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VETERINARY STUDIES ON OILED BIRDS FOLLOWING THE ERIKA WRECK: IMPROVEMENT OF KNOWLEDGE Mrs Sophie Le Dréan Quénech'du¹

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ABSTRACT

After the wreck of the tanker Erika off south Finistère in December 1999, all the care centres of the Atlantic coast received several thousand of oiled seabirds. From the first days of the oil spill, TotalFinaElf contacted the National Veterinary School of Nantes (ENVN), in order to manage studies on birds on care; the aim was to improve the veterinary knowledge on these birds and to evaluate the problems facing with the birds in such extended pollution. The management of the care must keep in account the primary risk linked to the oil but also the secondary risk linked to detention conditions. The authors studied the primary risk, which are the impact on feather and the toxic impact and the secondary risks, which are traumatic and infectious. Concerning the direct risk, the main impact was on the feathers: the results showed that the oil has an impact on microscopic characteristics of feathers and that, the longer the stay in oil is, the more damaged the feathers are. However, toxic target organs and tissues did not show modifications in relation with toxic phenomena problems. In the same way, the vanadium dosage (carried out by spectrometry of atomic absorption), trace element present in oil, did not show high values. Concerning the indirect risk, the main traumatic risks are lesions of legs, feet and breast: the authors describe a treatment. The management of infectious risk (mainly due to *Salmonella* and *Aspergillus fumigatus*) allows us to adapt better the treatment and then to protect in the same time the seabirds and people in the care Centre and the domestic animals around de the centre.



INTRODUCTION

After the wreck of the Erika tanker, the 12th of December 1999, the first oiled bird was found on a beach in Finistère the 14th of December. Then the birds would crowd in thousands on the shore. Facing to this rush, the existing care centres (Allouville Bellefosse (Seine Maritime), 1'Ile Grande (Cote d'Armor), Nantes (Loire Atlantique) and Lorient (Morbihan)) were quickly snowed under and new structures were opened in order to receive and to try to take care of these birds. From the first days of the oil spill, TotalFinaElf contacted the National Veterinary School of Nantes (ENVN), in order to manage studies on birds on care; the aim was to improve the veterinary knowledge on these birds and to evaluate the problems facing with the birds in such extended pollution.

The management of oiled birds must take into account not only the risks directly linked to the oil, but also the secondary risks linked to the detention conditions of these wild seabirds (Figure 1). Effectively, the wild animals received in care centres may suffer from secondary aggressions in relation with this status. Firstly they are placed in an unusual environment, that exposes them to traumatic risks, and secondly, they are stressed and confined, that expose them to infectious risks; and these expositions may be independent of the cause of their entrance in the centre. The staff who looks after the birds have to manage these two risks.

Moreover, in the case of the Erika oil spill, the affected birds belonged to poorly studied species, from the medical point of view, because they usually live far from the shore. The caring people were also confronted with the problem of the "normality" definition for numerous biological parameters usually used to evaluate the animal health.

In this paper, we are presenting the studies carried out in order to try to address these questions. This work was conducted with the help of National Union of Care Centres (UNCS), of several care centres, eight of them being included in this study: the centres of Allouville Bellefosse, Ile Grande, Trégunc (Finistère), Saint Vio (Finistère), Lorient, Nantes, Noirmoutier (Vendée) and La Rochelle (Charente maritime). The analyses were conducted in 10 specialized laboratories.

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I- MATERIALS AND METHODS

The analyses were carried out in 10 specialized laboratories: departmental veterinary laboratories (Cote d'Armor, Ille et Vilaine, Vendée, Loire Atlantique), the Compagnon Animal Laboratoire (CAL) from Troyes, the Agence Française de Sécurité Sanitaire des Aliments (AFSSA) from Maisons-Alfort, and laboratories of ENVN (comparative anatomy, pathological anatomy, pharmacy – toxicology, anti poison centre for animals).

1. Study of direct risks

2.1. Analysis of parameters influencing the feathers permeability

We studied the feathers and the uropygial glands.

A total of 85 uropygial glands from 10 birds species were collected, fixed in formaldehyde and analysed with classical histological techniques (hemalun – eosine-safran staining).

The feathers study was conducted with two observation types, but in each type, the feathers were watched one by one under an optic microscope in order to quantify some structure and abrasion characteristics:

-Feathers from birds dead during the care were collected according to a collecting grid: 16 feathers by individual, with 10 individuals by species (Common Murre and Black Scoter),

-Plots with 10 feathers from non oiled birds (coming from the north of France for Common Murre (J.P. Jacques, pers. comm.) and from the Netherlands for Eider Ducks (K. Camphuysen, pers. comm.) were examined then oiled (3 durations of stay in n°2 fuel), washed (3 shampoo with 2 durations of action) and then examined together: a total of 180 feathers from Murre and 180 feathers from Eider Ducks were watched before and after

being oiled.

Technical lessons learnt from the Erika incident and other oil spills - Brest, 13-16 march 2002



2.2. Analysis of target organs and tissues

The organs and tissues studied were:

- Blood: 199 blood samples were collected from 3 birds species (Common Murre, Black Scoter, Eider Ducks) in 6 care centres (Table I). The blood cells were counted and 2 biochemical parameters were estimated (the enzyme AST/SGOT and uric acid, these two parameters being indicators of liver and kidney functions).

- Gonads: 55 gonads were collected from 8 birds species coming from 3 care centres. They were analysed by histology.

- Livers and kidneys: these organs were used in one hand for histological analysis (114 livers from 6 birds species of 3 care centres and 16 kidneys from 2 birds species of 4 care centres). On the other hand, vanadium was quantified (122 livers and 44 kidneys from 3 birds species of 3 care centres) because this trace element, present in the Erika fuel, may be considered as a fuel tracer and because of its own toxicity. The quantitative measurement was made by atomic absorption spectrometry (AAS) (as described by Sroop *et al.*, 1982) with a preliminary validation period because it was never done on wild birds livers.

3. Studies of indirect risks

3.1. Traumatic risks

The pododermatitis, studied for Common Murre and sea ducks, were treated classically. We proposed a bandage (Le Dréan-Quénec'hdu and coll. on press), which allows to keep the mobility and to protect the wound. The sternum scraps, studied for seaducks, were sorted into type 1 (benign lesions, with no need of particular care), type 2 (more important scraps, with need of local care) and type 3 (more or less extended wounds with need of chirurgical care). We then made local care after eventually chirurgical dress, according to the lesion extension. A protection crossed dressing (Risi, Ferlaux and coll., 2001) was put on a perfusion tube winding the wound: it allows protecting the wound and in the same time displacing the pressure strengths that cause this winding. Moreover, the birds were put on a water mattress to prevent recurrences.



3.2. Infectious risk

A better evaluation and knowledge of the infectious risk (which pathogenic agents for which birds species) allows an appropriate treatment of the birds and a better protection of the birds "environment", i.e. the people dealing with the birds and the other animals. On the other hand, the survey of pathogenic agents of wild animals in general, and of migratory birds in particular, (because of the long way they flight) allows to contribute to the networks of epizooties survey: effectively, some pathogenic agents of wild animals may provoke epizooties of domestic animals or epidemic (example of influenza).

We mainly looked for pathogenic agents from domestic birds:

- Virus: type 1 paramyxovirus (PMV1) from which one type may be very pathogenic for domestic and wild birds, and influenza virus that causes avian grippes.

- Bacteria: most of the classically isolated bacteria were looked for in laboratories (*Salmonella, Staphylococcus, Pasteurella, Mycoplasma* ...) as well as *Chlamydia psittaci*, which is pathogenic for human and that required a specific analysis.

- Fungus: it is mainly Aspergillus fumigatus, but also Candida albicans.

- Unicellular parasites of intestine.

These researches were made (Table I)

- On blood samples by serology; we searched of the following infections: PMV1, Avian Influenza Virus, *Mycoplasma (Mycoplasma gallisepticum, M. synoviae* and *M. meleagridis*) and *Chlamydia psittaci*.

- On organs collected on dead birds: we search directly the pathogenic agent. The birds collected were mainly Common Murre. The organs were collected immediately after the death and put into dry and sterile pots. Bacteria and fungus were searched on lungs. *Chlamydia psittaci* were searched on livers. When bacteria were isolated, we were then able to test their sensitivity to antibiotics. In the same time, histological analyses were also carried out on livers, lungs, encephalon.

- On environment: we used drag-swab and dropping samples. On drag-swab, only Salmonella was searched.



II- RESULTS AND DISCUSSION

1. Direct risks

1.1. Parameters influencing the feathers permeability

The main lesion found on uropygial glands was a more or less important dilatation, on central tubes. However, it was impossible to conclude on a pathogenic characteristic of this lesion, without reference on this subject.

The feathers from oiled birds were significantly more worn and unstructured than the cleaned feathers, especially for Black Scoter. Moreover, concerning the experimentation on "normal" feathers, several results had to be quoted:

- The oil has an impact on the microscopic characteristics of the feathers

- The longer the stay in the oil is, the more worn the feathers are. Moreover, the oiled and washed feathers from the Eiders ducks, were significantly more worn, even after a single day in the oil, than the not oiled feathers.

- There is not impact of the type of shampoo, neither of the duration of the shampoo action on the abrasion or structure of the feathers.

Very short stay in n°2 fuel was sufficient to ruin in an important way the feather and then prevent them to become proof under a short term: this was particularly true for Eider Ducks. However, the experimentation conditions were not identical to the normal conditions in two ways: all the feathers of a bird are not necessarily oiled and especially not as oiled as the feather in the experimentation conditions. Despite this fact, it would be interesting to examine the feather under microscope in order to evaluate the degree of abrasion and the chances the bird have to become proof again.

1.2. Analysis of target organs and tissues

The quantification of vanadium had allowed showing that there were species differences. We did not find high values but the lack of reference values did not allow us to conclude to the "abnormal" characteristic of



theses values. However, they were globally low, agreeing with an absence of toxic resorption in the organism. But we need to compare them with reference values. It will also be interesting to follow these values during a long period, in relation with the vanadium quantification in the sea birds prey. Effectively, in our study, we were in an acute poisoning context and it would be interesting to follow the chronic exposure to the oil, *via* the food chain.

The histological analyses did not show lesions of toxic origin: the observed lesions were more probably from infectious origin.

Concerning blood analyses (Table II), the determined values, especially for red cells, were globally lower than the values found in the literature (Newman and Zinlkl, 1998). However, without clinical signs suggesting anaemia, and on the other hand without reference values on a sufficient number of birds from the same species, we considered these values as normal.

2. Indirect risks

2.1. Traumatic risks

Amongst the 58 birds with sternum lesions, 41 were treated: 24 for type 2 lesions, 17 for type 3 lesions and 17 were not treated. The treated birds had a significantly higher recovery levels than the non treated birds (41.5% vs 5.9%, $\chi^2 = 7.78$, p<0.01). In the case of the treated scoters, the recovery level was not significantly different of the recovery level from the one of the whole population examined (41.5% vs 51.9%, $\chi^2 = 1.53$, p>0.05). Moreover, the treated birds showed a good healing. They also had gained weight during their stay in the sick bay (Risi and Ferlaux, and coll. 2001).

The treatment results for the pododermatitis and the sternum scraps were very encouraging and allowed us to recommend it: the sternum wound could no longer be considered as a euthanasia criterion.

2.2. Infectious risk

There were positive serologies with PMV1 for the 3 species tested, in agreement with the other studies described in literature (Graves, 1996). Concerning Influenza virus, only the eiders ducks were found positive,

Technical lessons learnt from the Erika incident and other oil spills - Brest, 13-16 march 2002



ducks being known to be sensitive to this virus (Graves, 1992). On the contrary, the prevalence of *Chlamydia psittaci* was low, according to the direct isolation.

Concerning the direct isolation, numerous pathogenic agents were identified on birds, from which 5 strains of *Salmonella* (Table III). These strains are classically found in domestic birds. We emphasised the clear predominance of *Aspergillus fumigatus*, an outbreak germ, such as in numerous other care centres (Mikaelian and coll., 1997). This fungus is able to grow on bacterial lesions, particularly on respiratory tract, theses lesions being due for example to *Mycoplasma*, especially in the case of immuno-depressive birds. The isolation of *Mycoplasma gallisepticum* (fragile bacteria) confirmed this hypothesis, this pattern being usual in poultry farms.

There were differences between the species of birds: the Murre showed higher prevalence than sea ducks, for the whole agent, in relation with a higher prevalence of lung lesions.

In the care centres, 6 *Salmonella* serotypes were isolated (Table IV) with a predominance of *S*. Typhimurium and *S*. Hadar. *S*. Enteritidis, usually incriminated in collective food poisoning, was also present. The rooms showed a relatively high load of contamination (particularly due to the faeces), including the "clean" rooms. This contamination showed the importance of the prevention and training of the people working in the care centres. Actually, if it seems obvious to be protected during the manipulation of diseased or dead birds, it is less obvious during the maintenance works of the box: now, *Salmonella* are also present and highly resistant in environment (30 days to 1 year in the soils, without light, and up to 18 month in litter at 11°C for *S*. Typhimurium; Amand and coll., 2000).

On histological analyses, 17% of the Murre livers (N=100) and 12.5% of the scoter livers (N=8) did not show significant lesions. The main lesions found were lesions of multifocal necrotic hepatitis, (23% of the murre livers, 12.5% of the scoter livers), with a bacterial origin, and hepatic aspergillosis, according to the isolations. There were also lesions of hepatic hemosiderose, due to the addition of iron in the food of the birds (25% of the Murre livers showed a more or less important hemosiderose).

Technical lessons learnt from the Erika incident and other oil spills - Brest, 13-16 march 2002



CONCLUSION

These studies allowed:

- to suggesting improvement in the care protocol, especially concerning the management of the traumatic risk but also of the infectious risk,

- to evaluate the different types of risks for the birds dealing with this type of fuel: the impact on the feather seems to be very important and the microscopic observations allowed to estimate the range of the feather lesions. The secondary infections were a major cause of mortality of the oiled birds, and are more frequent and fatal as the stay increases

- to improve the knowledge on the birds from a medical point of view. Blood, histological, toxicological and anatomic analyses allowed to begin the constitution of a biomedical data bank on these species.

However, we also lack data to interpret the whole results. Moreover, the evaluation of the risk according to the type of oil and to species affected would allow a better organisation of the birds management after acute or chronic overflows and a decrease in the duration of the stay in the care centres, *sine qua none* condition for successful treatments.

BIOGRAPHY

Sophie Le Dréan-Quénec'hdu, veterinary doctor, graduated from the National Veterinary School of Nantes in 1994. She continued her studies with a post graduate thesis about animal ecology in the University of Rennes I. This thesis about Water birds, defended in 1999, resulted in numerous publications in scientific press, nationally and internationally, and also communications in international congresses. In parallel, Sophie Le Dréan-Quénec'hdu managed different ornithological studies for public and private organisms and coordinated the students in the triage center of the Veterinary School in Nantes. She set up as a veterinary consultant in breeding and wild life environment since March 1999.

Technical lessons learnt from the Erika incident and other oil spills - Brest, 13-16 march 2002



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LISTE DES TABLEAUX ET FIGURES

Table I: number of sampling made in the care centres

Table II: mean number of blood cells and biochemical values of sea birds in the care centres (mean, standard deviation SD): parameters of red lines (RBC: red blood cells, HGB = hemoglobin in g/dl, HCT = hematocrite = total volume of red cells / total volume of blood, MCV = mean cells volume = ht/red cells numbers), blood



platelets, parameters of white lines (WBC: white blood cells, from which HTC %: heterophile cells in percent, $L\Phi\%$: lymphocytes in percent, MONO% : monocytes in percent) and biochemical (UA: uric acid, AST).

Table III: isolated agents on birds

Table IV: prevalence of Salmonella in the different samples collected in the care centres

Figure 1: Direct and indirect effects of oil on seabirds (from Schiber, 1981). The framed parameters were studied.



	ENV Nantes	Lorient	St Vio-Trégunc	Ile Grande	CHENE	Noirmoutiers	La Rochelle
Serology	123	20	0	20	5	6	7
Lungs							
(bacteriology)	70	25	22	0	6	2	7
Lungs							
(histology)	25	7	0	0	0	0	0
Livers							
(Chlamydia)	33	0	0	0	0	0	0
Livers							
(histology)	80	17	17	0	0	0	0
Kidneys							
(histology)	6	7	1	0	0	0	2
Encephalon							
(histology)	11	0	0	0	0	0	0
Drag-swab	16	12	8	6	7	0	13
Droppings	4	1	0	3	0	0	4

Agent	prevalence
Aspergillus fumigatus	49.2%
Salmonella Typhimurium	13.6 %
Salmonella Enteritidis	2.3%
Salmonella Hadar	15.2%
Salmonella Bokley	3.8%
Salmonella Derby	3.0 %
Staphylococcus aureus	4.6 %
Mycoplasma gallisepticum	2.3 %
Escherichia coli	11.4%
Pseudomonas aeruginosa	12.1 %
Klebsiella pneumoniae	1.5%
Chlamydia psittacci	0 %

	Birds	Drag-swab	Droppings
Salmonella Typhimurium	13.64	19.35	16.67
S. Enteritidis	2.27	0.00	8.33
S. Hadar	15.15	19.35	0.00
S. Derby	3.03	4.84	0.00
S. Newport	0.00	9.68	33.33
S. Blokley	3.79	0.00	0.00

		RBC	HGB	НСТ	MCV	PLATELETS	WBC	НТС%	LΦ%	MONO%	UA	AST
Murre	Mean	2126991.2	10.8	31.7	144.8	77178.9	6779.91	75.8	23.7	0.5	81.0	108.4
n=113	SD	1454852.0	3.2	31.6	12.1	58721.9	3875.64	12.8	12.9	1.7	43.8	64.9
Scoter	Mean	2504102.6	13.8	36.8	146.9	56933.3	10384.4	76.5	21.8	1.7	113.1	25.5
n=41	SD	314723.9	2.2	5.6	11.7	26169.1	3648.12	8.7	8.3	2.9	50.4	36.6
Eiders	Mean	1430250.0	13.4	23.2	165.2	45075.0	8856.6	78.1	19.8	1.5	132.3	141.5
n=45	SD	579409.5	2.8	8.2	11.3	33622.6	4345.5	11.3	10.9	3.8	19.6	26.2

Figure 1

