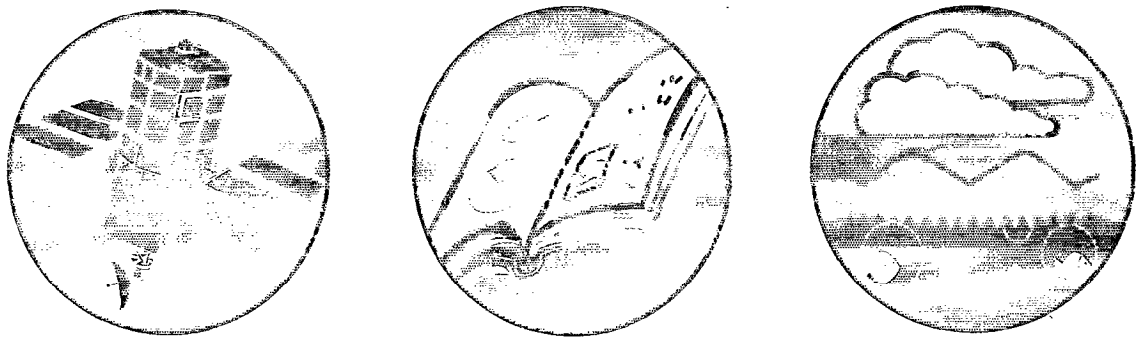
A Post Mortem Study of Otters (Lutra lutra) Found Dead in South West England



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Technical Report
W148

ENVIRONMENT AGENCY

A Post Mortem Study of Otters (*Lutra lutra*) Found Dead in South West England

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This R&D Technical Report contains the main findings of post mortems carried out on 77 otters found in south west England between 1988 and 1996, the majority of which died in road traffic accidents. Clear trends in the concentrations of certain environmental pollutants within the otter carcasses are shown by this report which will be of interest to ecologists, veterinarians, toxicologists, environmental scientists and highway engineers. The recommendations made within this report will be discussed by the Conservation Technical Group. It should be noted that this is one report of a number which have been produced on otter ecology (Reference otter reports/projects).

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EXECUTIVE SUMMARY

Post mortem examinations were carried out on 77 otters found dead in south west England between 1988 and 1996. The majority of deaths were due to road traffic accidents and most of these occurred in the winter months. Bite wounds, apparently caused by other otters, were the other main cause of death. There was no evidence of significant infectious disease and most otters were in good physical condition. Evidence of breeding was seen in almost half the females but in three cases the uteri appeared unusual, and were possibly abnormal.

The concentrations of certain PCB congeners and OC pesticides in the otter's livers declined significantly over the eight year study period. These declines coincided with increased liver Vitamin A levels, but a causal relationship was not established. PCB levels in males were generally much higher than in females. There was a relationship between the levels of certain pollutants and some organ weights.

Future work should concentrate on the relationship between the levels of specific pollutants and the histopathology of endocrine organs and reproductive tracts.

KEYWORDS

Otter, post mortem, road traffic accidents, pesticides, organochlorines, polychlorinated biphenyls (PCBs), Vitamin A, toxicology, endocrine organs, pathology.

SUMMARY OF MAIN FINDINGS

1. Post mortem examinations were carried out on 77 otters between December 1988 and March 1996. With the exception of two cases from Hampshire, all the otters came from Cornwall, Devon and Somerset. None were received from Dorset, Wiltshire or South Gloucestershire.

2. The numbers submitted increased noticeably from 1992. In part, this was due to increased submissions from NRA/Agency staff. From 1993 to 1996, twice as many males as females were submitted.

3. There was a marked pattern of seasonal mortality, with very few deaths in the summer months. Female mortality peaked in February and male mortality in March and December. Seasonality in mortality may be influenced by rainfall patterns, with otters frequently killed on roads a day or so after periods of heavy rain when rivers are in spate. Such rainfall events most commonly occur between September and March.

4. Road traffic accidents were responsible for 83 % of mortality. Bite wounds, apparently caused by other otters; were seen in 12 animals (16 %), and five died as a result of their injuries.

5. On average, males were larger than females but they were in overall poorer condition. There was no evidence of deaths due to starvation.

6. Dental condition was generally good. Examination of teeth from 28 animals indicated a mean age of two years for both males and females.

7. Very few gross pathological lesions were seen, indicating a general absence of significant bacterial, viral or parasitic diseases. Streptococcal infections were seen in animals suffering from bite wounds.

8. Occasional focal lesions were seen in lungs and were mostly due to inhaled fungal spores. Some lesions resembled tuberculous foci, but specific stains showed no evidence of mycobacteria. *Sarcocystis* infection was seen in the external eye muscles of one otter.

9. Adrenal hypertrophy and splenic atrophy were seen in males dying of bite wounds. Enlarged adrenal glands were also seen in females in late pregnancy and early lactation. There was a separate, strong, positive correlation between adrenal weight and liver PCB levels, particularly for congeners 138, 153 and 180.

10. Evidence of reproduction was seen in 11 out of 25 female otters, with two being pregnant and four lactating.

11. Three otters, one of which was in early pregnancy, had convoluted uteri. Although these appeared abnormal their significance is uncertain. One stunted male was a cryptorchid with a single, undescended testis and histological examination showed no spermatogenesis in either testis.

12. Samples of liver were analysed for a wide range of organic pollutants and for heavy metals. The liver levels of PCBs, Dieldrin and pp' DDE were generally higher in males than females, although this only achieved significance in the case of PCBs.

13. There was a relationship between pollutant levels and body weight, with higher levels in heavier animals. There was also a positive correlation between thyroid weight and body weight, but no significant connection between thyroid weight and pollutant levels after the relationship with body weight had been taken into account.

14. The heavy metal concentrations in most cases were considered to be of little or no significance. One otter from Hampshire had an elevated arsenic level. Mercury levels were generally low but, as this metal accumulates with age, these levels could be a reflection of the young age of the animals in this study.

15. PCB levels were mostly lower than those reported in previous studies but some animals had levels which were considered to be high. The results show a significant decline in the concentration of congeners, 118, 138, 153, and 180, but not of total PCB, over the period of the study.

16. Two otters had high Dieldrin levels. One came from Newbridge, west Cornwall, where very high levels had been reported in eels two years previously. The source was believed to have been Aldrin used on daffodil crops. The second animal came from near Falmouth, Cornwall but in this case no source was apparent. Dieldrin levels declined significantly between 1988 and 1996.

17. DDT levels were considered to be low in the majority of animals but, as with Dieldrin, the results show a significant decline over the period of the study. The levels of γHCH were all low but also declined significantly. There were no significant residues of other organochlorine pesticides, organophosphates or herbicides.

18. Analysis of liver samples for Vitamin A content showed a wide range of values. Seven animals had less than 7 μmol/kg and in most species this is considered to be evidence of deficiency. Five animals had values of around 1,000 μmol/kg or more.

19. Very low Vitamin A levels were typically seen in animals which also had high levels of PCBs, Dieldrin and DDT. Although the results provide evidence of an inverse relationship between Vitamin A and some of these pollutants, a causal connection was not demonstrated.

20. Most of these animals with low Vitamin A levels and high PCB and OC levels were seen in the period 1988-1992.

21. Gonadal hypoplasia and cryptorchidism are recognised features of Vitamin A deficiency, and the Vitamin A level in the cryptorchid otter in this study was below the limit of detection. Apart from this case, no other otter had gross lesions indicative of Vitamin A deficiency.

22. Histopathological examination of livers showed bile duct hyperplasia and fibrosis of varying degree in almost all the otters. In most animals the thyroid glands had small or very small follicles with sparse, pale staining, colloid. In some cases there was little or no colloid and the glands appeared very vascular. In others the follicles were well developed and full of pink staining colloid. Splenic atrophy and adrenal cortical nodular hyperplasia were seen in several otters. Although these observations may be related to the levels of pollutants present, too little is known about the normal appearance of these tissues in otters to draw definite conclusions. No specific lesions of Vitamin A deficiency were seen.

ACKNOWLEDGMENTS

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1 INTRODUCTION

1.1 Background

During the 1950s and 1960s there was a marked decline in the otter population of Britain and much of western Europe. The otter became rare or absent from most of England and in some countries, such as The Netherlands, it is now extinct (Macdonald and Mason, 1990). The reasons for the decline are uncertain but there is strong circumstantial evidence to suggest that the organochlorine pesticides (OCs) and polychlorinated biphenyls (PCBs) were responsible (Jefferies et al 1974, Chanin and Jefferies 1978, Mason 1989). However, although very considerable resources have been invested in analysing otter tissues for these chemicals, only a negligible number of otters have been subjected to detailed veterinary pathological examination (Keymer 1991). As a consequence, very little is known about the pathological conditions affecting otters, whether caused by organochlorine pesticides, PCBs or any other agent. In the absence of such data, those organisations concerned with otter conservation and/or environmental pollution have few criteria by which to monitor the success or failure of their strategies.

The author had carried out *ad hoc* post mortem examinations on a small number of wild otters in Cornwall and west Devon since 1988 and had also examined specimens which had died in captive breeding projects. However, from late 1992 the National Rivers Authority (NRA) (now the Environment Agency), provided funding for the post-mortem examination of all otters found dead in south west England. This report summarises the results of these investigations up to March 1996.

1.2 Objectives

The overall objective was to establish the health status of otters in south west England. More specific objectives were, firstly, to examine otters for evidence of significant infectious, metabolic or nutritional disease, and secondly, to examine them for evidence of exposure to environmental pollutants.

Previous studies on otters have recorded the pollutant levels in their tissues but have not related these levels to any pathological changes which could have been present. (Olsson et al 1981, Mason 1989, Kruuk and Conroy 1991.) A component of the second objective, therefore, was to examine organs for evidence of the gross and histopathological lesions which are known to occur following exposure to pollutants, particularly PCBs and OCs. As these compounds are known to affect organ weight, Vitamin A metabolism, thyroid activity and steroid hormone levels, a post mortem protocol was adopted which, it was hoped, would reveal whether the pollutants present were at physiologically significant levels.

2 MATERIALS AND METHODS

2.1 Post Mortem Procedure

Seventy-seven wild otters were examined between December 1988 and March 1996. With the exception of two specimens from Hampshire, all the otters came from Cornwall, Devon and Somerset. Where carcases were submitted in a fresh state, post mortem examinations were normally carried out on the day of receipt, or failing that, within 24 hours. Carcases submitted frozen were held at -20°C until they could conveniently be thawed and examined. Each carcase was given a unique laboratory reference number.

The post mortem examinations were carried out to a standard protocol which included recording the sex, body weight, overall length and the weight of most organs (Appendix I). In addition to a visual assessment of the animal's nutritive state, a Condition Index was calculated (Kruuk and Conroy 1991, following LeCren 1951) using the formula:

$$\text{Condition Index} = \frac{W}{aL^n}$$

Where W = body weight in kilograms and L = nose to tail tip length in metres. The constants a and n were 5.02 and 2.33 respectively for females and 5.87 and 2.39 for males (Kruuk et al 1987).

2.2 Laboratory Examinations

Where lesions suggestive of a bacterial infection were seen tissue samples were inoculated on to 5 % sheep blood agar and MacConkey agar. Incubation was at 37°C for up to 48 hours. In selected cases intestinal contents were inoculated into Selenite F liquid medium, incubated at 37°C for up to 48 hours and sub-cultured onto brilliant green agar for a further 24 hours.

A range of organs was routinely sampled for histological examination (Appendix Ia). Samples were fixed in 10 % buffered formol saline, embedded in paraffin wax, sectioned at 6 μm and stained by haematoxylin and eosin (H&E). Samples of liver taken for toxicological examination were wrapped in aluminium foil and held at -20°C until they could be submitted to the Environment Agency laboratory, Exeter. They were examined by gas liquid chromatography and by mass spectometry for a range of pollutants, including halogenated hydrocarbons, and for heavy metals by atomic absorption spectrophotometry. Provided they were not autolysed, duplicate liver samples were analysed for Vitamin A at Shrewsbury Veterinary Investigation Centre.

An incisor tooth was collected from each case and placed in 10 % buffered formal saline. Teeth samples collected between 1988 and 1994 (n=28) were submitted to Drs. H Kruuk and J Conroy, Institute of Terrestrial Ecology, Banchory, who determined the animals' ages by counting the incremental rings of cementum (Mc Laren 1992).

3 RESULTS OF POST MORTEM EXAMINATION

3.1 Numbers Submitted and Origins

The first otter was received in December 1988. None had been received in the previous nine years, despite the fact that many other wildlife specimens were received during that period. No otters were received again in 1989 but numbers increased from 1990, and this trend became more apparent from 1992 onwards (Table 1).

Table 1 - The Number of Otters Submitted Each Year.

Year	1988	1989	1990	1991	1992	1993	1994	1995
Number	1	0	6	3	12	11	20	17

Note: 1) The total for 1990 includes a new born cub, found in May, which had drowned in a fishing net. This case is excluded from the rest of the report.

2) Seven specimens were submitted in the first three months of 1996.

The increase in submissions from 1992 was undoubtedly influenced by the NRA decision to pay for the post mortem examinations and also by the efforts of NRA, later Environment Agency, staff in collecting specimens from more distant areas, such as Somerset and much of Devon. The geographical distribution of 73 of these specimens is shown in Figure 1.

It will be seen that no otters were received from Dorset, Wiltshire or south Gloucestershire. A list of all specimens is given in Appendix II, together with their laboratory reference number, National Grid Reference and the river catchment in which they were found. Apart from the newborn cub mentioned above, and a juvenile weighing 2 kgs, they were all well-grown immatures or adults.

In order to assess whether there had been a significant increase in the number of otters submitted annually, the submissions were analysed over two periods, firstly from July 1987 to June 1992, secondly from July 1992 to March 1996, i.e. before and after the NRA started collecting carcases. A Poisson regression model showed a significantly increasing trend up to mid-1992 ($p<0.01$), but only weak evidence for a further increase after that ($p<0.1$).

Until 1993, the ratio of males to females was about equal (10 males, 11 females) but from 1993 to March 1996, there were 38 males and only 17 females (see Figure 2). Although these figures might suggest a trend towards an increase in the proportion of males killed after 1992, the results are not statistically significant ($\chi^2 = 2.3$). However, there was a significant overall tendency for more males to be killed than females.

Distribution of Otters Submitted for Post-Mortem Examination.

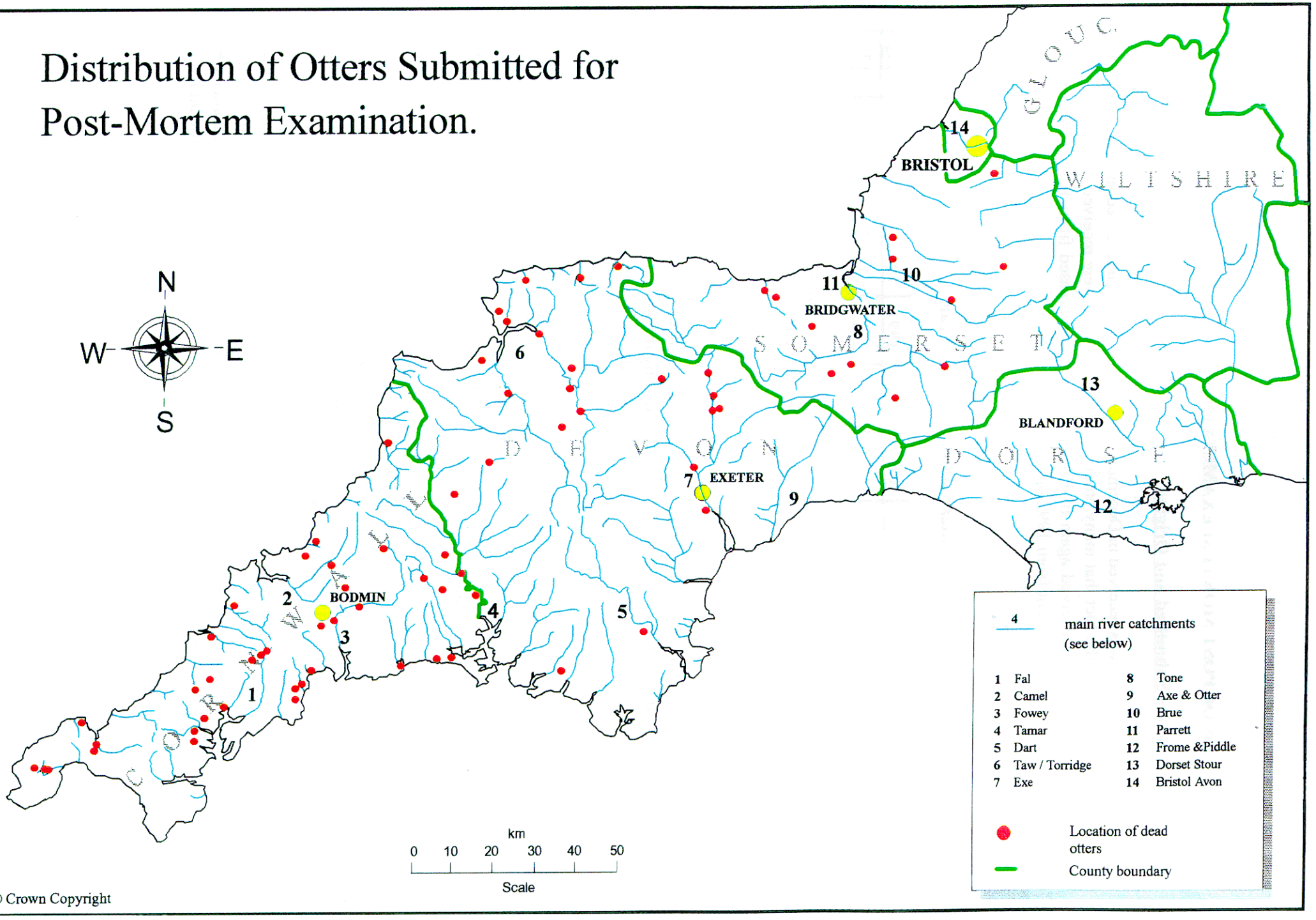


FIGURE 1

The number of otters submitted varied markedly according to weather conditions and also the time of the year. Many animals were submitted after periods of heavy rainfall, when rivers were in spate and almost no otters were submitted during the summer months of any year. However, although this generalisation applies almost equally to males and females, the limited data does suggest that there may be differing peaks of mortality between the sexes (Figure 3).

Female deaths occurred most frequently in the month of February whereas only one male died at this time. Male mortality peaked in March and again in December. However, although a Poisson regression model showed that the rate of mortality varied very significantly ($p<0.001$) between months, the difference in temporal pattern between males and females was significant only at the 5 % level.

Figure 2 - Total number of male and female otters submitted per year

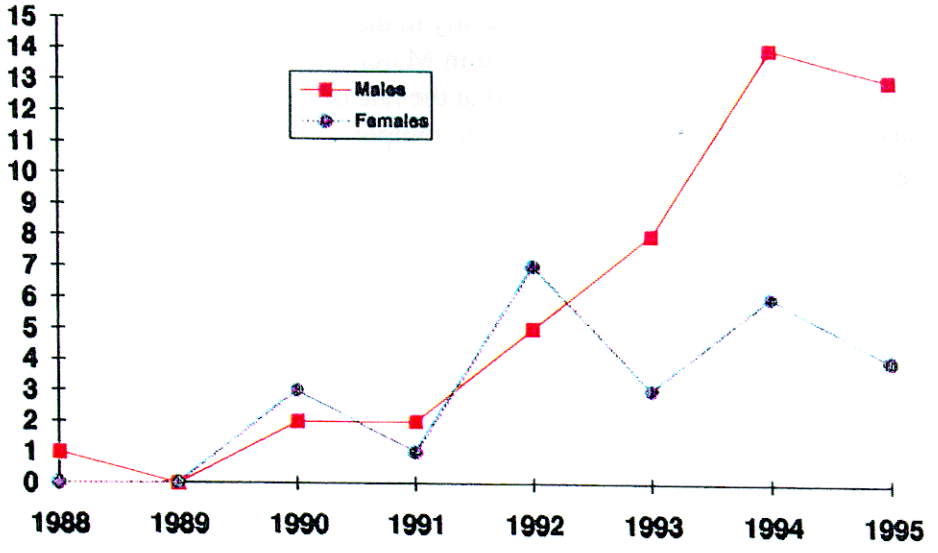
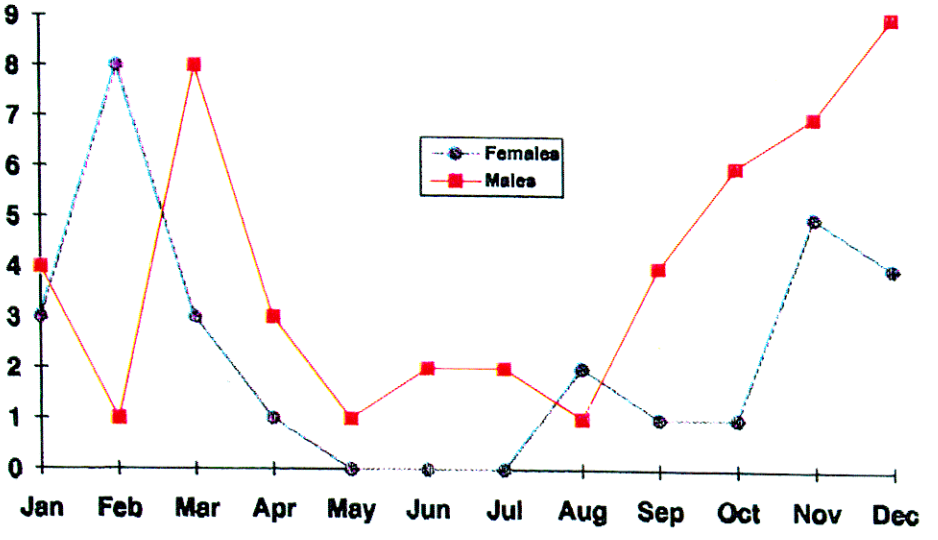


Figure 3 - Number of otters submitted each month, 1988-1996



3.2 Gross Pathology

The state of preservation of the otters varied considerably. A few were freshly dead, most were slightly or moderately decomposed and some were badly decomposed. Many specimens, irrespective of their state of freshness, had also been frozen which reduced their diagnostic value. The great majority were found dead on or near major roads and, not surprisingly, many of these showed evidence of severe trauma with fractured bones, ruptured organs and extensive haemorrhage. In some cases, the internal organs were totally crushed and no meaningful examination could be carried out.

With few exceptions the physical condition of the otters was good. The fur was in good condition, there were adequate subcutaneous fat deposits and the skeletal muscles were well developed. Males were generally larger than females, with median values for body weight and total length of 7.5 kg and 110 cm, compared with 5.75 kg and 100 cm for females. The respective mean values were 7.26 kg/107.6 cm and 5.65 kg/99.4 cm. These differences were highly significant (Student's t, $p \leq 0.01$).

Measurement of both body weight and length was not possible in every case, usually because of severe trauma. Therefore, the Condition Index could not be calculated for 16 otters. The Condition Index for the female otters was consistently high with 17 of 21 (81 %) having a value of 1.0 or more, and ranging from 0.96 to 1.45 (mean = 1.17). The animal with the index of 1.45 was exceptional and its high body weight (8 kg) was almost certainly anomalous. It had extensive subcutaneous bruising and oedema but was badly affected by autolysis. The next highest value in a female was 1.23. In contrast, the males were in overall poorer condition with only 21 of 39 (54 %) having an index of 1.0 or more (mean = 0.99). They also showed much greater variation in condition, with the index ranging from 0.54 to 1.34. In many cases the loss of condition was clearly associated with bite wounds.

The following notes briefly describe the individual organ systems and any lesions, but do not describe lesions judged to have been caused by trauma at, or after, the time of death.

3.2.1 External Features

Ixodid-type ticks were present on three otters, with larvae and nymphs present in two cases and an adult female *Ixodes hexagonus* in one case. An animal submitted from Hampshire (M290/6/94) had two discrete patches of white fur, 2-3 cm diameter, on the left ventral surface of the thorax, and a further two corresponding patches dorsally. It is thought that this animal had suffered localised skin damage sometime previously with non-pigmented hair growing over scar tissue. This otter also had abnormal feet, with the third toe of both front feet turned inward, and each with an unusually long toe nail. The animal was thought possibly to have been a captive release but there was no evidence of a microchip when examined by radiography and by scanner (AVID Microchip I.D. Systems).

Injuries to the feet were common, with cuts, puncture wounds, scar tissue and amputated digits recorded in 18 animals. Cuts mostly affected the plantar surface of the pads and/or interdigital webs. In some cases they were severe, resembling those seen in dogs which have cut their feet on broken glass. It is possible that these otters had also cut their feet on glass, whilst scrabbling in stream beds. In other cases the lesions were punctures or small tears and were considered to be bite wounds.

Two otters (M31/9/93 and M139/12/94) with bite wounds elsewhere on the body also had raw, eroded foot pads which were similar to those seen occasionally in stray dogs which have travelled long distances on roads. Neither the pads nor skin elsewhere on the body showed evidence of hyperkeratosis, although one had interdigital papillomata on a fore foot. The toe nails on the hind feet were typically shorter than those on the fore feet but in a few cases the hind toe nails were almost completely worn away. The proportion of animals showing foot damage was 30 to 40 % in most years, but in 1994, when the numbers submitted increased markedly, only two out of 20 animals had foot lesions.

Apart from the feet, bite wounds were also recorded around the cheek, lip, lower jaw (Figure 4) and around the perineum (Figure 5). In some cases these lesions were severe. A few animals had bite wounds elsewhere, e.g. hock or shoulder. Interpretation was sometimes made difficult due to the effects of decomposition and/or severe crushing but unequivocal bite lesions were present in 12 animals (nine males, three females). In the case of four males and one female it was considered that the animal had died solely as a result of being bitten. A fifth male, affected by autolysis, had also probably died from bite wounds. The other six affected cases were all animals killed by road traffic.

Only one otter had a visible eye defect. This was a young female, killed in a road accident, which had recently given birth or aborted and had colostrum in its mammary glands (M219/1/96). The right eye was not visible externally but was located deep within the orbit. It was about half normal diameter, white and very firm. The zygomatic process had also been fractured at both ends and was attached to the skull by fibrous tissue only. The other eye was grossly normal (but see 5.6). It is presumed that the otter had been involved in a previous road accident where the eye had been ruptured and the cheek bone fractured.

The dental health of the otters was very good, with only one animal showing a significant amount of calculus. The degree of wear was mostly slight to moderate, suggesting a young population. The 28 teeth examined up to 1994 showed that both males and females (see Appendix VI for dental ages) were on average about two years old. Only three animals were four or more years of age. Two animals had pink discoloured canine teeth, and in one of these cases the enamel also showed white pitting. It was observed that eight animals had, at some time, suffered fractures to a canine tooth, often resulting in exposure of the pulp cavity. An animal caught in a snare and the suspected captive release animal from Hampshire both had damage to the left and right canines, but in five cases a left canine tooth only was damaged. One immature animal had upper and lower canines on the right side worn down, but not broken.

Fig 4. Bite wounds to the right cheek of a male otter. There is facial swelling and haemorrhage into the mouth.

Fig 5. Bite wounds (arrows) around the anus (A) and scrotum (S) of a sub adult male.

3.2.2 Internal Organs

Road traffic accidents were responsible for 64 (83 %) of the submissions and, in the great majority of cases, the only lesions in the internal organs were those associated with physical trauma. In a few cases there was evidence of trauma in animals found well away from roads. Two of these were alongside railway tracks, but the internal lesions resembled those seen in road traffic cases and it was apparent that the otters had been killed by trains. However, a pregnant female (M200/1/92) found dead on a sporting estate on the River Tamar had severe subcutaneous bruising and haemorrhage over the thorax but no fractures or organ damage. The cause of death was not established.

Similar lesions were seen in an adult female found near Chew Reservoir, Somerset (12/6/92) and in a cub found floating in the River Lyn, Cornwall (M290/9/94) but both these cases were too badly decomposed for detailed examination. An otter from the White River, St Austell (M129/3/92) which had a multiple-strand wire snare around its abdomen was also in partly decomposed condition. The snare had cut through the skin and deep into the flesh, perforating the abdomen at one point. The free end of the snare was frayed and it is possible that the animal had chewed through it, although there was damage only to the incisors and canines, and not the cheek teeth.

Alimentary System: No abnormalities were seen in the mouth or oesophagus of any of the otters. The simple stomach frequently contained food remains and, although these were not examined in detail, it was apparent that eels were the most common food item. Frogs were present in a few cases and what appeared to be salmonid ova were seen in one case. No avian or mammalian remains were recognised and no nematodes or other parasites were seen. An immature male otter (M153/3/95), which was emaciated and had suffered severe bite wounds to its hind feet, had scattered ulcers in the pyloric area of the stomach. There was bloody fluid in the stomach and intestines plus blackish scour material around the anus. The rest of the carcase appeared anaemic. One otter with severe bite wounds to the feet and scrotum also had haemorrhagic fluid in the stomach but no visible ulcers (M31/9/93).

Respiratory System: Otter lungs are unusual, with two lobes on the left side but four on the right. The fourth lobe lies medial to the diaphragmatic lobe and the apical lobe is served by an accessory bronchus. In no case were lesions seen in the larynx, trachea or bronchi. One animal (M216/11/92) had scattered 1 mm diameter, white, focal lesions in the diaphragmatic lobes of the lungs. No bacteria were isolated. A second animal (M153/3/95), (see earlier comments under Alimentary system), had a collapsed left diaphragmatic lobe which was believed to be a complication due to its other injuries, and not primarily a respiratory problem.

Thyroid Glands: The thyroid glands are flat, elongated and tapering. They are rather inconspicuous, lying closely applied to the lateral surfaces of the caudal larynx and proximal trachea. There is no isthmus. The left gland is often slightly longer and heavier than the right. Although no specific gross lesions were seen the glands varied in colour from pale pinkish tan to reddish brown. The glands were generally larger in males, with a mean combined weight of 0.65g compared with 0.47g for females. This difference was highly significant; Student's t, $p \leq 0.01$).

Thymus Gland: The cardiac thymus was present in the majority of otters but a cervical thymus was only seen in one specimen. The glands varied considerably in size, and the largest were not necessarily in the youngest animals. One adult female had a thymus which weighed 50g but in most cases it weighed less than 10g.

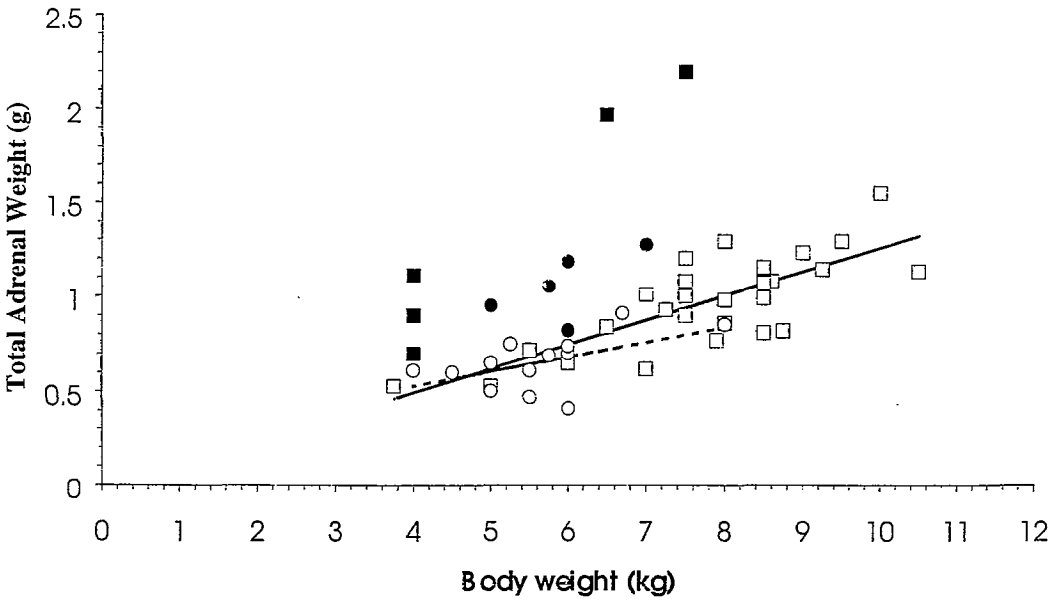
Cardiovascular System: One animal (M153/3/95, see earlier) had a gelatino-fibrinous mass adherent to the left atrio-ventricular valve but apart from this case no cardiovascular defects were seen. The left ventricle of the otter is exceptionally muscular and, in comparison, the right ventricle is rather thin walled. These features, together with the unusually globular form of the ventricles, could be interpreted as hypertrophy by anyone not familiar with the otter's heart.

Spleen: The otter's spleen is elongated and roughly triangular in section. No specific lesions were seen but there was considerable variation in size. In most males the spleen weighed between 40 and 50g but in nine animals it weighed less than 25g, often only 10-15g. Six out of these nine otters were suffering from bite wounds. Although many otters died of hypovolaemic shock as a result of road accidents such cases did not have small spleens.

Pancreas: The diffuse pancreas is attached to the anterior mesentery and is bilobed. No specific lesions were seen, although it was an organ that was typically badly damaged by trauma and/or autolysis.

Adrenal Glands: The otters' adrenal glands are symmetrical, flattened and bean shaped. In four animals there was evidence of abnormality. Two otters with severe bite wounds had both glands visibly enlarged, one markedly so, and a third otherwise normal animal, had an enlarged right gland only. In the fourth case, the glands were approximately normal size but the cortex had a very nodular appearance. In several other animals the adrenals were slightly nodular. A female in late pregnancy and three lactating females also had relatively enlarged glands. Figure 6 shows the weight of adrenal glands in relation to body weight, and illustrates the degree of adrenal hypertrophy seen in some of these cases.

Figure 6 - Relationship of Body Weight to Adrenal Weight



Males which died due to bite wounds are demonstrated by ■ and females in late pregnancy or lactating are demonstrated by ●. The solid regression line is calculated from the remaining males (□) and the broken line from the remaining females (○).

Liver The multilobular liver has well defined lobes and a gall bladder is present, lying on the right side. The cystic bile duct discharges into an ampulla on the wall of the duodenum. Alongside this is a common hepatic bile duct, formed by an unusual complex of ducts coming direct from the individual lobes. Anastomoses occur between the various ducts. Scattered white focal lesions 1-2 mm diameter were seen in the liver of one animal but apart from this case no liver lesions were seen.

Kidneys The otter's kidneys are multilobular and are normally of equal size. In two females, one lactating and one pregnant, the right kidney was approximately 30 % heavier than the left but no pathology was apparent. Although renal calculi are frequently seen in captive otters, and were systematically looked for in this study, none were seen on gross examination.

Ureters and Bladder One animal had a diverticulum to the wall of the bladder but this was considered to be a development defect and of no pathological significance. No abnormalities were seen in ureters.

Reproductive System One male otter had a vestigial undescended left testicle. The other testicle appeared rather small and the animal itself was unusual in that it looked immature and weighed only 3.7 kg, yet the wear on its teeth resembled that of an older animal (M340/10/94). Despite severe bite wounds to the scrotum in several males the testicles were normally undamaged.

Out of 25 female otters which were suitable for examination, 13 had what appeared to be immature genital tracts with no evidence of current or earlier sexual activity, and three of these uteri were unusual in that they were dark reddish black in colour. Two otters were pregnant, both with a single foetus in the right uterine horn. The first foetus, seen in January 1992, was well developed and had a crown rump length of 13.2 cm (Fig. 7a). The dam (M200/1/92) was killed near Tavistock.

The second foetus was much younger, with a crown rump length of 1.5 cm; its dam was killed near Delabole, north Cornwall in October 1992 (M192/10/92). However, the uterus in this second case was unusual in that both horns were severely convoluted, with multiple cyst-like dilations, approximately 5 mm in diameter, along their length (Figure 7b). Very similar uterine changes were seen in another female (M455/2/90) killed on the road at Newbridge, west Cornwall. It had seven glassy nodules/cysts, approximately 4-5 mm diameter plus one of 6-7 mm diameter in each horn (Figure 8a).

A less severely convoluted, mature uterus was seen in an otter killed in Somerset in 1993 (M206/12/93) but in that case the cysts or nodules were poorly defined. This otter also had what appeared to be a corpus rubrum in the right ovary, but no visible follicles. The uterus of all the other otters was in the form of a straight "Y" with a very short body (Figure 8b).

A young female killed in January 1996 (M219/1/96) had colostrum-like fluid in its mammary glands. Its uterus was large and flaccid (5-6 mm diameter) but no placental scars were visible. Similar colostrum-like fluid was seen in the pregnant otter from Delabole (M192/10/92).

Four animals were lactating. One of these (M216/11/92), killed in November 1992 had a placental scar in each uterine horn. In another female, examined in November 1995 (M81/11/95) the uterus was still large but detailed examination was not possible because it had been severely damaged in a road accident. In the other two lactating females the uterus had regressed to normal size. Both of these animals were killed in February, one in 1994 (M301/2/94), the other in 1995 (M332/2/95), and in each case the nipples were large, suggesting that they had been lactating for some time. A further three adult females were examined which, although not lactating, showed evidence of having bred, with large, well developed uteri (17/11/91, 12/6/92 and M36/12/92). One also had two enlarged nipples.

A summary of significant gross post mortem findings and causes of death, together with data on sex, body weight, length, thyroid weight and adrenal weight, is given in Table 2.

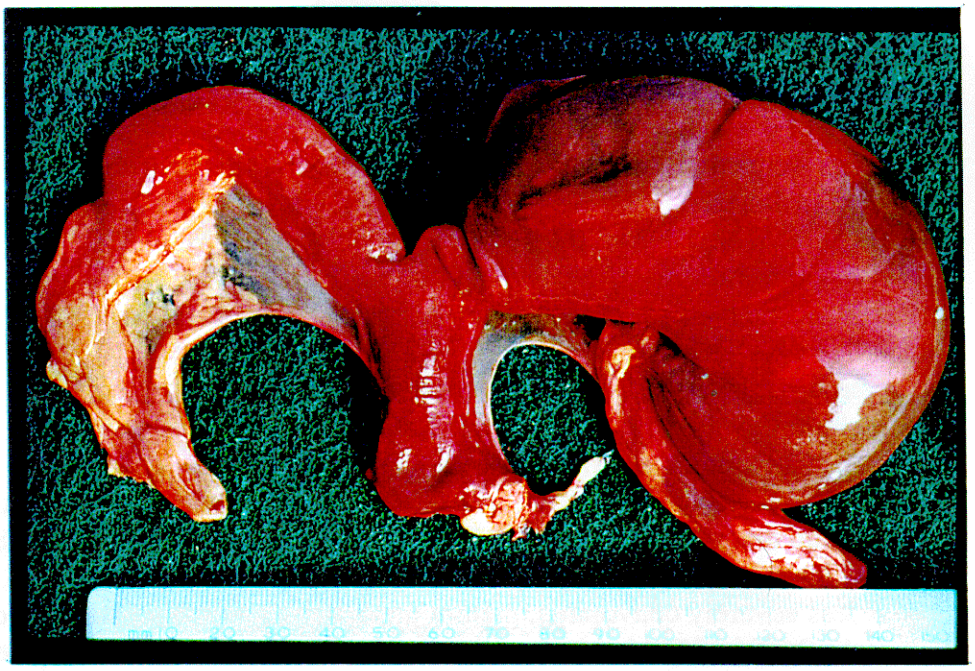


Fig 7a. Dorsal view of a reproductive tract containing a single large foetus in the right uterine horn (M200/1/92).

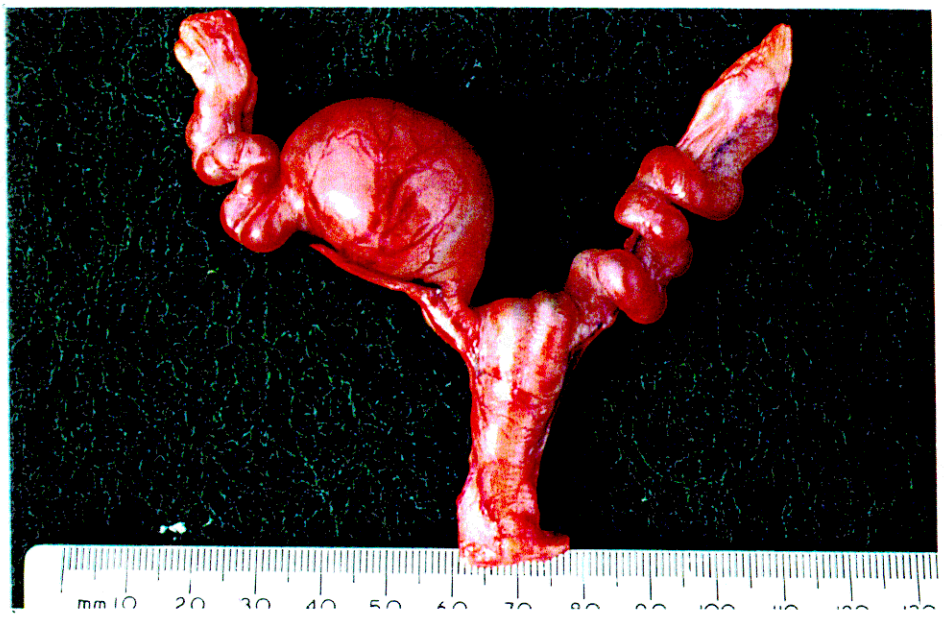


Fig 7b. Ventral view of a reproductive tract showing convoluted uterine horns and multiple nodular swellings. The larger swelling in the right horn contained a small foetus (M192/10/92).

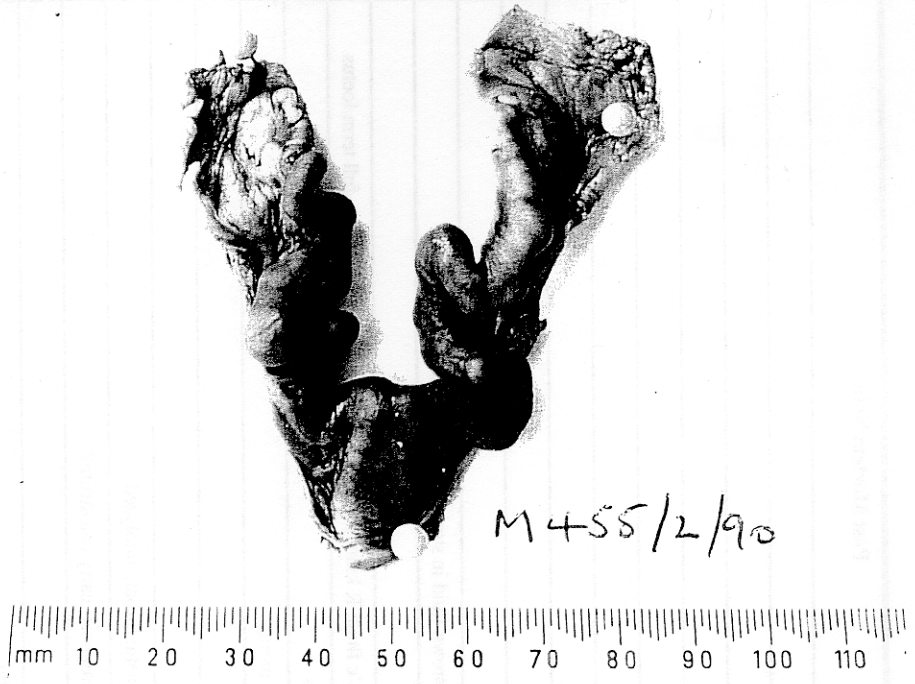


Fig 8a. Ventral view of a reproductive tract showing multiple nodular swellings in convoluted uterine horns.

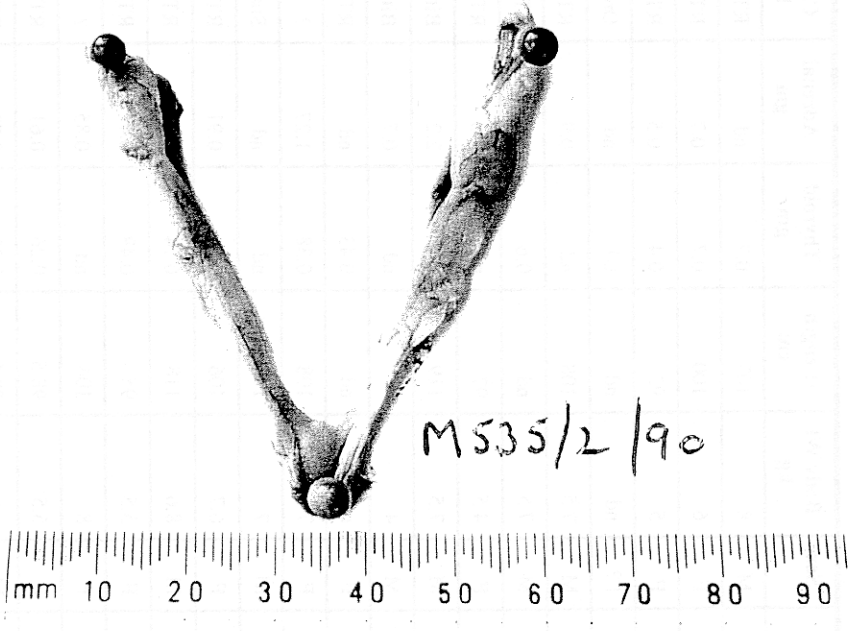


Fig 8b. Ventral view of a reproductive tract in a normal young adult.

Table 2 - Summary of causes of death, post mortem findings and some morphometric data

Case Ref.	Sex	Body Wt kg	Length cm	Thyroid gm	Adrenal gm	Cause of Death	Post Mortem Notes
M310/12/88	M	8	107	0.7	nd	RTA	NSL
M455/2/90	F	6	100	0.7	0.7	RTA	Convoluted mature uterus
M535/2/90	F	5	92	0.4	0.5	RTA	Immature uterus
M701/5/90	F?	nd	nd	nd	nd	Drowned	NSL Newborn cub
M125/6/90	M	7.5	108	nd	0.9	RTA	*Ixodes* ticks
M111/9/90	M	7.5	nd	0.6	1	RTA	NSL
M238/12/90	F	4.5	97	nd	0.6	RTA	Immature uterus
M616/1/91	M	7.5	119	1.1	2.2	Bites	Bite wounds: face; sepsis
23-03-91	M	4	87	nd	0.7	Bites	Bites: face; perineum; feet; brown fluid in stomach; small spleen
17-11-91	F	nd	nd	0.43	nd	RTA	Mature uterus
M200/1/92	F	7	108	0.38	1.27	?	S/c haem. Oedema; haem. L'c fluid; R kidney larger than L; adrenals large; single full term foetus
M129/3/92	M	7	114	nd	nd	Snared	Emaciated; fractured canines
M188/3/92	F	6.7	106	0.58	0.91	RTA	? Bites: face, feet; immature uterus
04-04-92	M	8.6	118	0.74	1.08	RTA?	Cut pads; lame hind leg
28-04-92	F	5.5	95	0.49	0.47	RTA	Cut pads; immature uterus
12-06-92	F	8	104	nd	0.85	?	Mature uterus; s/c oedema/haem: throat; autolysed
M192/10/92	F	5.5	98.5	0.26	0.61	RTA	Convoluted mature uterus; pregnant; early colostrum?
M193/10/92	M	5.5	99.5	0.32	0.72	RTA	Bites: scrotum
M177/11/92	M	9	113	0.84	1.23	RTA	Scarred pads; adrenals nodular
M216/11/92	F	6	99	0.52	1.18	RTA?	Mature uterus; lactating; R kidney larger than L; focal lung lesions
M36/12/92	F	5.75	107.5	0.44	1.05	RTA	Mature uterus; large nipples; worn teeth

Table 2 - Summary of causes of death, post mortem findings and some morphometric data

Case Ref.	Sex	Body Wt kg	Length cm	Thyroid gm	Adrenal gm	Cause of Death	Post Mortem Notes
M91/12/92	M	8.5	111	0.9	1.1	RTA	*Ixodes* ticks
M34/2/93	F	6	101	0.27	0.41	RTA	Immature uterus; autolysed
M30/3/93	M	4	89	0.77	0.9	?	? Bite wounds; small spleen
M224/3/93	M	7	106	0.77	0.62	RTA	NSL
M225/3/93	F	5	91.5	0.55	0.65	RTA	Immature uterus
M115/4/93	M	9.5	126	nd	nd	RTA	NSL
M135/7/93	M	8.5	113	1.05	0.52	RTA	*Ixodes* ticks
M31/9/93	M	6.5	114	0.58	1.97	Bites	Bites: face, feet, scrotum; ulcerated pads, emaciated: blood in stomach; small spleen, large adrenals
M131/10/93	M	8.5	115	0.96	0.99	RTA	NSL
M154/12/93	M	8.5	109.5	1.19	1.07	RTA	NSL
M206/12/93	F	5.75	101	0.66	0.69	RTA	Convoluted mature uterus; healed pad cuts
M281/12/93	M	8	108	0.77	1.29	RTA	Large R adrenal
M301/2/94	F	6	100	0.56	0.82	RTA	Mature uterus; lactating
M348/2/94	F	nd	nd	0.48	nd	RTA	NSL
M364/3/94	M	9.25	115	0.69	1.14	RTA	Teeth worn, pink, spotty
M161/4/94	M	8	118	0.56	0.86	RTA	Teeth pink, enamel eroded
M290/6/94	M	7	115	0.43	1.01	RTA	Medial deviation L & R front 3rd toe; thorax: two patches white fur; small spleen
M210/7/94	M	nd	nd	0.51	1.26	RTA?	NSL, skinned
M77/8/94	M	7.25	107	0.75	0.93	RTA	Bites; face, hock
M289/9/94	F	6	nd	nd	nd	RTA?	Bites - old: perineum; U/S
M290/9/94	M	2	62	0.17	nd	?	Cub, s/c bruising: thorax; autolysed

Table 2 - Summary of causes of death, post mortem findings and some morphometric data

Case Ref.	Sex	Body Wt kg	Length cm	Thyroid gm	Adrenal gm	Cause of Death	Post Mortem Notes
M311/9/94	M	6	103.5	0.71	0.65	RTA	Bites: head, perineum, small spleen
M211/10/94	M	9.5	117.5	1.13	1.29	RTA	NSL
M340/10/94	M	3.75	91	0.31	0.53	RTA	Stunted? Teeth worn: small kidneys and spleen, cryptochid
M13/11/94	M	10	114.5	0.57	1.15	RTA	Cut foot; lungs congested L oedema
M152/11/94	F	4	95	0.22	0.61	Bites	Immature uterus; bites: face, perineum, shoulder, stunted? autolysed
M180/11/94	F	6	107.5	0.66	0.7	Train	Immature uterus, pigmented
M219/11/94	M	6.5	110	0.37	nd	RTA	Dental calculus
M4/12/94	M	7	nd	0.4	nd	RTA	NSL, autolysed
M90/12/94	M	6.5	100	nd	0.81	RTA	NSL
M139/12/94	M	5	101.5	nd	0.53	RTA	Bites: feet, perineum, axilla; eroded pads, small spleen
M173/12/94	F	6	103	nd	nd	RTA	Immature uterus; large vulva
M44/1/95	M	10.5	124	nd	1.13	RTA	NSL
M110/2/95	M	7.5	112	0.78	1.2	Train	NSL
M332/2/95	F	5	96.5	0.55	0.95	RTA	Mature uterus; lactating
M112/3/95	M	8.75	nd	nd	nd	RTA	NSL
M153/3/95	M	4	110.5	0.29	1.11	Bites	Bites: feet; emaciated; stomach ulcers, blood in SI; heart valve lesion: small thymus, spleen, large adrenals
M157/5/95	M	4.7	98	0.67	nd	RTA	Large thymus, small spleen
M89/8/95	F	5.75	101	0.35	nd	RTA	Immature uterus; bites: feet, perineum
M90/8/95	F	6	100	0.51	0.74	RTA	? Immature uterus, pigmented; worn teeth
M71/10/95	M	8	108	0.6	0.98	RTA	NSL
M73/10/95	M	nd	nd	nd	nd	RTA	NSL U/S

Table 2 - Summary of causes of death, post mortem findings and some morphometric data

Case Ref.	Sex	Body Wt kg	Length cm	Thyroid gm	Adrenal gm	Cause of Death	Post Mortem Notes
M62/11/95	M	7	108	0.34	nd	RTA	?Bites: perineum, autolysed
M70/11/95	M	8.75	115	0.95	0.82	RTA	NSL
M81/11/95	F	5.25	103	0.47	0.75	RTA	Bites: feet; mature uterus; lactating
M82/11/95	F	8.5	118	nd	0.81	RTA	NSL
M153/11/95	M	7.5	106.5	0.7	1.08	RTA	Bites: feet, hock, perineum
M62/12/95	M	nd	nd	nd	nd	RTA	NSL, worn teeth, U/S
M100/12/95	M	8.5	117	0.57	1.15	RTA	NSL, worn teeth, U/S
M134/1/96	M	9.5	111	0.62	nd	RTA	NSL, U/S
M190/1/96	M	nd	92.5	0.39	nd	RTA	NSL, U/S
M191/1/96	F	nd	89	0.48	0.46	RTA	Immature uterus, pigmented
M219/1/96	F	nd	106	0.44	0.59	RTA	Mature uterus; colostrum; fractured cheek; shrunken R eye
M71/2/96	F	nd	87.5	0.53	0.41	RTA	Immature uterus
M50/3/96	F	nd	nd	nd	nd	RTA?	? Immature uterus
M111/3/96	M	7.9	112	0.5	0.77	RTA	NSL

List of Abbreviations

RTA:	Road Traffic Accident
NSL:	No Significant Lesions
S/c:	Subcutaneous
?:	Denotes uncertainty
U/S:	Unsuitable for examination

SI:	small intestine
p.c.:	pericardial
haem:	haemorrhage/ic
L:	left
R:	right

KEY (Table 2)

RTA:	Road Traffic Accident
MAT UT:	Mature Uterus
IMM UT:	Immature Uterus
CONVOL:	Convoluted
PERIN:	Perineum
S/C:	Subcutaneous
HAEM:	Haemorrhage/ic
P/C:	Pericardial
R:	Right
L:	Left
ADR:	Adrenal Gland
LACT:	Lactating
CONG:	Congestion/ed
F:	Front
H:	Hind
?BITES:	Possible bite wounds
SI:	Small intestine
HT:	Heart
U/S:	Unsuitable for Examination
?	Indicates uncertainty
NSL:	No significant internal lesions
>	Greater than
-	No value available
AUTOL:	Autolysed

4 BACTERIOLOGY RESULTS

Apart from the lesions associated with bite wounds there was, in most cases, little justification for carrying out bacteriological examination. For this reason, and because of various constraints, samples of heart blood, lung and/or liver were cultured from only eleven otters. Fifteen animals were checked for possible salmonella infection by culturing intestinal contents. All proved negative.

Pasteurella haemolytica was isolated in pure culture from the lung, but not the liver, of an otter which had apparently been run over. It seems unlikely that this infection was causing clinical disease as no typical lung pathology was present. Of the other ten animals cultured, six were suffering from bite wounds. A sample collected directly from a wound yielded a mixed growth of *Streptococcus equisimilis* and a coagulase negative *Staphylococcus* sp. *Streptococcus equisimilis* together with *Streptococcus dysgalactiae* was isolated from heart blood in another case and *Streptococcus canis* from both liver and heart in a third case. The animal which died in a snare yielded a mixed growth of *E. coli* and *Streptococcus faecalis* and the same organisms were recovered from two other otters. However, these carcases were not in fresh condition and the organisms could well have invaded the animal's tissues after death. In five cases, including two suffering from bite wounds, no organisms were isolated from any major organ.

5 HISTOPATHOLOGY RESULTS

Tissues from 46 otters were examined between December 1988 and March 1996. (See Appendix III). In many cases, a combination of trauma, autolysis and freezing meant that only a few tissues were suitable for processing. Wherever possible, selected, or 'target' tissues were routinely placed in fixative. Other tissues were examined less systematically. However, because of various constraints not all fixed tissues were processed and some samples remain archived. The following account is a brief overview of the results of the histopathological examinations. A more detailed description of certain tissues will be reported elsewhere.

5.1 Liver

Liver samples were examined from 31 otters. In only a few animals, e.g. a young female from Lelant (M71/2/96) were the bile ducts 'normal' in appearance when compared to other species. In the majority of otters they were convoluted, hyperplastic and surrounded by fibrous tissue. In some cases, e.g. M646/1/91, Gunnislake and M219/1/96, St Austell, the changes were pronounced. The epithelial cells lining the bile ducts were normally tall columnar although in a few cases they were low columnar. The hepatic parenchyma was generally unremarkable, although areas of focal necrosis and replacement fibrosis were seen in an otter from Newquay (M70/11/95) and herpes-like inclusions were present in hepatocyte nuclei of an otter from the River Brue (M206/12/93). In some otters there was fatty vacuolation of the hepatocytes and this was most noticeable in an animal with a very low Vitamin A status (M340/10/94).

5.2 Kidneys

Although no otters showed gross lesions of renal calculi, one animal from Devon (M89/8/95) had a small calculus in the lumen of a calyx. A second otter from Devon (M332/2/95, Holsworthy) had an irregular mineralised body within a medullary pyramid. However, this animal had an extensive, mononuclear cell, interstitial nephritis affecting both kidneys plus cellular debris and a single small nematode larva lying free within the renal pelvis. In many of the 39 kidneys examined the glomeruli and/or proximal convoluted tubules were distended with acidophilic proteinaceous fluid. It was thought that this was possibly due to reflux of tubular fluid occurring at the time of death.

Unexplained basophilic bodies, often granular or sometimes laminated, were seen in the medulla of many otters' kidneys, particularly from 1994 onwards. Their exact location is uncertain but they appear to be within the lumen of very small tubular structures lined by flattened epithelial cells, possibly the descending (thin) loop of Henle. There was no evidence of pathology associated with these bodies although the convoluted tubules sometimes appeared dilated and compressed against one another. The smaller basophilic bodies possibly represent salts which have precipitated out after death, or are a fixation artefact, but this is unlikely in the case of the larger bodies, especially the laminated ones.

Cortical interstitial fibrosis was seen in the kidneys of an immature female from Torrington which died of bite wounds (M152/11/94). At the time of post mortem examination this animal had a stunted appearance and its condition index was low for a female (0.90). Another stunted animal, the cryptorchid male from Goss Moor (M340/10/94) had widespread cortical tubular and glomerular degeneration. Even allowing for its low body weight and low condition index (0.77) the kidneys of this animal were unusually small and this is consistent with the pathology. An unidentified protozoal schizont was present in the cortex of one otter (M364/3/94). Unfortunately, the carcase had been frozen and detailed structure of the parasite had been lost.

5.3 Thyroid Gland

Thyroid glands from 35 otters were examined. In almost every case the majority of the follicles appeared small, although there was some variation both within and between lobules. Colloid was generally sparse and in most follicles it stained very pale greyish pink by haematoxylin and eosin. In some cases the follicles were uniformly very small, with almost no visible lumen, and colloid was minimal or absent (e.g. M219/1/96, M89/8/95). The follicular epithelial cells were either cuboidal or low columnar and the cytoplasm often stained poorly. Many glands had a few extremely large, irregular follicles which were lined by flattened epithelial cells. The colloid in these atypical follicles stained dark pink and often contained detached epithelial cells.

A small number of otters (e.g. M332/2/95 from Holsworthy) had consistently larger, more well developed follicles which contained pink-staining colloid. The epithelium in these cases was mostly low cuboidal, although in the larger, more rounded, follicles it was flattened and the colloid stained darker. Most glands were well vascularised and in some cases the capillary supply to the follicles was prominent. Lymphocytic foci were seen in one case.

5.4 Adrenal Gland

A common feature of adrenal glands (n=28) was the presence of focal accumulations of lymphocytes either within, or beneath, the capsule. In many cases, the outer layer of the cortex, the zona glomerulosa, was sub-divided into distinct pockets by fibrous extensions of the capsule. In some instances (e.g. M90/12/94) there were proliferative foci of cortical cells which were enclosed completely within capsular tissue. The cells of the zona glomerulosa and zona fasiculata often appeared effete and without lipid droplets. In some otters, the columns of cells in the zona fasiculata were severely disrupted and normal architecture was lost.

Cortical necrosis was seen in an otter from the River Taw (M71/10/95) and similar changes were seen in one from Somerset. This animal (M153/3/95) had enlarged adrenals and had died from bite wounds. However, interpretation was difficult in this case due a degree of autolysis.

Even in animals with visibly enlarged adrenals there was no obvious hypertrophy of the cortex relative to the medulla. The capsular and cortical changes referred to above were considered to be responsible for the nodular appearance of some of the glands. Haemorrhage at the base of the inner cortical layer, the zona reticularis, and within the medulla was commonly seen. Other lesions seen in the medulla were focal mineralisation, necrosis and, in one case, focal infiltration by lymphocytes (M157/5/97). Unfortunately, because of autolysis or freezing, some of the larger adrenal glands were unsuitable for histopathology and others, although they have been held in fixative, have not been processed.

5.5 Footpad

The other tissue regularly sampled was foot pad and sections were examined from 26 animals. Mild epidermal hyperplasia and hyperkeratosis was seen in several cases and one of these was a male which had eroded pads and bite wounds to the feet (M31/9/93). Apart from the changes to the epidermis, there were also bacterial colonies developing within the scent, or modified sweat, glands. The cryptorchid otter (M340/10/94) also had a bacterial infection of the epidermis. Changes of both a proliferative and a degenerative nature were seen on a few occasions affecting the scent glands in the foot pad.

5.6 Other Organs

Other organs which were examined less consistently were lung, thymus, spleen, uterus, salivary gland, eye and urinary bladder. Occasional lesions only were recorded in these tissues. The white focal lesions, 1-2 mm diameter, seen in the lungs of M216/11/92 were granulomas, with an outer rim of mononuclear cells, eosinophils and a central core of macrophages. Within this core there was often a hollow structure with an eosinophilic staining wall. The lesions were consistent with a diagnosis of adiaspiromycosis, caused by inhalation of fungal spores. In other lesions no central body was visible but Ziehl-Neelsen stained sections showed no evidence of mycobacteria. Routine examination of lungs showed the presence of fungal spores in several more cases. Peribronchial accumulations of lymphocytes were seen in several otters. An otter which died of bite wounds (M646/1/91, see Fig. 4) had multiple thrombosed blood vessels containing colonies of bacteria which were surrounded by a heavy infiltration of neutrophils and macrophages.

Lesions in other organs included mineralised deposits in the collecting ducts of salivary glands with associated fibrosis (M77/8/94, M211/10/94), and follicular depletion of lymphocytes from spleens and thymus glands. In one otter the cornea had been perforated by a foreign body composed of plant material. There was associated inflammatory reaction in the anterior chamber of the eye. Corneal ulceration with evidence of earlier trauma was seen in the apparently sound eye of M219/1/96. This was the young female with a collapsed shrunken right eye and fractured cheek bone. Unidentified cells resembling adipose tissue were seen in the posterior chamber of an otter from the River Taw (M71/10/95). In many otters there was evidence of retinal folding but this was thought possibly to be a fixation artefact. Whilst examining another eye section (M70/11/95) several protozoal bodies were seen within the muscle fibres of the external eye muscles and these were identified as *Sarcocystis* sp. Testes were examined regularly from October 1994 and in only one case was there no evidence of spermatogenesis. This was the cryptorchid otter from Goss Moor (M340/10/94) and both the descended and the undescended testes were inactive. A variety of histological changes were seen in various uteri but a detailed description of these is beyond the scope of this summary and they will be reported at a later date.

5.7 Hypovitaminosis A Cases

Seven otters had hepatic Vitamin A levels below 7 μmol/kg and tissues were taken from five of them for histopathological examination. However, for economic reasons only a limited number of tissues were taken in each case and not all of these were processed (see Appendix III).

Unfortunately, in only one case was the salivary gland included, but the urinary bladders were examined from three and the kidneys from all five. The epithelium lining the salivary gland ducts, the renal calices and urinary bladders appeared normal in all cases. Eyes were retained in fixative from three out of the seven but none were processed. (However, see 9.1d.)

The thyroid glands were examined from five otters with low Vitamin A levels. In view of the relationship between Vitamin A and thyroid activity, (see 6.6 and 6.7) it might have been expected that the follicles in these cases would appear inactive, with accumulation of colloid and flattened epithelial cells. However, although there was some variation between the cases, the general appearance of these glands was not significantly different from that in many other otters. The predominance of small follicles, often with pale staining epithelial cells and meagre, pale staining colloid, may be a normal feature of otters. However, in a few cases, the follicles and colloid more closely resembled those seen in normal domestic animals. Furthermore, the author has observed much more "normal" looking thyroid glands in otters bred in captivity (Simpson, unpublished data). It is possible that the thyroid glands of all the wild otters examined have been influenced to some degree by the pollutants, such as PCBs and OCs, present in their tissues.

6 BIOCHEMISTRY AND TOXICOLOGY RESULTS

Liver samples from 56 otters, collected between December 1988 and February 1996, were analysed by NRA/EA in three batches. Determinands were fat, organochlorine pesticides and their metabolites, industrial organochlorines, PCBs, organophosphate pesticides, triazine herbicides and selected metals. The levels of fat, selected organochlorines and PCBs are shown in Table 3. PCB congeners are referred to by their IUPACS numbers (Ballschmiter and Zell, 1980) and total PCB is expressed as Aroclor 1260 equivalent.

6.1 Organochlorine Pesticides

Although pp' DDT and its derivatives, pp' DDE and pp' TDE, were present in almost every sample in batch I, their respective *ortho-para* isomers were recorded in only a single sample. For this reason, presumably, no results were received for op' DDE or op' TDE in batches II and III, and although op' DDT was analysed for in these batches it was not detected. These results are therefore excluded from Table 3. Similarly, α HCH was not detected in any sample and all but two were negative for the β isomer. Therefore, γ HCH values only are shown. Only one sample contained 1, 2, 3, trichlorbenzene, a breakdown product of γ HCH. No samples contained Endosulphan or Endrin. Aldrin was only detected twice and the Dieldrin levels in these two samples were not noticeably higher than in the rest.

6.2 Polychlorinated Biphenyls

PCB congener 28 was not detected in any sample and numbers 52 and 101 were each present in only three samples. Batches II and III only were analysed for congener 31 but it was not detected. For these reasons, none of these lower chlorinated congeners are included in Table 3. Congener 105 was not analysed for in batches I and II but was shown to be present in all samples in batch III. Similarly, congener 156 was not analysed for in batch I but was present in all but four samples in batch II and in all samples in batch III. The higher chlorinated congeners 118, 138, 153, and 180, were present in all but one sample (M216/11/92). No results were received for certain other potentially important congeners e.g. 126 and 169 and it is presumed that either they were not detected or were present at very low levels only.

6.3 Other Pollutants

Hexachlorobenzene (HCB), an organochlorine of petrochemical origin, was present in all samples and hexachlorobutadiene was detected in all samples in batch I but, somewhat surprisingly, was not detected in any of the samples in batches II and III. The herbicides Atrazine, Simazine and Trifluralin were absent from all but one sample, where Simazine was present at 67 μg/kg WM. No organophosphates were detected.

Table 3 - Halogenated Hydrocarbon Levels (µg/kg Wet Matter) and Vitamin A Levels (µmol/kg Wet Matter) in Liver. Batch 1

		OC Pesticides					PCB Congenors (IUPACS) Numbers								
Case Ref.	% LIPID	PP' DDE	PP' DDT	PP' TDE	Gamma HCH	Dieldrin	105	118	138	153	156	180	Total PCB (= 1260)	HCB	Vitamin A u mol/kg
M310/12/88	3.6	1005.0	11.2	159.0	N	288.0	N/A	21.3	78.9	67.7	N/A	40.0	540.0	25.4	2.1
M455/2/90	2.9	1087.0	17.3	172.0	N	2761.0	N/A	51.0	214.0	208.0	N/A	149.0	814.0	34.0	0.6
M535/2/90	2.0	261.0	4.8	42.7	N	161.0	N/A	23.8	48.6	60.9	N/A	39.7	123.0	24.8	47.5
M125/6/90	3.1	195.0	22.8	29.9	N	316.0	N/A	42.9	214.0	259.0	N/A	191.0	860.0	20.5	0.8
M111/9/90	2.4	343.0	10.7	53.2	N	645.0	N/A	20.5	140.0	132.0	N/A	93.8	546.0	16.2	0.6
M238/12/90	2.3	454.0	6.8	117.0	N	819.0	N/A	17.6	59.5	60.3	N/A	32.5	250.0	22.4	4.2
M646/1/91	1.9	407.0	22.4	61.3	N	100.0	N/A	147.0	329.0	434.0	N/A	233.0	1238.0	73.6	35.7
23/03/91	2.3	438.0	4.9	60.6	3.8	240.0	N/A	18.3	50.7	46.5	N/A	33.3	218.0	28.1	N/A
17/11/91	N/A	80.2	N	11.1	N	83.3	N/A	8.2	25.5	39.3	N/A	28.2	N/A	5.9	N/A
M188/3/92	2.0	323.0	5.6	69.3	8.3	549.0	N/A	43.4	142.0	172.0	N/A	84.0	591.0	23.2	N/A
04/04/92	8.3	242.0	12.1	28.9	17.7	168.0	N/A	41.4	225.0	307.0	N/A	297.0	1222.0	57.6	N/A
28/04/92	1.3	75.2	N	6.6	N	178.0	N/A	N	20.4	23.0	N/A	15.4	110.0	6.5	94.7
M216/11/92	N/A	35.6	N	N	N	18.6	N/A	N	N	N	N/A	N	N/A	3.3	N/A
M91/12/92	1.8	604.0	38.0	39.0	N	451.0	N/A	153.0	375.0	293.0	N/A	152.0	1265.0	47.8	11.9
M36/12/92	2.1	460.0	8.8	53.7	7.2	223.0	N/A	17.8	53.7	65.0	N/A	46.6	258.0	15.9	15.2
M30/3/93	1.4	632.0	5.1	153.0	6.1	148.0	N/A	21.0	41.0	52.2	N/A	33.7	180.0	27.6	544.0
M224/3/93	1.3	1894.0	3.5	528.0	3.7	921.0	N/A	24.5	80.9	78.9	N/A	47.3	316.0	36.5	867.0
M135/7/93	1.2	271.0	4.7	79.4	4.3	506.0	N/A	12.0	35.3	43.0	N/A	31.0	223.0	10.6	1230.0
M31/9/93	1.7	2397.0	28.2	261.0	2.7	430.0	N/A	230.0	407.0	542.0	N/A	299.0	1063.0	148.0	1333.0
M131/10/93	1.8	254.0	9.8	80.2	N	355.0	N/A	69.3	212.3	210.0	N/A	88.0	688.0	23.1	41.7
M154/12/93	N/A	491.0	11.2	78.0	N	371.0	N/A	137.0	340.0	347.0	N/A	209.0	N/A	35.6	4.9
M206/12/93	N/A	172.0	N	12.1	N	142.0	N/A	N	23.4	32.0	N/A	26.8	N/A	9.9	533.0
M281/12/93	N/A	189.0	N	N	N	151.0	N/A	32.5	177.0	199.0	N/A	97.4	N/A	20.7	19.5
SD	1.6	576.8	9.5	116.6	4.8	558.3	N/A	60.8	124.3	145.3	N/A	90.1	407.7	30.4	
Mean	2.4	535.2	12.7	99.8	6.7	435.9	N/A	56.6	149.7	166.9	N/A	103.1	583.6	31.2	
Number	18	23	23	23	23	23	N/A	23	23	23	N/A	23	18	23	
Range	1.2-8.3	35.6-2397	0-28.2	0-528	0-17.7	18.6-2761	N/A	0-230	0-375	0-542	N/A	0-299	110-1265	3.3-148	

Notes:

a) N = Nil Detected

b) N/A = No analytical result received. (Not analysed or measured.)

c) Total PCB expressed as Aroclor 1260 equivalent

d) HCB = Hexachlorobenzene

Table 3 - Halogenated Hydrocarbon Levels (µg/kg Wet Matter) and Vitamin A Levels (µmol/kg Wet Matter) in Liver. Batch II

		OC Pesticides					PCB Congenors (IUPACS) Numbers								
Case Ref.	% LIPID	PP' DDE	PP' DDT	PP' TDE	Gamma HCH	Dieldrin	105	118	138	153	156	180	Total PCB (= 1260)	HCB	Vitamin A u mol/kg
M301/2/94	3.8	21.4	1.0	6.9	1.4	40.5	N/A	4.3	10.8	17.2	N	13.7	132.0	2.8	80.2
M348/2/94	6.8	21.7	N	2.5	N	32.8	N/A	6.4	15.2	18.4	N	10.4	190.0	6.2	334.0
M364/3/94	2.5	140.0	5.8	40.7	N	198.0	N/A	37.2	N/A	N/A	28.7	N/A	1900.0	24.1	17.9
M161/4/94	2.5	541.0	1.3	255.0	N	392.0	N/A	13.9	45.2	46.6	5.3	24.4	305.0	15.4	78.0
M290/6/94	3.8	21.0	2.2	N	N	13.5	N/A	26.2	N/A	N/A	10.5	N/A	180.0	13.2	N/A
M210/7/94	4.2	681.0	2.2	394.0	1.7	588.0	N/A	N	N/A	N/A	29.6	N/A	1070.0	16.9	N/A
M77/8/94	6.2	214.0	1.0	15.5	N	164.0	N/A	13.0	122.0	142.0	10.0	36.6	323.0	8.9	30.4
M290/9/94	4.0	41.5	2.9	6.6	2.3	13.4	N/A	6.7	17.1	25.6	N	11.0	137.0	7.5	N/A
M311/9/94	2.4	78.2	0.6	10.3	N	70.4	N/A	5.2	13.7	18.2	N	8.3	134.0	6.0	73.7
M211/10/94	2.7	1025.0	2.2	217.0	1.8	629.0	N/A	39.2	553.0	625.0	28.1	433.0	1240.0	29.2	315.0
M340/10/94	5.8	72.9	1.0	5.4	N	95.8	N/A	19.7	36.5	45.6	6.7	29.5	348.0	13.8	N
M43/11/94	5.4	193.0	1.5	80.2	N	156.0	N/A	44.8	125.0	155.0	7.7	80.0	230.0	11.8	160.0
M180/11/94	5.7	43.4	N	4.5	N	26.5	N/A	19.8	47.9	61.9	7.6	23.0	283.0	11.4	1836.0
M219/11/94	3.3	13.8	N	4.2	N	30.1	N/A	4.1	16.9	22.0	3.4	14.4	76.7	3.4	48.0
M4/12/94	6.3	1374.0	4.0	960.0	N	2801.0	N/A	455.0	136.0	571.0	44.2	234.0	243.0	89.0	N/A
M139/12/94	4.9	376.0	0.6	272.0	N	267.0	N/A	28.7	103.0	108.0	13.2	36.4	391.0	28.4	25.5
SD	1.5	406.7	1.5	257.3	0.4	684.0	N/A	113.3	145.3	207.7	13.0	123.8	507.3	20.6	
Mean	4.4	303.6	2.0	151.7	1.8	344.9	N/A	48.3	95.6	142.8	16.3	73.4	448.9	18.0	
Number	16	16	16	16	16	16	N/A	16	13	13	16	13	16	16	
Range	2.4-6.8	13.8-1374	0-5.8	0-960	0-2.3	13.4-2801	N/A	4.1-455	10.8-553	17.2-625	0-44.2	8.3-433	76.7-1900	2.8-29.2	

Notes:

a) N = Nil Detected

b) N/A = No analytical result received. (Not analysed or measured.)

c) Total PCB expressed as Aroclor 1260 equivalent

d) HCB = Hexachlorobenzene

Table 3 - Halogenated Hydrocarbon Levels (µg/kg Wet Matter) and Vitamin A Levels (µmol/kg Wet Matter) in Liver. Batch III

		OC Pesticides					PCB Congenors (IUPACS) Numbers								
Case Ref.	% LIPID	PP' DDE	PP' DDT	PP' TDE	Gamma HCH	Dieldrin	105	118	138	153	156	180	Total PCB (= 1260)	HCB	Vitamin A u mol/kg
M152/11/94	2.4	49.9	N	N	N	25.1	4.5	16.5	40.8	44.6	8.7	32.2	394.0	6.9	970.0
M90/12/94	3.6	47.5	1.1	19.5	1.2	66.3	5.1	15.7	31.1	8.5	34.8	34.8	N	10.0	N/A
M44/1/95	3.4	52.8	5.4	19.7	N	33.7	14.6	31.0	111.0	70.5	36.8	38.6	472.0	11.3	N/A
M110/2/95	3.7	229.0	86.7	12.4	N	121.0	35.2	71.9	327.0	N	36.4	88.3	1930.0	14.7	N/A
M153/3/95	1.4	300.0	13.8	39.2	N	82.0	10.7	35.5	318.0	N	47.4	514.0	1560.0	11.6	541.0
M157/5/95	2.5	118.0	3.4	10.1	0.8	44.9	2.4	11.5	26.0	33.2	6.2	27.6	294.0	7.7	47.7
M89/8/95	1.7	73.4	34.6	14.4	N	39.4	3.0	12.7	49.1	57.6	5.8	17.1	551.0	7.6	N/A
M90/8/95	2.6	121.0	62.9	N	N	152.0	5.1	18.6	57.9	61.4	17.2	49.8	501.0	6.9	672.0
M71/10/95	1.5	125.0	1.8	48.3	N	46.4	4.2	15.7	54.3	63.8	9.3	66.5	247.0	10.2	125.0
M70/11/95	1.8	36.1	1.9	7.4	N	29.5	3.5	14.0	40.0	44.3	13.3	43.2	601.0	7.4	106.0
M81/11/95	1.8	95.7	14.4	25.3	N	59.8	N	4.1	15.4	23.2	5.2	29.4	244.0	2.7	N/A
M82/11/95	2.0	969.0	270.0	128.0	N	496.0	4.4	12.6	37.7	33.3	15.8	21.3	538.0	12.9	N/A
M153/11/95	1.8	17.7	2.5	5.8	N	22.7	3.7	14.6	61.4	78.4	18.5	74.0	265.0	4.1	74.0
M190/1/96	2.4	18.4	0.7	2.1	N	22.0	N	5.6	11.7	14.7	4.1	7.1	199.0	4.4	N/A
M191/1/96	2.1	345.0	1.7	18.5	N	39.7	4.1	15.3	25.3	31.4	7.6	27.1	111.0	16.9	575.0
M219/1/96	1.8	62.5	1.1	22.2	N	50.2	N	14.5	43.8	40.0	17.4	14.0	220.0	17.2	244.0
M71/2/96	1.0	48.3	1.9	25.6	N	23.1	3.0	8.6	21.7	26.0	9.0	25.1	N	3.9	139.0
SD	0.8	229.6	68.3919	30.6	0.3	113.1	8.7	15.7	114.2	20.7	13.4	117.7	515.2	4.5	
Mean	2.2	245.8	31.4938	26.6	1.0	79.6	7.4	18.7	74.8	42.1	17.3	65.3	541.8	9.2	
Number	17	17	17	17	17	17	17	17	17	17	17	17	17		
Range	1-3.7	17.7-969	0-270	2.1-128	0-1.2	22-496	0-35.2	4.1-71.9	11.7-327	0-78.4	4.1-47.4	7.1-514	0-1930	2.7-17.2	

Notes:

a) N = Nil Detected

b) N/A = No analytical result received. (Not analysed or measured.)

c) Total PCB expressed as Aroclor 1260 equivalent

d) HCB = Hexachlorobenzene

6.4 Fat Determination

The results of liver fat analysis are shown in Table 4. Values for other otters from south west England, as determined at two other laboratories between 1987 and 1993, are shown for comparison.

Table 4 - Liver fat content as percentage of wet weight.

Laboratory	n	Mean fat content %	SD
NRA: Batch I	18	2.41	1.60
NRA: Batch II	16	4.39	1.50
NRA: Batch III	17	2.21	0.77
University of Essex	9	3.88	0.78
ITE	7	3.33	0.48

It will be seen that there is quite a marked variation between batches I and II and also between batches II and III, but the reasons for this are unknown. However, liver samples from three otters examined in duplicate for fat content by both NRA/EA and Dr Mason, University of Essex showed poor agreement, viz 1.2 : 3.79, 1.08 : 4.03, 1.4 : 3.19. In many scientific reports on otters the concentration of organochlorine pesticides and PCBs are expressed as a proportion of the fat, or lipid, content of the tissue. Therefore, variations in fat content of the order seen above will have a profound effect on the OC and PCB values when they are expressed in weight of lipid. For this reason, all values in this report are expressed in weight of wet tissue.

6.5 Pollutant Trends Over Time

The concentrations of various important determinands in individual otters were plotted in chronological sequence (Figures 9 to 15). Although the X-axis in these figures is not on a true timescale, the results appeared to suggest a downward trend in values over the 1988 to 1996 period for pp' DDE, Dieldrin and possibly some of the PCB congeners, but not for total PCB.

The data were therefore re-examined statistically to determine whether there was evidence of trends through time for the principal pollutants. This was done by analysis of regression of the logarithms of the concentrations of the determinands against the dates of death. The results are shown in Figures 9a to 15a. When systematic differences were shown to exist for males and females separate regression lines are drawn. The decision to use the logarithms of the pollutant concentrations was taken because the distribution of the values appeared to be skew. One consequence of this was the exclusion of a small number of otters from the results where the level of the determinand was below the level of detection. This involved one animal each for PCB 138 and 180, three for 153 and four for 118.

Figure 9 - Total PCB Levels in Livers in Chronological Order (µg/Kg WM)

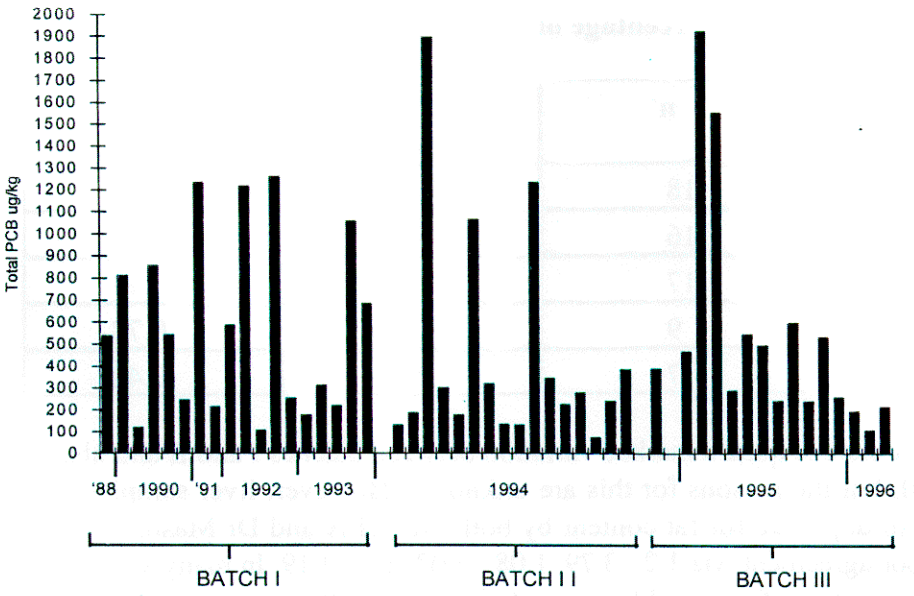


Figure 9a - Time series of log concentrations of Total PCB

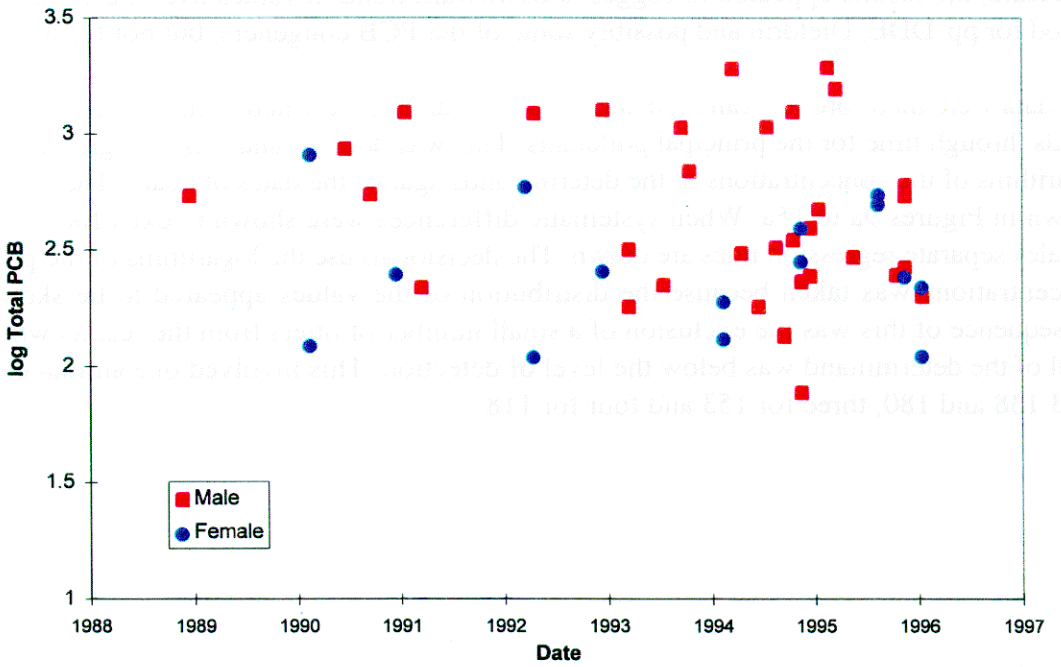


Figure 10 - PCB Congener 118 Levels in Livers in Chronological Order (µg/Kg WM)

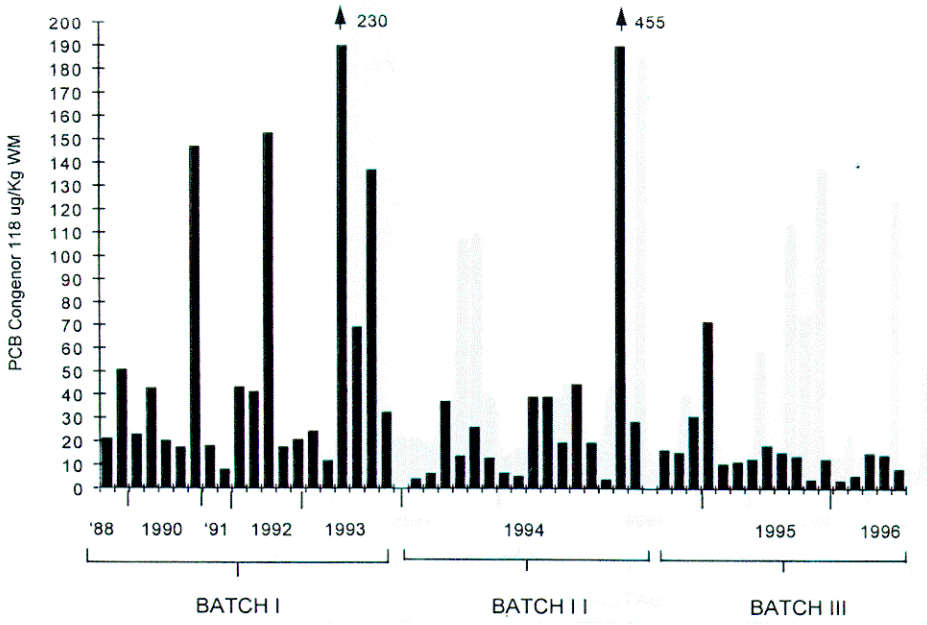


Figure 10a - Time series of log concentration of PCB 118

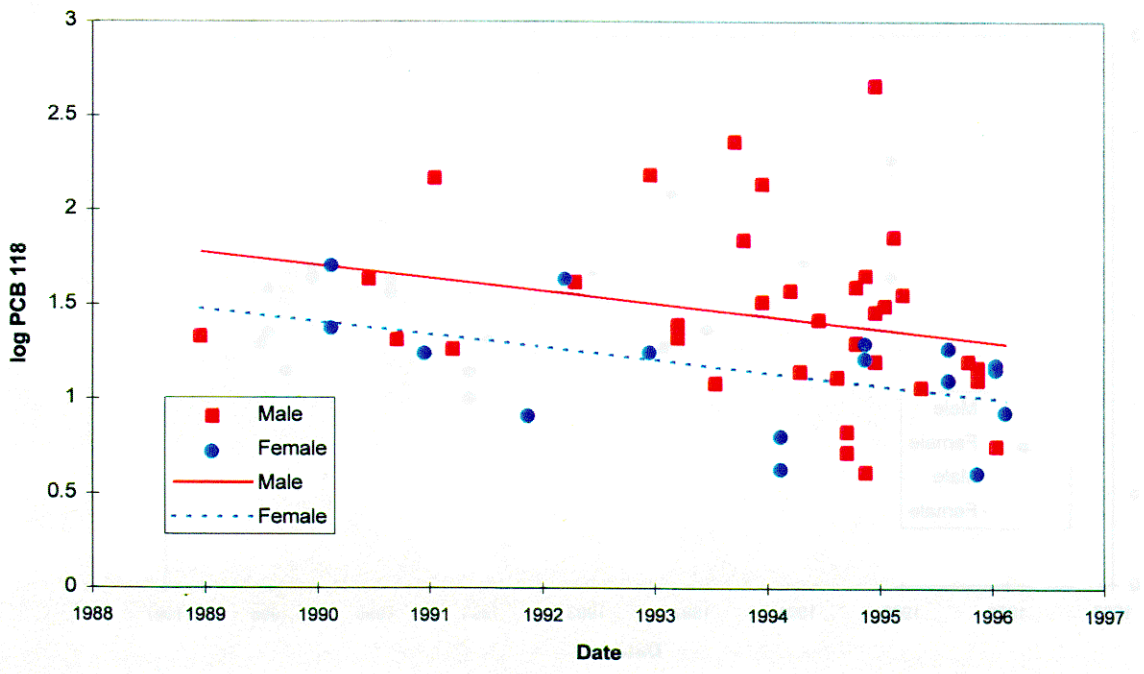


Figure 11 - PCB Congener 138 Levels in Livers in Chronological Order (µg/Kg WM)

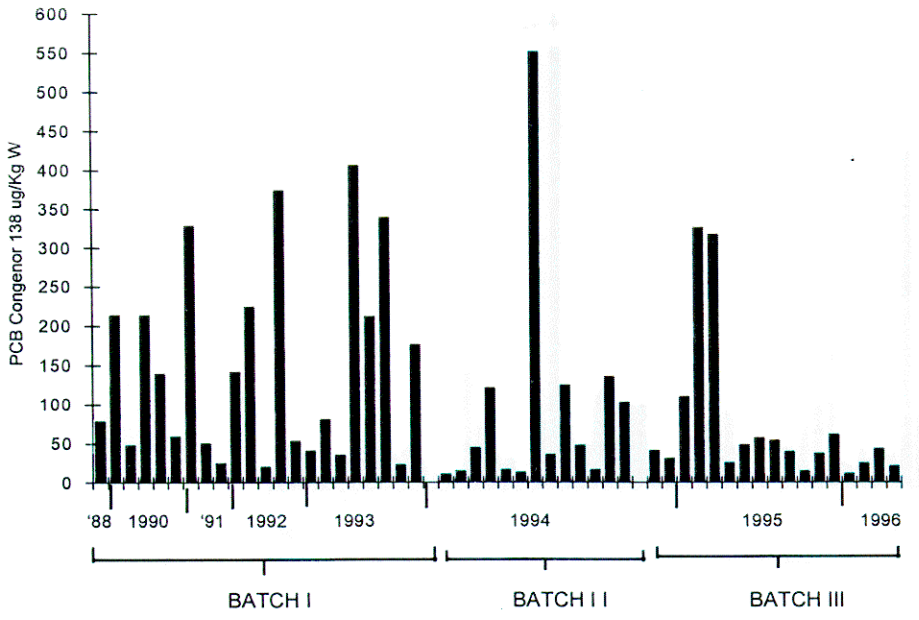


Figure 11a - Time series of log concentration of PCB 138

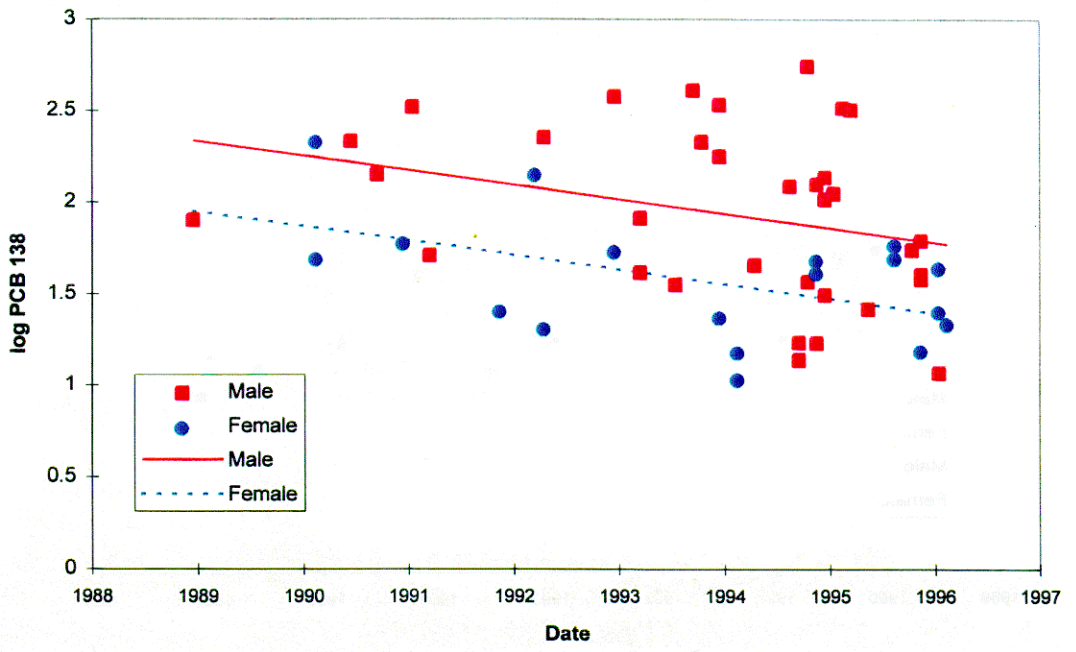


Figure 12 - PCB Congener 153 Levels in Livers in Chronological Order (µg/Kg WM)

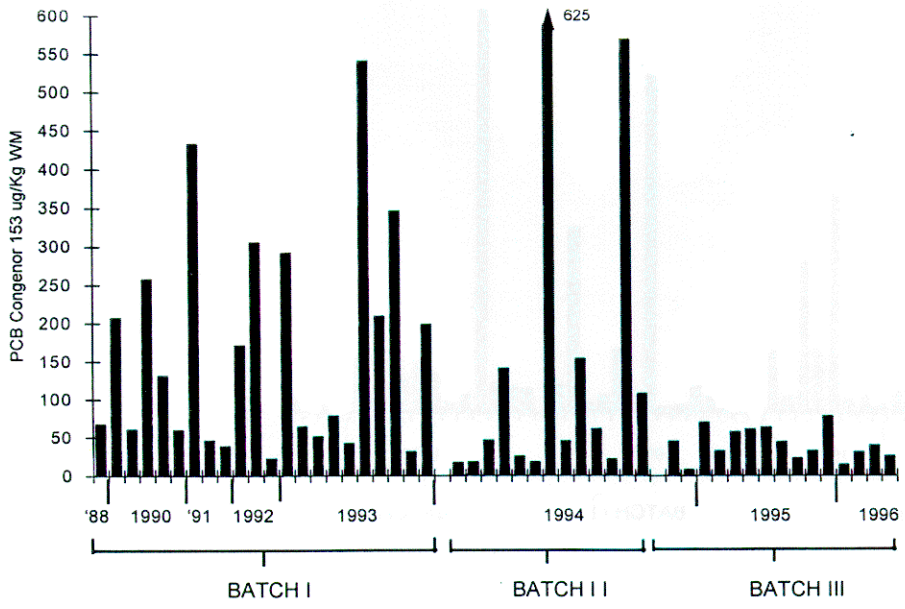


Figure 12a - Time series of log concentration PCB153

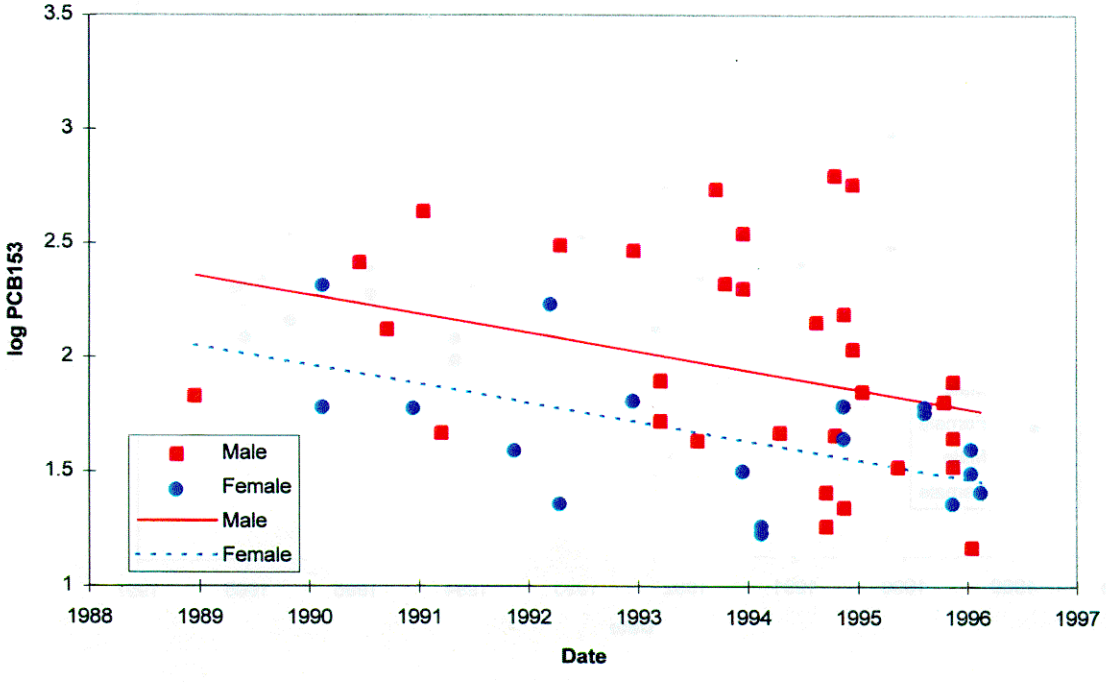


Figure 13 - PCB Congener 180 Levels in Livers in Chronological Order (µg/Kg WM)

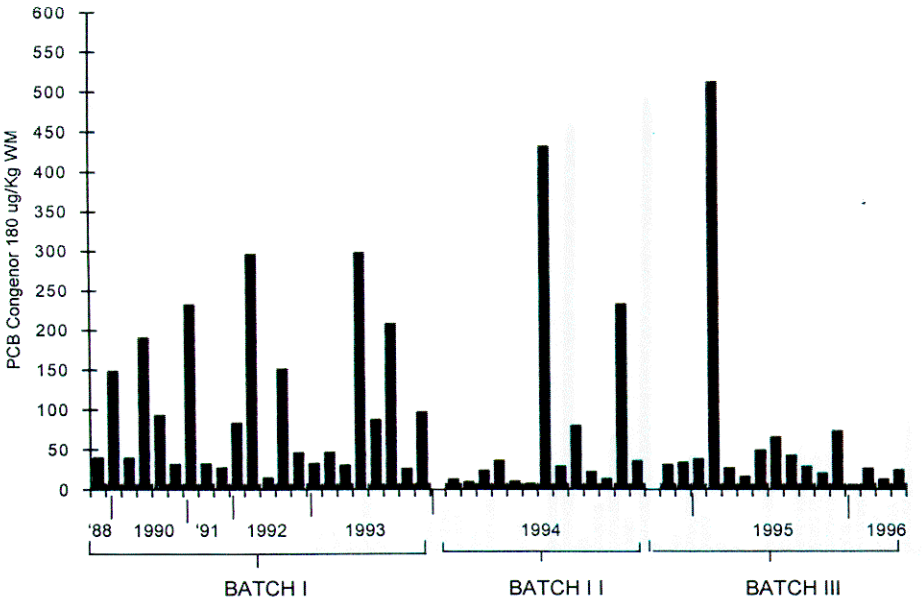


Figure 13a - Time series of log concentration of PCB 180

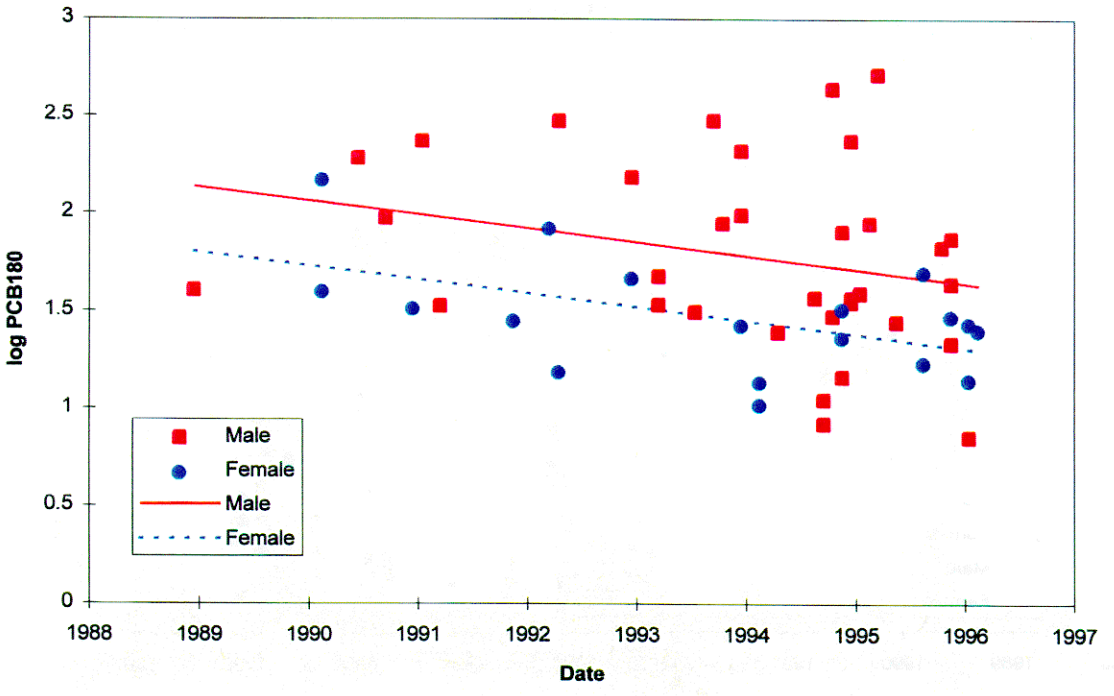


Figure 14 - **pp'DDE Levels in Livers in Chronological Order (µg/Kg WM)**

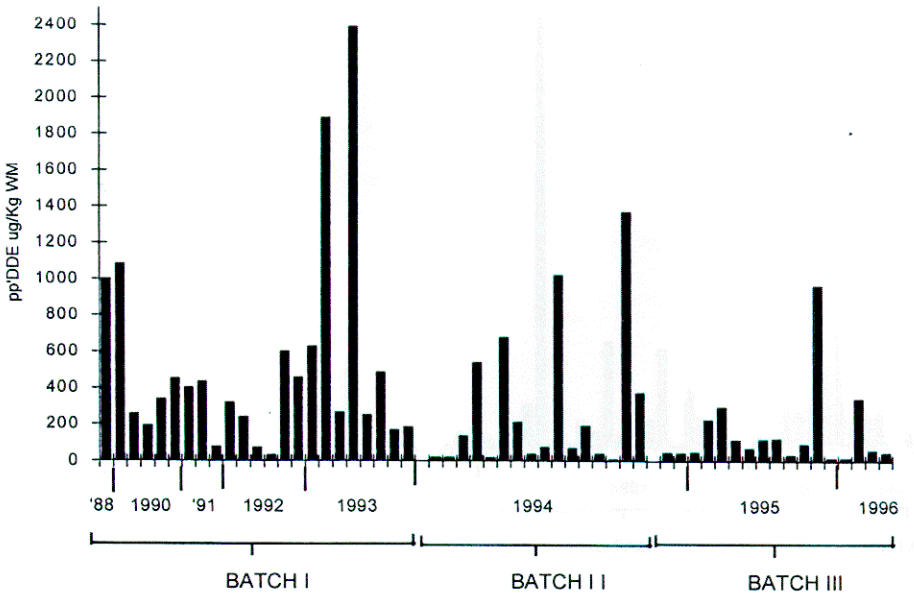


Figure 14a - Time series of log concentration of pp'DDE

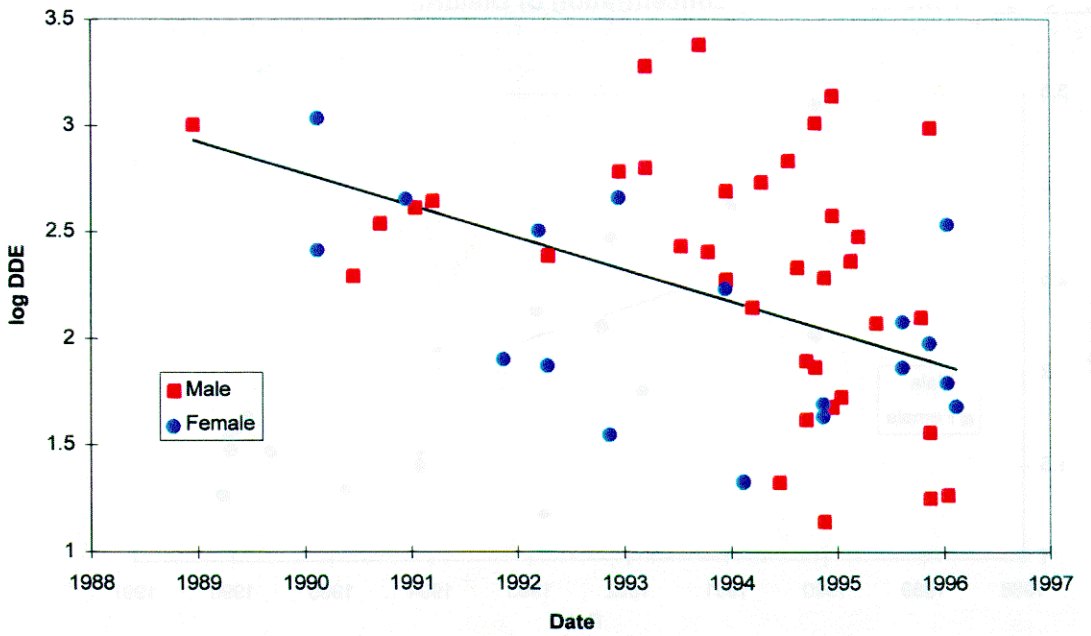


Figure 15 - Dieldrin Levels in Livers in Chronological Order (µg/Kg WM)

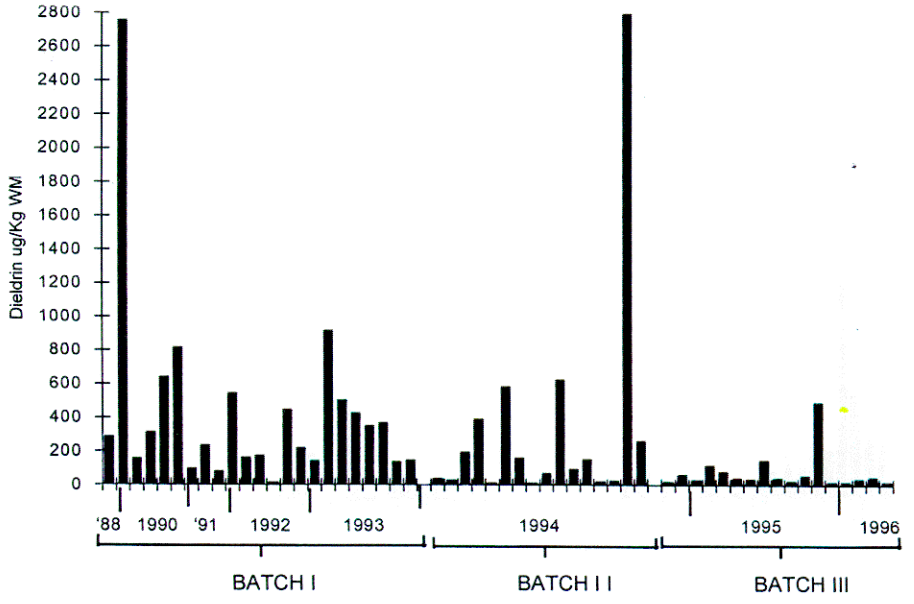
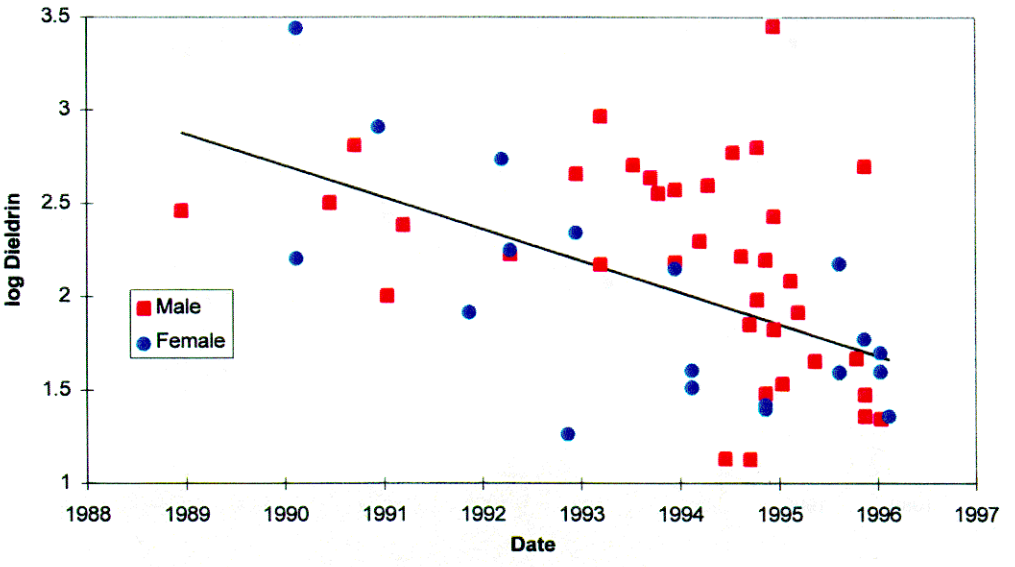


Figure 15a - Time series of log concentration of Dieldrin



The data in Figures 9a-15a, together with data for other important determinands, are summarised in Table 5. The regression coefficients are given for time expressed in years and re-expressed in the final column as the compound rate of annual change on the untransformed scale.

Table 5 - Regression Coefficient of Log Pollutant Concentrations on Time

	Regression Coefficient	Standard Error	Significance	Rate of Annual Charge
DDE	-0.150	0.039	***	-29 %
DDT	-0.083	0.049	#	-17 %
TDE	-0.131	0.042	**	-26 %
HCH	-0.243	0.061	***	-43 %
Dieldrin	-0.170	0.036	***	-32 %
PCB 118	-0.068	0.031	*	-14 %
PCB 138	-0.079	0.031	*	-17 %
PCB 153	-0.083	0.031	*	-17 %
PCB 180	-0.070	0.031	*	-15 %
Total PCB	-0.031	0.028	ns	-7 %
HCB	-0.112	0.024	***	-23 %

Key: *** $p \leq 0.001$
** $0.001 < p \leq 0.01$
* $0.01 < p \leq 0.05$
# $0.05 < p \leq 0.1$
ns $0.1 < p$

These results confirm that there were highly significant downward trends over the study period for pp' DDE, pp' TDE and Dieldrin and less pronounced, but nevertheless significant, downward trends for PCB congeners 118, 138, 153 and 180. However, there was no clear evidence of either an upward or downward trend for total PCB. The results for γHCH may not be valid because a high proportion of otters had nil detectable levels and, as explained earlier, these could not be ascribed log values and they were therefore excluded from the calculations.

6.6 Vitamin A Levels

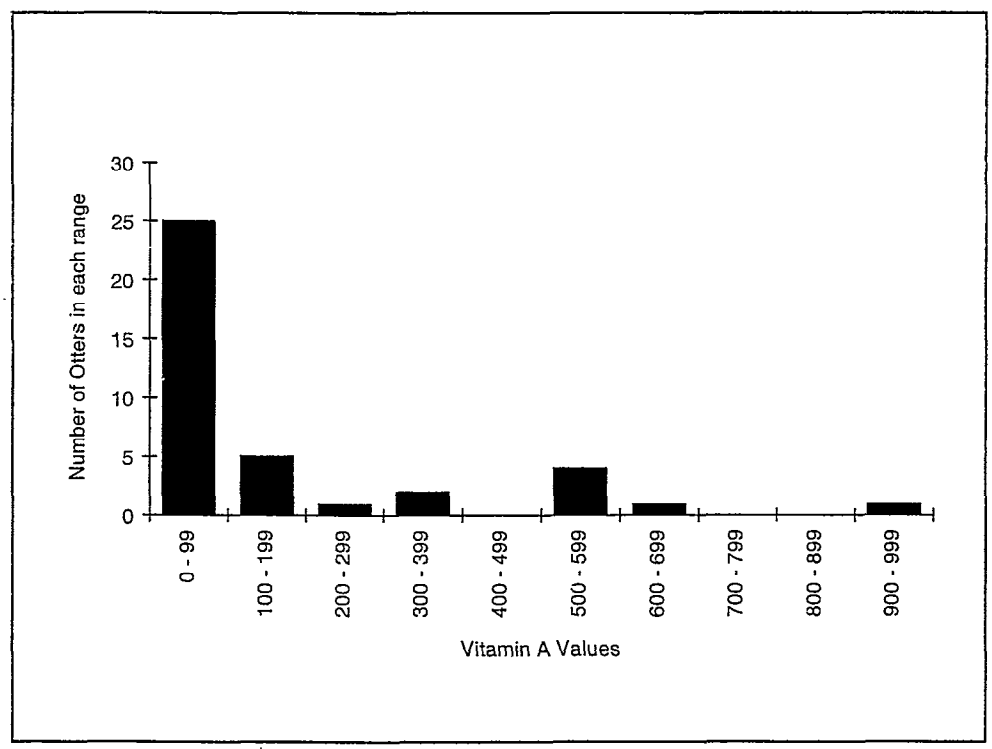
Many halogenated hydrocarbons, including PCBs, chlorinated napthalenes, PBBs and some organochlorine pesticides are known to cause low Vitamin A levels (Olafson 1947, Kimbrough 1974, Jefferies 1975 b, Darjano et al 1983). In some cases it is thought that these compounds block the conversion of β-carotene to Vitamin A (Kimbrough, 1974) but there is also evidence that they increase hepatic turnover of the vitamin (Darjano et al, 1983) as well as blocking its plasma transportation (Brouwer et al, 1986).

In view of the key role played by Vitamin A in both the development and function of many organs, and the fact that PCBs and OCs have been widely implicated in the decline of otter populations, all otter livers which were in fresh condition were analysed for Vitamin A.

A total of 44 livers was examined and the results show considerable variation with values below 1 μmol/kg in some cases and over 1,000 μmol/kg in others. However, there was a markedly skewed distribution, with most values below 100 μmol/kg, and this was apparent when the values were ranked (Figure 16).

In Figure 17 otters are shown in chronological sequence with Vitamin A values shown on a logarithmic scale. It will be seen that the majority of animals (68 %) had values <200 μmol/kg, which is the quoted threshold for normal domestic animals (Blood et al 1983). Seven had values <7 μmol/kg and this is regarded as a critical level (Blood et al 1983). Most of the animals with very low values occurred in the early years of the study and there also appeared to be an overall increase in values over time. The data were therefore analysed for evidence of a trend through time, as performed for PCBs and OCs, and the results are shown in Figure 17a. There was a highly significant rate of change of +94 %. (Regression coefficient 0.28, standard error 0.064, p <0.001).

Figure 16 - Vitamin A Levels in Livers Showing Distribution in the Population (μmol/kg)



Note: Four otters had values in excess of 1,000 μmol/kg.

Figure 17 - Vitamin A Values in Livers in Chronological Order (µmol/Kg WM)

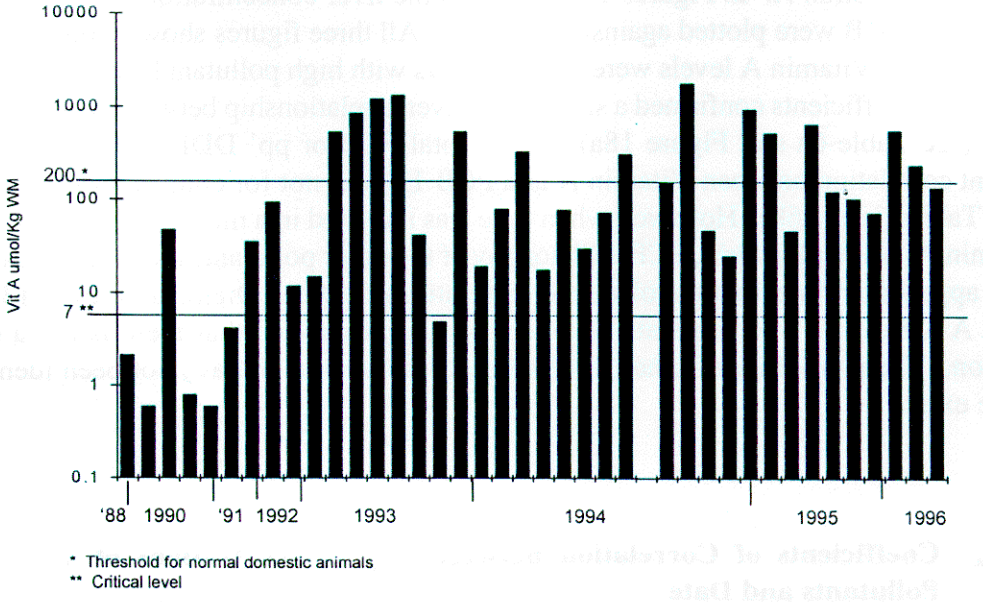
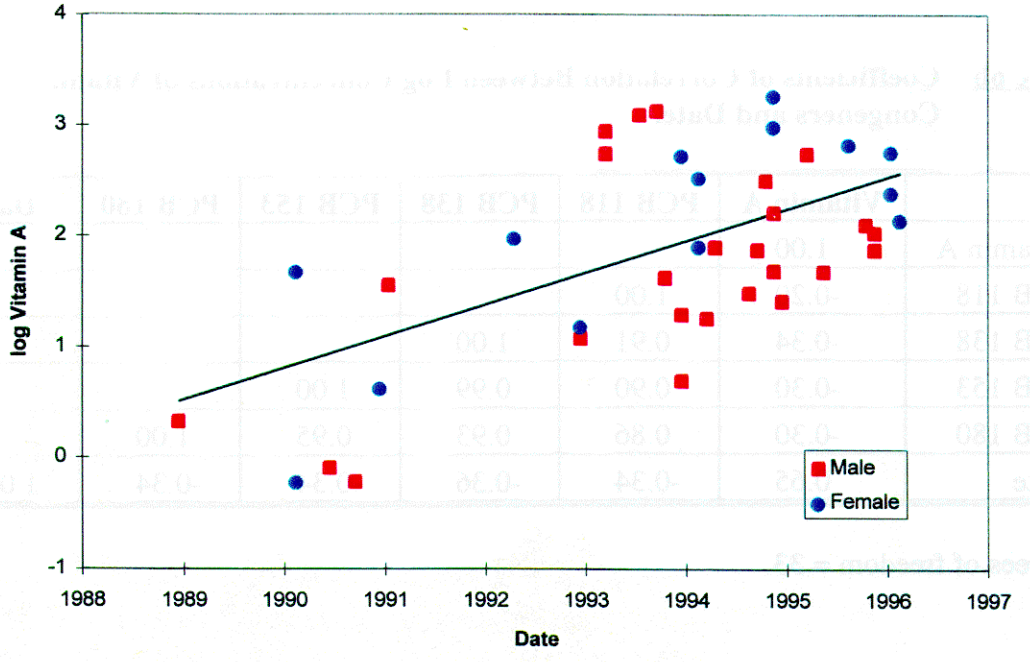


Figure 17a - Time series of log concentration of Vitamin A



6.7 Relationship Between Levels of PCBs, OCs and Vitamin A

As the declines in PCB and OC levels through time coincided with increased Vitamin A levels, the data were re-examined for evidence of an inverse relationship between these pollutants and Vitamin A. In Figures 18, 19 and 20 the liver concentrations of Dieldrin, pp' DDE and total PCB were plotted against Vitamin A. All three figures show a similar pattern, where the lowest Vitamin A levels were often in otters with high pollutant levels. Calculation of correlation coefficients confirmed a significant inverse relationship between Vitamin A and Dieldrin (see Table 6a and Figure 18a) but not total PCB or pp' DDE. There was also a significant correlation between Vitamin A and PCB 138 but not for congeners 118, 153 and 180 (see Tables 6a and 6b). However, when time was included in a multiple regression model for Vitamin A, neither Dieldrin, PCB 138 nor any of the other pollutants remained significant. It would appear therefore, that the observed correlations between Dieldrin, or PCB 138, and Vitamin A are due to their separate relationships with time and that there is not a causal connection. The possibility of other, confounding, factors which have not been identified, therefore exist.

Table 6a **Coefficients of Correlation Between Log Concentrations of Vitamin A, Pollutants and Date**

	Vitamin A	DDE	Dieldrin	Total PCB	Date
Vitamin A	1.00				
DDE	-0.17	1.00			
Dieldrin	-0.44	0.81	1.00		
Total PCB	-0.23	0.45	0.40	1.00	
Date	0.67	-0.46	-0.61	-0.17	1.00

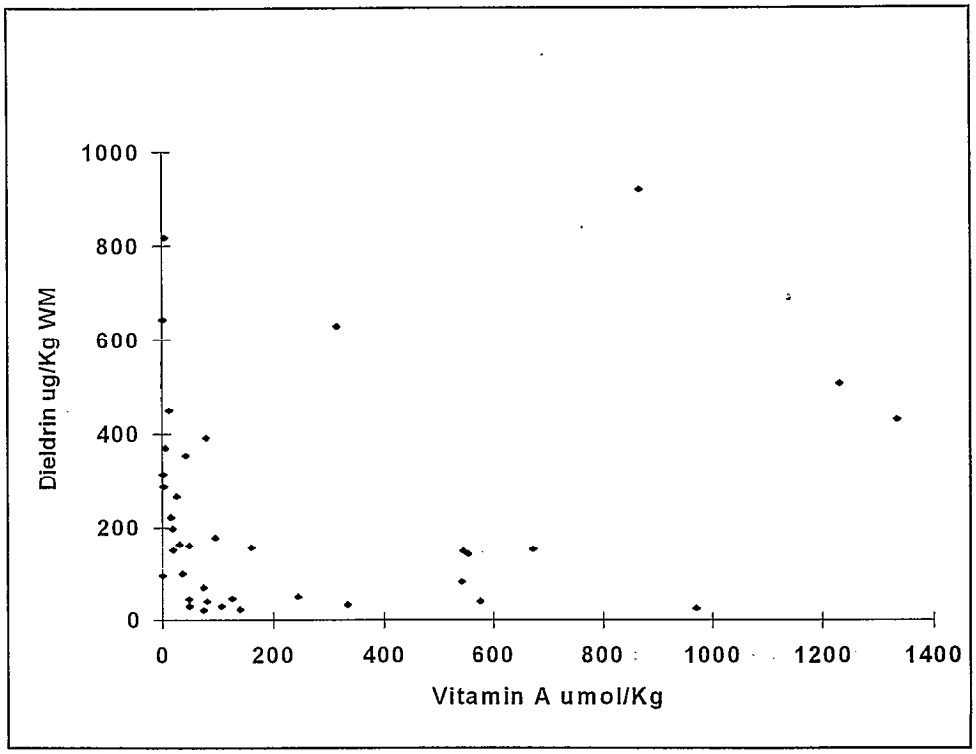
Degrees of freedom = 33.

Table 6b **Coefficients of Correlation Between Log Concentrations of Vitamin A, PCB Congeners and Date.**

	Vitamin A	PCB 118	PCB 138	PCB 153	PCB 180	Date
Vitamin A	1.00					
PCB 118	-0.20	1.00				
PCB 138	-0.34	0.91	1.00			
PCB 153	-0.30	0.90	0.99	1.00		
PCB 180	-0.30	0.86	0.93	0.95	1.00	
Date	0.65	-0.34	-0.36	-0.34	-0.34	1.00

Degrees of freedom = 33.

Figure 18 - Relationship Between Vitamin A and Dieldrin



Two animals with high values are not shown:
M455/2/90, Dieldrin 2761 µg/kg, Vit A 0.6 µmol/kg.
M180/11/94 Dieldrin 26.5 µg/kg, Vit A 1836 µmol/kg.

Figure 18a - Relationship between Log Vitamin A and Log Dieldrin

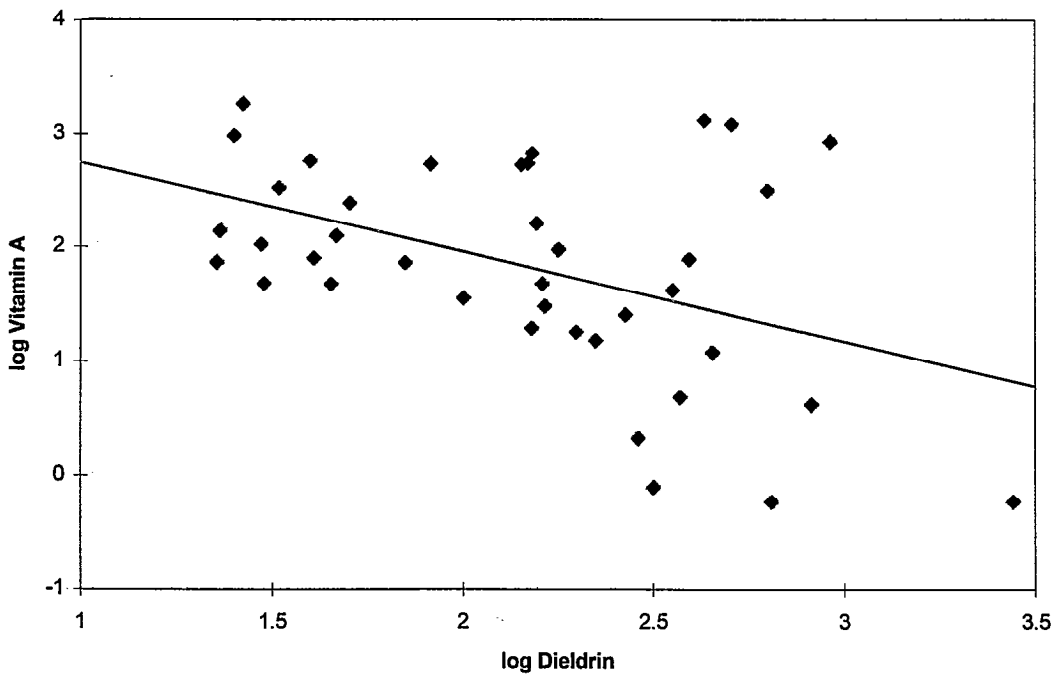
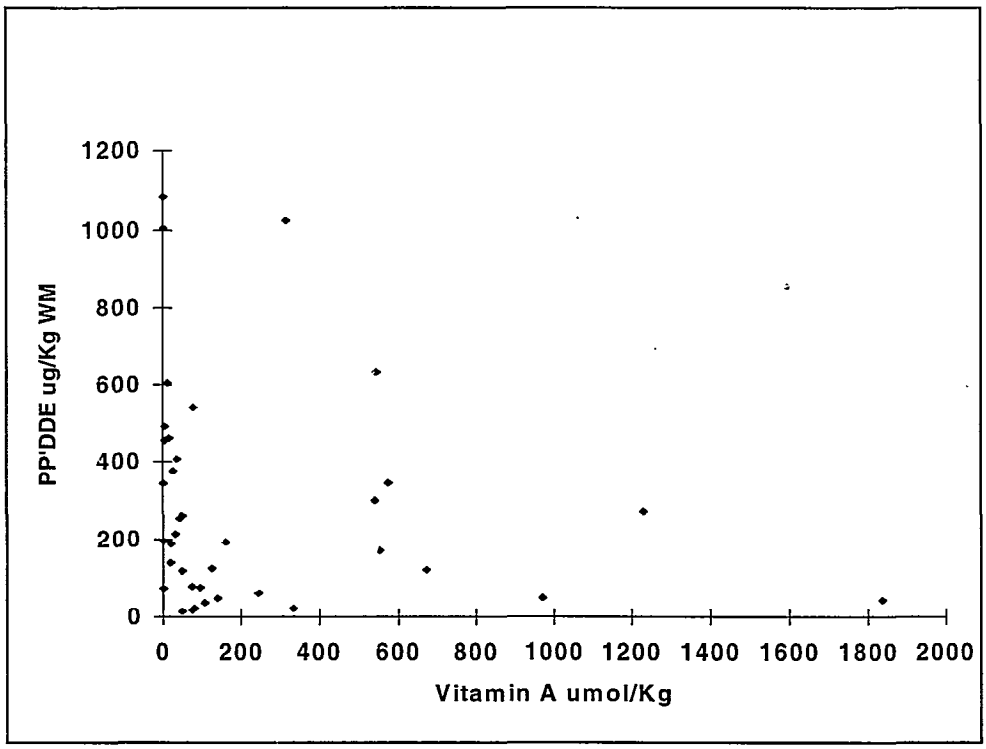
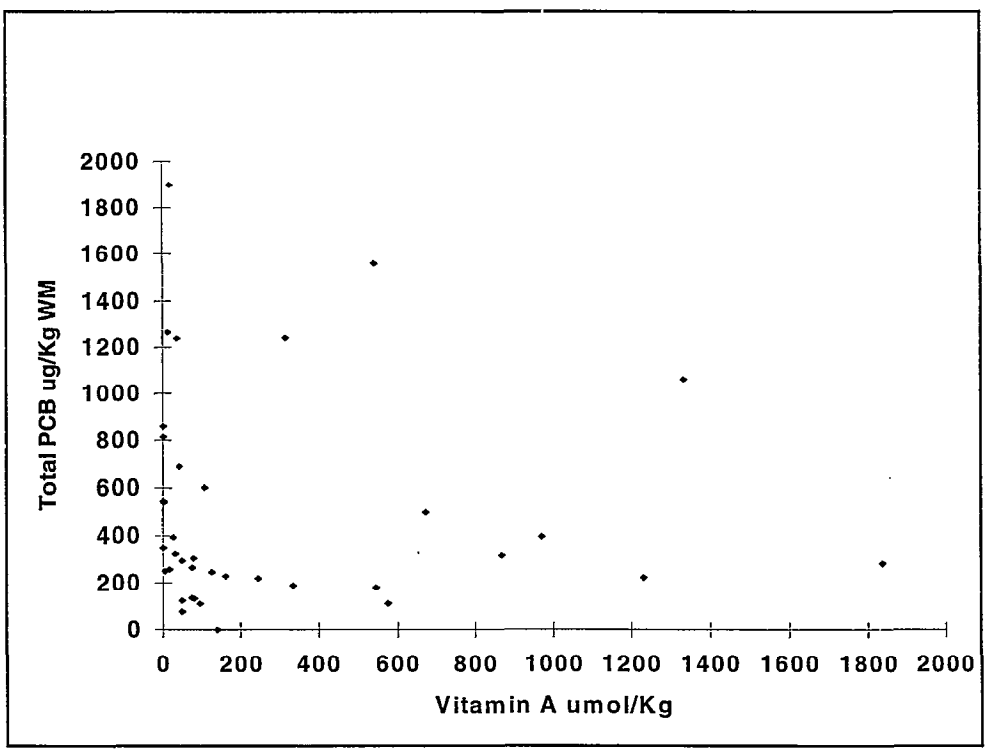


Figure 19 - Relationship Between pp'DDE and Vitamin A in Livers



Two animals with high pp'DDE values are not shown:
M224/3/93 pp'DDE 1893 μg/kg, Vit A 867 μmol/kg.
M31/9/93 pp'DDE 2397 μg/kg, Vit A 1333 μmol.kg.

Figure 20 - Relationship Between Total PCB and Vitamin A in Livers



6.8 Relationship Between Levels of PCBs, OCs and Thyroid Weight

Secretions from the thyroid gland control the body's metabolic rate but it is known that PCBs and OCs can have a profound effect on thyroid gland function and therefore influence the metabolic rate. Moreover, Vitamin A metabolism is closely linked to thyroid activity and experiments with pigeons (*Columba livia*) showed that Dieldrin and pp' DDT both caused initial hyperthyroidism and increased Vitamin A levels. On prolonged exposure this was followed by hypothyroidism and decreased liver Vitamin A levels. These effects occurred even at very low levels of exposure (Jefferies, 1975 b). Not all species respond in the same way however, and in some cases initial hyperthyroidism does not occur. Hypothyroidism also occurs as a result of PCB toxicity (Kimbrough 1974, Jefferies and Parslow 1972).

Both hyperthyroidism and hypothyroidism can lead to increased thyroid gland size. In the case of hyperthyroidism, there may be an increase in the mass of follicular secretory cells and in the number of small follicles but a decrease in colloid. Increased gland size in hypothyroidism may be due to accumulation of colloid in the follicles, and can therefore resemble colloid goitre. However, hypothyroidism may also result from thyroid atrophy and, in this case, there is a decrease in gland size.

Otters were examined for a possible relationship between thyroid weight and some of these pollutants by plotting organ weight against total PCB, Dieldrin and pp' DDE (Figures 21 to 23). In each case, the figures suggest a positive correlation. They also illustrate the fact that males tended to have higher pollutant levels, and that their thyroid glands were mostly heavier than those of females. As thyroid weight was shown to be closely linked to body weight ($p<0.001$), and males were significantly heavier than females ($p<0.01$), this last observation was not unexpected. However, after the dependence of thyroid weight on body weight (and sex) had been allowed for in a multiple regression model, none of the pollutants listed above had a significant effect on thyroid weight.

When the logarithm of pollutant concentration was taken as the dependent variable, thyroid weight and body weight each exhibited significant effects that acted as alternate explanatory variables and were not both needed. The pp' DDE levels were slightly better explained in terms of thyroid weight than of body weight, but for Dieldrin and Total PCB there was nothing to choose between the two alternative explanations (Table 7). The relationships of pollutant levels to thyroid weight are illustrated in Figures 21a to 23a.

Table 7 **Partial Regression Coefficient of Log Concentration of OCs & PCB on Body Weight and on Thyroid Weight Allowing for Date and Sex**

	Regression Coefficient	Standard Error	t	Significance
For pp'DDE on:				
Body Weight	0.043	0.044	0.96	ns
Thyroid Weight	0.743	0.336	2.21	*
For Dieldrin on:				
Body Weight	0.105	0.038	2.74	**
Thyroid Weight	0.921	0.299	3.08	**
For Total PCB on:				
Body Weight	0.074	0.033	2.25	*
Thyroid Weight	0.587	0.279	2.10	*

Key: ** $0.001 < p \leq 0.01$
* $0.01 < p \leq 0.05$
ns $0.1 < p$

Figure 21 - Relationship Between Thyroid Weight and Total PCB in Livers

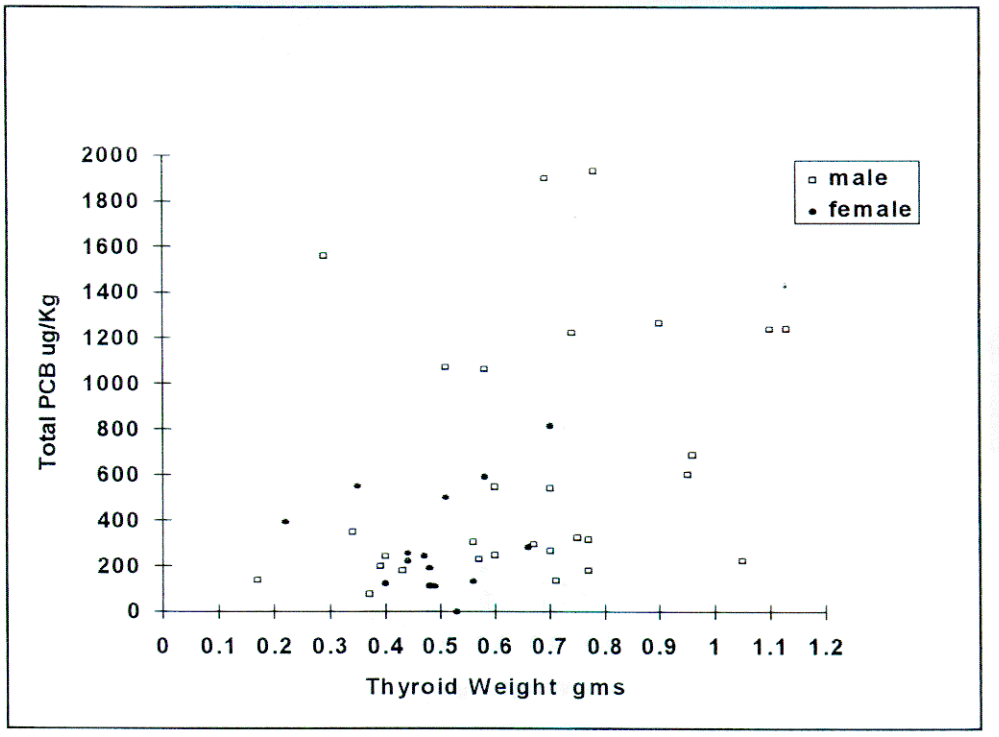


Figure 21a - Relationship between log Total PCB concentration and Thyroid weight

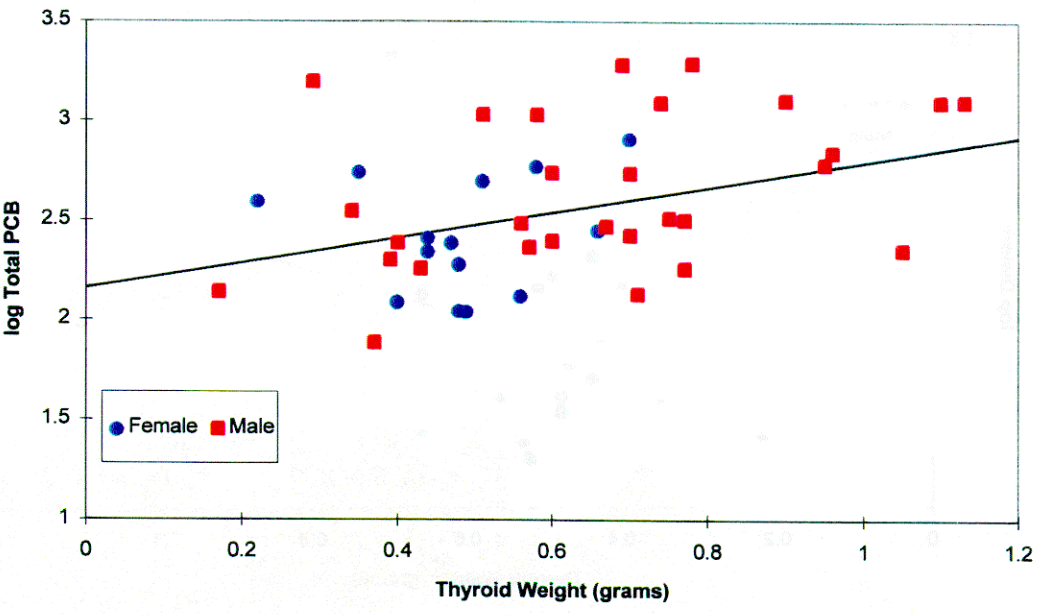
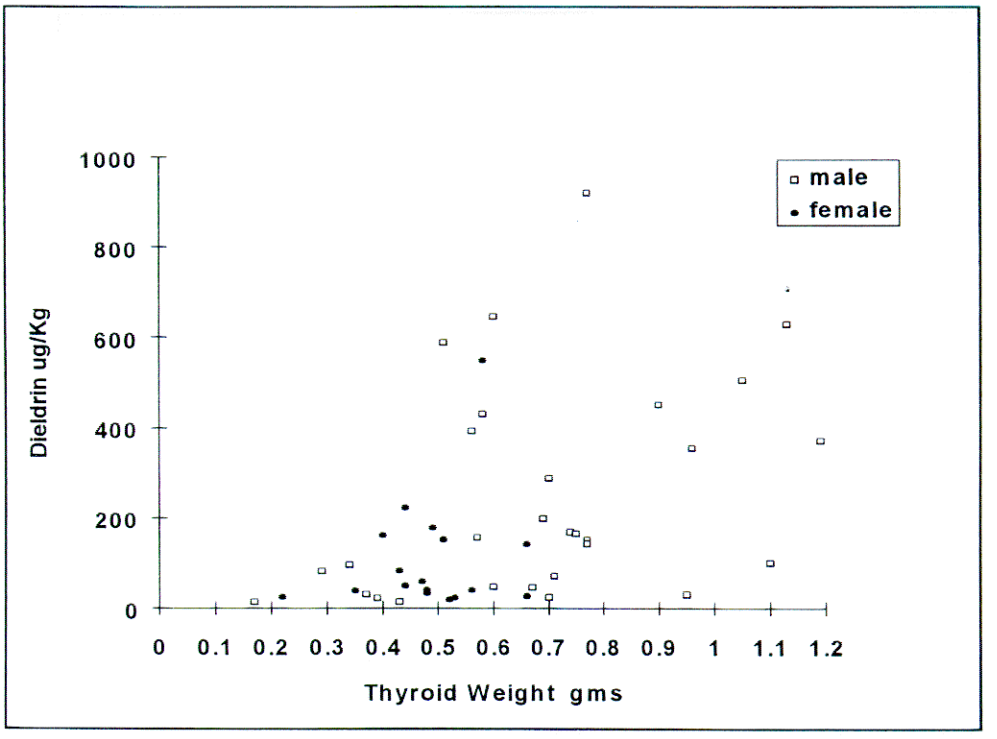


Figure 22 - Relationship Between Thyroid Weight and Dieldrin in Livers



Two animals with very high Dieldrin values are not shown:
M455/2/90, Dieldrin 2761 µg/kg: thyroid 0.7g.
M4/12/94, Dieldrin 2801 µg/kg, thyroid 0.4g.

Figure 22a - Relationship between log Dieldrin concentration and Thyroid weight

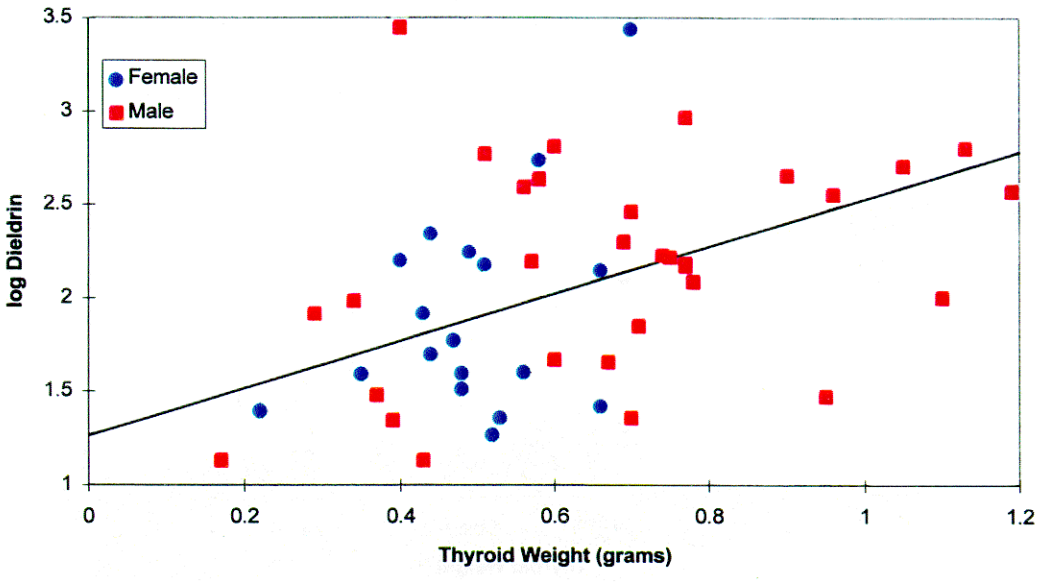


Figure 23 - Relationship between Thyroid Weight and pp'DDE in Livers

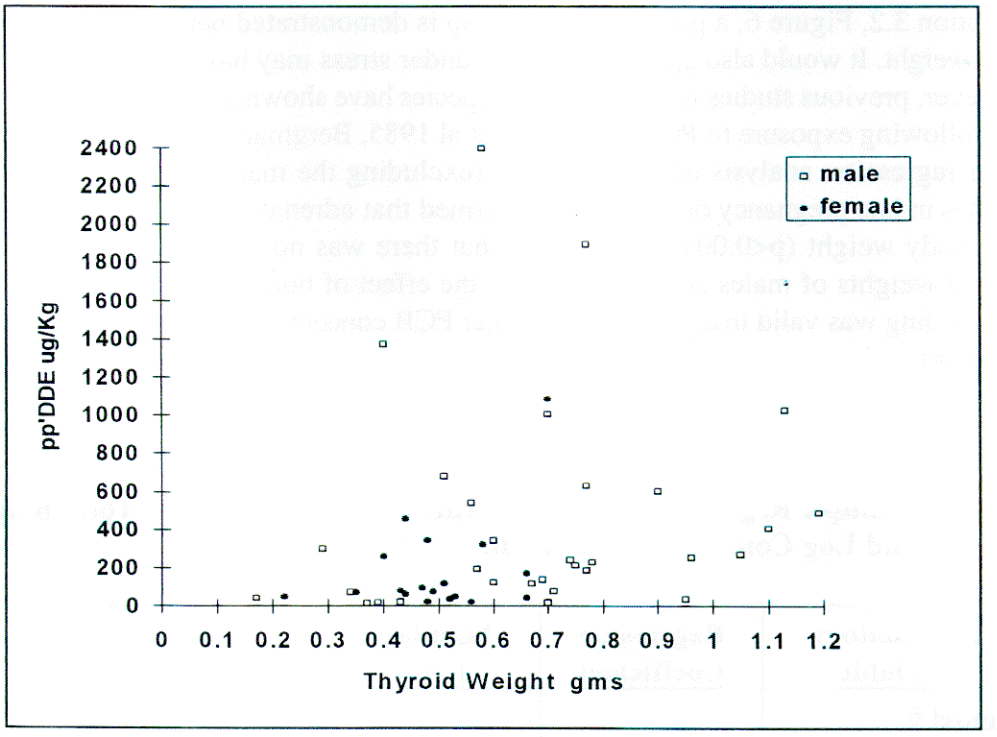
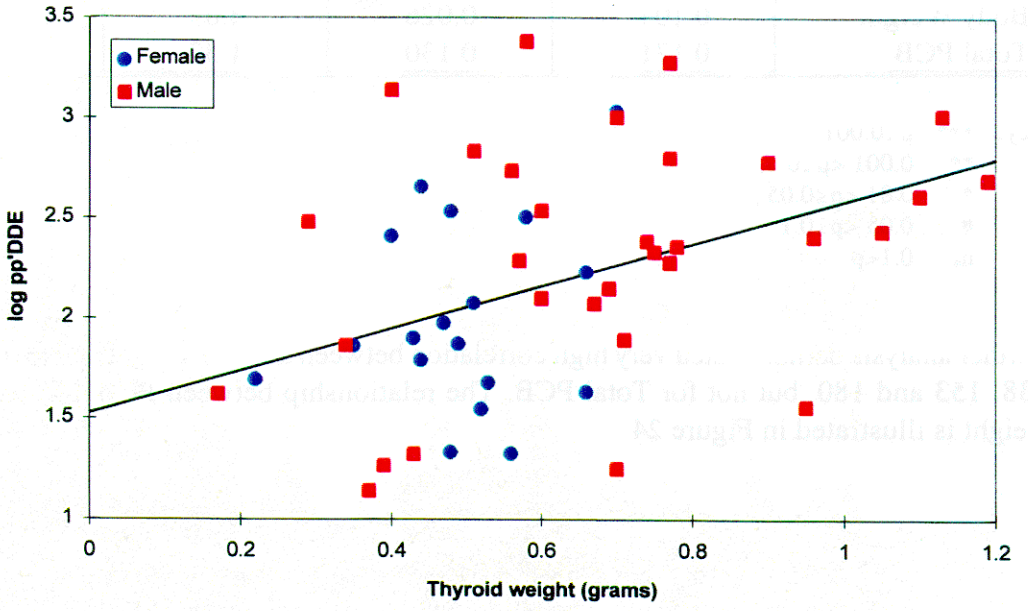


Figure 23a - Relationship between log pp'DDE concentration and thyroid weight.



6.9 Relationship Between Levels of PCBs, OCs and Adrenal Weight

In section 3.2, Figure 6, a positive relationship is demonstrated between adrenal weight and body weight. It would also appear that otters under stress may have enlarged adrenal glands. However, previous studies in mink and seal species have shown evidence of increased adrenal size following exposure to PCBs (Aulerich et al 1985, Bergman and Olsson 1985). Multiple linear regression analysis of the otter data (excluding the males dying of bite wounds and females in late pregnancy or lactating) confirmed that adrenal weight increased significantly with body weight ($p<0.001$) but showed that there was no significant difference between adrenal weights of males and females once the effect of body weight had been allowed for. This finding was valid irrespective of whether PCB concentration was included in the model (Table 8).

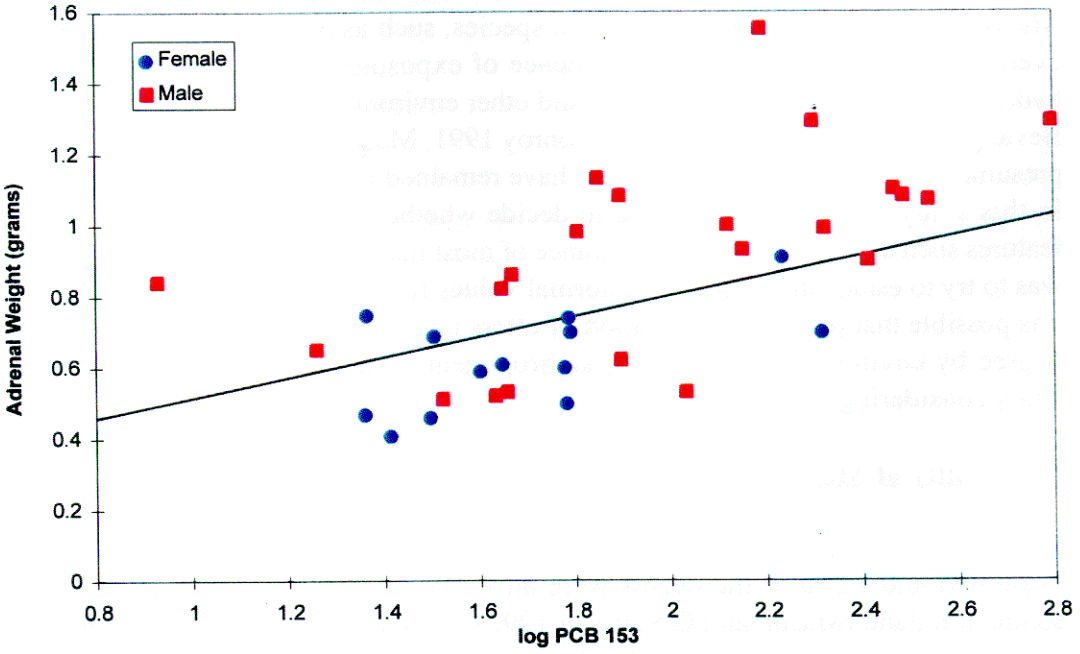
Table 8 Multiple Regression Models for Adrenal Weight, On Sex, Total Body Weight, and Log Concentration of PCB.

Explanatory Variable	Regression Coefficient	Standard Error	t	Significance
Model 0				
Constant	-0.024	0.195		
Sex F	0.062	0.095	0.65	ns
Body Weight	0.116	0.024	4.80	***
Model 1				
Constant	-0.378	0.198		
Sex F	0.060	0.081	0.74	ns
Body Weight	0.089	0.022	3.99	***
PCB 153	0.285	0.087	3.29	**
Model 2				
Constant	-0.382	0.333		
Sex F	0.067	0.094	0.72	ns
Body Weight	0.104	0.026	4.05	***
Total PCB	0.171	0.130	1.32	ns

Key: *** $p \leq 0.001$
** $0.001 < p \leq 0.01$
* $0.01 < p \leq 0.05$
# $0.05 < p \leq 0.1$
ns $0.1 < p$

Further analysis demonstrated very high correlation between adrenal weight and levels of PCB 138, 153 and 180, but not for Total PCB. The relationship between PCB 153 and adrenal weight is illustrated in Figure 24.

Figure 24 - Scatter plot of Adrenal weight against log PCB 153 concentration, with partial regression line for otters of average weight (6.5 kg)



6.10 Heavy metal levels

Analysis for selected metals was carried out on batches II and III only. The results are summarised in Table 9. (Individual values are given in Appendix IV.)

Table 9 Heavy Metal Levels in Liver. Values in mg/kg (Dry Matter) Batches II and III only

	Zn	As	Hg	Pb	Cr	Cu	Cd
Mean	125.5	0.57	9.6	0.3	0.08	40.9	0.59
SD	114.7	2.6	6.6	0.74	0.43	17.2	0.44
Range	64.7-711	0-14	0.2-31.9	0-3.2	0-2.3	15-90.6	0-1.5
Number	29	29	29	29	29	29	29

These results show no evidence of significant heavy metal levels (see section 7.15).

7 DISCUSSION

7.1 General

It is unfortunate that no long term mortality studies were carried out prior to the collapse of the European otter population. Without a sound knowledge of what is normal with regard to the anatomy, physiology, population structure and pathological conditions in a species, one cannot pronounce on what is abnormal. Limited data are available for comparison from areas where the otter has remained a common species, such as Scotland, Ireland and Norway, but even in those areas there is clear evidence of exposure to pollutants, such as halogenated hydrocarbons, heavy metals, acid rain and other environmental pollutants (Olsson et al 1981, Bevanger and Albu 1986, Kruuk and Conroy 1991, Mason and O'Sullivan 1992). One cannot presume that the animals in these areas have remained unaffected and are therefore "normal". In this study an attempt was made to decide whether each animal was healthy or not, and features such as the weight and appearance of most major organs were recorded. The objective was to try to establish a baseline of normal values for use in future investigations. However, it is possible that the entire population of otters in south west England is influenced to some degree by environmental factors of anthropogenic origin, and this needs to be recognised when considering the data.

7.2 Seasonality of Mortality

In a study of otter mortality in Shetland (Kruuk and Conroy, 1991) 113 animals were examined and 42 % of the deaths were attributed to road accidents. In a similar study in southern Ireland (Mason and O'Sullivan 1992), road accidents were responsible for 70 % of the deaths and this compares with 83 % in the present study. It is likely that these figures reflect the relative levels of traffic density in the respective areas.

In Shetland it was considered that the otters were equally likely to die in road accidents at any time of the year and a similar conclusion was reached in another study, combining otters collected in the Outer Hebrides and Denmark (Mason and Madsen, 1990). These results are in marked contrast to those obtained in the present study, where there were few road fatalities in the summer months and a seasonal effect was quite clear. One factor which influenced this in south west England was that otters were frequently killed on roads a day or so after a period of heavy rain, when rivers were in spate. Such rainfall most commonly occurs between September and March. In badgers (*Meles meles*) there is also a marked seasonal variation in deaths due to road accidents, with peaks in the spring and autumn (Neal 1977, Davies et al, 1987). However, these peaks are mostly attributed to patterns of sexual and social behaviour, rather than weather conditions. Furthermore, the males and females may have different seasonal peaks of mortality (Jefferies 1975a, Neal 1977).

7.3 Seasonality of Reproduction

The pattern of spring and autumn mortality observed in the otters in this study resembles that seen in badgers and it is possible that it is linked to seasonal reproductive activity, for example oestrus, mating or parturition. Although otters are believed to breed all the year round in Britain (Stephens 1957, Mason and Macdonald, 1986), they breed seasonally in Shetland (Kruuk et al, 1987), Sweden (Erlinge, 1967), and Germany (Reuther, 1980). Unfortunately, neither Kruuk and Conroy (1991) nor Mason and Madsen (1990) differentiated between males and females in their analyses of the seasonality of road traffic casualties, and therefore comparisons cannot be made with the data in the present study. Apparently differing patterns of road traffic mortality for males and females could be related to reproductive events and more specimens need to be examined from south west England before any firm conclusions can be drawn.

The gestation period of the Eurasian otter is 61-63 days (Stephens 1957, Mason and Macdonald 1986) and delayed implantation of the blastocysts is thought not to occur (Heggberget 1988), although it is a feature of the American river otter (*Lutra canadensis*) (Hamilton and Eadie 1964). There do not appear to be published growth tables for otter foetuses, but newborn cubs are said to measure about 12 cm in length (presumably nose to tail tip) (Mason and Macdonald 1986). Using these data, it is clear that the foetus with a crown-rump length of 13.2 cm, seen on 31 January 1992, would have been born in February, and the smaller foetus measuring 1.5 cm on 22 October 1992, would have been born in November or December. Attempts to estimate the time of parturition for the four lactating females is more difficult. Although cubs remain dependent on the dam for about ten months (Kruuk et al 1987) the duration of lactation does not appear to have been established. Judging by the appearance of their uteri, the two females lactating in November (1992 and 1995) had only recently bred, probably in September or October. The other two, which were killed in February, appeared to have been lactating for some months. Although these results show evidence of breeding during the winter period, they do not prove that this is the sole, or even the main, breeding season, because very few animals were examined during the summer months.

7.4 Ratio of Males to Females

Roughly equal numbers of male and female otters were submitted up until 1993 but thereafter there were more than twice as many males as females. Statistically, this change in the ratio of males to females was not significant but, overall, males were more likely to be killed than females. The reason for this is not clear but a higher percentage of males dying in road accidents has been reported in southern Ireland (Mason and O'Sullivan 1992) and in Devon (Marshall 1991). As males range over much larger areas than females, it is likely that they must cross roads more frequently than females, and are therefore more likely to be killed by road traffic. The risk is likely to be even greater where there is territorial conflict between males.

7.5 Deaths Due to Natural Causes

The otters examined in this study almost certainly represent a biased sample, as those dying on busy roads are much more likely to be noticed and presented for examination than those dying in remote situations. For this reason, Kruuk and Conroy (1991) suggested that, although more than half the otters they examined in Shetland died from physical injury (road accidents, fish traps etc), deaths from natural causes, especially starvation, were more significant. However, in that study 16 out of 52 animals thought to have starved were cubs, and the deaths of these animals could well have been secondary to the deaths of their mothers in road accidents or traps. In the present study, several lactating otters were examined and their young would inevitably have died from starvation.

Kruuk and Conroy (1991) noted that deaths due to non-violent causes were more common in the spring (March-June) and that these coincided with a period of reduced food availability. They therefore considered that most of these 'natural' deaths were due to starvation. However, the authors also observed that otters killed in road accidents at the same time of the year were in good condition. If the shortage of food had been sufficiently severe to cause some otters to die of starvation, it is perhaps surprising that the rest of the population did not show a significant loss of condition. The starving animals had empty stomachs, often with ulcers and/or blood in the alimentary tract. In this respect the cases resembled two otters in the present study. One animal (M153/3/95) was emaciated and had pyloric ulcers, blood in the intestines and slightly enlarged adrenal glands. The second case (M31/9/93) was also emaciated, had blood in the intestines and enlarged adrenals. However, both these animals had severe bite wounds.

Only four animals in this study had possibly died from unexplained causes and none was thought to have starved. Two females and a cub had evidence of severe subcutaneous haemorrhage and bruising and a male had probably died due to bite wounds to the anus and genitals (M30/3/93). All four carcases were partially decomposed and this prevented detailed examination.

Kruuk and Conroy (1991) observed that the average condition index in the spring was lower due to the high proportion of natural deaths in that period. Mason and Madsen (1990) observed a seasonal loss of condition in July/August but they recorded that natural deaths were evenly distributed throughout the year. In the present study, very few animals were examined during the spring and summer months and, therefore, it was not possible to determine whether there was a seasonal loss of condition, or whether bite wounds occurred seasonally. However, many of the male otters with a condition index of less than 0.95 were suffering from bites. Conversely, only two out of nine males with bite wounds had an index greater than 0.95.

7.6 Bite Wounds

Bite wounds in otters have been commented on by Kruuk and Conroy (1991) and by Mason and O'Sullivan (1992) and in each case the wounds have been attributed to dogs. In the present study, bite wounds were a significant finding, affecting 16 % of the animals and being directly responsible for the deaths of at least five otters. However, it is the author's opinion that in these cases the wounds were not caused by dogs but by other otters.

The uniform distribution of the lesions suggests a stylised form of fighting such as occurs, for example, in domestic cats and in badgers. However, although in those species the location of the cranial lesions is similar to that in the otters, the caudal lesions are typically dorsal to the tail base and do not involve the anus and genitals. In most of the otters, bite wounds around the head resulted in marked tissue swelling, with bloody subcutaneous oedema. Bites to the scrotum, vulva and around the anus generally resulted in the formation of deep necrotic tracts or sinuses.

The isolation of streptococcal species from these cases supports the suggestion that the bites are not caused by dogs, because dog bites are typically infected with *Pasteurella multocida.* This hypothesis needs verification; if it is true, it contrasts strongly with the observation in Shetland that interactions between neighbours are generally peaceful and that otter ranges overlap (Kruuk et al, 1987). It is interesting to note that Stephens (1957) also recorded anecdotal evidence of bite wounds to the testes being caused by other otters.

7.7 Adrenal Hyperplasia

Adrenocortical hyperplasia has twice been recorded in otters in Britain and tentatively attributed to ageing, although the possible involvement of PCBs was also considered (Keymer et al 1988, Wells et al 1989). PCBs have been shown to cause adrenal hyperplasia in mink (*Mustela vison)* (Aulerich et al 1985), and have been linked with cortical hyperplasia in two species of seal (Bergman and Olsson 1985). They have also been recorded at what are considered to be significant levels in otters in south west England (Mason and Macdonald, 1994).

Although the PCB levels in the present study were generally lower than have been reported in otters elsewhere, the results show a clear relationship between the levels of congeners 138, 153 and 180 and adrenal gland size. PCBs are known to interfere with steroid hormone levels and it is therefore possible that the histological changes seen in the adrenal cortices are related to the presence of these pollutants. However, although PCBs may be a contributory factor, the more obviously enlarged adrenal glands seen in the male otters in this study were associated with severe bite wounds and were probably a result of stress caused by territorial fighting (Figure 6).

Three of the four lactating females and the single animal in late pregnancy also showed some evidence of adrenal hyperplasia (Figure 6). In both sexes, these changes in the adrenal glands could be interpreted as a normal response to natural stress factors, but the stress could also be influenced by the effects of PCBs. Some human activities, such as flood alleviation measures and aquatic recreation, may create additional stresses. It is unlikely that the adrenal changes in these animals were age-related because in many cases the ages of the otters had been determined and ranged from one to four years.

7.8 Absence of Infectious Disease

When compared with most other wildlife species, the otters were remarkable for the general absence of gross pathological lesions, especially those associated with parasitic, bacterial or viral infections. In part, this may be because the great majority were healthy animals killed by road traffic and few sick animals were found. Overall, there was no evidence of malnutrition and the situation in south west England may be unlike that in Shetland and Denmark. The stomach contents of many otters confirmed that eels and frogs are important prey items in the area and, although expert analysis of ingesta is required, it appears that, unlike Shetland, marine fish seldom represented a significant part of the diet.

7.9 Uterine Abnormalities

Apart from the importance of bite wounds, the most significant observations were probably those relating to the uteri. It is possible that the cystic, convoluted horns seen in three animals could represent an early stage of development in normal pregnancy in otters, but there do not appear to be any records of this in literature. The author has not observed similar uterine changes in other mustelids, for example, badgers, and it seems more likely that they are abnormal.

Furthermore, in a study of 27 adult female otters in Norway, Heggberget (1988) also recorded a similar case which she regarded as pathological. Each uterine horn had three cyst-like structures approximately 5 mm in diameter which resembled early implanted embryos. Uterine stenosis and cystic dilations have been recorded in up to 70 % of seals in the Baltic and attributed to the effects of PCBs (Helle et al 1976, Bergman and Olsson 1985).

Although the cystic and convoluted uteri seen in three otters in this study could have a similar aetiology, no such lesions have been reported in mink following exposure to PCBs (Aulerich and Ringer 1977, Backlin and Bergman 1992). However, mink fed diets containing even very low PCB levels either failed to breed or produced small, weakly litters (Aulerich and Ringer 1977). Further studies on mink have shown that PCBs do not suppress ovulation or implantation but interrupt gestation (Platonow and Karstad 1973, Jensen et al 1977, Kihlstrom et al 1992).

Whilst the decline of the otter population has been attributed to the effects of PCBs and organochlorines, there has been very little evidence of them causing direct toxicity. It has been suspected that their principal effect has been on reproduction (Olsson et al 1981, Mason 1989) but physical evidence for this has been lacking. In this respect, the convoluted, nodular uteri seen in three otters in this study are of considerable interest. However, the data do not indicate a relationship between the appearance of the uteri and the levels of PCBs and OCs.

The first otter with a convoluted uterus (M455/2/90), had very high Dieldrin and pp' DDE levels but unremarkable PCB levels. The second case, which was also pregnant, (M192/10/92) was not analysed by the Agency laboratory but by Dr. Chris Mason, Essex University, and the levels of Dieldrin, pp' DDE and PCB were all very low. The third case (M206/12/93) also had low OC and very low PCB levels. At the present time, therefore, the reason for the appearance for these uteri is unknown.

Otters do not breed until they are about two years old and normally produce litters of one to three young (Mason and Macdonald 1986). As the mean age of females in this study was only two years, it is clear that factors affecting reproduction could seriously affect population recruitment. Although there is general agreement that the otter population in south west England is increasing, the nature and causes of the changes seen in the uteri in this study merit further investigation.

7.10 PCB Levels

There are 209 theoretically possible PCB isomers or congeners and, although some of these are known to be of low toxicity, others, especially the coplanar congeners, are extremely toxic (Tanabe et al, 1987). Commercial PCB preparations are composed of imprecise mixtures of these congeners, together with contaminants such as chlorinated naphthalenes, dibenzofurans and dibenzodioxins (Vos et al 1970, Kihlström et al 1992). The variable composition of such products has made it very difficult to evaluate their toxicity.

Furthermore, data derived from experimental dosing studies, where commercial PCB preparations are added to an animal' diet, are of limited value, especially when making comparisons with total PCB levels found in the tissues, or diet, of a wild animal. This is because, weight for weight, the PCBs present in wild prey species such as fish, are much more toxic than those in the original commercial product (Aulerich and Ringer 1977, Platonow and Karstad 1973). On entering the food chain, the less toxic congeners are metabolised, whereas the more toxic ones, which tend to be the higher chlorinated compounds, are not. The result is the accumulation in tissue of the more toxic congeners.

Therefore, the total PCB level in the tissues of a wild animal which has fed on prey in a contaminated environment may be the same as that in a laboratory animal which has been experimentally dosed with a commercial PCB, but the congeners present are likely to be much more toxic. The biological half-life for some of these congeners can be extremely long, e.g. 27.5 years for congener 153 in man (Yakushiji, 1988). In most wildlife species this period would exceed their life expectancy.

In this study the total PCB levels are expressed as Aroclor 1260 equivalent because the pattern of the higher chlorinated congeners present in the tissues of wildlife most closely match those present in this commercial product. The Aroclor 1260 equivalent has also been widely used in earlier reports on otters. When compared with previously reported values in *Lutra lutra* in Spain and in the closely related *Lutra canadensis* in North America the values obtained in this study appear low (Table 10).

Table 10 - Total PCB Levels in Liver mg/kg WM

Species	Country	Total PCB		n	Reference
		Mean	Range		
L. lutra	England	0.47	nd-1.9	47	This report
L. lutra	Spain	2.5	2.4-2.5	5	Hernandez et al 1985
L. canadensis	USA	0.8	0.04-7.3	61	Foley et al 1988
L. canadensis	USA	4.5	nd-23	20	Henny et al 1981
L. canadensis	Canada	0.02	nd-2.3	44	Somers et al 1987

nd = nil detected

However, 9 out of 47 animals analysed had values between 1000 and 2000 μg/kg WM(1-2mg/kg). If these are expressed in lipid concentration, seven had values in excess of 50 mg/kg. This is the level above which, it has been suggested, otter reproduction is likely to be impaired (Mason 1989, Mason et al 1986a, Mason and O'Sullivan 1992, Mason and Macdonald 1994, Olsson et al 1981). This much quoted "critical" value is based on two references.

In the first reference by Jensen et al (1977), mink were fed 3.3 mg/kg commercial PCB (+ 3.3 mg/kg DDT) in their diet and suffered impaired reproduction. However, the mean PCB level in the livers of the female mink was not 50 mg/kg, as normally quoted, but 86 mg/kg (lipid). The second reference (Olsson et al 1981) refers to reproductive failure in mink where the body fat PCB level exceeded 50 mg/kg. Unfortunately, this is not a proper scientific report as no laboratory data were given and there were no supporting references. It is therefore impossible to determine how a value of 50 mg/kg was arrived at.

In other experiments mink have shown impaired reproduction when their lipid PCB levels were well below 50 mg/kg. However, in these cases the PCBs were those retained in the tissues of the "prey" species used to feed the mink, and not a commercial product as used by Jensen et al (1977). Female mink fed on fish from the American Great Lakes containing 0.66 mg/kg (WM) PCBs suffered reproductive failure and the PCB concentration in their subcutaneous adipose tissue (fat) was only 24.8 mg/kg WM (Hornshaw et al, 1983).

In another experiment Aroclor 1254 was first fed to cattle and the resulting beef, containing 0.64 mg/kg (WM) PCB, was then fed to mink. The females failed to breed and their mean liver PCB level after three months' exposure was 1.3 mg/kg (WM) (Platonow and Karstad, 1973). Assuming a liver fat concentration of approximately 4 %, this equates to a PCB level of 32.5 mg/kg lipid. Thus, although PCBs can undoubtedly have a profound effect on reproduction in mink, particularly when derived from prey tissues, the basis for deciding that 50 mg/kg (lipid) represents a critical value for mink, or otters, is unclear. It cannot be assumed that otters are more, less or as sensitive to these compounds as mink, and although circumstantial evidence may link values in excess of 50 mg/kg (lipid) with locally declining populations, further supporting evidence is necessary before this "critical" value can be accepted as valid.

Total PCB values are, at best, a crude measurement of exposure or toxic risk. It is now recognised that there is a need to determine the concentrations of the individual congeners but as these vary considerably in their toxicity, simply measuring the sum of the weight of the congeners per gram of tissue is of limited value. However, these compounds all induce the liver to produce microsomal enzymes to a greater or lesser degree and the relative toxicity of each congener can be determined by the amount of enzyme which it is shown to induce compared with the amount induced by the highly toxic 2,3,7,8 tetrachlorodibenzodioxin (TCDD). This is referred to as the Toxic Equivalency Factor (TEF). The TEFs for the congeners present in a sample can then be used to calculate the toxic burden, and this is referred to as the Toxic Equivalent, or TEQ. The TEQ for a sample is the sum of the product of the concentration of selected congeners multiplied by their respective TEF's (Kannan et al 1988, Safe 1990).

Some species accumulate different congeners more readily, and at different ratios, than other species. Marine mammals, such as dolphins and porpoises, accumulate the toxic coplanar congeners 77, 126 and 169 more readily than some terrestrial mammals do (Tanabe et al 1987). In part, this appears to be due to their relative inability to produce sufficient amounts of the hepatic microsomal enzymes required to metabolise the PCBs.

This deficiency has also been observed in mink and may explain why they are so sensitive to the effects of PCBs (Tanabe 1988). The position with regard to otters is unknown but a study of wild polecats (*M. putorius*) in The Netherlands showed that the highest PCB concentrations in tissues were of congeners 138, 153, 170 and 180 (Leonards et al 1994). However, when the TEFs were calculated it was congeners 77, 105 and 126 which represented 90 to 99 % of the toxic risk. Similarly, experimental studies on mink have shown this species to be very susceptible to congener 169 (Aulerich et al 1985). Congeners 77, 126 and 169 were not recorded in the analytical results in the present study and congener 105 was only reported in batch III. It was not possible, therefore, to calculate TEQs based on these congeners.

There was a strong correlation between the levels of congeners 118, 138, 153 and 180 in the otters. The marked annual declines recorded in the levels of these compounds over the study period is encouraging but it also raises the question why there is no matching decline in total PCB levels.

The significance of the levels of the various congeners in the otters is uncertain but the strong relationship between congener 153 and adrenal weight (see 6.9) indicates the need to investigate the effects which different congeners may have on certain tissues.

In future it would be desirable to calculate TEQs for PCBs in individual otters and to try to relate these to the pathology. However, even if TEQs are calculated, interpretation will be complicated by the fact that some congeners may be of very low toxicity when administered individually, but when given as mixtures they show marked toxicity (Kihlström et al, 1992). Furthermore, other halogenated hydrocarbons may have a pronounced synergistic effect, for example, small concentrations of Dieldrin when added to mink diets containing Aroclor 1254 caused increased mortality (Aulerich and Ringer, 1977).

7.11 Dieldrin

The cyclodiene pesticide Dieldrin was responsible for mass-mortality of wild birds and mammals worldwide during the 1950s and 1960s. Birds of prey, such as sparrow hawk and peregrine falcon, were badly affected but, whereas DDT was primarily responsible for reproductive failure, Dieldrin caused direct toxicity (Newton and Haas, 1984).

Analysis of otter hunt records by Chanin and Jefferies (1978) also showed that the population of otters in Britain dropped sharply after 1956 - the year in which Dieldrin was introduced. It was used widely as an agricultural pesticide in crop dressings and sheep dips. It was also used in timber treatment and in various industries, including textile and carpet manufacturing and in electrical insulation material. The agricultural use of Dieldrin, (and of Aldrin, which is rapidly metabolised to form HEOD, the active principle in Dieldrin), was progressively restricted, starting in 1962 (spring cereals), then 1965 (sheep dip), 1975 (autumn cereals) and, with certain exceptions, was finally prohibited in 1981.

Industrial uses continued to be permitted and in 1986/7 a survey of eels from 62 sites in Britain showed significant residues of Dieldrin (MAFF 1988). The eel tissue levels at the 13 worst affected sites, which were principally rivers in industrial areas, ranged from 0.007 to 4.1 mg/kg (WM). In 1988, a follow-up study of eels in rivers in south west England showed high Dieldrin levels at Clyst Honiton, Devon, North Tamerton on the River Tamar and Truro in Cornwall. However, a sample of eels from the Newlyn River, near Newbridge in the far west of Cornwall contained up to 22 mg/kg (WM) (South West Water, 1988).

Experimental diets containing as little as 2.5 mg/kg Dieldrin caused almost 100 % mortality in mink (Aulerich and Ringer, 1970) and, even allowing for variation in species susceptibility, the level of 22 mg/kg in eels would almost certainly have been sufficient to cause acute toxicity in otters. Despite being prohibited as an agricultural chemical in 1981, Dieldrin had remained in use under an EEC derogation for various purposes, including the control of narcissus bulb fly. Both Dieldrin and Aldrin were extensively used on daffodil crops in mid and west Cornwall until this derogation was revoked in May 1989.

The half-life of Dieldrin in soil ranges from four to seven years depending on soil type (Anon 1964, Edwards 1966) and it would be instructive to know what the residue levels are now in eels in south west England, eight years after the agricultural use of Dieldrin and Aldrin was finally banned.

Two otters had liver levels in excess of 2 mg/kg (WM) and these are significant. The first was the non-pregnant adult female which had a highly convoluted uterus (M455/2/90). This animal came from Newbridge where, two years earlier, eels contained up to 22 mg/kg (WM) Dieldrin. The second animal was an adult male killed near the port of Falmouth in December 1994 (M4/12/94). This high value in a recent case, together with the presence of unaltered Aldrin, rather than Dieldrin, in two other otters (M224/3/93 and M31/9/93) is evidence that localised releases of these compounds into the environment have continued to occur.

The Dieldrin levels in the otters in this study show evidence of a significant decline over the eight-year period of sampling (Figures 15 and 15a). Regular sampling of river water in Great Britain over the last 20 years has shown evidence of a similar decline, and Dieldrin levels in eels also declined from 1988 to 1993 (Zaman 1997). Taken together, these results all provide strong evidence for declining levels of Dieldrin in the environment.

7.12 DDT

The other organochlorine pesticide of major concern is DDT. Over the last 50 years it has become a worldwide environmental contaminant and, although it was withdrawn from agricultural use in Britain in 1984, significant residues were detected in eels in the MAFF 1986/87 survey (MAFF 1988). Recent analyses of eels in the St. Austell area have also shown some with high values (Environment Agency, personal communication).

Technical grade DDT is approximately 70 % pp' DDT and 15 % op' DDT. Following ingestion the *ortho-para* isomer is rapidly metabolised to form pp' DDT which is then converted to the more stable pp' DDE. For this reason, tissue levels of pp' DDT in excess of pp'DDE are usually regarded as evidence of recent ingestion (Jefferies and Walker 1966, French and Jefferies 1969). DDT residues were detected in all the otters in this study, but the values were generally low and the pp' DDT levels seldom exceeded 30 % of the pp' DDE concentration. The highest pp' DDE level was 2,397 μg/kg WM and this was in a male from Tiverton which died of bite wounds (M31/9/93). The high level was possibly influenced by the fact that the otter was emaciated. (CI = 0.81). It is known that where sick or starving animals have mobilised fat reserves, residues become concentrated in any fat that remains. Other notable cases, with DDE values over 1,000 μg/kg, were the two otters with Dieldrin residues of around 2,800 μg/kg (M455/2/90, Newbridge and M4/12/94, Falmouth).

The pp' DDE levels in the otters declined significantly over the study period and were closely correlated with the Dieldrin levels. However, whilst the decline of Dieldrin is consistent with it having been withdrawn from general use by 1981, and with its relatively short half life in the environment, the same cannot be said for DDT. Although it was withdrawn from general agricultural use in 1984, it is very much more persistent in the environment, with a half life in soil of around 57 years (Cooke and Stringer 1982). Therefore leaching from contaminated sites, especially landfill sites, could be expected to continue for many years. It is also notable that whilst pp' DDE and pp' TDE levels fell significantly over the study period, there was no clear evidence of a similar decline in pp' DDT. This is anomalous but it mirrors the situation with the PCB congeners and total PCB.

7.13 Gamma HCH

Gamma HCH, formerly known as γ BHC or Lindane, is another organochlorine pesticide used extensively in agriculture. Following the ban on Dieldrin in 1965, γ HCH was the active ingredient in the majority of sheep dips until it too was banned in 1984. However the compound is still used extensively as a pesticide on arable crops and in two earlier studies (Mason et al 1986a, Mason and Macdonald 1994) γ HCH was detected in the majority of otter samples in Great Britain, although at generally low levels. It was detected in only 25 % of the samples in this study, and then at very low levels only. The α and β isomers, which occur as contaminants in commercial preparations and which possess no insecticidal properties, were detected at trace levels only in three otters. Interpretation of analytical results for γ HCH is complicated by the fact that the compound is rapidly destroyed in tissues undergoing post mortem putrefaction. This can result in the true levels being underestimated (French and Jefferies, 1968). Nevertheless, 1,2,3 trichlorbenzene, which is a breakdown product of γ HCH, was only detected once and it seems unlikely that the otters in this study were exposed to significant levels of γ HCH.

7.14 Other Halogenated Hydrocarbons

Hexachlorobenzene is a persistent global pollutant and is a by-product of the petrochemical industry. It also occurs as a contaminant in some pesticides and has been used as a fungicide. In experimental studies it has been shown to affect the parathyroid gland, causing osteosclerosis in rats (Andrews et al, 1989), and to induce hepatic porphyria in various species (Vos et al 1968, Courtney 1979). It was present in all the otters in this study and although the levels are very low they are, nevertheless, 10 to 15 times greater than those recorded in *L. canadensis* in north west Canada (Somers et al, 1987). As the petrochemical industry worldwide continues to expand, and as HCB is distributed primarily by atmospheric fallout, one might expect the levels in the environment to increase. The marked decrease recorded in the otters from 1988 to 1996 was, therefore, unexpected (Table 5). However, it would be prudent to continue to examine otters in order to monitor the levels of this compound in aquatic ecosystems.

7.15 Heavy Metals

In general, the liver metal levels in the otters give little cause for concern (Table 9). The copper and zinc concentrations show only small variations and appear to be consistent with normal physiological values. Kidney would have been the preferred tissue when monitoring for lead and cadmium levels but, even so, the liver levels are so low that it is possible to conclude that neither of these heavy metals were having a significant adverse affect on the otters.

Mercury is extremely toxic and there are numerous reports of it causing poisoning in both man and animals. Historically, most cases were due to inorganic mercury but, in more recent times, organic mercurial compounds have been extensively used in agriculture and have proved equally dangerous. However, fish eating species of birds and mammals characteristically have tissue mercury levels much higher than would be tolerated by other species and this needs to be borne in mind when reporting levels in otters.

It is thought that naturally occurring inorganic mercury, particularly in marine environments, is converted by micro-organisms to methyl mercury. Unlike inorganic mercury, this is fat soluble and therefore readily absorbed. However, the levels of mercury in the environment have increased as a result of human activities, and in some areas they are thought to represent a risk to man as well as wildlife. Fish contaminated with mercury were thought to have been responsible for the death of a wild mink in Canada (Wobeser and Swift, 1976). It had a liver level of 58.2 mg/kg, whereas mink experimentally poisoned with methyl mercury had liver levels of 18-37 mg/kg (WM) (Wobeser et al , 1976).

In another experiment, river otters (*L. canadensis*) died when they were fed fish containing methyl mercury at 2 mg/kg (WM). The mercury concentrations in the river otters' livers were in the range 25 to 39 mg/kg (WM) and half of this was still present in the organic form (O'Connor and Nielsen, 1981). In Britain, Mason et al (1986 b) recorded a mean value of 5.37 mg/kg WM in 19 wild otters, with an upper value of 20.5 mg/kg WM. Similar values were recorded in Shetland by Kruuk and Conroy (1991) with a maximum concentration in that case of 21 mg/kg WM.

There can be little doubt that liver values of around 20 mg/kg WM are likely to be significant in otters, but the highest value recorded in the present study was approximately 8 mg/kg (WM) (31.9 mg/kg DM) and most values were well below this. However, mercury is known to accumulate in tissues over time and in Shetland the higher concentrations were in the older animals (Kruuk and Conroy, 1991). The otters examined from south west England were predominantly young specimens, with a mean age of around two years, and this may have influenced the mercury results.

The 1986/87 survey of eels in Great Britain (MAFF 1988) unfortunately did not include Cornwall. However, eels from the rivers Taw and Torridge in Devon and Tone and Huntspill in Somerset had mean mercury levels ranging from 0.04 to 0.17 mg/kg WM. These are very similar to the levels recorded in prey species in Shetland by Kruuk and Conroy (1991) where levels of up to 21 mg/kg WM were present in the older otters. There is, therefore, no reason to be complacent about mercury levels in south west England, particularly if, in time, the average age of otters in the area increases.

Cornwall has a long history of mining for metals, including arsenic, and large areas of the county are contaminated with arsenical waste. As a result, cases of acute arsenic poisoning are occasionally seen in cattle which have ingested contaminated soil and many streams receive discharges rich in arsenic from disused mines.

Arsenic was detected in two otters, one at 2.4 mg/kg DM and the other at 14 mg/kg DM. These equate with wet tissue levels of approximately 0.6 and 3.5 mg/kg. In a domestic animal this latter value would be suggestive of arsenic poisoning, provided that the relevant pathological lesions were seen. In the case of this otter (M290/6/94) they were absent. However, as is the case with mercury, certain fish-eating species may carry what appear to be elevated levels without ill effect. In such species it is thought that organic arsenical compounds of low toxicity are derived from food items such as shellfish and crustacea. It is ironic that this otter came from Hampshire, not Cornwall. However, it was also the suspected captive release and as such may have been fed a diet rich in arsenic, e.g. marine shellfish.

Overall there is no evidence from this study that heavy metals are causing health problems in otters in south west England, but the position with regard to mercury merits further monitoring.

7.16 Pathology of PCBs, OCs and Related Compounds

At the start of this study it was apparent that, although many hundreds of otters in both Europe and North America had been analysed for pollutants, there was no clear evidence that the levels detected were causing health problems. At best, it could be said that where PCB levels were low, otter populations were stable, but where they were high, the populations were low or absent (Mason 1989, Olsson et al 1981). For these reasons, it was decided that a post mortem protocol was necessary which included not only analysis of tissues for pollutants, but also examination for pathological lesions, especially those known to be associated with halogenated hydrocarbons and heavy metals.

Numerous experimental studies have shown that the PCBs, along with other halogenated hydrocarbons, are capable of causing a great range of pathological effects. These are too numerous to describe in detail but include the following:-

Body weight:	Initial increase, followed by wasting and death.
Liver:	Increase in size, fatty infiltration, hepatocyte necrosis and bile duct hyperplasia.
Kidney:	Enlargement with distended convoluted tubules.
Heart:	Initial hypertrophy, then regression and atrophy.
Spleen, thymus:	Atrophy, with reduced lymphoid follicles.
Alimentary system:	Mucosal ulceration, haemorrhage, tarry faeces.
Adrenal glands:	Cortical hyperplasia.
Thyroid glands:	Hyperplasia or atrophy or colloid goitre-like state. (Response depends on compound, species, dose and duration.)
Skin:	Epidermal hyperplasia, hyperkeratosis.

In addition, the halogenated hydrocarbons cause significant physiological disorders, particularly to Vitamin A metabolism and steroid hormone balance. These compounds all stimulate the liver to produce microsomal enzymes which are then involved in the metabolism and excretion of the pollutants. Possibly because of structural similarities between the halogenated hydrocarbons and the steroid hormones, the induced enzymes destroy hormones such as testosterone, oestrodiol, progesterone and desoxycorticosterone (Kimbrough 1974, Conney et al 1967, Peakall 1967). Further hormonal dysfunction is caused by compounds such as op' DDT and some PCBs which mimic oestrogens (Bitman et al 1968, Levin et al, 1968).

The lesions listed above have been observed in a wide range of mammalian and avian species, and it will be seen that some, such as splenic atrophy, adrenocortical hyperplasia and bile duct hyperplasia, were also present in otters in this study. However, none of the lesions is pathognomonic, i.e. caused by PCBs and related compounds but by nothing else. Splenic atrophy, for example, also occurs in cases of starvation and adrenocortical hyperplasia may result from chronic stress. The bile ducts of many otters appeared hyperplastic but it is not known if this is a normal feature of healthy otters. Therefore, although several of the gross and histopathological changes observed were suggestive of PCB or OC toxicity, they were not conclusive. Comparisons with tissues from otters in both more and less polluted environments would be worthwhile.

7.17 Pathology of Vitamin A Deficiency

The pathology of Vitamin A deficiency varies to some degree between species but typically it includes:

a) foetal resorption, abortion, stillbirth and malformations, including hypoplasia of the gonads and cryptorchidism,
b) defects in bone modelling,
c) xerophthalmia, keratitis, corneal ulceration, retinal degeneration, night blindness
d) increased predisposition to infections, and
e) formation of renal calculi.

In many instances, the pathology of Vitamin A deficiency is due to its effect on epithelial development, with atrophy followed by squamous metaplasia and keratinisation. In secretory organs, such as salivary glands, lachrymal glands or pancreas, the result may be obstruction of the ducts and reduced secretions.

As otters eat mostly fish, which should be rich in Vitamin A, one would not expect them to have low Vitamin A reserves. Liver values in domestic animals vary but are normally in excess of 200 μmol/kg (Blood et al, 1983). Values below 7 μmol/kg are considered to be critical (Blood et al, 1983) and below 2 μmol/kg are evidence of severe deficiency (Doxey, 1983).

These quoted figures should only be applied to otters with caution but, fortunately, there is a report of Vitamin A values in apparently normal wild otters (Stephens, 1957). Although the sample was small, the results are particularly valuable, as the analyses were carried out in 1952 to 1954, i.e. immediately before the decline of the otter population in Britain. Six samples were analysed, including two from Cornwall, and the values ranged from 27.5 to 515 iu./g, with a mean of 168.5 iu./g. When expressed in SI units these equate to a range of 28.8 to 540 μmol/kg and a mean of 176.6 μmol/kg. For comparison, the values in the present study ranged from less than 1 μmol to 2433 μmol/kg, with a mean of 317 μmol/kg. Within this group there were seven animals with values lower than 7 μmol/kg and five with values of approximately 1,000 μmol/kg or more.

In view of the ability of the halogenated hydrocarbons to increase as well as decrease Vitamin A levels, this extreme range of values is of interest, particularly as very high Vitamin A levels may also cause physiological effects. These include foetal resorption, congenital abnormalities, bone defects, exophthalmia and thickening of the skin.

Considering the analytical results, it was surprising that so few pathological changes were present which could be attributed to a deficiency, or excess, of Vitamin A. The stunted, cryptorchid otter from Goss Moor (M340/10/94) was the only animal with gross defects which were consistent with hypovitaminosis A. In this case the changes would probably have occurred during foetal development *in utero* - where the dam was deficient in Vitamin A - or during its early life as a cub.

It is of interest to note that both the other otters killed in the Goss Moor area also had very low Vitamin A levels (M310/12/88 and M111/9/90). More detailed histopathological examinations of eyes and reproductive tracts are currently being carried out (see 9.1) and it is possible that these may yield further evidence of Vitamin A deficiency.

Renal calculi are commonly found in captive otters and have occasionally been recorded in wild ones. Their aetiology is unknown, but it has been suggested that they could be caused by Vitamin A deficiency (Keymer 1981). However, the results of this study do not provide any support for this hypothesis. Conversely, the author has repeatedly seen cases in captive Eurasian otters and Asian small clawed otters (*Aonyx cinerea*) where the Vitamin A levels have been in the range of 1,000 to 3,000 μmol/kg (Simpson, unpublished data). Only one otter in the present study had a renal calculus (M89/8/95) and it was not in fresh enough condition for Vitamin A analysis to be carried out.

Recent work in The Netherlands, carried out on otters killed in Denmark, showed an inverse relationship between hepatic Vitamin A levels and PCBs (van der Weiden et al 1996, Murk A J, personal communication). The results in the present study were similar to those obtained by the Dutch workers and it was surprising therefore, that the Vitamin A and PCB levels were more strongly related to time than to one another. However, the Dutch reported that the correlation was much clearer when PCBs were expressed as the TEQ of seven of the most toxic congeners, and not as the concentration of total PCB, or of individual congeners (Ahlborg et al 1994).

7.18 Thyroid Size and Relationship to PCB and OC Levels

Histological examination of thyroid glands did not reveal any obvious relationship between the degree of follicular activity, or the amount of colloid, and the levels of these pollutants. In other species of mammals or birds, DDT or Dieldrin-induced increases in thyroid gland size have typically been associated with follicular hyperplasia and depletion of colloid, whereas PCB-induced increases in gland size have resulted in increased colloid. In the otters it is likely that the thyroid glands were being influenced by both groups of chemicals but, in the absence of any glands from 'non-polluted' otters, it is not possible to say whether the appearance, or function, of the glands has been affected. A quantitative assessment of follicle size or colloid area might make it possible to relate the degree of activity to the level of certain pollutants.

The male otters in this study had, on average, heavier thyroid glands than the females. This is consistent with the fact that thyroid weight is related to bodyweight and males were usually heavier than females. When the effect of body weight was taken into account, there no longer was a significant difference between the thyroid weights of males and females. Although there was a significant relationship between thyroid weight and the levels of certain pollutants, notably pp' DDE and Dieldrin, there was no evidence of a causal connection once the effect of body weight had been taken into account. Overall, heavier otters generally had higher pollutant concentrations.

Although males generally had larger thyroid glands and higher PCB and OC levels than females, it can be seen in Figure 22 that six males had unusually small thyroids (< 0.5 gm) and low Dieldrin levels (<100 μg/kg). The smallest of these was a cub weighing only 2 kg but it was in good condition, with a CI of 1.07. However, the other five were all animals in poor condition, with CIs ranging from 0.54 to 0.88.

Only one of these (M153/3/95) was suffering from bite wounds and the cause of the poor conditions of the other otters was not established, although one was the cryptorchid case from Goss Moor (M340/10/94). In view of the poor physical condition of these animals, their low Dieldrin levels were surprising. It is a well-established fact that residue concentrations tend to rise in animals which, because of stress, illness or migration, have metabolised body fat reserves. If the animals in question had had low or average Dieldrin levels prior to their loss of body condition, it would be expected that the concentration would have risen to average or above, after body fat reserves had been metabolised.

One further anomalous case was the male M4/12/94 from Penryn. This had by far the highest Dieldrin level in a male and also had a very high pp' DDE level. It weighed 7 kg but its thyroid glands weighed only 0.4 gm. It is possible that this case represents thyroid exhaustion and involution, following prolonged exposure to high levels of Dieldrin and DDT.

7.19 PCB and OC Levels in Males and Females

The hepatic PCB and OC concentrations were higher in males than females, although this difference was only statistically significant in the case of PCBs. Similar observations have been made in other wild species, especially marine mammals. It is believed that this occurs because females excrete a large proportion of their total body PCB burden in the fat fraction of colostrum and milk (Tanabe 1988). A consequence of this is that the new born animal is likely to receive a heavy burden of PCBs and OCs at a particularly vulnerable stage in its development.

The residue levels in female otters which showed evidence of having bred were, on average, lower than in those which had not bred. However, the number of animals involved was very small (7 and 10 respectively for each group) and the results have not been analysed statistically.

Recent work on polecats in The Netherlands revealed that excretion of PCBs also occurs via secretions from the anal glands (Leonards et al 1994). The authors suggest that PCBs accumulate in immature animals and then, when the animal becomes sexually mature, excretion occurs due to increased anal gland activity associated with territorial behaviour. As otters are also mustelids, one might expect them to excrete PCBs by this route. However, it is unlikely that females would excrete more PCBs by this route than males and, therefore, this is unlikely to provide an explanation for the lower levels in females.

Female otters may have different dietary preferences, but in polecats this was not thought to be of major significance (Leonards et al 1994). Furthermore, eels were observed to be the predominant food item in the stomachs of both male and female otters. At the present time it seems likely that the lower levels of pollutants in female otters are due to excretion via milk and colostrum. Although this may benefit the dam, it could have serious consquences for the cubs. The possible effects of the higher concentrations of pollutants on the physiology of males, particularly with regard to organs such as thyroids and adrenal glands, merit further consideration.

8 CONCLUSIONS

8.1 The results of this study suggest a young, generally healthy population of otters in south west England with very little evidence of infectious disease. Road traffic represents a major, and increasingly important, cause of mortality and suffering. Bite wounds, apparently caused by other otters, are the other main cause of death, but these may be regarded as natural events. The unusual uteri seen in three females of breeding age are cause for concern. Although they could represent a normal stage of development in early pregnancy their appearance is unusual and could be due to the effects of pollutants.

8.2 Low Vitamin A levels were seen predominantly in the period 1988 to 1992 and, although these were related to the hepatic levels of Dieldrin, DDT and PCBs, a causal relationship was not established. Several of the histopathological lesions recorded in the otters are consistent with those known to be caused by PCBs, OCs and related compounds, but there is no proof that the lesions seen were caused by the levels of pollutants present. Some of the lesions could possibly have been due to naturally occurring processes in otters. However, the strong correlation between adrenal weight and hepatic concentrations of some PCBs, especially congener 153, suggest that these pollutants may be interfering with otters' endocrine systems. There is also evidence that pollutant levels, in particular, pp'DDE and Dieldrin, are related to body weight.

8.3 There is now widespread agreement that the otter population in Britain is increasing and this study has shown that in south west England there has been a marked increase in submissions in recent years. During this period the levels of Dieldrin in the otters have decreased significantly along with levels of DDT and some PCB congeners. At the same time there has been a marked reduction in the number of cases of otters showing very low Vitamin A levels.

The decline in the populations of otters and birds of prey in Britain followed immediately after Dieldrin was introduced in 1956. The recovery in the numbers of birds of prey in this country started three to four years after Dieldrin was banned as a seed dressing and the current recovery of the otter population in south west England also appears to have started only a few years after Dieldrin was banned altogether. These events are consistent with the relatively short half-life of Dieldrin and are unlikely to be related to reduced levels of DDT and PCBs in the environment, as they are so much more persistent.

Whilst there can be no doubt about the potential for PCBs and related compounds to induce toxic changes in most living organisms, the combined circumstantial and analytical evidence suggests that Dieldrin, possibly acting synergistically with such compounds, has had a major effect on Britain's otter population.

9 RECOMMENDATIONS

9.1 The contract brief included an agreement to examine/re-examine archived histology material. However, the investigations produced more data than anticipated and it was not possible to examine all the archived material in detail in the time available. It is therefore recommended that:

a) Histology sections of tissues already processed should be reassessed in the light of the various analytical results obtained. This applies in particular to the adrenal and thyroid glands and how their appearance and structure might relate to the levels of PCBs and OCs.

b) In some cases, tissues have been stored in fixative but not processed. Where appropriate, selected tissues from such cases should be sectioned and examined in the same manner as those described in a).

c) Female reproductive tracts collected during this study, together with those from otters collected up to May 1997, have been subjected to gross and histological examination by Sonja Rivers at the Royal Veterinary College, London. Her preliminary results suggest the possibility that delayed implantation may be occurring. The Physiology Department of the RVC is keen to extend these investigations and, in view of the importance of this area of work, the Environment Agency should continue to give its full support.

d) Eyes collected during this study, and up to mid 1997, have been submitted to Dr. David Williams at the Animal Health Trust, Newmarket, for specialist examination. His report is expected shortly and, when available, the results should be collated with those in this report.

9.2 The data and methodology relating to the levels of fat in liver need to be subjected to critical evaluation.

9.3 All existing frozen, archived, tissues should be retained for possible future studies, particularly with regard to levels of specific PCB congeners and related compounds, and the partition of these between various tissues. (See also 9.7).

9.4 The mean age of the sample of otters in this study was much lower than has been reported previously from elsewhere in Europe. This may have been due to a high proportion of young, inexperienced animals being killed by traffic. However, the Environment Agency should consider trying to establish the true age structure of the population. Teeth collected from 1994 onwards are currently being examined by Dr. Ranald Munro, Lasswade Veterinary Laboratory, in order to establish the age of each otter. The results, when available, may indicate whether the otters examined show evidence of an increase or decrease of age over time. It may also be possible to determine whether the pollutant levels or some of the histological findings are age-related.

9.5 No otters were received from areas such as east Devon or Dorset, even though the habitat there appears suitable. Every effort should be made to collect dead otters from such areas. The Environment Agency should also consider comparing the results of analyses of silt and eel flesh for PCBs, OCs and mercury from such areas with contemporary results from other areas which hold good numbers of otters. It would be particularly valuable to repeat the eel sampling exercise of 1986/87.

9.6 The Environment Agency should consider possible sources of PCBs and OCs, particularly in areas where otters have shown high levels of pollutants or low levels of Vitamin A, e.g. Goss Moor. The possibility of contamination from landfill sites, electricity sub-stations, timber treatment works, sewage works, industrial estates, etc. should be considered.

9.7 It is recommended that the Environment Agency continues to pay for post mortem examinations on the present basis, in order to maintain continuity of data and to establish trends over time. When submitting carcases, increased effort should be made to ensure that those in fresh condition are kept chilled and reach the laboratory within 24 hours. Archived histology specimens should be stored in wax blocks, rather than as wet tissue, as they remain much more stable in this form.

Tissues should continue to be analysed in year batches for organic pollutants but unused tissues should be retained for future reference, particularly between batches. The total PCB, expressed as Aroclor 1260 equivalent, should continue to be calculated. However, a TEQ for each sample should also be calculated and it is recommended that this should be based on an internationally recognised list of toxic congeners (Ahlborg et al 1994). Levels of HCB should continue to be monitored.

10 REFERENCES

Ahlborg U G, Becking G C, Birnbaum L S, Brouwer A, Derks H J G M, Feeley M, Golor G, Hanberg A, Larsen J C, Liem A K D, Safe S H, Schlatter C, Wacru F, Yonnes M, Yrjanheikki E (1994). Chemosphere 28, 1049-1067.

Andrews J E, Courtney K D and Donaldson W E (1989). Fundamentals of Applied Toxicology. 12, 242.

Anon (1964). Report of the Research Committee on Toxic Chemicals. Agricultural Research Council, London.

Aulerich R, Bursian S, Breslin W, Olson B and Ringer R (1985). Journal of Toxicology and Environmental Health. 15, 63-79.

Aulerich R and Ringer R (1970). American Fur Breeder. 43, 10-11.

Aulerich R and Ringer R (1977). Archives of Environmental Contamination and Toxicology. 6, 279-292.

Backlin B and Bergman A (1992). Ambio. 21, 596-601.

Ballschmiter K and Zell M (1980). Fresenius Z. Anal. Chem. 302, 20-31.

Bergman A and Olsson M (1985). Finnish Game Research. 44, 47-62.

Bevanger K and Albu O (1986). Biological Conservation. 38, 75.

Bitman J, Cecil H C, Harris S J and Fries G F (1968). Science. 162, 371-372.

Blood D, Radostits O and Henderson J (1983). Veterinary Medicine. Sixth Edition. Bailliere Tindall.

Bröuwer A, van den Berg K, Blaner W and Goodman D (1986). Chemosphere. 15, 1699-1706.

Chanin P and Jefferies D (1978). Biological Journal of The Linnean Society. 10, 305-328.

Conney A H, Welch R M, Kuntzman R and Burns J (1967). Clinicial Pharmacology and Therapeutics. 8, 2-10.

Cooke B K and Stringer A (1982). Pesticide Science. 13, 545-551.

Courtney K D (1979). Environmental Research: A Review. 20, 225-266.

Darjano S, Stowe H and Aust S (1983). Toxicology and Applied Pharmacology. 71, 184-193.

Davies J M, Roper T J and Shepherdson D J (1987). Journal of Zoology, London. 211, 525-529.

Doxey D (1983). Clinical Pathology and Diagnostic Procedure. Second Edition. Bailliere Tindall.

Edwards C A (1966). Residue Review. 13, 83-132.

Erlinge S (1967). *Oikos*. 18, 186-209.

Foley R E, Jackling S J, Sloan R J and Brown M (1988). Environmental Toxicology and Chemistry. 7, 363-374.

French M C and Jefferies D J (1968). Nature. 219, 164-166.

French M C and Jefferies D J (1969). American Association for the Advancement of Science. 165, 914-916.

Hamilton W J and Eadie W R (1964). Journal of Mammalogy. 45, 242 - 252.

Heggberget T (1988). Journal of Mammalogy. 69, 164-167.

Helle E, Olsson M and Jensen S (1976). Ambio 5, 261-263.

Henny C, Blus L, Gregory S and Stafford C (1981). Proceedings of the Worldwide Furbearers Conference. Frostburg, MD. Eds. J Chapman and D Pursley, pp 1763-1780.

Hernandez L M, Gonzalez M J, Rico M C, Fernandez M A and Baluja G (1985). Journal of Environmental Science and Health. 20, 633-650.

Hornshaw T, Aulerich R and Johnson H (1983). Journal of Toxicology and Environmental Health. 11, 933-946.

Jensen S, Kihlström J, Olsson M, Lundberg C and Orberg J (1977). Ambio 6, 239.

Jefferies D, French M and Stebbings R (1974). Pollution and mammals. Monks Wood Experimental Station Report 1972-73. 13-15. N.E.R.C., Huntingdon.

Jefferies D and Walker C (1966). Nature. 212, 533-534.

Jefferies D and Parslow J (1972). Bulletin of Environmental Contamination and Toxicology. 8, 306-310.

Jefferies D (1975a). Journal of Zoology, London. 177, 504.
Jefferies D (1975b). The Role of the Thyroid. In: Organochlorine Insecticides: Persistent Organic Pollutants. Ed. F. Moriarty. Academic Press, London.
Kannan N, Tanabe S and Tatsukawa R (1988). Bulletin of Contamination and Toxicology. 41, 267.
Keymer I (1981). Sonderdruck aus Verhandlungsbericht des XXII Internationalen Symposiums über die Erkrankungen der Zootiere, Halle/Saale, 391-401.
Keymer I (1991). Otters in East Anglia, Past, Present and Future. Norfolk Naturalists Trust, 72 Cathedral Close, Norwich. 18.
Keymer I, Wells G, Mason C and Macdonald S (1988). Veterinary Record 122, 153-155.
Kihlström J, Olsson M, Jensen S, Johansson A, Ahlbom J and Bergman A (1992). Ambio 21, 563-569.
Kimbrough R (1974). CRC Critical Reviews in Toxicology. 2, 445-495.
Kruuk H, Conroy J and Moorhouse A (1987). Symposia of the Zoological Society of London. 58, 263-278.
Kruuk H and Conroy J (1991). Journal of Applied Ecology 28, 83-94.
Le Cren E (1951). Journal of Animal Ecology. 20, 201.
Leonards P, van Hattum B, Cofino W and Brinkman U (1994). Environmental Toxicology and Chemistry. 13, 129-142.
Levin W, Welch R M and Conney A H (1968). Federation Proceedings. 27, 649.
MAFF (1988). Report of Working Party on Pesticides and Residues (1986-1987).
Macdonald S and Mason C (1990). Action Plan for European Otters. Otters, an Action Plan for their Conservation. I.U.C.N. 29.
McLaren G W (1992) BSc Thesis, Aberdeen University.
Marshall, H (1991). Report of the Transactions of the Devon Association for the Advancement of Science. 123, 137-147.
Mason C (1989). Lutra 32, 97-131.
Mason C, Ford T and Last N (1986a). Bulletin of Environmental Contamination and Toxicology. 36, 656-661.
Mason C, Last N and Macdonald S (1986 b). Bulletin of Environmental Contamination and Toxicology. 37, 844-849.
Mason C and Macdonald S (1986). Otters: Ecology and Conservation. Cambridge University Press.
Mason C and Macdonald S (1994). The Science of the Total Environment. 144, 305-312.
Mason C and Madsen A (1990). Natura Jutlandica 22, 217-220.
Mason C and O'Sullivan W (1992). Bulletin of Environmental Contamination and Toxicology. 48, 387-393.
Neal E (1977). Badgers. Poole, Dorset; Blandford Press.
Newton I and Haas M (1984). British Birds. 77, 44-70.
O'Connor D and Nielsen S (1981). Proceedings of the first Worldwide Furbearers Conference, American Forestry Association. pp 1728-1745.
Olafson P (1947). Cornell Veterinarian. 37, 279-291.
Olsson M, Reutergardh L and Sandegren F (1981). Sveriges Natur. 6, 234-240.
Peakall D B (1967). Nature. 216, 506.
Platonow N, Karstad L (1973). Canadian Journal of Comparative Medicine 37, 391-400.
Reuther, C (1980). Natursh. Landschaftspf. Niedersachsen. 11, 1.0
Safe S (1990). C.R.C. Critical Reviews in Toxicology. 21, 51-88.
Somers J, Goski B and Barrett M (1987). Bulletin of Environmental Contamination and Toxicology. 39, 783-790.
South West Water (1988). South West Water News, SWW, Peninsula House, Rydon Lane, Exeter, EX2 7HR.
Stephens M (1957). The Otter Report. Pub: UFAW, 230 High Street, Potters Bar, Herts.
Tanabe S (1988) Environmental Pollution 50, 5-28.

Tanabe S, Kannan N, Subramanian An., Watanabe S and Tatsukawa R (1987). Environmental Pollution. 47, 147-163.

Van der Weiden M E, Hendriks A J, Klein M H, Leonards P E, Smit M, Murk T A, Plassche E, Crommentuijn T, Luttik R and Hattum B (1996). Organohalogen Compounds. 29, 126-131.

Vos J, Breeman H and Benschop H (1968). Meded. Rijksfac. Landvouwwetensch. Gent. pp 1263-1269.

Vos J, Koeman J, Van der Maas H L, Ten Noeveer De Brauw M and De Vos R (1970). Food Cosmet. Toxicol. 8, 625-633.

Wells G, Keymer I and Barnett K (1989). Veterinary Record 125, 232.

Wobeser G, Nielsen N and Schiefer B (1976). Canadian Journal of Comparitive Medicine. 40, 34-45.

Wobeser G and Swift M (1976). Journal of Wildlife Diseases. 12, 335-340.

Yakushiji T (1988). Reviews of Environmental Contamination and Toxicology. 101, 139-164.

Zaman N (1997). Review of organochlorine pesticide and polychlorinated biphenyl bioaccumulation in fish and fish-eating mammals. Report No. TAPS/Tox/201/00NWZ. Environment Agency.

GLOSSARY OF TECHNICAL TERMS

The following list of veterinary terms is based on those given in Baillière's Comprehensive Veterinary Dictionary by D C Blood & Virginia P Studdert, Baillière Tindall.

Acidophilic	Readily stains with acid dyes.
Atrophy	Decrease in size, or wasting of, a normally developed tissue.
Basophilic	Readily stains with basic dyes.
Calculus, pl. calculi	Abnormal concretion of mineral salts, e.g. dental: tartar; renal: kidney stones.
Calyx, pl. calices	Cup-shaped recess in the kidney pelvis which encloses a medullary pyramid.
Cardiac	Pertaining to the heart.
Colloid	Thyroid: thick secretion in follicles.
Cortex	1) Adrenal: outer, firm, layer comprising the larger part of the adrenal gland. 2) Renal: the smooth textured outer layer of the kidney.
Cortical	Pertaining to, or emulating from, the cortex.
Epidermis	The outermost, non-vascular, layer of the skin.
Epithelium	The cellular covering of the internal and external surfaces of the body.
Eosinophilic	Readily stains with eosin.
Focus	The chief centre of a morbid process, (a small, defined, lesion).
Follicle	1) Spleen: small clusters of dividing lymphocytes. 2) Thyroid: cyst-like units filled with a colloid substance.
Glomerulus	Renal: encapsulated tuft of capillaries in kidney involved in blood filtration.
Gonad	Testis or ovary.
Hepatic	Pertaining to the liver.
Hepatocytes	Predominant cells of liver.
Hyperkeratosis	Increase deposition, or accumulation, of keratin in a tissue.
Hyperplasia	Increase in size of organ or tissue caused by increased number of normal cells.
Hypertrophy	Increase in size of organ or tissue caused by increase in size of existing cells.
Hypoplasia	Incomplete or under-development of organ or tissue.
Hypovolaemic shock	Shock associated with severe blood loss.
Keratinisation	Development of, or conversion to, keratin.
Keratitis	Inflammation of the cornea.
Medulla	Central or inner portion of a gland, e.g. adrenal gland or kidney.
Metaplasia	Change in the type of adult cells in a tissue to an abnormal form.
Mucosa	Mucous membrane lining, e.g. alimentary tract.
Necrosis	Morphological changes indicative of cell death.
Osteosclerosis	The hardening, or abnormal density, of bone.
Parenchyma	The essential or functional elements of an organ, as opposed to its supporting framework etc.
Plantar surface	Pertaining to the sole of the foot/digit.
Porphyria	Abnormal accumulation in the tissues, or increased excretion of, porphyrins.
Renal	Pertaining to the kidney.
Schizont	A stage in the development of a protozoal parasite where the nucleus divides into many.
Spermatogenesis	The formation in the testes of sperm.
Splenic	Pertaining to the spleen.
Xerophthalmia	Abnormal dryness and thickening of the surfaces of the conjunctiva and cornea.

Basic PM Protocol for Otters (*Lutra lutra*)

1. Weigh the animal (preferably dry) and give it a unique laboratory reference number.
2. Examine external features, including eyes, nose, ears, genitals, anus and feet.
3. With the animal on its back, and using a rigid rule, measure.
   a) nose to anus.
   b) anus to tail tip.
4. Examine mouth and teeth. Remove an upper incisor (complete) for age estimation. Place in 10 % BFS.
5. Carry out detailed gross examination of all internal organs, including reproductive system. Photograph any abnormalities. Weigh all major organs (see Appendix Ia).
6. Collect 3-4 mm thick samples of any organ showing pathological lesions. Place samples in 10 % BFS for histological examination.
7. In cases where organs show lesions suggestive of an infectious process, e.g. pus formation or inflammation, culture onto appropriate media. If there are lesions suggestive of a viral infection, e.g. CDV or Aleutian disease place, small uncontaminated, samples in sterile containers (e.g. universal pots) and hold at 4°C whilst contacting a virology reference laboratory.
8. Collect samples of liver and body fat (minimum of 20g in each case), wrap in aluminium foil, label and store in deep freeze pending toxicological examination.
9. Collect any stomach contents and store as in paragraph 8.
10. Specifically check kidneys for renal calculi and if present, collect them (dry) for biochemical analysis. Retain kidney tissue (minimum 10g) in deep freeze for possible future toxicological examination.
11. If possible collect a serum sample for future antibody studies. Hold in deep freeze.

Appendix Ia

POLWHELE VETERINARY INVESTIGATION UNIT
Otter *Post Mortem* Data

VI Ref No: .....

Total Length: cms
Nose to Vent: cms
Vent to Tail: cms
Body Weight: kgs

Date Submitted:
Date of PM:
Sex:
Fresh or Frozen:

Organ	Weight gms	Hist	Freeze	Notes
Heart				
Liver				
Spleen				
Right Kidney				
Left Kidney				
Right Thyroid				
Left Thyroid				
Right Adrenal				
Left Adrenal				
Lung				
Cardiac Thymus				
Pancreas				
Foot Pad	---		---	
Eye	---		---	
Fat	---	---		
Muscle	---			
Uterus/Gonads	---		---	
Brain	---			
Sub Mand S/G	---		---	
Bladder	---		---	
Stomach Contents		---		
Rib	---	---		
Incisor Tooth	---		---	
Clotted Blood	---	---		
Liver: Vitamin A	---	---		
Urine	---	---		

Appendix II

LIST OF OTTERS SUBMITTED AND ORIGIN

No.	Ref. No.	NGR	Origin	No.	Ref. No.	NGR	Origin
1	M310/12/88	SW 901 571	Tresillian	40	M77/8/94	SW 812 514	Allen
2	M455/2/90	SW 42 31	Drift	41	M289/9/94	SX 031 799	Amble
3	M535/2/90	SX 01 47	St. Austell	42	M290/9/94	SS 732 488	Lyn
4	M701/5/90	Unknown	N/A	43	M311/9/94	SW 400 315	Tidal Hayle
5	M125/6/90	SX 33 55	Lynher	44	M211/10/94	ST 447 236	Parrett/Yeo
6	M111/9/90	SW 94 60	Fal	45	M340/10/94	SW 945 604	Fal
7	M238/12/90	SX 37 55	Lynher	46	M43/11/94	SS 631 249	Taw
8	M646/1/91	SX 43 71	Tamar	47	M152/11/94	SS 499 179	Torridge
9	23-3-91	SS 93 24	Exe	48	M180/11/94	Hampshire	Unknown
10	17-11-91	SX 35 71	Lynher	49	M219/11/94	SS 215 036	Bude
11	M200/1/92	SX 386 643	Lynher	50	M4/12/94	SW 775 357	Penryn
12	M129/3/92	SX 008 499	St. Austell	51	M90/12/94	ST 625 451	Sheppey
13	M188/3/92	SX 25 53	Looe	52	M139/12/94	ST 245 229	Tone
14	4-4-92	SS 55 33	B'staple Yeo (Sea)	53	M173/12/94	SX9003 9678	Creedy
15	28-4-92	SW 42 31	Drift	54	M44/1/95	SS 956 123	Exe
16	12-6-92	ST 636 643	Chew	55	M110/2/95	SW 535 349	Hayle
17	M192/10/92	SX 05 84	Allen	56	M332/2/95	SS 443 013	Torridge
18	M193/10/92	SX 043 528	Crinnis	57	M112/3/95	ST 343 154	Isle
19	M177/11/92	SX 082 794	Camel	58	M153/3/95	ST 100 394	Doniford Stream
20	M216/11/92	SX 371 926	Carey	59	M157/5/95	SW 508 398	Stennack
21	M36/12/92	SW 814 609	Gannell	60	M89/8/95	SS 511 445	Taw
22	M91/12/92	SW 81 41	Fal	61	M90/8/95	SS 464 377	Taw
23	M34/2/93	ST 38 53	Axe	62	M71/10/95	SS 829 217	Middle Exe
24	M30/3/93	SS 663 147	Taw	63	M73/10/95	SX 215 803	Lynher
25	M224/3/93	SS 48 36	Caen	64	M62/11/95	Unknown	Unknown
26	M225/3/93	SS 635 446	B'staple Yeo	65	M70/11/95	SW 867 681	Porthcothan
27	M115/4/93	SX 774 617	Tidal Dart	66	M81/11/95	ST 165 298	Tone
28	M135/7/93	SW 77 48	Kenwyn	67	M82/11/95	ST 073 409	Doniford
29	M31/9/93	SS 949 112	Exe	68	M153/11/95	SX 087 648	Camel
30	M131/10/93	SW 772 386	Kennal	69	M62/12/95	SX 121 716	Camel
31	M154/12/93	SX9488 8798	Exe	70	M100/12/95	SS 620 199	Taw
32	M206/12/93	ST 485 378	Brue	71	M134/1/96	SX 156 653	Fowey
33	M281/12/93	SX 608 519	Yealm	72	M190/1/96	SX 347 798	Lower Tamar
34	M301/2/94	SW 83 43	Fal	73	M191/1/96	SS 946 169	Exe
35	M348/2/94	SX 057 637	Camel	74	M219/1/96	SX 007 497	St. Austell
36	M364/3/94	SX 295 732	Lynher	75	M71/2/96	SW 543 364	Tidal Hayle
37	M161/4/94	SS 613 093	Okement	76	M50/3/96	SS 427 262	Tidal Torridge
38	M290/6/94	Hampshire	Unknown	77	M111/3/96	ST 381 481	Mark/Yeo
39	M210/7/94	ST 20 20	Tone				

Histopathology - Tissues Examined

Appendix III

Ref	Ht	Liv	Spl	Kid	Thyr	Adr	Lng	Thym	Panc	F.pad	Eye	Ut/ov	Test	Brain	S/G	Blad	LN
M455/2/90		✓		✓	✓	✓						✓✓	-		✓	✓	✓
M535/2/90		✓		✓	✓	✓		✓		✓		✓✓	-			✓	
M111/9/90				✓	✓	✓											
M238/12/90		✓	✓	✓	✓	✓		✓		✓		✓	-			✓	✓
M646/1/91	✓	✓	✓	✓	✓		✓	✓	✓	✓						✓	
23-3-91		✓		✓	✓	✓				✓	✓					✓	✓
M200/1/92																	✓
28/4/92					✓												
M192/10/92	✓	✓		✓	✓	✓	✓	✓	✓		✓	✓✓	-		✓	✓	
M193/10/92	✓	✓	✓	✓	✓	✓		✓		✓	✓				✓	✓	✓
M225/3/93					✓	✓		✓	✓						✓		
M31/9/93										✓							
M131/10/93		✓		✓	✓	✓	✓	✓							✓	✓	
M154/12/93			✓	✓	✓		✓	✓								✓	
M206/12/93		✓		✓	✓		✓									✓	✓
M281/12/93					✓												
M301/2/94		✓		✓	✓					✓					✓	✓	
M364/3/94		✓	✓	✓													
M348/2/94					✓					✓							
M77/8/94		✓		✓	✓	✓									✓		
M311/9/94		✓		✓	✓	✓									✓	✓	
M211/10/94		✓		✓	✓	✓		✓					✓		✓		
M340/10/94		✓		✓	✓	✓		✓		✓			✓				
M43/11/94		✓		✓	✓	✓				✓			✓				
M152/11/94		✓		✓													
M180/11/94		✓		✓	✓	✓	✓	✓			✓					✓	✓

Histopathology - Tissues Examined

Appendix III

Ref	Ht	Liv	Spl	Kid	Thyr	Adr	Lng	Thym	Panc	F.pad	Eye	Ut/ov	Test	Brain	S/G	Blad	LN
M219/11/94										✓				✓			
M90/12/94				✓		✓				✓							
M110/2/95		✓		✓	✓	✓				✓	✓		✓			✓	
M332/2/95				✓	✓	✓				✓	✓	✓✓					
M112/3/95				✓									✓				
M153/3/95		✓		✓	✓	✓	✓	✓								✓	
M157/5/95		✓		✓	✓	✓	✓	✓	✓		✓					✓	
M89/8/95		✓		✓	✓							✓					
M90/8/95		✓		✓			✓	✓		✓							
M71/10/95		✓		✓	✓	✓		✓		✓	✓		✓			✓	
M70/11/95		✓		✓	✓	✓	✓	✓		✓	✓		✓				
M81/11/95				✓	✓	✓				✓		✓					
M153/11/95		✓		✓	✓	✓	✓	✓		✓	✓		✓			✓	
M100/12/95				✓	✓	✓				✓			✓				
M134/1/96				✓						✓			✓				
M190/1/96		✓		✓						✓							
M219/1/96		✓		✓	✓	✓	✓			✓	✓	✓✓				✓	
M191/1/96		✓		✓				✓		✓	✓	✓✓				✓	
M71/2/96		✓		✓	✓	✓	✓	✓	✓	✓	✓	✓✓				✓	
M111/3/96		✓		✓	✓	✓	✓	✓		✓	✓		✓				

HEAVY METAL LEVELS IN LIVERS (Values in mg/kg DM)

Case Ref	Zn	As	Hg	Pb	Cr	Cu	Cd
M301/2/94	86.6	ND	3.9	ND	ND	44.9	0.7
M348/2/94	108	ND	7.9	1.2	ND	43.1	1.5
M364/3/94	93.9	2.4	7.8	1.4	ND	39.6	0.9
M161/4/94	116	ND	8	ND	ND	29.6	0.5
M290/6/94	113	14	6	ND	ND	15	0.3
M210/7/94	107	ND	9	ND	ND	32.5	0.7
M77/7/94	123	ND	1.9	3.2	ND	45.2	1.2
M290/9/94	N/A	N/A	N/A	N/A	N/A	N/A	N/A
M311/9/94	N/A	N/A	N/A	N/A	N/A	N/A	N/A
M211/10/94	119	ND	9.1	ND	ND	33.5	0.5
M340/10/94	N/A	N/A	N/A	N/A	N/A	N/A	N/A
M43/11/94	88.6	ND	7.9	ND	ND	42.7	1.1
M180/11/94	N/A	N/A	N/A	N/A	N/A	N/A	N/A
M219/11/94	64.7	ND	19.8	ND	ND	17	0.6
M4/12/94	136	ND	4.5	ND	ND	55.2	0.3
M139/12/94	96	ND	5.6	1.7	ND	36.3	0.1
M152/11/94	129	ND	11.2	ND	ND	25.8	0.4
M90/12/95	98	ND	7.4	1.1	ND	19.6	ND
M44/1/95	174	ND	11.9	ND	2.3	60.4	1.5
M110/2/95	121	ND	8.6	ND	ND	68.2	0.6
M153/3/95	711	ND	31.9	ND	ND	90.6	1.3
M157/5/95	89.5	ND	9.5	ND	ND	33.7	0.3
M89/8/95	77.1	ND	12.5	ND	ND	27.8	0.4
M90/8/95	84.6	ND	9.7	ND	ND	35.4	0.1
M71/10/95	119	ND	19.3	ND	ND	41.1	0.8
M70/11/95	86.1	ND	3.8	ND	ND	46.9	0.3
M81/11/95	95.8	ND	17.4	ND	ND	35.6	0.3
M82/11/95	135	ND	15.9	ND	ND	73.6	0.3
M153/11/95	91.9	ND	13	ND	ND	60.3	0.9
M190/1/96	94.4	ND	0.2	ND	ND	24.6	0.1
M191/1/96	85.6	ND	2.9	ND	ND	30.4	0.1
M219/1/96	104	ND	11.4	ND	ND	45.6	1.1
M71/2/96	92.4	ND	1.3	ND	ND	31.1	0.1
Mean	125.5	0.57	9.6	0.3	0.08	40.9	0.59
SD	114.7	2.6	6.6	0.74	0.43	17.2	0.44
Range	64.7-711	0-14	0.2-31.9	0-3.2	0-2.3	15-90.6	0-1.5
Number	29	29	29	29	29	29	29

ND = Nil Detected
N/A = Not analysed

NOTES AND ABBREVIATIONS

1. The term 'Halogenated Hydrocarbons' is used in the report to embrace all the organochlorine pesticides, polychlorinated or polybrominated biphenyls, furans, dioxins, chlorinated naphthalenes and allied compounds.

2. DDT: 1,1,1 - Trichloro-2-2 di (4-chlorophenyl) ethane.

3. γHCH: Gamma Hexachlorohexane.

4. Aroclor 1254 and Aroclor 1260. Commercial PCBs produced by Monsanto Chemical Company and containing 54 % and 60 % chlorine respectively.

5. IUPACS: International Union for Pure and Applied Chemistry.

6. Zn: Zinc, As: Arsenic, Hg: Mercury, Pb: Lead, Cr: Chromium, Cu: Copper, Cd: Cadmium.

7. ITE: Institute of Terrestrial Ecology.

8. CDV: Canine Distemper Virus.

9. BFS: Buffered Formol Saline.

DENTAL AGE OF OTTERS SUBMITTED UP TO DECEMBER 1993

Year	Ref.	Males			Ref.	Females	
		Age	BW			Age	BW
1990	M125/6/90	2	7.5		M455/2/90	2	9.0
					M535/2/90	1	5.0
					M238/12/90	<1	4.5
1991	M646/1/91	4	7.5		17-11-91	4	-
	23-3-91	<1	4.0				
1992	4-4-92	2	8.6		M188/3/92	2	6.7
	M193/10/92	2	5.5		28-4-92	<1	5.5
	M177/11/92	1	9.0		12-6-92	4	8.0
	M91/12/92	1	8.5		M192/10/92	1	5.5
					M216/11/92	4	6.0
					M36/12/92	4	5.75
1993	M30/3/93	2	4.0		M/34/2/93	1	6.0
	M115/4/93	2	9.5		M225/3/93	1	5.0
	M224/3/93	2	7.0		M206/12/93	2	5.75
	M135/7/93	1	8.5				
	M31/9/93	3	6.5				
	M131/10/93	1	8.5				
	M154/12/93	3	8.5				
	M281/12/93	3	8.0				
	MEAN	2			MEAN	2	