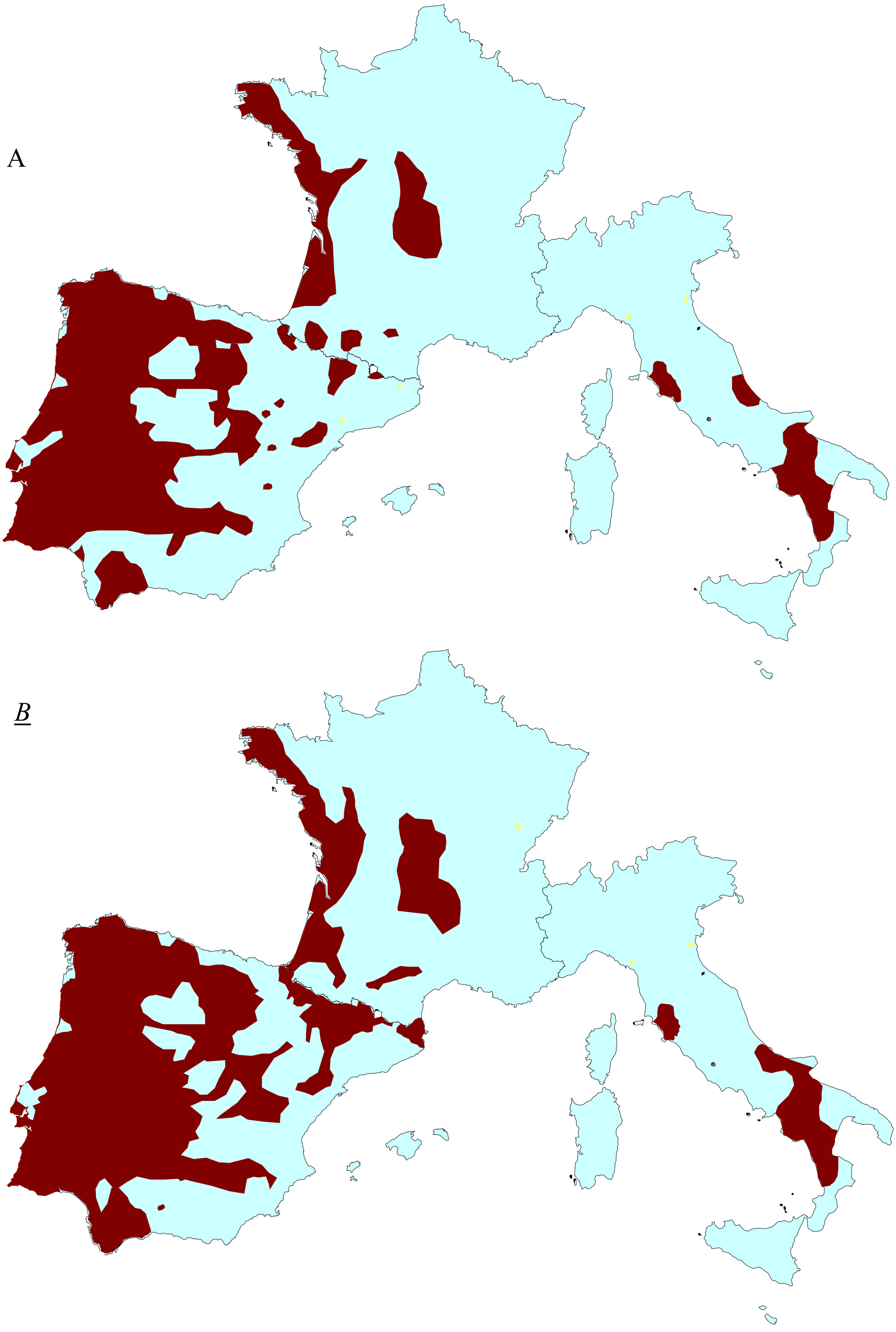


Figure 1. Distribution of the otter in SW Europe: a) 1980-85 and b) 1995-2000 (GREEN & GREEN, 1981; MACDONALD & MASON, 1982, 1983; ELLIOT, 1983; SANTOS-REIS, 1983; DELIBES & CALLEJO, 1983; BOUCHARDY, 1986; CASSOLA, 1986; DELIBES, 1990; ROSOUX *et al.*, 1996; PRIGNIONI, 1997; RUIZ-OLMO & DELIBES, 1998; TRINIDADE *et al.*, 1998).

In order to assess the impact of pollution and to explain the decline of the otter populations, as well as endowing managers with conservation measures, several research projects were begun in the 1980s. This paper is a synthesis of the current knowledge on the effect of pollution on otter populations and their principal prey in South-west Europe. At the same time, an analysis of the effect that pollution dynamics and its control could have on the evolution in the distribution of this species was carried out.

2 STUDY AREA

The zone under consideration includes France, Italy, Portugal, Spain, Andorra, Monaco and San Marino. In the remaining countries of continental Western Europe (Netherlands, Belgium, Luxembourg, Switzerland and West Germany) otters disappeared, with only small populations surviving in the north of Germany (MACDONALD and MASON, 1994). The area covered in this paper is nearly 1,418,450km^2, and is bounded by the Atlantic Ocean and the Mediterranean Sea. This determines the climate notably, with a different climate in the Mediterranean area in the south (dry, especially in summer), in the Atlantic area to the west and north (wet). There is also a continental trend in the higher zones; the maximum altitudes of which are in the Alps (4,807m), the Pyrenees (3,404m) and the Sierra Nevada (3,478m). This determines a strong altitudinal, microclimatic and biogeographical gradient. The Atlantic and continental hydrographic networks are dense in watercourses with more or less permanent river flow. In the Mediterranean area, watercourses have a tendency to be of low density and of medium or low flow (or even dry), with minima in summer. Such rivers often show catastrophic flows, especially in autumn or spring, and have a great capacity for movement and cleaning of the river bed.

Temperatures are higher in Mediterranean rivers and lakes, especially during the summer. This means that great rates of metabolism could occur that in extreme cases determine the occurrence of anoxy situations.

All these differences could be important in pollution tolerance capacity or could mean its dilution, with the situation in Northern and Central Europe being different from that in Southern Europe, and also with differences in Southern Europe itself.

3 OTTER DISTRIBUTION 1980-85

Formally, the otter was widespread in most of aquatic European ecosystems up to the first half of the 20th century. Between these dates and the beginning of the 1980s a dramatic decline took place in Western Europe, and resulted in this species becoming extinct in some areas and/or restricted to small areas of previously occupied zones (MACDONALD and MASON, 1994). Otter surveys, based on the search for indirect signs of the species' presence (spraints [faeces] and tracks) (MASON and MACDONALD, 1986), allowed for an improvement in the precision and standardisation of the study of its distribution. Thereby, the distribution of the otter in our study areas has been described by several authors (Fig. 1a) (GREEN and GREEN, 1981; MACDONALD and MASON, 1982, 1983; ELLIOT, 1983; SANTOS-REIS, 1983; DELIBES and CALLEJO, 1983; BOUCHARDY, 1986; CASSOLA, 1986; DELIBES, 1990). The otter had disappeared from Andorra, where it was cited before (RUIZ-OLMO and GOSÁLBEZ, 1988), and other small states. In France there were only stable populations in the Departments on the Atlantic Ocean to the south of Normandy, including Normandy and the Massif Central, with isolated cores in the adjacent zones. In Italy, the otter had practically become extinct, having been reduced to some rivers in the south of the country (it was only found in 8.2% of the surveyed points in the southern half), and two rivers to the north of Rome (Fiora and Farma-Merse). There were also some isolated individuals in other zones near these. In

Portugal, the otter was distributed throughout the country (found at 70% of the survey sites). Finally, in Spain the otter was found in 40% of the surveyed sites in 1981 and in 33% in 1984-85. Otters were more abundant in the western half of the country, being absent from the more industrialised zones and the big cities and their surroundings and from the main intensive agriculture areas.

The process of decline has been well reported in various areas, mainly occurring between 1950 and 1985 (RUIZ-OLMO and GOSÁLBEZ, 1988; LODÉ, 1993; ROSOUX, TOURNEBIZE and MAURIN, 1996).

4 OTTER DISTRIBUTION AND POLLUTION: THE OTTER AS A BIOINDICATOR SPECIES

As causes of decline, various factors have been suggested that could affect *L. lutra*. These include: persecution; habitat changes; disturbance; decrease in food and water availability or illness, although in the last 15 years the possible role of pollution has grown in importance as a global factor that could explain this simultaneous decline, in a relatively short period of time (OLSSON and SANDEGREN, 1983; MASON and MACDONALD, 1986; MASON, 1989, 1997).

The effect of the pollution on the otter's distribution in our study areas has been demonstrated by several works (ADRIÁN, WILDEN and DELIBES, 1985; RUIZ-OLMO, 1985; LAFONTAINE, FORTUMEAU and MAINSANT, in press; RUIZ-OLMO, LÓPEZ MARTÍN and DELIBES, 1998). These authors found otters at significantly lower frequencies in many polluted areas.

For this reason, the otter has been suggested as a good bioindicator species of water quality and riparian habitat conservation, because of its sensitivity to pollution, to the transformation of its habitats and to the changes in the availability of prey species. RUIZ-OLMO *et al.* (1998) compared the distribution of the otter with those of the main orders of macroinvertebrates and the most-used indices: BMWP (Biological Monitoring Working Party) and ASPT (Average Score per Taxon) (HELLAWELL, 1978; ARMITAGE, 1983) in order to test if both behave in the same way (macroinvertebrates are commonly used as bioindicators of contamination and of quality of water; see VALENTINE, 1973; WESTMAN, 1978). According to these results, in northeast. Spain, otters act as a pollution bioindicator species. However, they found differences related to the biological, ecological and ethological characteristics between otter and macroinvertebrates. The use of the otter as a bioindicator thus needs revision. Invertebrates, otters and, surely, other species could be used as bioindicators in a complementary fashion.

5 LEVELS OF ORGANOCHLORINES AND HEAVY METALS IN OTTER TISSUES

What types of pollutants are therefore affecting otters?. A wide range of different pollutants could affect them directly or through their prey (MASON, 1989). Among these, organochlrine compounds and heavy metals should be highlighted (OLSSON and SANDEGREN, 1983; MASON, 1989, 1997; MASON and MACDONALD, 1986). MACDONALD (1991) linked pollutant distribution, in particular the polychlorinated biphynels (PCBs), to wind circulation (that transports micropollutants subsequently deposited by rain), the distribution of the main sources of pollution and the distribution of the otter. Otters have healthy populations in the territories next to the Atlantic Ocean or further south, away from these pollutant carrying winds. These tend to be located more to the west (Portugal, Galicia, Extremadura, Western France, Ireland, Scotland, Norway and an area of Denmark).

In Table 1 the levels of organochlorine compounds and heavy metals found in the muscular tissues of otters from France and Spain are shown. The levels of PCBs are the more elevated in all cases. However, high levels of some pesticides were found in Spain (mainly DDTs in the south, an average of nearly 80mg/kg: lipid weight) and also mercury in France (although the geometric mean is not very high, 4.71mg/kg), with a maximum of 41mg/kg, whereas the threshold of 30mg/kg has been proposed, beyond which neuronal damages are produced in mammals (GUTLEB, 1995). Locally, there are high levels of most of the analysed compounds (especially some heavy metals, oxychlordane or lindane), but these are not related to a possible decline of the species on a national/ international scale.

For these reasons, PCBs should have the greatest effects on otter survival from the studied regions, although locally other pollutants could have more influence and, in general, the effects should be added.

The arithmetic means of PCB levels (between 6.15 – 33.94mg/kg: lipid weight) are lower in three of the four studies than the threshold of 50 mg/kg, a level proposed as an upper limit for the presence and distribution of the otter (see MASON and MACDONALD, 1993). This is based on the results from JENSEN *et al*. (1977) for American mink (*Mustela vison*). They are also lower or similar to the 30mg/kg for the same species proposed by LEONARDS *et al.* (1994) and SMIT *et al*. (1996). The exception concerns data from our Spanish sample collected during the period 1981–1993 and from Doñana (1982–1983). The arithmetic mean of the whole country data (78.30mg/kg: lipid weight) exceeded both thresholds. However, the sample from the whole of Spain is very heterogeneous due to the geographical amplitude and difference in uses between zones; in the second case the sample is too small. In fact, RUIZ-OLMO, LÓPEZ-MARTÍN and DELIBES (1998) highlight important differences between the diverse regions (greater contamination in the southwest than in the north of the country), and even between river basins. In France, there are also differences between basins (LAFONTAINE, 1995). The presentation of results as arithmetic means could mask the real situation, since extreme values (of up to 1005mg/kg in South-west Spain) can affect the average values. For this reason LAFONTAINE (1995), following the arguments of NEWTON and WYLLIE (1992) and SMIT *et al*. (1994), used the geometric mean for his samples from France, while using both means as part of a pan-European comparative synthesis (see also Table 1). In Spain, only 28% of the otters analysed in the period 1981-1993 exceeded the threshold of 50mg/kg (RUIZ-OLMO, LÓPEZ-MARTÍN and DELIBES, 1998), being almost the same (29%) as in French otters from 1987-1995 (according to the sample studied by LAFONTAINE, 1995).

Table 1. Arithmetic ([a]) and geometric ([g])mean levels (mg/kg lipid basis in muscular tissues) of organochlorine compounds and heavy metals (mg/kg dry weight in liver) of otters from France and Spain (the range is given between brackets, except for France b: standard deviation). For Spain a (Doñana), levels are expressed as mg/kg wet weigh), and heavy metals were analysed in muscular tissue. Underlined values are expressed as wet weigh.

	France a (5 regions) (*n* = 8 to 22). Mostly 21 or 22	**France b (Western Marshes, 3 regions) (*n* = 32)**	**Spain a (Doñana) (*n* = 5)**	**Spain b (whole) (*n* = 41: *n* = 19 for heavy metals)**	**Spain c (whole) (*n* = 10)**
PCBs	33.94[a] 13.99[g] (1.24 - 145.31)	26.19[a] (± 21.74)	<u>2.44</u> [a] (<u>2.40</u>- <u>2.45</u>)	78.30[a] 25.06 g (1.49 - 1005.59)	6.15 [a] (n.d. – 20.60)
DDTs	1.10[a] 0.38[g] (0.01 - 6.08)	(not detailed)	<u>3.50</u> [a] (<u>2.25</u> - <u>5.62</u>)	14.79[a] 5.78 [g] (0.19 - 82.95)	1.28 [a] (0.38 – 2.97)
Oxychlordane	0.55[a] 0.28[g] (0.002 - 1.95)	-	-	-	-
BHC	0.17[a] 0.08[g] (0.005 - 0.67)	0.08[a] (± 0.07)	-	-	-
HEPO	0.124[a] 0.068[g] (0.009 - 0.546)	0.003[a] (± 0.010)	-	0.26[a] (n.d. - 1.53)	-
Dieldrin	0.70[a] 0.33[g] (0.12 - 2.91)	0.75[a] (± 1.45)	-	-	-
Aldrin	-	0.02[a] (± 0.07)	-	0.24[a] (n.d. - 5.84)	-
Lindane / HCH-γ	0.47[a] 0.10[g] (0.01 - 3.27)	0.18[a] (± 0.46)	0.02 [a] (0.01 -0.02)	1.95[a] 2.50 [g] (0.03 - 9.92)	-
Hg	9.46[a] 4.71[g] (0.50 - 41.00)	-	1.33 [a] (1.25 - 1.41)	0.99[a] (n.d. - 2.80)	-
Pb	0.55[a] 0.42[g] (0.12 - 1.58)	-	0.64 [a] (0.51 - 0.80)	0.09[a] (n.d. - 0.34)	-
Cd	0.35[a] 0.17[g] (0.01 - 2.03)	-	0.13 [a] (0-10 - 0.17)	0.04[a] (n.d. - 0.22)	-
Cu	28.78[a] 26.00[g] (11.00 - 53.00)	-	-	-	-
As	0.070[a] 0.055[g] (0.02 - 0.20)	-	-	-	-
Cr	1.51[a] 0.63[g] (0.07 - 4.90)	-	-	0.17[a] (n.d. - 0.49)	-
Ni	2.80[a] 1.51[g] (0.10 - 4.34)	-	-	-	-
Zn	77.33[a] 77.00[g] (70.50 - 90.70)	-	-	-	-
Reference	Lafontaine, 1995	Tans *et al.*, 1995	Hernández *et al.*, 1985	Ruiz-Olmo *et al.*, 1997	Ruiz-Olmo & López-Martín, unpublished
Period	1987-1995 except fpr one idividual in 1981	1987-1994	1982 -1983	1981-1993	1994-1999

On the other hand, the most recent studies on the effect of the pollutants on the American mink (KIHLSTRÖM *et al*., 1992; LEONARDS *et al*., 1994) have shown that the effect of PCBs on reproduction is progressive and asinthothic. Some American mink can breed with low success even with high pollutant levels in tissues. For this reason it seems appropriate to carefully consider the use of such static thresholds, especially when remembering that resistance to these levels could be variable in each species, according to the individuals and/or the populations (SMIT *et al*., 1994). Organochlorine compounds can also be eliminated by female otters during gestation and weaning periods and by glandular secretions in both sexes. In addition, the body condition and the mobilisation of fat reserves and intoxication of the organism for these lipophilic pollutants must be considered in each case.

RUIZ-OLMO, LÓPEZ-MARTÍN and DELIBES (1998) found no correlation between the levels of any organochlorine compound or heavy metals, and the age of the otters (neither as a whole, nor by sex). This negative result could be affected by the great heterogeneity of the environments and to the great geographical dispersion of samples. LAFONTAINE (1995), found no significant correlation between the age and the levels of several organochlorine compounds (samples also originating from different zones). However, he found a positive significant correlation between the age of the otters and the levels of some heavy metals (mercury: $r^2 = 0.988$: $p < 0.09$ in liver; cadmium: $r^2 = 0.994$: $p < 0.05$ in kidney; lead: $r^2 = 0.976$: $p < 0.13$ in kidney).

6 CORRELATION BETWEEN POLLUTION LEVELS AND OTHER VARIABLES

In Table 2, the functions between the levels of main pollutants are shown. In the case of significant results, the high correlation found, allows us to use, for discussion, a single tissue of the most representative compounds (in this case, the PCBs).

Table 2. Correlation between the body condition index (KRUUK et al., 1987; LAFONTAINE, 1995; RUIZ-OLMO, 1995) and levels of several pollutants in otter tissues from France and Spain (after LAFONTAINE, 1995, and RUIZ-OLMO, LÓPEZ MARTÍN and DELIBES, 1998). (*) significant differences.

		France	Spain
PCBs	**Muscle**	$r = 0.580$ $p < 0.05$*	n.s.
	Liver	$r = -0.501$ $p < 0.05$*	n.s.
HCB	**Muscle**	$r = -0.498$ $p < 0.05$*	n.s.
	Liver	$r = -0.680$ $p < 0.01$*	n.s.
Cu	**Liver**	$r = -0.674$ $p < 0.01$*	n.s.
Hg	**Liver**	n.s.	$r = 0.561$ $p = 0.19$

LAFONTAINE (1995) found a significant correlation between the body condition index, *k* (KRUUK, CONROY and MOORHOUSE, 1987), for French otters (LAFONTAINE, 1995) and the levels of some pollutants (Table 3). However, RUIZ-OLMO, LÓPEZ-MARTÍN and DELIBES (1998) found no similar significant correlation. Again, the geographical dispersion of the samples and their heterogeneity could explain the lack of correlation for this study.

Table 3. Functions, correlation coefficients and signification between the levels of some pollutants in the otter tissues from France and Spain (after LAFONTAINE, 1995 and RUIZ-OLMO, LÓPEZ MARTÍN and DELIBES, 1998). First pollutant is *y* and second *x*. (*) significant differences.

y / x	France	Spain
PCBs liver / PCBs muscle	log y = 0.967 log x + 0.012 $r = 0.57$: $p < 0.01$	log y = 0.758 log x + 0.441 $r = 0.81$: $p < 0.0001$
DDTs liver / DDTs muscle	log y = 0.426 log x + 1.729 $r = 0.50$: $p < 0.05$	log y = 0.667 log x + 0.585 $r = 0.65$: $p < 0.0001$
PCBs / DDTs (muscle)	$p > 0.25$	log y = 0.475 log x + 1.026 $r = 0.55$: $p < 0.0003$
PCBs / DDTs (liver)	$p > 0.25$	-
DDTs (muscle) / Hg (liver)	log y = 0.599 log x + 2.136 $r = 0.44$: $p < 0.05$	$p < 0.05$
DDTs (liver) / Hg (liver)	$p > 0.25$	$p < 0.05$

LAFONTAINE (1995) also found a positive correlation ($p < 0.02$) between the levels of lindane (γ-HCH) in the muscle of individual otters from Brittany, France, and the rate of maize culture (on a local scale), where lindane was widely used. A similar case could be found in South-west Spain, with a large agricultural area and higher use of some pesticides and where the levels of some pesticides (DDTs, lindane, etc.) were greater than those used in the north (RUIZ-OLMO, LÓPEZ-MARTÍN and DELIBES, 1998).

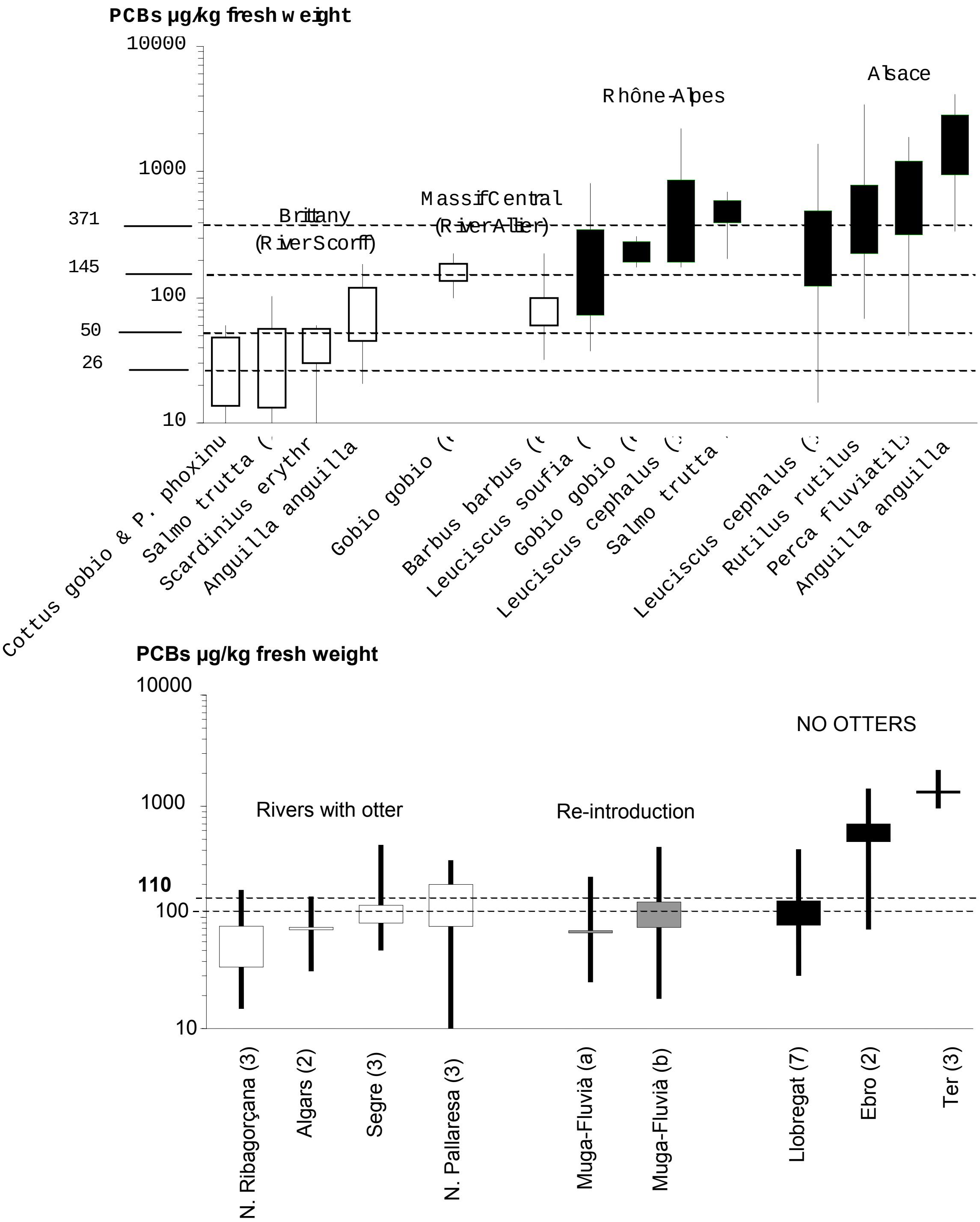
7 LEVELS OF ORGANOCHLORINES AND HEAVY METALS IN TISSUES OF FISH AND CRAYFISH AS OTTER PREY

The accumulation of organochlorines in otter tissues could only come from food, specifically fish, the main diet of the species. RUIZ-OLMO and LÓPEZ-MARTÍN (1994) found that the populations of otters from Catalonia (North-east Spain) were distributed in zones with less than the average level (arithmetic mean) of 0.1mg/kg: wet weight of PCBs in the muscle of fish (referring to the group of the fish consumed by the otter in each site).

LAFONTAINE and DE ALENCASTRO (2000) found quite a close relationship between the levels of PCBs in fish from different regions of France (Brittany, Massif Central, Rhône-Alpes and Alsace) and otter occurrence (Figure 2) (also see MICHELOT *et al*., 1998).

After these findings, the critical thresholds for contamination effects on fish could be between 0.15-0.20mg/kg: fresh weight of PCBs. In most areas, the average levels of PCBs in fish tissues from sites used by otters were under 0.10mg/kg, and are similar to those found in the previous work, and are comparable to the 0.145mg/kg proposed by LEONARDS *et al*. (1994); however, we need to keep in mind that there are differences between levels in muscle and the whole fish. They are higher than 0.026 - 0.05mg/kg levels presented by MACDONALD and MASON (1994), for the whole data, and for eels, respectively.

Figure 2. a) Synthesis of the average values, ranges ans interquartiles of PCBs in 11 fish species from four French regions, showing samples coming from the otter range (grey) and outside (white) (after LAFONTAINE & DE ALENCASTRO, 2000). b) Average values, ranges and interquartiles of PCBs levels in fish muscle from nine basins in Catalonia (N.E. Spain), showing samples coming from the otter range (grey), outside (white) and from the re-introduction area (recalculated data from LÓPEZ-MARTíN *et al*., 1995, and MATEO *et al*., 1999).



In North-east Spain, levels found by LÓPEZ-MARTÍN, RUIZ-OLMO and BORRELL, (1995), from the different basins (unpublished data), and plotted in Figure 2, coincide again with the results of LAFONTAINE and DE ALENCASTRO (2000), and with all otter populations in sites under 0.14 mg/kg: fresh weight of PCBs in fish muscle. In rivers not used by otters, average levels were often over 0.2mg/kg, reaching up to 1.34mg/kg. We must highlight the basins of the Muga and Fluvià Rivers, where, despite the fact that the otter became extinct toward the end of the 1970s (RUIZ-OLMO and GOSÁLBEZ, 1988), levels in fish tissues found at the beginning of the 1990s were compatible with the presence of the mustelid. This has allowed for the development of a reintroduction project (SAAVEDRA and SARGATAL, 1998) in which nearly 50 individuals have been relocated.

Results from France and Spain confirm those from Italy, since levels of PCBs within the otter range are much lower that those from zones without otters (VIVIANI *et al*., 1974; GALASSI and GANDOLFI, 1981; GALASSI, GANDOLFI and PACCHETTI, 1981; CANTONI, CATTANEO and FABRIS, 1985; COZZANI and PIETROGIACOMO, 1985; TURSI, CONSTANTINO and MATARRESE, 1989). In the Ticino River, where there is a reintroduction project, PCBs levels seem to be appropriate for their return (NARDI *et al*., 1993), while they could be less compatible with those of DDTs.

RUIZ and LLORENTE (1991), in the 1980s, found high levels of PCBs and DDTs in tissues of fish from Ebro delta (Northeast Spain), where otters had become extinct in the 1970s.

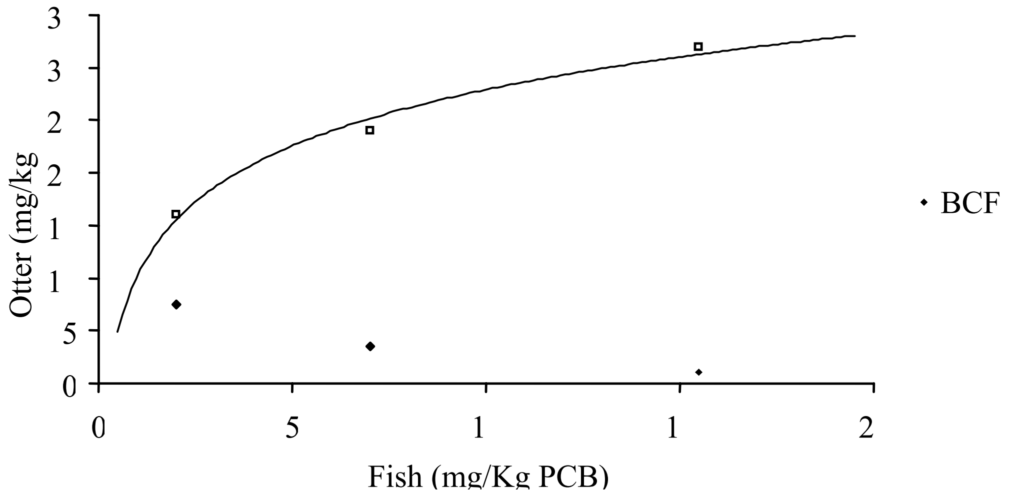
There are several studies on heavy metal levels in fish tissues from Italy; TURSI, CONSTANTINO and MATARESSE (1989) and FUMAGALLI and PRIGNIONI (1991) found very low levels in fish from Italian rivers with otters; the higher levels were mercury, being in almost all cases below the 0.2mg/kg: fresh weight. In other Italian rivers, with no otters, mercury levels were lower (BEGLIMONDI, FRAVOLINI and MOROZZI, 1975; LOCHT *et al.,* 1981). In Spain, RALDUA and PEDROCCHI (1996) found average mercury values between 0.74 and 2.80mg/kg: wet weight, in fish from Huesca (Aragon, Northeast Spain), outside the otter range or on the border of the otter distribution towards 1993. In Portugal, SANTOS-REIS, AFONSO and FREITAS, (1995), found high mercury levels in American crayfish, *Procambarus clarkii*, from the Tejo basin, this being the main otter prey in many areas; levels were highest in Almonda River (mean: 0.21mg/kg) and Server River (mean: 0.29mg/kg).

In some rivers, lead, chromium or nickel levels were very high (CAGGINO, 1982), and could have contributed in an important way to the decline of the otter.

8 BIOMAGNIFICATION FACTORS

In Spain, most of our studied otter populations feed mainly on fish (in the zones where we have studied bioconcentration factors: 90-95%; RUIZ-OLMO and PALAZÓN, 1997). This allowed us a better approach to the bioconcentration factors (considered as the ratio: levels in otter tissue/levels in fish tissue), than in other otter populations with a more complex diet. The only bioconcentration factors to PCBs and DDTs on the whole (LÓPEZ-MARTÍN and RUIZ-OLMO, 1996; RUIZ-OLMO, LÓPEZ-MARTÍN and DELIBES, 1998). These authors base their data on the analysis of the most-caught fish (the most abundant in the environment) and its importance in otter diet. Levels are drawn in Figure 3.

Figure 3. Biomagnification factors for otter and fish tissues in otters from NE Spain (RUIZ-OLMO *et al.,* 1995; LAFONTAINE & DE ALENCASTRO, 2000).



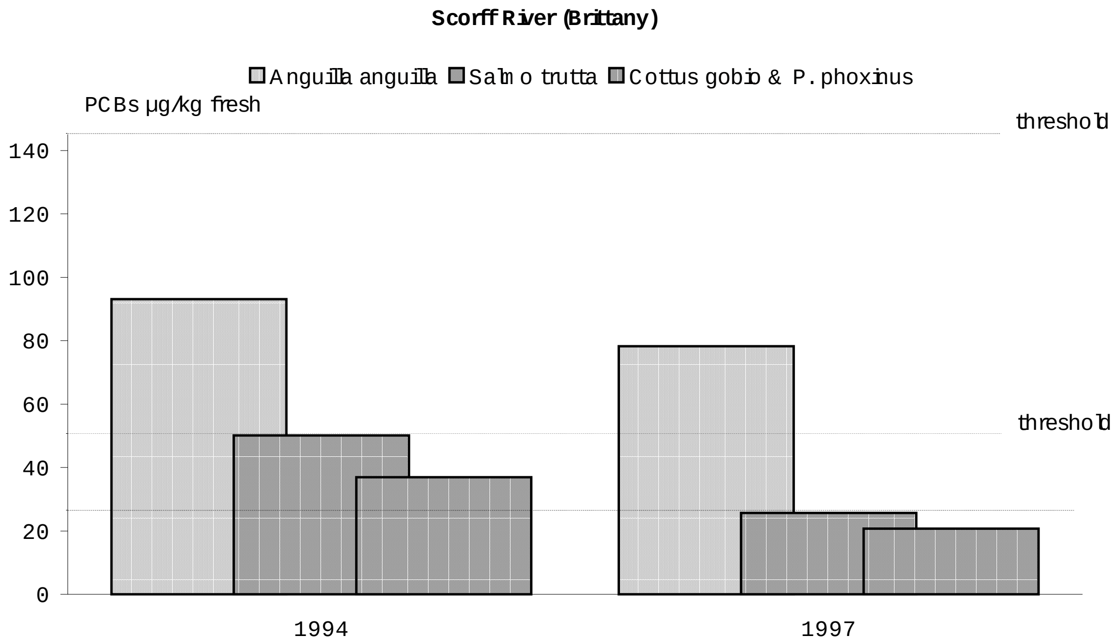
Biomagnification factors in PCBs were double those of DDTs (i.e., they accumulated double). For PCBs, a logarithmic function is followed ($r = 0.99$: $p = 0.021$), very similar to the results from the Canadian river otter *Lutra canadensis* (FOLEY *et al*., 1988). Bioconcentration values ranged between 2.2 and 8.2 in the case of PCBs (lipid weight), and between 0.9 and 4.8 in the case of DDTs.

For the River Scorff, Brittany (North-west France), from 1994 to 1997, LAFONTAINE and DE ALENCASTRO (2000), found PCBs biomagnification factors varying from 22.2 to 44.9 (wet weight) for otter tissues *vs* all the fish species together, and from 584 to 3236 (wet weight) for otter tissues *vs* sediments.

RUIZ-OLMO, LÓPEZ-MARTÍN and DELIBES, (1998) found PCB congeners No.138, 153, 170, 180, 194, 195, 196/ 203 and 201, in greater proportion in otter tissues than in fish, there being a significant bioaccumulation among fish in congeners No. 101, 138, 141, 153, 170, 174 and 180. LAFONTAINE (1995) found greater prevalence of congeners 138, 153, 170 180, for the 12 analysed, the data agree with the Spanish results. MASON and RATFORD (1994) found that the congeners No 138, 153, 163, 180 and 187 are the most prevalent in British otters and Nos 118, 138, 153, 163 and 180 in Danish otters. For otters from Denmark, SMIT *et al*. (1996) found a greater bioconcentration of the congeners Nos. 138, 180, 156, 157, 189, 126 and 169. On consideration that they are the most bioacumulated congeners and could have greater weight in the toxicity, they utilise an index that includes these seven congeners only (∑ 7 PCBS), or with the Toxicity Equivalent Concentration (TEQ) (SAFE, 1994; LEONARDS, 1997). These methods allow the real power of toxicity for each concentration of PCB congeners, but differences between studies must be explained.

For the River Scorff, Brittany (North-west France), from 1994 to 1997, LAFONTAINE and DE ALENCASTRO (2000), analysed for 17 CB congeners, including three coplanars, and found the cumulative contribution ($p < 0.001$) of the most toxic CBs (groups 1A + 1B + 2, according to the typology of MACFARLAND and CLARKE (1989) along the food chain, as follows: sediments > *Scardinius* *erythrophtalmus* > *Salmo trutta* > *Cottus gobio* & *Phoximus phoxinus* > *Anguilla anguilla* > otter spraints > otter tissues (Figure 4).

Figure 4. Evolution of pollution levels in fish tissues from Scorff River (Brittany).



The proportion of congeners in each zone depends on the type of PCB (mixture) that the otters/fish have ingested (with the congeners 138, 153, and 180 found in great proportion in all the studies). The several industrial uses of the zones could characterise the proportion of each PCB in each zone.

9 EVOLUTION OF POLLUTION LEVELS

Unfortunately, otter carcasses were not analysed for contaminants between1950 and 1970 in the countries discussed here. This was the period when many of the organochlorine compounds and heavy metals were being used indiscriminately, widely and without control. The only data available refer to fish from the Po delta, Northern Italy. They show high levels (VIVIANI *et al*., 1974), with maxima up to 12mg/kg: wet weight for PCBs in the liver of *Gobius paganellus* and 6.13mg/kg in the liver of *Squalus acanthias*. If these species were consumed in significant numbers by otters, they could give rise to high concentrations in otter tissues (remember the thresholds for otter presence of 0.05-0.2mg/kg in fish). These authors found high levels of DDTs (up to 9mg/kg in some species) and lindane (up to 1.3mg/kg).

Lack of historical information prevents us from carrying out an analysis of the evolution and dynamics of several compounds in otters and their prey from those decades. Also, analytical methods and accuracies have changed during this time. However, we found a decrease in the levels of PCBs, DDTs and other pollutants in otter tissues (Table 1) and fish tissues (Figure 4: Table 4). These data agree with MASON (1998) in that they demonstrate a decrease in these pollutants.

Table 4.- Comparison between the average PCB levels in fish tissues from some rivers of NE Spain in two different periods (after LÓPEZ-MARTÍN & RUIZ-OLMO, unpublished).

		1990-92		**1998-99**		
Basin	**Site**	**PCBs**	**DDTs**	**PCBs**	**DDTs**	**Fish species**
N. Pallaresa	Gerri de la Sal	0.145 (0.06-0.30)	0.087 (0.02-0.2)	0.065 (0.04-0.09)	0.030 (0.01-0.05)	*S. trutta*
Segre	Seu d'Urgell (1)	0.207 (0.04-0.45)	0.235 (0.05-0.16)	0.212 (0.191-0.436)	0.128 (0.003-0.253)	*S. trutta* *C. gobbio* *C. toxostoma*
	Balira (1)			0.13 (0.07-0.19)	0.072 (0.037-0.107)	*S. trutta*
	Ponts	0.095 (0.05-0.24)	0.052 (0.02-0.07)	0.040 (0.005-0.08)	0.102 (0.01-0.02)	*S. trutta* *C. carpio* Others

(1) Sampling stations downstream Southern Andorra border

Heavy metal and other organochlorine compound levels found in Spain, France and Italy since the 1980s have been generally low, with the exception of mercury in some areas in France (LAFONTAINE, 1995) and Portugal (SANTOS-REIS, ALFONSO and FREITAS., 1995), in some small rivers of Tejo Basin and in Aveiro Ria, (Nuno Gomes, *pers. comm.*). This decrease in pollution levels (not only of PCBs but in general), would contribute to an explanation of otter recovery in several areas.

10 OTTER DISTRIBUTION 1995-2000: THE RESPONSE OF A BIOINDICATOR SPECIES

Otter distribution has changed since 1985, with a tendency towards increase and recovery in wide areas (ROSOUX, TOURNEBIZE and MAURIN, 1996; PRIGNIONI, 1997; RUIZ-OLMO and DELIBES, 1998; TRINDADE, FARINHA and FLORENCIO, 1998; CONROY and CHANIN, 2001). Figure 1 shows this increase in otter distribution, with an evident spread in several areas of central France and in zones of Spain (Pyrenees, Centre, Andalucia, etc.). Even in Portugal, where the otter was previously widespread, better results were recorded in 1995 (89% of positive sites).

However, the animal has not recovered in other areas of these countries, nor in Andorra or small states. In Andorra, the Balira Basin crosses the country and has high pollution levels which could explain the absence of otter in 1999, when we carried out an otter survey for the new Otter Action Plan (RUIZ-OLMO, unpublished). High contaminant values obtained in 1991 in fish tissues from the Segre River, just beyond the junction with Balirain Spanish stretch (LÓPEZ-MARTÍN, RUIZ-OLMO and BORRELL, 1995), are in accordance with the lack of otters. Even in 1998-99 we found 0.13-0.21 mg/kg: wet weight in muscle of brown trout from the Rivers Segre and Balira, just downstream of Andorra. The Balira River was determined to be the most polluted in the Spanish Pyrenees by both the Water Suitability Index and water analysis (CONFEDERACIÓN HIDROGRÁFICA DEL EBRO, 2000). This could explain the absence of otters.

This otter recovery in wide areas is attributable to assistance in and an improvement of the general conditions for the species, especially pollution levels. On the other hand, the lack of otter recovery in other parts (with a good otter habitat index), could be a result of these conditions not improving enough or even worsening conditions, especially pollution.

11 DISASTERS AND OTHER CONTAMINANTS

Up to now, the effects of more widespread pollution types have been analysed. However, big disasters, producing negative effects on otters and their prey, should be emphasised. In analysing these types of occurrence, we can divide them into two types:

a) Oil spills. They have taken place mainly along the Atlantic coasts after ship accidents. In Spain, those from the *Urquiola* (May 1976) with 28.1 million gallons, and the *Amocco Cadiz* (1992) with 21.9 million gallons, can be highlighted. They affected the coast of Galicia. Recently (2000) an unidentified oil slick impacting several kilometres was found off the east coast of Cadiz (Southern Andalucia). Although in the first two instances, effects on some fish species, invertebrates and plants were important, no dead shore-living otters were found. These types of accidents have also occurred in rivers, like the Tajo River in Central Spain, in August 2000. There are no studies on the effects of these accidents on otter populations. However, years later, otters still inhabit some of these zones.

   A similar disaster occurred in December 1999, along the North-west coast of France (Brittany). The *Erika* oil spill polluted several hundreds of kilometres of sea-shore. Invertebrates, fish and especially several thousands of seabirds were affected. No dead shore-living otter was found however. This does not mean that this oil spill had nor will have no impact on otter survival, and, in terms of accumulation in the food chain, a study is now starting to analyse PAHs in both otter spraints (from coastal areas) and marine mammal tissues (LAFONTAINE and HASSANI, in progress).

b) Break-up of toxic reservoirs. This is especially highlighted by the case of the Aznalcollar mine (May 1998), on the Guadimar and Guadalquivir Rivers, Huelva (Southern Spain). A high quantity of heavy metals was released into these rivers as a result of mineral cleaning. Arsenic was the main metal, with more than 4000 mg/kg in sediments in three of nine sampling sites. Manganese, cadmium, chromium, copper, lead (more than 3000mg/kg at three of the sites), zinc and iron were also found at high levels. A monitoring program on physical and chemical parameter effects and on biotic elements has been carried out (Spanish Ministry of the Environment, in progress). In sediments, a decrease in levels was found in arsenic, iron, manganese and nickel, although no decrease was shown in the remainder. An increase in chromium, copper and lead was registered.

c) Mines. Heavy metals are found in high levels in waters used for mining activities, in Northern Portugal, for example, these affected pH levels and sediments (TRINDADE, FARINHA and FLORENCIO, 1998).

On the other hand, perhaps different pollutants are still active or could well begin to be so in the near future. A study of the effect of dioxines, furans, PVCs and organophosphorates, etc., should be made.

12 CONCLUSIONS

The results show the negative effect of contamination on otter distribution in France, Italy, Portugal, Spain and Andorra. This could explain the great decline during a relatively short period (three decades) and could have been coincidental with other factors such as habitat transformation, decrease of food availability and persecution, and other.

The study area is very extensive and is characterised by a high diversity of weather as well as hydrological, ecological and human characteristics. This is of great importance in understanding how pollution could affect otters. In fact, there are examples of pollution from several sources that have determined the distribution of the otter in some specific zones (organic pollution, acidification of water, detergents, etc.). However, only a limited series of compounds (micropollutants) producing lethal effects at a population level and sublethal (for example, affecting breeding) could have had a more global effect, in accordance with a disappearance on a regional scale (MASON, 1989; MACDONALD, 1991). These, especially, are the organochlorine compounds (from industrial sources and agriculture) and heavy metals. Of these, PCBs seem to be the most widespread in fish and otters, attaining higher levels of bioaccumulation in tissues. However, in specific regions, other biocides (mainly DDTs, oxychlordane, dieldrin and lindane) (CHANIN and JEFFERIES, 1978; MASON, 1989) and heavy metals (mainly mercury) (GUTLEB, 1995; KRUUK and CONROY, 1996; KRUUK, 1997) could have had a more important effect than PCBs.

The differences in toxicity need a more thorough analysis of pollutants, both with reference to the type of mercury (GUTLEB, 1995), and with reference to the congeners that make up PCBs (SMIT *et al*., 1994, 1996; LEONARDS, 1997).

There is increasing evidence of the effect on reproduction, and the pathological effects of these compounds on mammals (DELONG, GILMARTIN and SIMPSON, 1973; TANABE, 1988), and more specifically on otters and American mink (JENSEN *et al*., 1977; AULERICH and RINGER, 1977; KEYMER *et al*., 1988; MASON and O'SULLIVAN, 1992; MASON and MACDONALD, 1993). There is a correlation between PCB levels and the index of body condition, *k*, that has been demonstrated to determine the probability of mortality (KRUUK, CONROY and MOORHOUSE, 1987; LAFONTAINE, 1995). Lately, the effect on vitamin A (and subsequent effects) was shown (SMIT *et al*., 1996; SIMPSON *et al*., 2000).

This approach to PCBs congeners could bring with it some results applicable in the management and real understanding of the effects of these compounds. Our results also contribute to the idea of the existence of some thresholds of pollutant levels in prey, of some 0.1-0.2mg/kg: wet weight for PCBs, although important differences between the fish species exist (LAFONTAINE and DE ALENCASTRO, 2000).

Organochlorine levels have been diminishing world-wide, since preventive measures were undertaken (STOUT, 1986; BINGNERT *et al.,* 1993; NEWTON & WILLEY 1992). This decline has also occurred in the areas studied here, both in pollution levels of otter prey as well as in otter tissue levels. This decrease in levels coincides with a recovery in otter distribution. This fact tends to convince us of the effect of pollutants on otters, and also the bioindicator property of this mustelid. It answers positively to the improvement in water quality, in pollution levels in prey tissues and in the environment in general. However, it is necessary to stay on guard since in many areas this recovery is not happening and, other new compounds or substances could have similar effects to those of organochlorine compounds and heavy metals.

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