Science of the Total Environment 610–611 (2018) 462–468

Contents lists available at ScienceDirect

Science of the Total Environment

journal homepage: www.elsevier.com/locate/scitotenv

Variation in organochlorine and mercury levels in first and replacement eggs of a single-egg clutch breeder, the thick-billed murre, at a breeding colony in the Canadian Arctic

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HIGHLIGHTS

- Contaminants in first-laid and replacement eggs of thick-billed murres were compared.
- Total mercury levels were lower in the replacement eggs.
- Organochlorines levels were generally higher in the replacement eggs.
- Organochlorines and PCB profiles varied between first-laid and replacement eggs.

GRAPHICAL ABSTRACT

ARTICLE INFO

Article history:
Received 11 May 2017
Received in revised form 8 August 2017
Accepted 9 August 2017
Available online 14 September 2017

Editor: Adrian Covaci

Keywords:
Organochlorines
Mercury
Seabirds
Eggs
Replacement eggs

ABSTRACT

Contaminant concentrations may vary among sequentially-laid eggs in multi-egg clutches, and this variation has implications for the interpretation of contaminant concentrations in monitoring programs. The thick-billed murre (*Uria lomvia*) is a key species for monitoring contaminants in the Canadian Arctic and lays only a single egg per year. Therefore, the potential issue of intra-clutch variation in contaminant concentrations is avoided. However, if the egg is removed or lost early in the incubation stage, the adult female murre will relay. In this study, we examined contaminant concentrations and patterns in first-laid and replacement eggs of thick-billed murres breeding in northern Hudson Bay in order to determine whether or not these eggs could be sampled interchangeably. Concentrations of the major legacy organochlorines (e.g. PCBs, DDT, chlordanes) were generally higher, and total mercury concentrations lower, in the replacement eggs compared with the first-laid eggs. The organochlorine profile was comprised primarily of ΣDDT and Σ_{70}PCB, and Σ_{70}PCB was comprised primarily of hexa-hepta PCBs in both first-laid and replacement eggs. As both concentrations and organochlorine patterns showed differences between first-laid and replacement eggs, we recommend that randomly selected first-laid eggs of thick-billed murres be consistently sampled for contaminant monitoring in the Canadian Arctic.

1. Introduction

Concentrations of organochlorines and total mercury (Hg) can vary among sequentially-laid avian eggs (Ackerman et al., 2016; Akearok et al., 2010; Becker, 1992; Evers et al., 2003; Kennamer et al., 2005;

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http://dx.doi.org/10.1016/j.scitotenv.2017.08.076
0048-9697/

Mineau, 1982; Morera et al., 1997; Nisbet, 1982; Pastor et al., 1995; Sanpera et al., 2000), although not all studies have found a relationship between contaminant concentrations and laying order (see Custer et al., 1990; Scharenberg and Ebeling, 1998; Van den Steen et al., 2011; Verreault et al., 2006). Where eggs are used as sampling units for contaminants monitoring, and where only one egg is taken from each clutch (to reduce population effects), which egg is sampled from the clutch may have implications for the interpretation of results. An advantage of monitoring species which lay only a single egg per year is that potential intra-clutch variation in contaminant concentrations is avoided. Some seabirds, such as the thick-billed murre (*Uria lomvia*) lay only a single egg per year, but if the egg is removed or lost early in the incubation stage, the adult female will relay (Gaston and Hipfner, 2000), in which case variation in contaminant concentrations between first-laid and replacement eggs may also have relevance for contaminant monitoring programs. Bignert et al. (1995) showed that concentrations of total DDT and PCBs were significantly higher in replacement eggs of guillemots (*Uria aalge*) from the Baltic Sea, suggesting that date of egg laying may need to be considered in monitoring studies of single-egg clutch breeders.

The thick-billed murre (a.k.a. Brünnich's guillemot) is one of the key seabird species monitored for contaminants in the Canadian Arctic (Mallory and Braune, 2012), but whether there is variation in contaminant concentrations between first-laid and replacement eggs in this species is not known. To test whether first-laid and replacement eggs may be interchangeably sampled, we compared contaminant concentrations and patterns between first-laid and replacement eggs of thick-billed murres breeding at a low-Arctic monitoring site at Coats Island in northern Hudson Bay.

2. Materials and methods

2.1. Sample collection

The study was carried out in 2003 at a breeding colony of thick-billed murres (*Uria lomvia*) on Coats Island, northern Hudson Bay, Nunavut (62°98′N, 82°00′W), in the eastern Canadian low Arctic (Fig. S1). Upon arrival at the breeding colony, positions of breeding sites were mapped for an accessible part of the colony. Breeding sites were then monitored daily and eggs removed on the day of egg laying or replacement egg laying. Adult birds were captured prior to each egg removal as part of another study and the identity of the adults confirmed using unique leg band numbers. Additionally, breast feathers of adult birds were marked with a permanent marker during the first capture to facilitate identification of replacement eggs.

First-laid eggs ($n = 13$) of thick-billed murres were collected on 28 June 2003 immediately after they were laid. Replacement eggs ($n = 13$) were laid by the same adult females, on average, 15.5 days later (11–20 July), and second replacement eggs ($n = 4$) were laid, on average 14.5 days after the first replacement eggs were removed (24–27 July). The replacement eggs and second replacement eggs were also collected immediately after they were laid. All eggs were collected by hand.

Eggs were cracked open in the field and the contents of each egg homogenized in a metal bowl using a standard, medium-sized egg beater until the egg contents appeared homogenous. The mixture was then beaten for an additional 2 min to ensure homogeneity. The homogenate from each egg was then transferred into a series of cryovials using a plastic, disposable pippette and immediately frozen in a propane freezer (−20 °C). All instruments were cleaned between individual egg homogenizations using soap and water followed by a methanol rinse. Samples were shipped to the National Wildlife Research Centre (NWRC) in Ottawa in a nitrogen vapour (dry) shipper. Upon arrival at NWRC, the cryovials were transferred into liquid nitrogen storage tanks until retrieved for chemical analyses.

2.2. Chemical analyses

All of the eggs ($n = 30$) were individually analyzed for legacy organochlorines, PCBs, and total mercury (Hg). All chemical residue analyses were carried out at NWRC.

2.2.1. Organochlorine analysis

Egg homogenates were analyzed for legacy organochlorines (OCs) including chlorobenzenes (ΣCBz = 1,2,4,5-tetrachlorobenzene, 1,2,3,4-tetrachlorobenzene, pentachlorobenzene and hexachlorobenzene), hexachlorocyclohexanes (ΣHCH = α-, β- and γ-hexachlorocyclohexane), chlordane-related compounds (ΣCHLOR = oxychlordane, *trans* chlordane, *cis* chlordane, *trans* nonachlor, *cis* nonachlor and heptachlor epoxide), DDT and its metabolites (ΣDDT = *p,p′*-DDE, *p,p′*-DDD and *p,p′*-DDT), octachlorostyrene (OCS), mirex (ΣMirex = photomirex and mirex), dieldrin and PCB congeners (ΣPCB). ΣPCB consisted of 70 congeners identified according to IUPAC numbers (Ballschmiter et al., 1992) and included congener numbers 16/32, 17, 18, 20/33, 22, 28, 31, 42, 44, 47/48, 49, 52, 56/60, 64, 66, 70/76, 74, 85, 87, 90/101, 92, 95, 97, 99, 105, 110, 114, 118, 128, 130, 137, 138, 141, 146, 149, 151, 153, 156, 157, 158, 167, 170/190, 171, 172, 174, 176, 177, 178, 179, 180, 183, 187, 189, 194, 195, 196/203, 200, 201, 202, 206, 207, and 208. Congeners separated by a slash co-eluted during the chromatography process and were therefore reported together.

Samples were analyzed for organochlorines by gas chromatography using a mass selective detector (GC/MSD) according to CWS Method No. MET-CHEM-OC-04 (Won et al., 2001). All solvents (pesticide grade) and sodium sulfate (Na_2SO_4) were obtained from EM Science (Mississauga, ON, Canada), as was the pesticide grade Florisil. Isotopically-labelled ^{13}C internal standards were obtained from Cambridge Isotope Laboratories (Andover, MA, USA). Briefly, samples were ground with anhydrous sodium sulfate, spiked with ^{13}C–labelled internal standards (chlorobenzenes, PCB congeners) and extracted with dichloromethane:hexane (50:50% v/v). Sample clean-up was performed by gel permeation chromatography followed by water-deactivated Florisil chromatography. Chemical analysis was performed using a Hewlett-Packard (HP) 5890 Series II gas chromatograph (GC) equipped with HP-7673A autosampler and coupled with HP-5971A mass selective detector (MSD) run in selected ion monitoring (SIM) mode. The column was a 30-m × 0.25 mm DB-5 fused silica capillary column (0.25-μm film thickness; Chromatographic Specialties, Brockville, ON, Canada). Method blanks and in-house reference materials (HGQA-E4005, DCCO13) were run for quality control. Internal standard recoveries were ≥80%. Residue results were not corrected for internal standard recoveries. The nominal detection limit was 0.1 ng g^{-1} wet weight (ww). Lipids were determined by gravimetric methods.

2.2.2. Mercury analysis

Sample homogenates were freeze-dried, then homogenized again and weighed into nickel combustion boats. Total Hg was analyzed using an Advanced Mercury Analyzer (AMA-254) equipped with an ASS-254 autosampler for solid samples (see EPA Method 7473, 2007 and Salvato and Pirola, 1996). The method employs direct combustion of the sample in an oxygen-rich atmosphere. Analytical accuracy for total Hg was determined by analyzing blank samples with each sample set, as well as three standard reference materials (DOLT-2 and TORT-2 from the Canadian National Research Council and Oyster Tissue 1566b from the National Institute of Standards and Technology). Recoveries of reference materials averaged 98.9% and results were within the certified range of values. Analytical precision was checked by analyzing replicate samples, averaging one replicate sample for every five samples analyzed. Standard deviation for replicate readings averaged 7%. The nominal detection limit was 40 ng g^{-1} dry weight sample.