Appendix 5

Literature review on the use of bio-monitoring programmes

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5.1 Bio-monitoring tools

Studies of mercury in wild birds can be divided into three categories: eggs and reproduction, liver and other organs, and feathers. In the 1960's great concern over reproductive failures of many avian species prompted studies of various contaminant levels in eggs, particularly in eggs that failed to hatch. Many of these studies were "positive", showing that unhatched eggs had higher contaminant levels (particularly chlorinated hydrocarbons), than hatched eggs and in many cases the contaminants levels were linked to eggshell thinning. Although mercury also causes eggshell thinning, most field studies did not analyse for mercury, and those that did often did not find significant increases in mercury. Studies of mercury in organs such as the liver were often made on moribund or dead birds to ascertain a cause of death. Widespread surveys of mercury in organs required the killing of large numbers of individuals. This was both inconvenient and undesirable. Hence many studies relied on measuring mercury in feathers.

Bird feathers offer several advantages as bio-indicators of metal exposure, as well as being non-invasive. The elimination of methylmercury (MeHg) via moulted feathers is a well-known process (Monteiro & Furness, 2001). The direct relationship between the concentration of MeHg in feathers and the body burden of MeHg during the time over which the feather was grown makes feathers an effective and commonly used indicator of overall MeHg body burden (Furness *et al*. 1986). A substantial part of the body burden of MeHg is found in feathers (Braune and Gaskin 1987), which, like human hair, have repeatedly proven their value as a means for monitoring mercury concentrations in birds (Malik and Zeb, 2009; Zamani-Ahmadmahmoodi *et al*. 2010). Moreover, feathers from newly born chicks indicate local contamination, derived mostly from food collected locally by their parents during the short period of egg formation and chick development (Boncompagni *et al*., 2003; Muralidharan *et al*., 2004). Waterbirds that breed in colonies provide additional advantages as bio-indicators of pollution: easy sampling (Burger and Gochfeld, 2000a, b); a limited foraging range around their colony site, thus allowing inference about the source of contaminants (Burger *et al*., 2004); and dependence on specific habitat and prey resources (Fasola *et al*., 1998).

Interpreting concentrations and origins of contaminants in wild birds presents unique challenges. In migratory animals, for example, exposure may occur at breeding, migratory, or overwintering sites. Persistent organic pollutants (POPs) and mercury accumulate in body tissues and are readily transported by migratory animals from one region to another. In birds, contaminants stored within endogenous energy reserves (e.g., lipids) can be mobilised to eggs, and therefore, eggs have been used for monitoring contaminants, especially in colonial water birds. Feather type (e.g. flight versus body) and species' moult patterns also affect the correlation between feather and body MeHg (Monteiro & Furness 2001; Furness *et al.* 1986; Thompson *et al.* 1998). The sex of a bird can also, in principle, affect exposure and accumulation of mercury. One conventional explanation for sex differences in mercury burden suggests female birds should have lower concentrations than conspecific males, because breeding females can depurate MeHg to their eggs. But meta-analysis across several species has shown that sex differences in body burden of mercury among birds are not consistent (Robinson *et al.* 2012). Therefore, gender is of interpretive concern, particularly when evaluating mercury exposure for species exhibiting sexual dimorphism and niche partitioning (Evers *et al*. 2005).

Walsh (2017) & ICES (2003) published guidelines on using seabirds to monitor marine pollution. Criteria are outlined for selecting species as good candidates for biomonitoring, in areas where there is concern about possible contamination of marine food chains by mercury or organochlorines. These

criteria are a) accumulation of the contaminant to high concentrations; b) resistance to toxic effects due to the contaminant (unless these are what is being monitored); c) known, or preferably no, migratory habits; d) a foraging range consistent with the spatial scale over which the contaminant is to be monitored; e) a large population size with known breeding biology and ecology, and with large numbers of colonies throughout the area where contaminant monitoring is required; f) be easy to collect without major disturbance to the breeding colony, and have easily identifiable life-stages if a particular category is to be sampled; g) have known physiology; h) have a narrowly defined and consistent diet; i) feed predominantly or exclusively on prey in the food web under investigation.

Several studies have followed these criteria either implicitly or explicitly (Gilbertson *et al.* (1987); Becker (1989); Becker et al. (1991); Becker *et al.* (1998)). However, the majority of published studies reporting contaminant concentrations in seabirds, or even reporting spatial or temporal patterns of contaminant levels in seabirds, have used varying collection methods are therefore difficult to accurately compare (ICES 2003).

Thompson *et al.* (1998) demonstrated the utility of feather mercury in documenting the temporal increase in mercury in the marine environment. Spalding *et al*. (2000a), Braune and Gaskin (1987) and Thompson and Furness (1989) found that the mercury levels in the feathers of provided a good indication of the mercury dose.

While feathers have been extensively used in bio-monitoring studies for heavy metal pollution, until very recently few studies had been performed on the use of feathers for monitoring of organic pollutants. In a recent review Garcia-Fernandez *et al*. (2013) examined the use of feathers for monitoring polyhalogenated compounds (PHCs). They discussed how some authors had found strong and significant correlations between the concentrations of PHCs in feathers and internal tissues, providing positive expectations for their future use in the field of ecotoxicology. However, changes in diet, time elapsed between the previous moult period and sampling, sample size, and/or external contamination have been suggested as possible causes to explain the lack of correlations reported in some studies. They concluded further studies with newly grown feathers and blood samples would be required in order to clarify this issue.

Jaspers *et al*. (2011) investigated the variation in concentrations and profiles of various classes of organohalogenated compounds (OHCs) in different feather types, muscle tissue and preen oil from 15 White-tailed Eagle (*Haliaeetus albicilla*) carcasses from Greenland. The influence of moult patterns and potential external contamination onto the feather surface was examined. Concentrations of sum polychlorinated biphenyls (PCBs) in feathers from White Tailed Eagles ranged from 2.3 ng/g in a primary wing feather to 4200 ng/g in body feathers. Using body feathers, they found almost 50 different OHCs could be quantified and median concentrations in body feathers were tenfold higher than concentrations in tail feathers or primary wing feathers. Furthermore, the effects of confounding variables such as feather size, moult and age were also minimised using body feathers.

While the use of feathers is still relatively new, several non-destructive bio-monitoring strategies have been developed for monitoring OHCs (Furness 1993, Burger 1993, Bustnes *et al*. 2005, Van den Steen *et al*. 2006). Eggs have been extensively used in bio-monitoring studies for organic pollutants (Becker *et al*. 1992, Lindberg *et al*. 2004, Van den Steen *et al*. 2006, Dauwe *et al*. 2006). One egg has been shown to reflect the contamination of the whole clutch (Van den Steen *et al*. 2006) and the collection of one egg is only expected to have a minor effect on the population level in species producing large clutches. However, the use of eggs does have some drawbacks, since they can only be collected during the breeding season from adult females. Therefore, levels in eggs do not represent concentrations in the general population during the year. Moreover, sampling of viable eggs may have an impact on the

population, while concentrations in unhatched eggs may be susceptible to microbiological degradation (Herzke *et al*. 2002). Other non-destructive bio-monitoring strategies are the collection of plasma or blood (Henriksen *et al*. 1998, Verreault *et al*. 2004) and the sampling of preen oil (Yamashita *et al*. 2007) that can be collected of both adults and nestlings. However, both techniques involve some sampling expertise and require the capturing of the birds. Furthermore, the amount of blood that can be collected is limited (species-dependent) and thus analytical problems may be of concern. In addition, levels in the blood only present a snapshot and are subject to variations in the diet, season and condition of the bird.

While feathers have been commonly used to measure heavy metal levels in birds, a number of factors have been identified which can cause variations in the levels of deposition including species, age, sex, life history. Becker *et al*. (1994) tested eggs, feathers (down, body feathers from side/shoulder and back) and some dead chicks (liver) from broods Herring Gull, Black-headed gull and Common Terns to examine inter-sibling differences in mercury contamination and elimination into the growing feathers. The mercury contamination in eggs, feathers, and liver of the terns was about four times that of the gulls; Black-headed Gulls had the lowest mercury concentrations. Body feathers, which grow when the chicks became older, had lower mercury levels than down feathers in the more contaminated species (11% lower in Herring Gulls, 49% in Common Terns), indicating the advancing decontamination of the body by the plumage development. The elimination of mercury was greater in chicks with higher mercury levels. Furthermore, down of the first hatched Herring Gull and Common Tern chick contained more mercury than down of the siblings hatched later, because of its higher burden derived from the first laid egg.

Tavares *et al*. (2013) examined levels of mercury in Wandering Albatrosses (Diomedea exulans) Mercury concentrations in tissues of the wandering albatross are greater than in any other vertebrate, including closely related species. In order to explore the alternative explanations for this pattern, total mercury concentrations in feathers, plasma and blood cells of wandering albatrosses of known age, sex and breeding status were sampled. Mercury concentrations were low in feathers and blood components of chicks, and higher in the feathers of young pre-breeders than in feathers or blood of older pre-breeders and breeding adults. There was no effect of sex on mercury concentrations in the feathers of pre-breeders or breeding adults, whereas levels were significantly higher in blood cells of breeding females than males. The high feather mercury concentrations of young pre-breeders compared with older birds suggest an increase in moult frequency as birds approach maturity.

As discussed organic contaminants and mercury accumulate in body tissues and are readily absorbed and therefore will be transported by migratory animals from one region to another. Therefore interpreting concentrations and origins of contaminants in tissues of migratory animals presents unique challenges in that exposure may occur at breeding, migratory, or overwintering sites (Hargreaves *et al*. 2010, Dietz *et al*. 2009). Feathers may be an effective mode of mercury bio-transport and deposition, as they are often moulted at sites far removed from the initial source of mercury exposure. For example, long-lived, piscivorous seabirds may transport marine sourced mercury to inland colonies, where concentrations in aquatic sediments from feathers, eggs, carcasses, and faeces may reach toxic level (Blais *et al*. 2007, Blais *et al*. 2005) Alternatively, species such as the Common Loon, Common tern and Roseate Tern (Sterna dougallii) accumulate more mercury in summer-grown feathers and may therefore transport mercury from northern breeding sites to the wintering grounds (Burgess *et al*. 2005, Burger *et al*. 1992, Nisbet *et al*. 1992).

5.2 Conclusions

Bio-monitoring is an important method for examining the impact of pollutants in the wild. While studies on heavy metals, in particular mercury, are well established, non-invasive bio-monitoring for dioxins is still relatively new. Bird feathers and eggs have been successfully used to monitor mercury levels in birds for many years. In the case of dioxins, bird livers and muscle are commonly used, although several studies have successfully used eggs to monitor contamination levels. However, other factors, including age, sex, time of year, migratory status and, in the case of eggs, laying sequence, will affect the levels of mercury or dioxins detected. Therefore using such techniques to monitor mercury patterns in a single area, or from a single source (in this case the proposed Indaver facility) would be difficult.

It is noted that long-term bio-monitoring programmes of this nature are generally not carried out by private companies. The predicted levels of dioxins and mercury generated by the facility will be low and no significant impact on piscivorous bird species is predicted. Given the difficulties inherent in determining the source of dioxins and mercury in piscivorous birds and the difficulties in ascribing levels to any particular source, the use of cows milk is considered an adequate means of determining if problematic levels of dioxins are entering the food chain via atmospheric deposition.

5.3 References

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