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High resolution mass spectrometric suspect screening, wide-scope target analysis of emerging contaminants and determination of legacy pollutants in adult black-tailed godwit *Limosa limosa limosa* in the Netherlands – A pilot study

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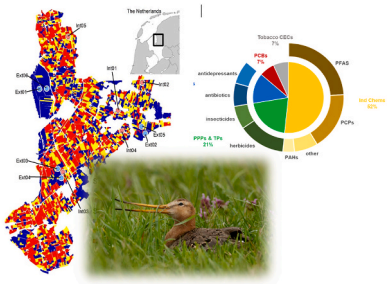
HIGHLIGHTS

- Dutch godwits contain residues of 29 emerging contaminants (ECs).
- Total EC load was higher in birds from intensive than from extensive grasslands.
- 20 additional substances were tentatively identified through suspect screening.
- Substances include industrial chemicals, PPPs and pharmaceuticals.
- Ni, Cd, Pb and Hg, 2 OFPRs, PCDDs, PCDFs and dl-PCBs were all detected.

GRAPHICAL ABSTRACT



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ABSTRACT

The Dutch breeding population of the black-tailed godwit *Limosa limosa limosa* has declined substantially over recent decades; the role of contaminants is unknown. We analysed liver samples from 11 adult birds found dead on their breeding grounds in SW Friesland 2016–2020, six from extensive, herb-rich grasslands, five from intensive grasslands. We carried out LC and GC wide-scope target analysis of more than 2400 substances, LC suspect screening for more than 60,000 substances, target analysis for Cd, Hg, Ni and Pb, organo-phosphate flame retardants (OPFRs), dechlorane plus compounds and selected polybrominated diphenyl ether flame retardants (PBDEs), and bioassay for polybrominated dibenzo-p-dioxins and dibenzofurans (PBDDs/PDBFs) and dioxin-like polychlorinated biphenyls (dl-PCBs). Residues of 29 emerging contaminants (ECs) were determined through wide-scope target analysis. Another 20 were tentatively identified through suspect screening. These

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contaminants include industrial chemicals (personal care products, surfactants, PAHs and others), plant protection products (PPPs) and pharmaceuticals and their transformation products. Total contaminant load detected by wide-scope target analysis ranged from c. 155 to c. 1400 ng g^{-1} and was generally lower in birds from extensive grasslands. Heatmaps suggest that birds from intensive grasslands have a greater mix and higher residue concentrations of PPPs, while birds from extensive grasslands have a greater mix and higher residue concentrations of per- and polyfluoroalkyl substances (PFAS). All four metals and two OPFRs were detected. All tested PBDEs were below the respective LODs. Bioassay revealed presence of PBDDs, PBDFs and dl-PCBs. Further research is required to elucidate potential health risks to godwits and contaminant sources.

1. Introduction

Chemical pollution from agricultural, industrial and urban sources is a recognised threat to wildlife (Badry et al. 2021). Contaminant monitoring in birds can qualify and quantify exposure of bird populations (and of other species in the same biotic communities) to both legacy and emerging contaminants and trigger any necessary remedial measures to safeguard species and ecosystems. Contaminant monitoring in birds can also be used to provide early warning of emerging contaminants (ECs), identify ongoing problems with legacy contaminants, highlight the need for further risk assessment of bio-accumulative substances, and assess the effectiveness of regulations and chemical risk management measures thereunder. By supporting chemicals' regulation and management, contaminant monitoring in birds can help deliver the objectives of chemicals regulations to improve wildlife and human health (Movalli et al. 2008, 2017, 2019).

Over 25 years ago, Kushlan (1997) identified wading birds as potential bioindicators of environmental change including environmental pollution, given their use of man-dominated landscapes and that, as primary carnivores, their population responses can serve as signals of environmental changes at lower trophic levels. Moreover, limitations in their feeding ranges makes them appropriate as local-scale sentinels (Matsinos and Wolff, 2003).

The black-tailed godwit *Limosa* is a philopatric ground-nesting shorebird, found throughout Eurasia in grassland, wetland, and marine intertidal habitats (e.g. Gill et al. 2007). It breeds from Iceland to eastern Russia and winters in Europe, Africa, the Middle East and Australasia. There are four subspecies, *L. l. islandica*, *L. l. limosa*, *L. l. melanuroides* and *L. l. bohai* (Zhu et al. 2021). The Dutch population belongs to the subspecies *L. l. limosa* and is the national bird of The Netherlands. It breeds in agricultural grasslands in western and central Europe to central Asia and Asiatic Russia. The Dutch godwit population has declined by 75% over the last 50 years (Kentie et al. 2016), contributing to classification of the species as a whole as 'Near Threatened' (BirdLife International, 2006, 2017). The Dutch population which numbers <30, 000 breeding pairs (mostly in the province of Friesland) still represents c. 85% of the world population (Kentie et al. 2016) and its status is therefore critical to the survival of this subspecies.

The ongoing decline in The Netherlands is associated with loss of habitat quality accompanying the intensification and industrialization of dairy farming and some loss of habitat to urbanisation. Adult godwit feed predominantly on earthworms (Lumbricidae), leatherjackets (crane fly larvae – *Tipula* sp.) and plant food (e.g. rice kernels), chicks exclusively on Arthropods. In intensified grasslands, devoid of flowers and insects, godwits have difficulty raising chicks (Kentie et al. 2018). Indeed, Kentie et al. (2013) found that chicks hatched on monocultures had lower growth and survival rates than chicks on meadows, which indicates that these chicks suffer a higher risk of starvation and/or predation.

The ecology and demography of the godwit population in relation to changing land use have been studied in Southwest Friesland over the last 20 years (Howison et al. 2019) and this is being augmented by research on the godwits' flyway (under an EU-funded LIFE Integrated Project). Southwest Friesland is dominated by dairy farming with some arable cropping. The area consists of polders, made up of herb-poor, artificially seeded, intensive grasslands (c. 80%) and remnants of botanically rich semi-natural grasslands (c. 20%) (Fig. 1). Godwits prefer herb-rich polders with high groundwater levels and the presence of foot drains (narrow ditches used for drainage in wetlands). In herb-rich areas, highest densities of breeding birds are found on soils of sandy clay loam and sandy clay (Groen et al., 2012).

While the intensification of agricultural practices has resulted in a substantial decline of Europe's farmland bird populations during the past 50 years (Busch et al. 2020; Donald et al. 2006; Emmerson et al. 2016; Badry et al. 2022), the increased use of agricultural chemicals has also been implicated in population declines (e.g. Emmerson et al. 2016). Exposure to agriculturally related chemicals has resulted in population declines in many wildlife species (Shore and Taggart, 2019). In addition to agricultural chemicals (plant protection products), a range of other classes of anthropogenic chemicals are increasingly detected in wildlife, including industrial chemicals, pharmaceuticals, personal care products, biocides, illicit drugs, coffee- and tobacco-related compounds and

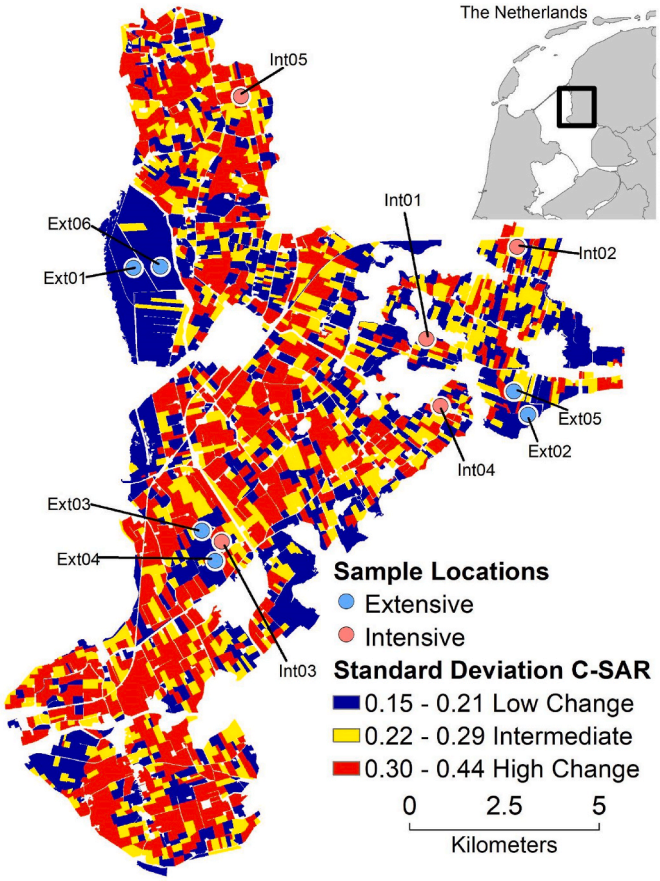

Fig. 1. Temporal stability of vegetation structure (2016), classed as low (blue), intermediate (yellow) & high (red) land-use change in SW Friesland with locations of godwit samples (EXT and INT – see also **Tables SI–1**). (Adapted from: Howison et al. 2018). (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

artificial sweeteners (e.g. Gkotsis et al. 2023). Even low-level exposure to contaminants may, in combination with other stressors, adversely affect the stability of wading bird populations (Rattner et al., 2000). Comprehensive biomonitoring approaches that screen for all classes of chemical and for respective chemical mixtures is therefore appropriate to better understand overall exposure.

The Dutch population of black-tailed godwit winters in West Africa with a small part remaining in southern Europe (Kentie et al. 2017), with staging along the Atlantic coast of France (especially the Bay of Biscay) and Portugal (Lourenço et al. 2010; Verhoeven et al. 2020) as well as the Doñana in Spain. These staging and wintering areas are inland wetlands (water storage basins, marshes, flooded plains, rice paddies). In West Africa rice farming is typically small-scale and traditional, in Spain and Portugal it is more intensive (Howison et al. 2019). In the staging and wintering grounds the adults feed predominantly on rice kernels but also on gastropods, oligochaete worms, dysticid insects and crayfish (Lourenço and Piersma, 2008). Black-tailed godwit is sexually dimorphic, the female being structurally bigger with a greater body mass (Schroeder et al. 2008), likely contributing to variation in contaminant concentrations between the sexes.

Although the decline of the black-tailed godwit is mostly blamed on habitat loss and changes in agricultural practices in the breeding areas (Beintema et al. 1985, Beintema and Müskens, 1987), conservation programmes have had limited success due to the scale of protection and other contributing factors (Kleijn et al. 2001; Kleijn and Van Zuijlen, 2004, Lourenço and Piersma, 2008), which may include contaminants. However, the extent to which contaminants – individually or as mixtures – may or may not contribute to this decline, either by reducing the insect food resources or through direct effects on adults or chicks, is not known. Adult godwits will be exposed to contaminants in breeding, staging and wintering grounds. Chicks are exposed to chemicals on the breeding grounds, and to some extent by maternal transfer to eggs. The godwit is an 'income-breeder' – i.e. breeding follows 'fattening' on spring staging grounds in southern Europe. Contamination of plant food and/or invertebrate prey in the staging grounds may therefore be important in any maternal transfer to eggs and chicks. Any observed hatching failure may be related to such maternal transfer, while effects in later chick development are more likely related to local exposures. However, hatching failure does not seem to be an issue in SW Friesland, while chick development does seem to be an issue (Kentie et al. 2013).

The objective of this pilot study was to investigate, for the first time, the presence of a wide range of legacy and emerging contaminants in liver tissues sampled from the Dutch breeding population of the black-tailed godwit. The study employs wide scope target analysis of 2400+ known ECs, suspect screening of 65,000+ environmentally relevant substances including their (bio)transformation products, as well as selected target analyses and bioassay for legacy contaminants.

2. Materials and methods

2.1. Study area and sampling

We analysed contaminants in livers of 11 adult black-tailed godwit specimens found dead in the period 2016–2020 and collected from SW Friesland, an area dominated by dairy grassland farming. The birds died from collisions with cars or as casualties of predators but were collected fresh. The collection sites were on polders which vary from average low land-use change (botanically rich semi-natural grasslands) ('low change' in Fig. 1) ('EXT' – 6 birds) to average high land-use change (herb-poor, artificially seeded, intensive grasslands) ('high change' in Fig. 1) ('INT' – 5 birds) (**Tables SI–1**).

2.2. Sample preparation

Livers were dissected at a state-of-the-art laboratory at the Royal Netherlands Institute for Sea Research (NIOZ) to minimize risk of cross-contamination. Sample preparation was carried out at the National and Kapodistrian University of Athens, Trace Analysis and Mass Spectrometry Group (TrAMS).

All samples were lyophilised and homogenized before analysis in order to enhance extraction efficiency, improve the precision and achieve lower detection limits. Validated generic sample preparation protocols, designed to retain compounds with a wide range of physicochemical properties, were used to extract both polar and non-polar ECs (Badry et al. 2022; Gkotsis et al. 2022). Detailed information on the lyophilisation and extraction of LC- and GC-amenable compounds can be found in **Supplementary Information, 1. Methods**.

2.3. HMRS wide-scope target analysis and suspect screening

2.3.1. Instrumental analysis

The samples were analysed by ultra-high performance liquid and gas chromatography hyphenated with a high resolution mass analyser (Maxis Impact, Bruker Daltonics, Bremen, Germany). The chromatographic separation was performed on an Acclaim RSLC C18 column (2.1 × 100 mm, 2.2 μm) from Thermo Fisher Scientific (Dreieich, Germany) preceded by a guard column of the same packaging material, ACQUITY UPLC BEH C18 1.7 μm, VanGuard Pre-Column, Waters (Ireland), thermostated at 30 °C for the LC-amenable compounds, whereas GC was operated in splitless injection mode (Restek Split liner w/Glass Frit (4 mm × 6.3 × 78.5)) and a Restek Rxi-5Sil MS column of 30 m (0.25 mm i.d. X 0.25 μm film thickness) was used with Helium as a carrier gas at the constant flow of 1.5 mL min^{-1}.

Detailed information on the instrumental analysis of the RPLC-ESI-QTOF and GC-APCI-QTOF system is provided in Supplementary Material, 1. Methods.

2.3.2. Quality assurance and quality control

Quality assurance and quality control (QA/QC) was applied during sample preparation and instrumental analysis. A mix of internal standards was added into each sample prior to extraction to assure satisfactory recovery of the target compounds and samples spiked with a mix of known ECs were also analysed in each batch of samples. Moreover, procedural blanks (reagent blanks) were prepared to assess any external contamination which might have been brought in during the sample preparation of the extracts and analysis. The detected compounds in blanks were subtracted from the samples, but most analytes were not present in the blanks. A mix of known analytes (RTI calibrant substances) was used to assess the stability of retention time during instrumental analysis (Aalizadeh et al. 2021). A QC sample was running every 10 injections to ensure the good operation and high sensitivity of the instrument.

2.3.3. Data treatment

2.3.3.1. HMRS wide-scope target analysis. Target screening was performed using in-house databases of 2402 contaminants, developed through the analysis of respective reference standards – the LC target list (https://doi.org/10.5281/zenodo.6012778) and the GC target list (https://doi.org/10.5281/zenodo.3753372), available respectively as S21 UATHTARGETS and S65 UATHTARGETSGC in NORMAN Suspect List Exchange (https://www.norman-network.com/nds/SLE/). The data treatment was performed using TASQ Client 2.1 and DataAnalysis 5.1 (Bruker Daltonics, Bremen, Germany) software.

The detection was based on specific screening parameters (mass accuracy <2 mDa, retention time shift ± 0.2 min, isotopic fitting <100 mSigma (only for confirmation of positive findings), whereas the presence of adduct and fragment ions confirmed the analytes. The quantification of the detected organic contaminants was performed using the standard addition method and representative structurally related isotope-labeled compounds (Internal Standards, IS) (Gkotsis et al.

2022). A five-point spiked calibration curve was used (1.0, 5.0, 10, 50 and 75 ng g-1 w. w.), whereas for very few cases for which the standard addition spikes were not satisfactory, additional concentration levels were prepared. For the quantification experiments, a pooled sample, composed of the less contaminated individual samples, in terms of both the number and the concentration levels of the detected chemicals, was used. For the detected contaminants, %Recovery and %Factor of Matrix Effect are provided in **Tables SI–7a** and compound-specific LODs/LOQs are provided in **Tables SI–8.** Method limits of detection (LODs) were calculated from standard addition curves (using relative peak areas of spiked samples) with the following equation: LOD = 3.3*(Sb/S), where S is the slope of the calibration curve and Sb is the standard deviation of the response. The contaminants that were detected in traces below the LOQ (concentration levels between the LOD and LOQ values) are reported as BQL (below quantification limit). For the statistical treatment of the results, BQL may be substituted by LOQ/2, as indicated by Directive (2009)/90/EC.

2.3.3.2. Suspect screening. Suspect screening for >60,000 substances was performed for environmentally relevant pollutants from NORMAN SusDat (https://www.norman-network.com/nds/susdat/) in all raw chromatograms. Chromatograms were imported into the NORMAN Digital Sample Freezing Platform (DSFP) (Alygizakis et al. 2019) (http://www.norman-data.eu/), a novel tool developed for revealing the presence of suspects and identification of unknown compounds in environmental samples. The suspect screening output was subjected to manual evaluation. This evaluation included visual inspection of the extracted ion chromatograms, search of adducts in the full-scan chromatogram and fragment ions in the bbCID-scans and HRMS/MS scans. The purpose for this additional evaluation was to minimize (if not eliminate) the possibility of false positive reporting. The calibrant masses were used to recalibrate the whole chromatogram using HPC fitting algorithm, which is embedded in DataAnalysis 5.1 (Bruker Daltonics, Bremen, Germany). This calibration method ensured mass accuracy below 2 mDa during the whole chromatographic run for *m/z* 50–1000. For exporting files in mzML format, CompassXport 3.0.9.2 (Bruker Daltonics, Bremen, Germany) was used. Chromatograms acquired under bbCID were separated in low and high collision energy layer chromatograms. All mzML files and their meta-data (instrumental, sample meta-data, matrix-specific meta-data, and retention time of RTI calibrant substances) were uploaded to DSFP. DSFP has integrated a standard operating procedure (SOP) to process the mzML files and all meta-data for the generation of Data Collection Templates (DCTs). This data reduction technique resulted in an automatic generation of DCTs, which includes condensed information from LC-HRMS files.

We carried out tentative identification and semi-quantification of ECs in our suspect screening results following Aalizadeh et al. (2022). The semi-quantification approach is based on electrospray ionization efficiency models transferred to the sample matrix by the use of dichlorvos as reference compound, atrazine-d5 as internal standard and 34 calibrant substances (**Tables SI–11**). The data were entered in a semi-quantification tool (http://trams.chem.uoa.gr/semiquantification/) which was applied following the instructions in the tool manual. The concentration of a suspected compound is derived from its predicted logIE according to the equation:

$$Pred.\ Conc. = \frac{\left(\frac{PeakArea_{Compound}}{PeakArea_{Atrazine-d5}}\right) * MW_{Dichlorvos}}{10^{Pred.\ logIE} * slope_{Dichlorvos} * MW_{Compounds}}$$

The innovation in this methodology is that it considers bias due to matrix effect. Moreover, the methodology provides declared uncertainty, which is one of the core subjects in Analytical Chemistry. Furthermore, the quantitative structure–property relationship (QSPR) model provides the applicability domain according to the chemical space limitations as required by OECD regulation.

2.4. Target analyses (metals, OPFRs, dechlorane plus compounds and PBDEs) and bioassay (PCDDs, PCDFs, dl-PCBs)

Elemental analysis was carried out using ICP-MS for the 4 Water Framework Directive heavy metals Ni, Cd, Hg and Pb.

Target analysis (on 4 samples only – Samples EXT 01, EXT 03, EXT 06, INT 01 – due to insufficient sample mass for other samples) was performed for 16 organo-phosphate flame retardants (OPFRs) and dechlorane plus compounds, and for 6 poly-brominated diphenyl ether flame retardants (PBDEs 28, 47, 99, 100, 153, 154) classified as priority substances under the Water Framework Directive (WFD-PBDEs).

Bioassay (on 4 samples only – samples EXT 03, EXT 04, INT 02, INT 04 – due to insufficient sample mass for other samples) was performed for poly-chlorinated dibenzo-p-dioxins (PCDDs) and poly-chlorinated dibenzo-furans (PCDFs) and 12 dioxin-like polychlorinated biphenyls (dl-PCBs: 3,3′,4,4′-T4CB (PCB 77, CAS 32598-13-3), 3,3′,4′,5-T4CB (PCB 81, CAS 70362- 50–4), 2,3,3′,4,4′-P5CB (PCB 105, CAS 32598-14-4), 2,3,4,4′,5-P5CB (PCB 114, CAS 74472-37-0), 2,3′,4,4′,5-P5CB (PCB 118, CAS 31508-00-6), 2,3′,4,4′,5′-P5CB (PCB 123, CAS 65510-44-3), 3,3′,4,4′,5-P5CB (PCB 126, CAS 57465-28-8), 2,3,3′,4,4′,5-H6CB (PCB 156, CAS 38380-08-4), 2,3,3′,4,4′,5′-H6CB (PCB 157, CAS 69782-90-7), 2,3′,4,4′,5,5′-H6CB (PCB 167, CAS 52663-72-6), 3,3′,4,4′,5,5′-H6CB (PCB 169, CAS 32774-16-6), 2,3,3′,4,4′,5,5′-H7CB (PCB 189, CAS 39635-31-9).

2.4.1. Metals

Following sample preparation as described in Nikolopoulou et al. (2022), all glassware and polypropylene bottles were cleaned with acidified Milli-Q grade water. 0.1 g of freeze-dried biota sample was weighed into a Teflon vessel and 5 mL of 65% HNO3 were added. The samples were digested with the MARS X- Press (CEM Corporation) microwave oven with a pre-selected programme (first stage: 1600 W, 2 min ramp time, from 25 to 165 °C, 0 min hold time; second stage: 1600 W, 3 min ramp time, from 165 to 175 °C, 5 min hold time). After digestion, each sample was diluted to a final volume of 20 mL with ultrapure water. The supernatant was diluted twenty times with Milli-Q grade water and was ready for injection to the Inductively coupled plasma mass spectrometry (ICP-MS) instrument. To ensure the quality control of the analysis for quantification purposes, the certified reference material ERM-CE278k (trace elements in mussel tissue) was also analysed.

2.4.2. OPFRs and dechlorane-plus compounds

For analysis of OPFRs and dechlorane-plus compounds, sample extracts were analysed by GC-MS/MS system using an electron impact (EI) ionization. The GC-MS system (Agilent 7890 (Waldbronn, Germany) has been upgraded to MS/MS by Chromtech, Germany. The GC was operated in large volume injection mode (Restek Split liner w/Glass Frit (4 mm × 6.3 × 78.5)). The injection volume was 10 μL. A Restek Rxi-5Sil MS column of 30 m (0.25 mm i. d. X 0.25 μm film thickness) was used with hydrogen as a carrier gas at the constant flow of 1.6 mL min^{-1}. The MS/MS system was operated in multiple reaction monitoring (MRM) mode. Quantification was based on external calibration curves established for the compounds of interest.

2.4.3. Priority PBDEs under the Water Framework Directive

For analysis of priority PBDEs listed under the Water Framework Directive (PBDEs 28, 47, 99, 100, 153, 154), sample extracts were analysed by GC-MS/MS system using a negative-ion chemical (NCI) ionization. The GC-MS system (Agilent 7890 (Waldbronn, Germany) has been upgraded to MS/MS by Chromtech, Germany. The GC was operated in large volume injection mode (Restek Split liner w/Glass Frit (4 mm × 6.3 × 78.5)). The injection volume was 10 μL. A Restek Rxi-5Sil MS column of 15 m (0.25 mm i. d. X 0.15 μm film thickness) was used with hydrogen as a carrier gas at the constant flow of 1.6 mL min^{-1}. The MS/MS system was operated in multiple reaction monitoring (MRM) mode. Quantification was based on external calibration curves

established for the compounds of interest.

2.5. Bioassay – PCDDs, PCDFs, dl-PCBs

Determining the sum of PCDDs, PCDFs and dl-PCBs in biota samples uses three stages of sample processing: (i) extraction of analytes from the matrix and its subsequent fractionation on PCDs-PCBs fraction, (ii) purification of extracts and (iii) DR-CALUX bioassay, analysed using a H4IIE rat hepatoma cell line stably transformed with an Ah-receptor-driven luciferase reporter, where subsequent luminescence is measured and the total concentration is calculated from the calibration curves. 2,3,7,8-TCDD is used as a calibration solution to determine fish concentrations. The established detection limits of the method, including standard deviations, are: (i) limit of detection (LOD) 0.03 ± 0.009 ng/kg and limit of quantification (LOQ) 0.09 ± 0.03 ng/kg CALUX wet weight equivalence.

3. Results and discussion

Results from the full range of analyses carried out are presented below, addressing a large universe of chemicals: target analyses for metals, OPFRs and WFD-PBDEs, bioassay for PCDDs/PCDFs/dl-PCBs, HMRS wide-scope target analyses and suspect screening. Concentrations are expressed in ng g^{-1} wet weight (ww) followed by median and interquartile range in parenthesis.

Our study demonstrates the presence of a wide range of contaminants in the livers of black-tailed godwit in The Netherlands, some of which have not been reported in the literature before.

Target analyses detected all four metals for which we did analyses (**Tables SI–6**), and two organo-phosphate flame retardants (OPFRs) (**Tables SI–7**). However, all six of the Water Framework Directive-listed PBDEs analysed for were below LOD (**Tables SI–7**). Bioassay detected the presence of PCDDs, PCDFs and dl-PCBs (**Tables SI–7**).

28 ECs were determined through wide-scope target analysis (**Tables SI–8**) from a wide range of chemical classes: 14 industrial chemicals (including six PFAS, five personal care products, and one PAH); six plant protection products (herbicides, insecticides) and their transformation products (TPs); four pharmaceuticals and their TPs; two PCBs; and two tobacco-related compounds.

In addition, 20 additional ECs were tentatively identified through suspect screening and semi-quantified (Aalizadeh et al. 2022) in the samples (**Tables SI–9** & **SI-10**): 11 industrial chemicals (including three compounds related to plastic production and two surfactants); two plant protection products (PPPs) or their TPs; and seven pharmaceuticals or their TPs. For compounds that were included in the target database but were not detected (i.e. <Screening Detection Limit [SDL]), their presence in godwit livers cannot be entirely ruled out.

We present and discuss our results below organized by class of contaminants, starting with metals, chemicals of the Stockholm Convention (persistent organic pollutants including PCBs, PBDEs, HCHs), industrial chemicals regulated by REACH (including PFAS, personal care products, PAHs, surfactants), plant protection products (herbicides, insecticides, fungicides, etc.), pharmaceuticals, and tobacco-related compounds.

We provide comparisons with other data in literature, where available, for each substance. For many of the ECs we detected, we could not find data in literature for other studies on the godwit or on other waders. We therefore also cite data from studies on raptors, though it is important to note that raptors are top predators and therefore may be subject to greater bioaccumulation of contaminants than godwits which are primary carnivores. In particular, we cite data from a recent review of ECs in raptors (González-Rubio et al. 2021) and a recent study of ECs in livers of 30 white-tailed eagles *Haliaeetus albicilla* collected in northern Germany 2015–2018 (Badry et al. 2022). While toxicokinetics may vary between godwits and raptors, one would in general expect to find higher residue levels in raptors as top predators than in godwits as primary carnivores.

A potential limitation in our study relates to the fact that all our samples are taken from birds found dead – in most cases due to predation at or near the nest (by fox, stoat). Predated birds may be less healthy birds (and thus more prone to predation) and therefore not representative of the wider population. Less healthy birds may be so due to higher levels of contamination. However, it is not ethically or legally possible to cull godwits for contaminant study, so we necessarily sampled birds found dead. Additional limitations relate to the methods applied. HMRS studies in biota matrices involve the use of generic sample preparation methods in order to extract compounds with a wide range of physicochemical properties. Consequently, the applied protocols are non-selective, and are not optimized for a specific group of organic compounds. This is a key difference between HRMS screening methodologies and common target methods using low resolution mass analyzers, where the sample preparation, as well as instrumental analysis, are optimized for a selected, but limited, number of compounds. Moreover, the full scan acquisition mode applied in HRMS instrumentation may have accounted for higher detection limits and lower FoA than would be the case using conventional LC- or GC- MS/MS targeted methodologies for a pre-selected and restricted number of compounds of the same chemical class using Selected Reaction Monitoring (SRM) mode (Badry et al. 2022).

Contaminants included in the databases but not detected when screened in the acquired HRMS chromatograms are reported as (<SDL. SDL is established as the lowest concentration level tested for which a compound is detected in all spiked samples, at the expected retention time and with a defined mass accuracy error of the precursor ion (Gago-Ferrero et al., 2020) and in this study is at 1.80 ng g^{-1} ww. Where we report a compound as < SDL by the applied screening methodology, this does not necessarily mean that the compound is not present in the sample. A non-detected compound might have been detected if a compound-oriented validation protocol has been applied or/and a more sensitive technique has been used for the analysis of the extract. Moreover, while we selected the liver as the matrix for analysis as it is metabolically the most competent organ for detection of a wide range of contaminants with differing physicochemical properties, not all target list substances are liver-specific; some may be more easily detected in other matrices.

Notwithstanding these limitations, HRMS screening methodologies usefully supplement conventional target analyses, allowing for more holistic investigations. While suspect screening inevitably involves time-consuming computer processing and may result in false positive identifications, these can be minimized by manual evaluation. Suspect screening will gradually become mainstream with advances in instrumentation, computational tools and increasing computational power.

3.1. Metals

Observed concentrations of Cd, Hg, Pb and Ni are shown in **Tables SI–6** expressed in ng g^{-1} ww. We convert these values here into μg g^{-1} ww for ease of comparison with values in literature. Our mean values for Ni (0.113 μg g^{-1} ww ≅ 0.45 μg g^{-1} dw), Cd (0.417 μg g^{-1} ≅ 1.6 μg g^{-1} dw), Hg (0.296 μg g^{-1} ww ≅ 1.2 μg g^{-1} dw) and Pb (0.084 μg g^{-1} ww ≅ 0.33 μg g^{-1} dw) are of a similar order of magnitude to the 1.43 ± 4.63, 0.59 ± 0.77, 1.97 ± 1.69 and 0.18 ± 0.26 μg g^{-1} dw respectively, detected by Lucia et al. (2012) in livers of black-tailed godwit in France. Roodbergen et al. (2008) detected Cd, Hg and Pb in godwit eggs and feathers in The Netherlands and in soils and inferred that these metals may be taken up through earthworm prey. Heavy metal detoxification may involve increased energetic needs (Lucia et al. 2012).

For a selection of different insectivorous bird species in southern Italy, Zaccaroni et al. (2011) found comparable (to our godwit) mean Cd levels of 0.56 μg g^{-1} ww and comparable mean Hg levels of 0.23 μg g^{-1} ww, but higher mean Pb levels of 0.28 μg g^{-1} ww.

Our mean value for Hg of 0.296 μg g^{-1} ww is below the adult liver Hg concentration in birds associated with adverse reproductive effects of 8.7 (range 2–52) μg g^{-1} ww (Shore et al. 2011). Our mean value for Pb of 0.084 μg g^{-1} ww is well below the adult liver concentration in birds associated with adverse effects of 2–6 μg g^{-1} ww (Franson and Pain, 2011).

It is important to note that the primary organ for Cd accumulation is the kidney, and Pb accumulates preferentially in bone, so analysis of these tissues in future can further elucidate Cd and Pb contamination in *L. l. limosa*.

3.2. Chemicals of the Stockholm Convention

Results of target analysis for OPFRs and dechlorane plus compounds, WFD-PBDEs and of Bioassay for PCDDs, PCDFs, dl-PCBs are given in **Tables SI–7**.

3.2.1. Organophosphate flame retardants (OPFRs)

Target analysis for 14 OPFRs detected just two of these, Tris (2-chloroethyl) phosphate (TCEP) and Triphenyl phosphate (TPHP), each in 50% of the four samples analysed with concentrations of 0.12 and 0.15 ng g^{-1} ww (TCEP) and of 1.11 ng g^{-1} ww and <LOQ (TPHP), in samples EXT 01 and EXT 06, respectively. Verreault et al. (2018) found TPHP in 71%, and TCEP in 73%, of liver samples of female glaucous gull *Larus hyperboreus* breeding off Cape Dorset (Eastern Canadian Arctic) with mean concentrations of 1.71 and 2.21 ng g^{-1} ww respectively.

3.2.2. Water Framework Directive-listed polybrominated diphenyl ethers (WFD-PBDEs)

Target analysis for 6 PBDEs (28, 47, 99, 100, 153, 154) determined values < LOD for all four samples analysed. This suggests that the godwit is not exposed to high concentrations of these substances in their breeding, wintering or passage sites. Other studies have noted a general decline in environmental concentrations of these substances over the last decade following the EU ban on PDBEs under the RoHS Directive (EC, 2003) and the listing of pentaBDE and octaBDE as persistent organic pollutants under the Stockholm Convention in 2009 (e.g. Eulaers et al. 2014).

3.2.3. PCDDs, PCDFs and dl-PCBs

PCDDs, PCDFs and dl-PCBs were detected by bioassay in all four samples (EXT 03, EXT 04, INT 02, INT 04), with values from 6.2 to 43 (14.5, 20.9) pg BEQ/g ww. Further analysis by GC-HMRS would be needed to determine residue concentrations of these substances but available sample mass was insufficient for this. PCDDs/PCDFs and dl PCBs were found in brown meat of Chinese mitten crabs in Dutch rivers at mean concentrations of 18.9 and 24.6 pg TEQ g-1 ww respectively (Hoogenboom et al. 2015).

PCDDs and PCDFs are produced as by-products during the burning of garbage and certain types of chemical manufacturing processes. Several congeners of PCDDs/PCDFs exhibit various biological and toxic actions, such as dermal toxicity, immunotoxicity, carcinogenicity and adverse effects on reproductive, neurobehavioral and endocrine functions and these actions are induced by low level exposure to these compounds (Uemura et al. 2008). Dioxins and PCBs are persistent in the environment. Some forms of PCBs, non-ortho- and mono-*ortho*-PCBs, are known to have a coplanar conformation and to exhibit toxic actions similar to 2,3,7,8-tetrachlorodibenzo-p-dioxin, and are called dioxin-like PCBs (dl-PCBs). PCDDs, PCDFs and PCBs are classified as persistent organic pollutants (POPs) because of their toxicity, carcinogenicity, and persistence in the animal body. While the manufacturing and transportation of PCDD/Fs are prohibited by the Stockholm Convention, they are produced unintentionally through the combustion of waste, or as impurities during the manufacturing of agricultural chemicals (Anezaki and Kashiwagi, 2021); Minomo et al. (2011) found in Japan that rice straw burning at paddy fields acts as a driving force in the transfer of PCDDs/PCDFs/dl-PCBs from paddy-field soil to the atmosphere. As black-tailed godwit occupy rice paddies in wintering and passage sites, rice straw burning may be a source of exposure to PCDDs/PCDFs/dl-PCBs for these birds.

3.2.4. PCBs

Wide-scope target GC-HRMS analysis detected PCB 138 in six samples, three of which at BQL and three at concentrations of 3.95, 4.24 and 5.82 ng g^{-1} ww, and PCB 153 in nine samples, two of which at BQL and the remainder at concentrations of 2.28–12.2 (2.89, 6.34) ng g^{-1} ww (**Tables SI–8**). Badry et al. (2022) detected these two PCB congeners in their white-tailed eagle livers with 100% and 80% frequency respectively and at much higher median concentrations of 238 (IQR 384) and 90.1 (IQR 217) ng g^{-1} ww respectively, as might be expected in a fish-eating apex predator.

Sakellarides et al. (2006) found several PCB congeners in four waterbird species in Greece; the higher chlorinated congeners PCB 118, PCB138, PCB 153 and PCB 180 dominated. Because of the lipophilic nature, stability and persistence of the high chlorinated biphenyls, a higher rate of bioaccumulation is favored in avian body tissues than the low chlorinated ones. For PCB138, their mean values for white stork *Ciconia*, greater flamingo *Phoenicopterus roseus*, Dalmatian pelican *Pelecanus crispus* and pygmy cormorant *Phalacrocorax pygmeus* liver were 2.66; 85.8; 0.85 and 9.38 ng g^{-1} ww for PCB 138 and 2.56, 33.2, nd (no data) and 0.725 ng g^{-1} ww for PCB 153 respectively. Our levels are therefore most similar to those found for *C. ciconia* (a primary carnivore).

3.3. Industrial chemicals regulated under REACH

Industrial chemicals manufactured in and/or imported to the EU above one tonne per annum are regulated under the EU REACH regulation (EC, 2006). This includes PFAS, personal care products, PAHs, surfactants and other substances.

3.3.1. Per- and polyfluoroalkyl substances (PFAS)

Per- and polyfluoroalkyl substances (PFAS) are a group of ECs that have proved to be persistent and highly bioaccumulative. They are broadly used in various applications and are known for their long-distance migration and toxicity (Androulakakis et al. 2022). Three PFAS (PFOS, PFDeA and PFHxS) were detected at quantifiable levels by LC-HRMS wide-scope target analysis and a further three (PFHpS, PFNA and PFOA) at BQL (Tables SI–8).

PFOS was the predominant PFAS compound, detected in 100% of samples at concentrations ranging from BQL (<2.79) to 48.3 (median 16, IQR 21.89) ng g^{-1} ww. Kannan et al. (2002) detected slightly higher PFOS concentrations of <3.9 to 127 (median 29) ng g^{-1} ww in livers of white-tailed eagles collected two to four decades ago (1979–1999) from Eastern Germany, and concentrations of 32–150 ng g^{-1} ww. In liver of great cormorant *Phalacrocorax carbo* in Sardinia, Italy, as might be expected in a fish-eating species. Badry et al. (2022) reported higher PFOS concentrations of 47.2–2440 (450, 518) ng g^{-1} ww for liver in more recently collected white-tailed eagles, suggesting an increasing trend of PFAS contamination over the past 20–40 years. The fact that PFOS concentrations in livers of present-day godwit (a species that is a primary carnivore feeding on earthworms and invertebrate larvae) are of a similar order of magnitude to those found in the 1980s–1990s in livers of white-tailed eagle (a fish-eating top predator) is of concern (though modern day PFOS levels in white-tailed eagles are now an order of magnitude higher).

Compared to detected PFOS concentrations in godwits, Meyer et al. (2009) found substantially higher liver PFOS concentrations ranging from 66.2 to 1489.1 (mean 476) ng g^{-1} ww in grey heron *Ardea cinerea*, from 52.3 to 676.6 (mean 292) ng g^{-1} ww in herring gull *Larus argentatus*, from 47.6 to 775 (mean 236) ng g^{-1} ww in Eurasian sparrowhawk *Accipiter nisus*, and liver PFOS concentrations similar to those for our

godwit, ranging from 8.5 to 37.1 (mean 17) ng g^{-1} ww in Eurasian magpie *Pica* and <2.5 to 39.5 (mean 12) ng g^{-1} ww in Eurasian collared dove *Streptopelia decaocto*. However, they sampled these species from an area in the close vicinity of the city of Antwerp (Belgium) where a major PFAS chemical plant is located, so higher PFOS concentrations are to be expected in their samples. A priori, we would expect godwit to have lower PFOS concentrations than grey heron (which eats fish and amphibians), herring gull (a scavenger), sparrowhawk (which eats birds) and magpie (which scavenges and predates eggs and nestlings) but higher than collared dove (a seed-eater). The fact that our godwit samples had similar levels to magpie sampled close to a PFAS factory is of concern.

PFOS is typically found as the dominant PFAS in wildlife (e.g. Robuck et al. 2021) and has a wide range of health effects in animals including developmental effects, reduced birth mass and higher infant mortality. However, the lowest observed adverse effect level (LOAEL) for PFOS for mallard *Anas platyrhynchos* is 61,000/11,000 (male/female) ng g^{-1} ww. (Park et al. 2021), well above our PFOS residue levels for godwits.

Perfluorodecanoic acid (PDDeA) was detected in five samples (all EXT birds) of which four at BQL and one with a concentration of 9.83 ng g^{-1} ww. We could not find comparable data in the literature for bird livers for this PFAS.

Perfluorohexanesulfonic acid (PFHxS) was detected in four samples (all EXT birds) of which three at BQL (<0.352 ng g^{-1} ww) and one with a concentration of 0.416 ng g^{-1} ww. Meyer et al. (2009) found higher PFHxS concentrations ranging from <3.2 to 120.7 ng g^{-1} ww. In grey heron, <3.2 to 9.0 ng g^{-1} ww. In herring gull and <3.2 to 40.6 ng g^{-1} ww in Eurasian sparrowhawk, <3.2 to 6.7 ng g^{-1} ww in magpie and <3.2 to 6.1 ng g^{-1} ww in Eurasian collared dove – though as previously noted these birds were sampled near a PFAS manufacturing plant. Badry et al. (2022) detected PFHxS in 23% of white-tailed eagle samples at a median concentration of 0.05 (IQR 0.12) ng g^{-1} ww. Kannan et al. (2002) detected PFOS at <7 ng g^{-1} ww in great cormorants in Sardinia, Italy.

Perluoroheptaesulfonic acid (PFHpS), Perfluorononanoic acid (PFNA) and Perfluorooctanic acid (PFOA) were each detected at BQL in two of the 11 samples (not always the same samples for all three substances). PFOA and PFNA are reported by Meyer et al. (2009) in the five species they studied, ranging from <7.3 to 28.2 and < 7.6 to 17.5 ng g^{-1} ww respectively, with higher values in livers of grey heron, herring gull and Eurasian sparrowhawk and lower values in magpie and collared dove. Kannan et al. (2002) detected PFOA at much higher concentrations of 29–450 ng g^{-1} ww in great cormorants in Sardinia, Italy.

Androulakakis et al. (2022) detected a wide range of PFAS in 65 samples of apex predators (marine mammals, otters, raptors) across Europe. For common buzzard *Buteo* liver (n = 12), they obtained a PFAS residue concentration range of 218–1092 ng g^{-1}, well above our values for godwits.

3.3.2. Personal care products (PCPs)

Three parabens (methylparaben, ethylparaben and butylparaben) were detected by LC-HRMS wide-scope target analysis (**Tables SI–8**). Parabens are a class of chemicals derived from *para*-hydroxybenzoic acid, used as preservatives in many cosmetics, personal care products, pharmaceuticals and foods. They are endocrine-disrupting chemicals associated with a range of health effects in humans (Hendryx and Luo, 2022). Methylparaben, a known endocrine disruptor (Kim et al. 2018) with developmental toxicity (Merola et al. 2020) was omnipresent in the l samples at concentrations ranging from BQL (<25.3) in one sample to 452 (median 98, IQR 125) ng g^{-1} ww. Methylparaben had high detection frequency (75%) in 20 liver samples of white-tailed eagles from the Baltic Sea coast at concentrations that ranged from <8.01 to 657 ng g^{-1} ww (Xue and Kannan, 2016). More recently, Badry et al. (2022) determined methylparaben in just one of their 30 liver samples of white-tailed eagles at a concentration of 7.95 ng g^{-1} ww.

We detected Butylparaben, previously widely used as a cosmetics preservative but banned in cosmetic products the EU in 2014 (EC, 2014), in three samples at concentrations of 36.6–65.2 ng g^{-1} ww and in four further samples at BQL (<28.4 ng g^{-1} ww). We detected Ethylparaben, an antifungal preservative, in two samples at concentrations of 4.97 and 7.36 ng g^{-1} ww and in a further two samples at BQL (<3.00 ng g^{-1} ww). Buylparaben and ethylparaben were not reported by Badry et al. (2022) in their study of white-tailed eagles.

Galaxolide (HHCB), a synthetic musk used in a wide range of cosmetics, was omnipresent (FoA 100%) at concentrations ranging from BQL (<29.8) to 199 (6.5, 34.0) ng g^{-1} ww. The ubiquity of galaxolide, which has lipophilic, persistent and highly bio-accumulative properties and is known for its negative effects on soil microorganisms and wildlife (Ehiguese et al. 2020). Contrasts with Badry et al. (2022) who found galaxolide in only 30% of their white-tailed eagle liver samples (median concentration 11.3 ng g^{-1} ww)., Galaxolide and Cd, both detected in some samples, have been found to have a combined greater effect on soil microorganisms than Galaxolide alone (Lv et al., 2021).

The PCP lauryl diethanolamide, used as a foam booster (wetting agent) in cosmetic products, was detected in two samples at 374 and 1180 ng g^{-1} ww. This substance was not reported by Badry et al. (2022) in their study of white-tailed eagles and is not reported by González-Rubio et al. (2021) in their recent review of ECs in raptors.

3.3.3. Polycyclic aromatic hydrocarbons (PAHs)

Chrysene was detected in one sample at a concentration of 5.22 ng g^{-1} ww (**Tables SI–8**). The substance is not reported in Badry et al. (2022) or in the review by González-Rubio et al. (2021).

3.3.4. Surfactants

Two surfactants were tentatively determined by suspect screening (**Tables SI-9, SI-10**). Surfactants are chemical products consumed in large quantities every day on a worldwide scale.

(9Z)-*N*-(2-Hydroxyethyl)octadec-9-enamide occurred in our samples with 100% FoA at a concentration of 5–170 ng g^{-1} ww. It is manufactured in and/or imported to the EEA for intermediate use only. This substance causes serious eye damage and causes skin irritation (ECHA Substance Information: https://echa.europa.eu/substance-information/-/substanceinfo/100.002.075). It is a ceramide-like molecule used in cosmetic and pharmaceutical formulations.

N,*N*-Bis(2-hydroxyethyl)dodecanamide (DDA) occurred in our samples with 27% FoA at an estimated concentration range of 7–80 ng g^{-1} ww. It is manufactured in and/or imported to the EEA at >100 tonnes per annum. This substance causes serious eye damage and skin irritation. It is used in a wide range of products in both consumer and industrial contexts with multiple sources for release to the environment. (ECHA Substance Information: https://echa.europa.eu/substance-information/-/substanceinfo/100.003.994).

3.3.5. Other industrial chemicals

3.3.5.1. Substances detected by LC-HRMS wide-scope target analysis. The industrial chemical Benzododecinium, a quaternary ammonium compound (QAC), was detected by wide-scope target analysis only in Sample INT 02 at a concentration of 11.8 ng g^{-1} ww (**Tables SI–8**). This substance is not reported as detected in raptors by either Badry et al. (2022) or González-Rubio et al. (2021). Benzododecinium is an antiseptic and disinfectant used as a topical agent. It is increasingly used in agriculture as an insecticide and known to be toxic to a wide range of organisms (Li et al. 2018).

N,*N*-Dimethyltetradecylamine was detected by wide-scope target analysis only in Sample INT 01 at a concentration of 2.18 ng g^{-1} ww (**Tables SI–8**). This substance is not reported as detected in raptors by either Badry et al. (2022) or González-Rubio et al. (2021). It is an antistatic agent and corrosion inhibitor used in solvents as well as hair

conditioning.

*3.3.5.2. Industrial substances identified by LC-HRMS suspect screening (**Tables SI-9, SI-10**).* 1,3-Benzenedimethanamine (1,3-BDMA) occurred in all our samples with an estimated concentration range of 4–20 ng g^{-1} ww. It is manufactured in and/or imported to the European Economic Area (EEA) at 10–100 tonnes per annum. This substance is used in a variety of industrial applications including amine-based curing agents for epoxy resins which may then be formulated into coatings, adhesives, sealants and elastomers. It is very toxic to aquatic life with long lasting effects. It is harmful if swallowed, causes severed skin burns and serious eye damage, may cause damage to organs through prolonged or repeated exposure and may cause an allergic skin reaction. (ECHA Substance Information: https://echa.europa.eu/substance-information/-/substanceinfo/100.104.036).

3-Methylbenzoic acid occurred in our samples with 90.9% frequency of appearance (FoA) at an estimated concentration range of 0.4–9 ng g^{-1} ww. It is manufactured in and/or imported to the EEA at 10–100 tonnes per annum. It is used at industrial sites and is known to cause serious eye damage.

Benzyl alcohol occurred in our samples with 90.9% FoA at an estimated concentration range of 30–280 ng g^{-1} ww. It is manufactured in and/or imported to the EEA at ≥10,000 tonnes per annum. It has a wide range of uses. (ECHA Substance Information: https://echa.europa.eu/substance-information/-/substanceinfo/100.002.600). It is harmful if swallowed or inhaled and causes serious eye irritation.

3-(4-*tert*-Butylphenyl)propanal (BHCA) occurred in our samples with 81.8% FoA at an estimated concentration range of 40–170 ng g^{-1} ww. It is manufactured in and/or imported to the EEA at 10–100 tonnes per annum. It is used by consumers and professional workers in a wide range of products. It may cause damage to organs through prolonged or repeated exposure, is harmful to aquatic life with long lasting effects, causes skin irritation and may cause an allergic skin reaction. (ECHA Substance Information: https://echa.europa.eu/substance-information/-/substanceinfo/100.038.182). It adversely affects sperm formation in rats (Laue et al. 2017) so may have similar reproductive effects in birds.

5,7-Dihydroxy-4-methylcoumarin occurred in our samples with 45.5% FoA at an estimated concentration range of 0.01–1 ng g^{-1} ww. It meets REACH Annex III criteria for substances manufactured and/or imported into the EEA at 10–100 tonnes per annum and '*predicted as likely to meet criteria for category 1A or 1B carcinogenicity, mutagenicity, or reproductive toxicity, or with dispersive or diffuse use(s) where predicted likely to meet any classification criterion for health or environmental hazards, or where there is a nanoform soluble in biological and environmental media.*' It may cause serious eye and skin irritation (ECHA Substance Information: https://echa.europa.eu/substance-information/-/substanceinfo/100.016.627).

3-Methylpyrazole is an alkylpyrazole which occurred in our samples with 9.09% FoA (1 sample) at an estimated concentration of 10 ng g^{-1} ww. It is manufactured in and/or imported to the EEA at 100–1000 tonnes per annum. It is used by professional workers in fertilizers and in agriculture, forestry and fishing. Pyrazoles are nitrification inhibitors used in agriculture to reduce nitrogen losses (Shi et al., 2012). In humans, it causes severe skin burns and eye damage, may damage fertility or the unborn child, is harmful if swallowed, causes serious eye damage and may cause damage to organs through prolonged or repeated exposure. (ECHA Substance Information: https://echa.europa.eu/substance-information/-/substanceinfo/100.014.478).

Three industrial compounds related to plastic production were detected; nylon-6, hexanedioic acid 2,2,4-trimethyl- and erucamide. Since the 1950s, the production and use of plastics has increased globally with increased amounts of microplastics released into aquatic environments (Monteleone et al. 2019). An estimated 4.9 billion tons of plastics have been discarded so far in the natural environment and landfills, with 600 million tons as polyester, polyamide, and acrylic fibers. Worldwide, the most commonly used plastic materials are polyamide nylon 6 (PN6), polyethylene (PE) and polyvinyl chloride (PVC), materials having a high resistance to biological degradation (Spyridakis et al. 2022). Nylon polymer particles are used in personal care products such as face powder and eyeshadow as pacifying and skin-improving agents (Timm et al. 2011; Burnett et al. 2014). They can be introduced into freshwater environments through human activities such as swimming as well as the influx of domestic wastewater and are common in aquatic environments (Mizukami-Murata et al. 2021).

Nylon-6 (polyamide 6) occurred in our samples with 100% FoA at an estimated concentration range of 0.5–40 ng g^{-1} ww. It meets REACH Annex III criteria for substances manufactured and/or imported into the EEA at 10–100 tonnes per annum and '*predicted as likely to meet criteria for category 1A or 1B carcinogenicity, mutagenicity, or reproductive toxicity, or with dispersive or diffuse use(s) where predicted likely to meet any classification criterion for health or environmental hazards, or where there is a nanoform soluble in biological and environmental media.*' https://echa.europa.eu/substance-information/-/substanceinfo/100.124.824). Nylon 6 is a micrometer-sized nylon polyamide and is used to make not only textile, carpet and technical fibers but also engineering plastics and packaging films. DSM has been producing caprolactam, the key raw material for Nylon-6, in The Netherlands for 50 years and expanded its plant at Geleen in 2007. A key environmental source of Nylon-6 is discarded fishing gear (Hongthong et al. 2021).

Hexanedioic acid 2,2,4-trimethyl- occurred in our samples with 9.09% FoA at a concentration range of 20–120 ng g^{-1} ww. The substance is manufactured in and/or imported to the EEA for intermediate use only. It is known to cause serious eye damage in humans. (ECHA Substance Information: https://echa.europa.eu/substance-information/-/substanceinfo/100.053.216). This and other n-Octyl esters of alkanoic and hexanedioic acids are found in urban wastes. They are commonly used in cosmetics, pharmaceuticals, and the food and textile industries (Chaler et al. 2004).

Erucamide occurred in our samples with 36.4% FoA at a concentration range of 0.1–6 ng g^{-1} ww. It is manufactured in and/or imported to the EEA at 10,000–100,000 tonnes per annum (ECHA Substance Information: https://echa.europa.eu/substance-information/-/substanceinfo/100.003.645) and is used in a wide range of products. Erucamide has potential toxicity to cause skin irritation and is hydrolysed to erucic acid, which can cause heart damage.

3.4. Plant protection products (insecticides, herbicides, fungicides, etc.)

4,4′-DDE, the transformation product of the banned insecticide DDT, was determined in 100% of our samples at concentrations from 3.33 to 73.0 (46.2, 47.1) ng g^{-1} ww (**Tables SI–8**). Badry et al. (2022) found 4, 4′-DDE in all 30 white-tailed eagle liver samples, at a higher median concentration of 169 ng g^{-1} ww (IQR 159) as might be expected for a fish-eating apex predator.

Benfluralin and Trifluralin were each detected in the same four samples (Samples INT 02, INT 03, INT 04, INT 05 – all from intensive grasslands) at concentrations of 0.226–1.31 (0.545, 0.797) and 0.399 to 4.05 (0.9155, 2.27) ng g^{-1} ww respectively (**Tables SI–8**). Benfluralin is an herbicide of the dinitroaniline class, used to control grasses and weeds and is known to cause thyroid effects in rats and classified by EFSA as 'suspected of causing cancer' (Strupp et al. 2020). Trifluralin is a commonly used herbicide used to control grasses and broadleaf weeds. It was banned in the EU in 2008. The fact that these substances were detected only in the INT birds suggests local exposure in the Dutch breeding grounds rather than at wintering or passage sites.

The herbicides Endothall (7-oxabicyclo [2.2.1]heptane-2,3-dicarboxylic acid) and Sulcotrione were each detected in one sample (INT5 and INT3 respectively) at BQL levels (**Tables SI–8**). Neither substance is reported by Badry et al. (2022) or in the review of González-Rubio et al. (2021). Endothall is a selective, contact herbicide.

It is highly soluble in water and semi-volatile but generally non-persistent in soils. Endothall is highly toxic to mammals, moderately toxic to fish and aquatic organisms, but less toxic to birds. It is not expected to bioaccumulate (Pesticides Properties Database: http://sitem.herts.ac.uk/aeru/ppdb/en/Reports/265.htm). Sulcotrione is a selective triketone herbicide marketed for use in maize culture since 1993 (Goujon et al. 2014).

Ethiofencarb-sulfone was detected at 41.5 ng g^{-1} ww in one sample only (INT 05) (**Tables SI–8**). Badry et al. (2022) detected the substance in 33% of their white-tailed eagles with a much lower median of 2.85 (IQR 2.52) ng g^{-1} ww and the substance is not reported in the review of González-Rubio et al. (2021). Ethiofencarb-sulfone is a metabolite of the non-authorised pesticide simazine, which has low P and B scores, so its presence in birds suggests recent illegal use (Badry et al. 2022).

It is notable that, other than 4,4′-DDE (a legacy contaminant), the detected herbicides were found only in birds sampled from intensive pastures, not in those sampled from extensive, herb-rich pastures. This would appear to indicate local use of these herbicides in these intensive grasslands.

One plant protection product was tentatively identified by suspect screening. Fenpropimorphic acid occurred in our samples with 63.6% FoA at an estimated concentration range of 0.3–0.9 ng g^{-1} ww (**Tables SI-9, SI-10**). It is a fungicide used to control various fungal pathogens in cereals and approved for use in some EU Member States including the Netherlands.

One insect control agent was tentatively identified by suspect screening. Epofenonane acid occurred in our samples with 90.9% FoA at an estimated concentration range of 10.0–50.0 ng g^{-1} ww (**Tables SI-9, SI-10**). It is a juvenile hormone mimic used in mosquito control.

3.5. Pharmaceuticals, their transformation products and metabolites

Pharmaceuticals are known to potentially alter the feeding rate of fish and affect behavioural traits such as activity, aggression, boldness, exploration and sociality (see review in Brodin et al. 2014). However, there is very little field-based information for higher vertebrates such as birds and mammals (Taggart et al. 2016; Distefano et al. 2022). A wide range of pharmaceuticals enter freshwaters globally and may be transferred from aquatic to terrestrial ecosystems via aquatic primary producers (Previšić et al., 2021).

Venlafaxine was detected in seven samples at concentrations of 14.8–76.0 (18.2, 24) ng g^{-1} ww along with its transformation product, D,L-*N*,*N*-didesmethyl-venlafaxine at concentrations of 1.84–15.3 (18.2, 24) ng g^{-1} ww (**Tables SI–8**). Badry et al. (2022) found only the TP, in 38% of liver samples of white-tailed eagles, and at a lower median concentration of 5.38 μg/kg ww. (IQR 4.45). Venlafaxine, a widely used antidepressant, is a serotonin and noradrenaline re-uptake inhibitor, routinely detected in aquatic environments. It has been found to affect courtship behaviour in adult zebrafish (Tang et al. 2022).

Sulfadoxine, a persistent substance with a P score >0.6 (Badry et al. 2022), occurred in only one sample (Sample EXT 04) at a concentration of 50.7 ng g^{-1} ww (**Tables SI–8**). It is used in combination with pyrimethamine to treat malaria and isused as a veterinary drug. It is possible that the birds are exposed to this substance on their wintering grounds in Africa. Sulfobenzamide, an antimicrobial drug, was also detected in one sample (INT 02) at a concentration of 26.3 ng g^{-1} ww. Sulfadoxine and Sulfobenzamide are sulfonamide antibiotics, one of the most widely used class of antibiotics worldwide and used in clinical practice since 1968 (Sampaio et al., 2016). Neither substance was presented in raptor samples reported by Badry et al. (2022) and González-Rubio et al. (2021).

Seven further pharmaceutical substances or their metabolites were tentatively identified by suspect screening (**Tables SI-9, SI-10**). Their presence in godwits suggests entry of these substances into the godwit food web via urban wastewater.

Zolmitriptan was tentatively detected with 100% FoA at an estimated concentration range of 3.0–50.0 ng g^{-1} ww. It is a selective serotonin receptor agonist used to treat the symptoms of migraine headaches (https://medlineplus.gov/druginfo/meds/a601129.html).

Amiterol was tentatively detected with 100% FoA at an estimated concentration range of 0.5–9 ng g^{-1} ww. It is an oral anti-asthma drug (bronchodilator). It is manufactured in and/or imported to the EEA at 1–10 tonnes per annum. It is identified under REACH as a '*substance predicted as likely to meet criteria for category 1A or 1B carcinogenicity, mutagenicity, or reproductive toxicity, or with dispersive or diffuse use(s) where predicted likely to meet any classification criterion for health or environmental hazards, or where there is a nanoform soluble in biological and environmental media.*'

Telbivudine was tentatively detected with 90.9% FoA at an estimated concentration range of 0.8–50.0 ng g^{-1} ww. It is an antiviral drug used in the treatment of hepatitis B infection. The marketing authorisation ceased to be valid in November 2020 and it is therefore no longer authorised for use in the EU (EMA, 2021).

5-([(Dimethylamino)methyl]furan-2-carboxylic acid), a metabolite of ranitidine, was tentatively detected with 72.7% FoA at an estimated concentration range of 2–6 ng g^{-1} ww. Ranitidine is a histamine-2 blocker used to reduce stomach acid and is known to cause a range of serious side effects and was removed from the market in the EU in April 2020 (our samples mostly pre-date this restriction).

(2-(4-{[(1*R*, 2*S*)-2-Hydroxycyclopentyl]methyl}phenyl)propanoic acid), a metabolite of the non-steroidal anti-inflammatory drug loxoprofen, was tentatively detected with 72.7% FoA at an estimated concentration range of 1–2 ng g^{-1} ww.

Aminolevulinic acid was tentatively detected with 54.6% FoA at an estimated concentration range of 0.07–3 ng g^{-1} ww. It is a naturally occurring compound in animals (in the pathway that leads to production of heme) and plants (in the pathway that leads to production of chlorophyll) and is also used in medicine in the photodynamic detection and surgery of cancer.

Zilpaterol was tentatively detected with 36.4% FoA at an estimated concentration range of 0.1–0.4 ng g^{-1} ww. It is a veterinary growth hormone, more specifically a β_2 adrenergic agonist, used to increase the size of cattle and the efficiency of feeding them (Arcella et al. 2016). It is produced in France but banned for use in the EU. It is also used as an illicit drug in athletes and body builders and administered to racehorses (Hepworth-Warren *et al.* 2014). It causes damage to organs through prolonged or repeated exposure.

3.6. Tobacco-related compounds

Two tobacco-related compounds were detected by wide-scope target analysis in sample INT05; Harman was detected at 275 ng g^{-1} ww and Harmaline at BQL (<13.9 ng g^{-1} ww) (**Tables SI–8**). Neither substance was reported by Badry et al. (2021) or the review by González-Rubio et al. (2021). Harman (1-methyl-β-carboline) is a hetero-cyclic aromatic amine with potential mutagenicity (Zhang et al. 2020). It is a common natural compound in several plants and is also found in various cooked foods, beverages and cigarettes. It has mutagenic, co-mutagenic and carcinogenic effects (El Gendy and El-Kadi, 2010).

3.7. Total number of substances and percentage of total number in each chemical class

This study is the first to apply wide-scope target and suspect screening, as well as a range of target analyses and bioassays, to godwits. Target analyses, LC- and GC-HRMS wide-scope target analysis and LC-HRMS suspect screening have determined 55 different contaminants, and bioassay has additionally determined the presence of PCDDs, PCDFs and dl-PCBs, in liver samples of adult black-tailed godwits found dead on their breeding grounds in SW Friesland in The Netherlands, a breeding stronghold for this declining subspecies.

Wide scope target analysis revealed the presence of 29 CECs, of

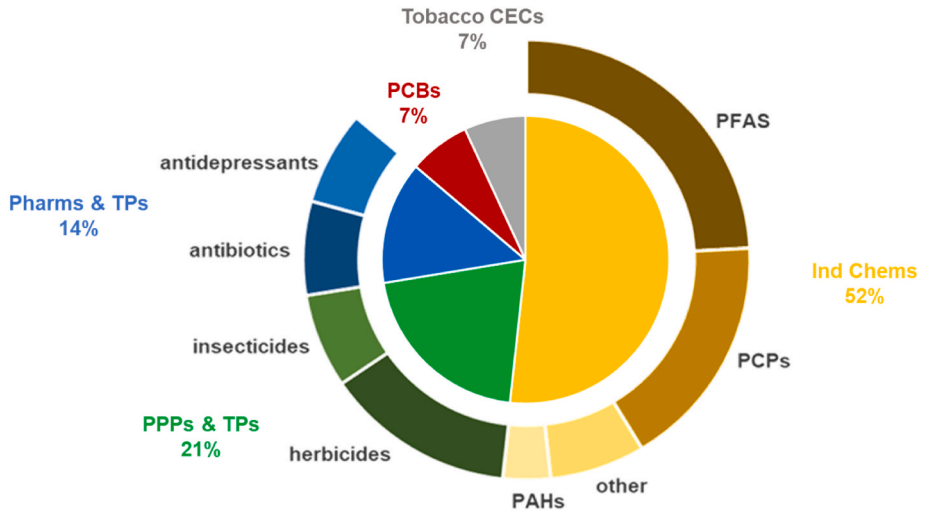

Fig. 2. Classification of the compounds detected through wide-scope target analysis, based on their main use. *PFAS = Per- and polyfluoroalkyl substances; PFSAs = Perfluoroalkane sulfonic acids; PFCAs = Perfluoroalkyl carboxylic acids; PPPs = Plant Protection Products; PCPs = Personal Care Products; Ind Chems = Industrial Chemicals; PCBs = Polychlorinated biphenyls; PAHs = Polycyclic aromatic hydrocarbons; Pharms = Pharmaceuticals; ECs = emerging contaminants; TPs = Transformation Products.*

which 17 industrial chemicals (including seven PFAS, six PPPs (or their TPs), two PCBs, one PAH and two other substances), four pharmaceuticals (or their TPs), six PPPs (or their TPs), and stimulants (tobacco-related compounds). Fig. 2 shows the percentage of these 29 CECs falling within each class of contaminant.

Suspect screening tentatively identified a further 20 substances: 11 industrial chemicals (of which two surfactants), seven pharmaceuticals (or their TPs) and two PPPs.

Target analyses additionally detected four metals and two OPFRs (but no PDBEs). Bioassay detected PCDDs, PCDFs and dl-PCBs. Residue concentrations of detected substances range from <1 to >450 ng g^{-1} ww.

3.8. Total contaminant load per bird and differences between EXT and INT samples

We obtained six of our 11 samples from extensive, herb-rich grasslands (EXT01 – EXT06) and five from intensive grasslands (INT 01 – INT 05) in their breeding grounds in SW Friesland.

Fig. 3 shows total contaminant load for each of the 11 samples, for the substances determined by HRMS wide-scope target analysis, with the proportions of each class of chemical shown in each bar. It does not include those determined by suspect screening (which only tentatively identifies substances and gives only estimated, semi-quantitative concentrations), and does not include substances determined by separate target analyses. The figures show that the total contaminants load for these 29 substances ranged from c. 160 to c. 330 ng g^{-1} for birds from extensive grasslands and from c. 350 to c. 1400 ng g^{-1} for birds from intensive grasslands.

3.9. Differences between EXT-INT samples for selected classes of contaminants

Figs. 4–7 show the relative concentrations of substances detected by wide-scope target analysis (for which we have firm concentration values) for the six EXT and five INT birds. Fig. 4 suggests that INT birds

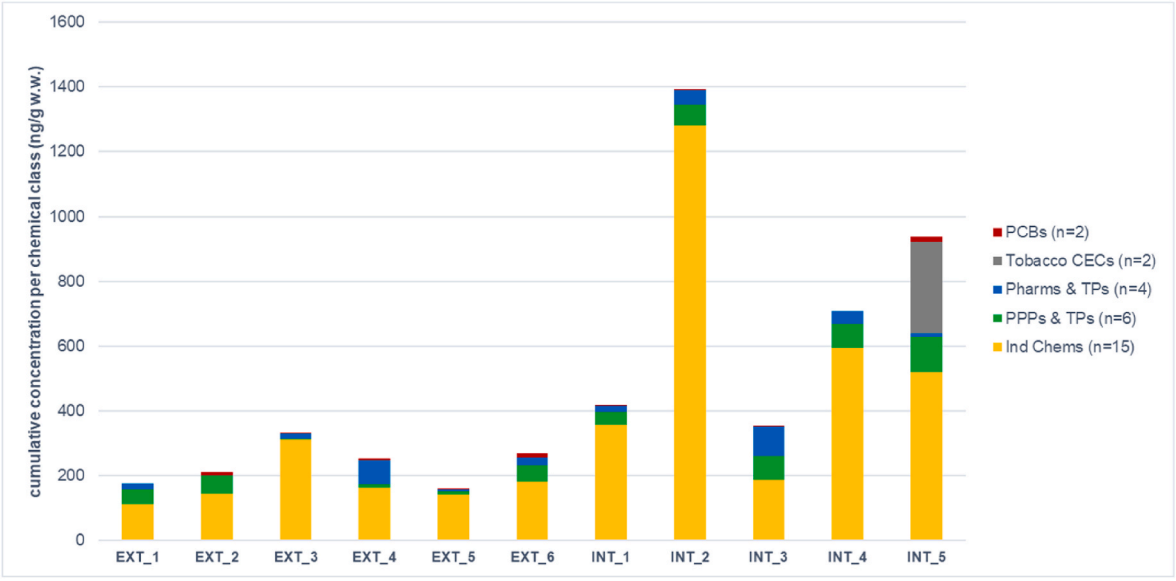

Fig. 3. Cumulative concentrations per chemical class (ng g^{-1} ww) of the compounds detected through wide-scope target analysis. Industrial chemicals include PFAS, PAH, personal care products, surfactants and others falling within the scope of REACH (EC, 2006).

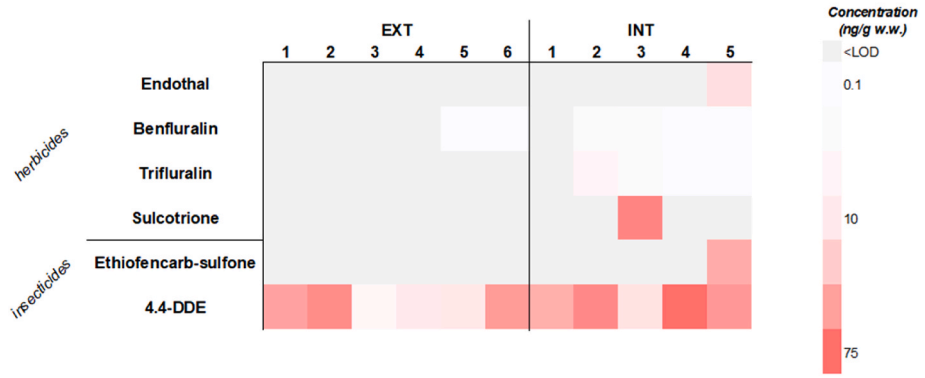

Fig. 4. Heatmap showing concentration levels (ng g^{-1} ww) of plant protection products detected in the EXT and INT samples.

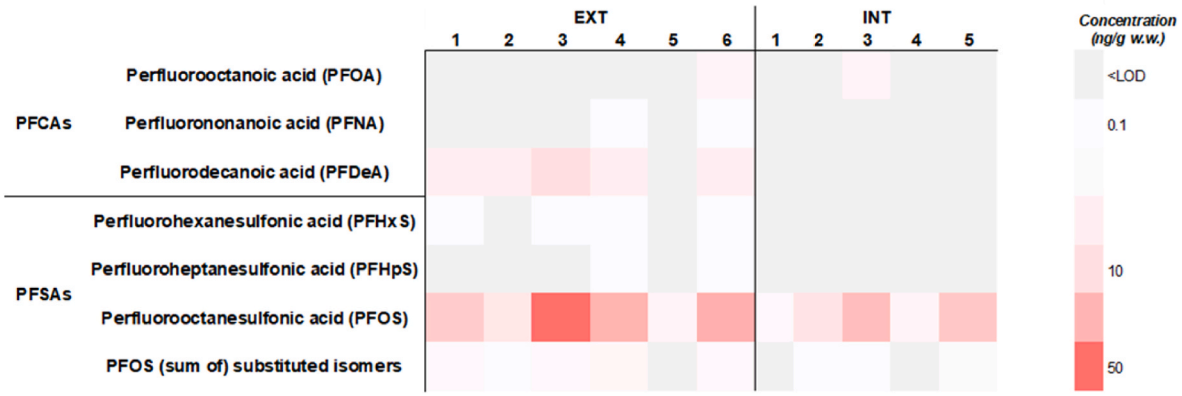

Fig. 5. Heatmap showing concentration levels (ng g^{-1} ww) of PFAS detected in the EXT and INT samples.

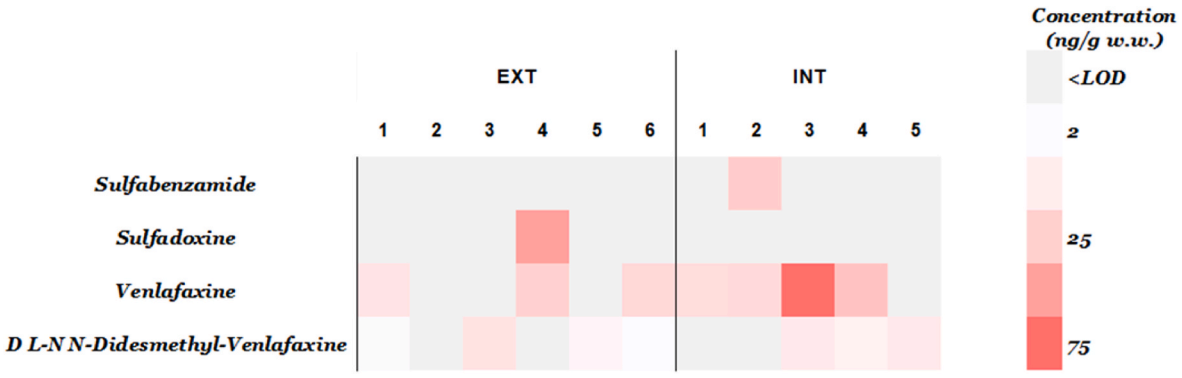

Fig. 6. Heatmap showing concentration levels (ng g^{-1} ww) of Pharmaceuticals & TPs detected in the EXT and INT samples.

have a greater mix of PPP substances in their livers and higher residue concentrations of PPPs than EXT birds. On the contrary, Fig. 5 suggests that EXT birds have a greater mix of PFAS substances in their livers and higher residue concentrations of PFAS.

Fig. 6 suggests no substantial difference between EXT and INT samples in the mix and concentrations of pharmaceuticals and their TPs, while Fig. 7 similarly suggests little substantive difference between EXT and INT samples in the mix and concentrations of industrial chemicals (other than PFAS). The most notable difference in Fig. 7 is perhaps the high concentrations of lauryl diethanolamide in INT birds and its absence from EXT birds.

While the contaminant profiles appear to indicate that for some substances, the detected substances are at higher residue concentrations for godwits from intensively managed (INT) grasslands and for other substances (PFAS) at higher concentrations in those from extensively managed (EXT) grasslands, suggesting local exposure to the respective substances within INT or EXT breeding grounds respectively (rather than exposure in the wintering and passage grounds), the NT and EXT sample sizes are small. This indicates the value of follow up analysis with additional samples.

3.10. Detection/non-detection of expected agricultural pesticides

We expected a priori to find a number of agricultural pesticides that we know have been widely used in the study area in SW Friesland, including DDT (a persistent organic pollutant, widely used before it was banned in 1979 under Directive 79/117/EEC), glyphosate (still authorised for use in the EU) and the neonicotinoid pesticides

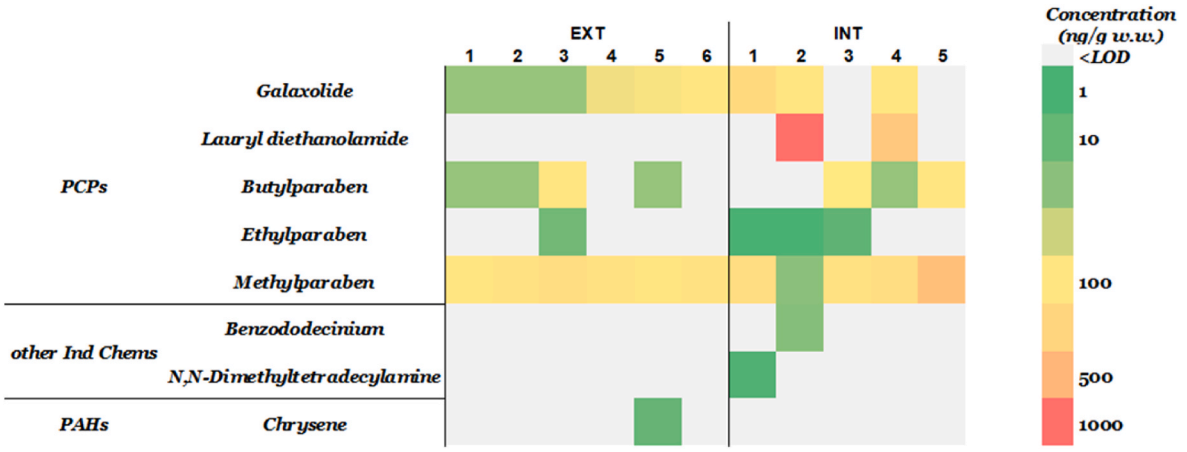

Fig. 7. Heatmap showing concentration levels (ng g^{-1} ww) of Industrial Chemicals (other than PFAS) detected in the EXT and INT samples.

thiamethoxam, imidacloprid or clothianidin (partially restricted in the EU since 2013 and banned since September 2020). We found 4,4′-DDE (the breakdown product of DDT) in all samples, reinforcing the findings of a recent study on DDT in Dutch godwits and their eggs (https://nos.nl/artikel/2378864-al-jaren-verboden-insectenverdelger-nog-steeds-gevonden-in-grutto-s). We were not able to detect glyphosate in our samples as this would have required specific sample preparation and an additional sample mass. We did not detect any neonicotinoid pesticides in our samples. Although the neonicotinoids imidacloprid, acetamiprid, dinotefuran and thiamethoxam are included in the LC target list, the concentrations in our samples are below SDL. Acetamiprid is also on the GC target list but the concentration is again below SDL. However, a generic sample preparation protocol was followed for wide-scope target and suspect screening purposes; an optimized extraction protocol with a more sensitive analytical technique is required to confirm presence and concentrations of these neonicotinoids.

4. Conclusion

Our results from target analyses, wide scope target analysis and suspect screening, reveal the presence of 55 contaminants in godwit livers, together with the positive bioassay for PCDDs, PDCFs and dl-PCBs, suggesting that these may pose a risk to godwit health. Further research is required to corroborate our results on contaminants in adult godwit livers, to extend this research to vulnerable life stages (eggs, chicks) and to godwit food items (invertebrates, plant food) in their wintering, staging and breeding grounds, and to elucidate potential health risks to godwits and contaminant sources. Such knowledge might usefully inform remedial measures for improved godwit conservation, as well as for further regulatory risk assessment of detected substances with a view to possible introducing appropriate chemical risk management measures.

Credit author statement

PM: Conceptualisation, Data Curation, Writing – Original Draft, Writing – Review & Editing, KB: Writing – Review & Editing, GG: Investigation, Formal analysis (HRMS analysis, wide-scope target analysis), Data Curation, Visualization, Writing – Review & Editing, NA: Investigation, Formal analysis (suspect screening, statistical analysis), Data Curation, Visualization, Writing – Review & Editing, MCN: Investigation, Data Curation (wide-scope target analysis), Writing - Review & Editing; KV: Formal analysis (samples' pre-treatment and extraction of organic contaminants), MK: Formal analysis (metals analysis), NST: Methodology (metals, wide-scope target analysis and suspect screening), Resources (ICP-MS, HRMS, reference standards of CECs), Supervision, Writing – Review & Editing, PO: Laboratory analysis (PDBEs, OPFRs, bioassay), Writing – Review & Editing, MO: Laboratory analysis (PDBEs, OPFRs, bioassay), JS: Laboratory analysis (PDBEs, OPFRs, bioassay), NG: Administration of laboratory analysis (PDBEs, OPFRs, bioassay), JCEWH: Specimen collection, Writing – Review & Editing, RAH: Specimen collection, Writing – Review & Editing, RWRJD: Writing – Review & Editing, funding acquisition, NvdB: Writing – Review & Editing, TP: Conceptualisation, Resources (specimen collection, sample extraction and shipping), Writing – Review & Editing, Funding Acquisition.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data provided in SI and in online database (NORMAN DFSP)

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.chemosphere.2023.138145.

References

Aalizadeh, R., Alygizakis, N.A., Schymanski, E.L., Krauss, M., Schulze, T., Ibanez, M., et al., 2021. Development and application of liquid chromatographic retention time indices in HRMS-based suspect and nontarget screening. Anal. Chem. 93 (33), 11601–11611.

Aalizadeh, R., Nikolopoulou, V., Alygizakis, N., Slobodnik, J., Thomaidis, N.S., 2022. A novel workflow for semi-quantification of emerging contaminants in environmental samples analyzed by LC-HRMS. Anal. Bioanal. Chem. 1–16 https://doi.org/10.1007/s00216-022-04084-6.

Alygizakis, N.A., Oswald, P., Thomaidis, N.S., Schymanski, E.L., Aalizadeh, R., Schulze, T., Oswaldova, M., Slobodnik, J., 2019. NORMAN digital sample freezing platform: a European virtual platform to exchange liquid chromatography high resolution-mass spectrometry data and screen suspects in "digitally frozen" environmental samples. Trends Anal. Chem. 115, 129–137. https://doi.org/10.1016/j.trac.2019.04.008.

Androulakakis, A., Alygizakis, N., Gkotsis, G., Nika, M.C., Nikolopoulou, V., Bizani, E., et al., 2022. Determination of 56 per-and polyfluoroalkyl substances in top predators and their prey from Northern Europe by LC-MS/MS. Chemosphere 287, 131775.
Anezaki, K., Kashiwagi, N., 2021. Daily variations and factors of atmospheric PCDD/Fs in post-harvest paddy fields: PCDD/F source estimation using a Bayesian semi-factor model. Chemosphere 268, 129292.
Arcella, D., Baert, K., Binaglia, M., Gervelmeyer, A., Innocenti, M.L., Ribo, O., Steinkellner, H., Verhagen, H., 2016. Review of proposed MRLs, safety evaluation of products obtained from animals treated with zilpaterol and evaluation of the effects of zilpaterol on animal healthand welfare. EFSA J. 14 (9), 4579.
Badry, A., Schenke, D., Treu, G., Krone, O., 2021. Linking landscape composition and biological factors with exposure levels of rodenticides and agrochemicals in avian apex predators from Germany. Environ. Res. 193, 110602.
Badry, A., Treu, G., Gkotsis, G., Nika, M.C., Alygizakis, N., Thomaidis, N.S., et al., 2022. Ecological and spatial variations of legacy and emerging contaminants in white-tailed sea eagles from Germany: implications for prioritisation and future risk management. Environ. Int. 158, 106934 https://doi.org/10.1016/j.envint.2021.106934.
Beintema, A.J., Beintema-Hietbrink, R.J., Miiskens, G.J.D.M., 1985. A shift in the timing of breeding in meadow birds. Breeding ecology of meadow birds (Charadriiformes); implications for conservation and management 29.
Beintema, A.J., Muskens, G.J.D.M., 1987. Nesting success of birds breeding in Dutch agricultural grasslands. J. Appl. Ecol. 743–758.
Burnett, R.T., Pope III, C.A., Ezzati, M., Olives, C., Lim, S.S., Mehta, S., et al., 2014. An integrated risk function for estimating the global burden of disease attributable to ambient fine particulate matter exposure. Environ. Health Perspect. 122 (4), 397–403.
BirdLife International, 2006. Limosa limosa limosa. The IUCN Red List Of Threatened Species 2006 e.T22693150A30075190.
BirdLife International, 2017. Limosa limosa limosa (Amended Version of 2016 Assessment). The IUCN Red List of Threatened Species 2017 e.T22693150A111611637.
Brodin, T., Piovano, S., Fick, J., Klaminder, J., Heynen, M., Jonsson, M., 2014. Ecological effects of pharmaceuticals in aquatic systems–impacts through behavioural alterations. Philos. Trans. R. Soc. B Biol. Sci. 369, 20130580 https://doi.org/10.1098/rstb.2013.0580.
Busch, M., Katzenberger, J., Trautmann, S., Gerlach, B., Droeschmeister, R., Sudfeldt, C., 2020. Drivers of population change in common farmland birds in Germany. Bird. Conserv. Int. 30 (3), 335–354.
Chaler, R., Cantón, L., Vaquero, M., Grimalt, J.O., 2004. Identification and quantification of n-octyl esters of alkanoic and hexanedioic acids and phthalates as urban wastewater markers in biota and sediments from estuarine areas. J. Chromatogr. A 1046 (1–2), 203–210.
Distefano, G.G., Zangrando, R., Basso, M., Panzarin, L., Gambaro, A., Ghirardini, A.V., Picone, M., 2022. Assessing the exposure to human and veterinary pharmaceuticals in waterbirds: the use of feathers for monitoring antidepressants and nonsteroidal anti-inflammatory drugs. Sci. Total Environ. 821, 153473.
Donald, P.F., Sanderson, F.J., Burfield, I.J., Van Bommel, F.P., 2006. Further evidence of continent-wide impacts of agricultural intensification on European farmland birds, 1990–2000. Agric. Ecosyst. Environ. 116 (3–4), 189–196.
EC, 2003. Directive 2002/95/EC of the European Parliament and of the Council of 27 January 2003 on the Restriction of the Use of Certain Hazardous Substances in Electrical and Electronic Equipment.
EC, 2006. Regulation (EC) No 1907/2006 of the European Parliament and of the Council of 18 December 2006 Concerning the Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH), Establishing a European Chemicals Agency. *amending Directive 1999/45/EC and repealing Council Regulation (EEC) No 793/93 and Commission Regulation (EC) No 1488/94 as well as Council Directive 76/769/EEC and Commission Directives 91/155/EEC, 93/67/EEC*, 93/105/EC and 2000/21/EC.
EC, 2014. Commission regulation (EU) no. 358/2014 of 9 April 2014 amending annexes II and V to regulation (EC) No 1223/2009 of the European parliament and of the council on cosmetic products. Off. J. Eur. Union 107, 5–9.
El Gendy, M.A., El-Kadi, A.O., 2010. Harman induces CYP1A1 enzyme through an aryl hydrocarbon receptor mechanism. Toxicol. Appl. Pharmacol. 249 (1), 55–64.
Ema, 2021. Public Statement: Sebivo. Withdrawal of the Marketing Authorisation in the European Union. EMA/594353/2020. Available at: https://www.ema.europa.eu/en/medicines/human/EPAR/sebivo. Accessed 03/07/2022.
Emmerson, M., Morales, M.B., Oñate, J.J., Batary, P., Berendse, F., Liira, J., et al., 2016. How agricultural intensification affects biodiversity and ecosystem services. In: Advances Ecol. Res., vol. 55. Academic Press, pp. 43–97.
Eulaers, I., Jaspers, V.L., Pinxten, R., Covaci, A., Eens, M., 2014. Legacy and current-use brominated flame retardants in the Barn Owl. Sci. Total Environ. 472, 454–462.
Ehiguese, F.O., Alam, M.R., Pintado-Herrera, M.G., Araújo, C.V., Martin-Diaz, M.L., 2020. Potential of environmental concentrations of the musks galaxolide and tonalide to induce oxidative stress and genotoxicity in the marine environment. Mar. Environ. Res. 160, 105019.
Franson, J.C., Pain, D.J., 2011. Lead in birds. In: Beyer, W.N., Meador, J.P. (Eds.), *Environmental Contaminants In Biota: Interpreting Tissue Concentrations* 2. CRC Press, Boca Raton, pp. 563–592.
Gago-Ferrero, P., Bletsou, A.A., Damalas, D.E., Aalizadeh, R., Alygizakis, N.A., Singer, H. P., et al., 2020. Wide-scope target screening of> 2000 emerging contaminants in wastewater samples with UPLC-Q-ToF-HRMS/MS and smart evaluation of its performance through the validation of 195 selected representative analytes. J. Hazard Mater. 387, 121712.
Gill, J.A., Langston, R.H.W., Alves, J.A., Atkinson, P.W., Bocher, P., Vieira, N.C., Crockford, N.J., Gelinaud, G., Groen, N., Gunnarsson, T.G., Hayhow, B., Hooijmeijer, J., Kentie, R., Kleijn, D., Lourenço, P.M., Masero, J.A., Meunier, F., Potts, P.M., Roodbergen, M., Schekkerman, H., Schroeder, J., Wymenga, E., Piersma, T., 2007. Contrasting trends in two Black-tailed Godwit populations: a review of causes and recommendations. Wader Study Group Bull. 114, 43–50.
Gkotsis, G., Nika, M.C., Nikolopoulou, V., Alygizakis, N., Bizani, E., Aalizadeh, R., Badry, A., Chadwick, E., Cincinelli, A., Claßen, D., Danielsson, S., Dekker, R., Duke, G., Drost, W., Glowacka, N., Göckener, B., Jansman, H.A.H., Juergens, M., Knopf, B., Koschorreck, J., Krone, O., Martellini, T., Movalli, P., Persson, S., Potter, E.D., Rohner, S., Roos, A., Rourke, E.O., Siebert, U., Treu, G., van den Brink, N.W., Walker, L.A., Williams, R., Slobodnik, J., Thomaidis, N.S., 2022. Assessment of contaminants of emerging concern in European apex predators and their prey by LC-QToF MS wide-scope target analysis. Environ. Int. 170, 107623.
Gkotsis, G., Nika, M.C., Athanasopoulou, A., Vasilatos, K., Alygizakis, N., Boschert, M., Osterauer, R., Hopker, K.-A., Thomaidis, N.S., 2023. Advanced throughput analytical strategies for the comprehensive HRMS screening of organic micropollutants in eggs of different bird species. Chemosphere 312, 137092.
González-Rubio, S., Ballesteros-Gómez, A., Asimakopoulos, A.G., Jaspers, V.L., 2021. A review on contaminants of emerging concern in European raptors (2002−2020). Sci. Total Environ. 760, 143337.
Goujon, E., Sta, C., Trivella, A., Goupil, P., Richard, C., Ledoigt, G., 2014. Genotoxicity of sulcotrione pesticide and photoproducts on Allium cepa root meristem. Pestic. Biochem. Physiol. 113, 47–54.
Groen, N.M., Kentie, R., de Goeij, P., Verheijen, B., Hooijmeijer, J.C., Piersma, T., 2012. A modern landscape ecology of Black-tailed Godwits: habitat selection in southwest 100 (1), 19–28.
Hendryx, M., Luo, J., 2022. Association between exposure to parabens and total mortality in US adults. Environ. Res. 205, 112415.
Hepworth-Warren, K.L., Alcott, C.J., 2014. Treatment and resolution of zilpaterol hydrochloride toxicity in a Quarter Horse gelding. Equine Vet. Educ. 26 (2), 81–85.
Hongthong, S., Leese, H.S., Allen, M.J., Chuck, C.J., 2021. Assessing the conversion of various nylon polymers in the hydrothermal liquefaction of macroalgae. Environments 8 (4). https://doi.org/10.3390/environments8040034.
Hoogenboom, R.L., Kotterman, M.J., Hoek-van Nieuwenhuizen, M., van der Lee, M.K., Mennes, W.C., Jeurissen, S.M., van Leeuwen, S.P., 2015. Dioxins, PCBs and heavy metals in Chinese mitten crabs from Dutch rivers and lakes. Chemosphere 123, 1–8.
Howison, R.A., Piersma, T., Kentie, R., Hooijmeijer, J.C., Olff, H., 2018. Quantifying landscape-level land-use intensity patterns through radar-based remote sensing. J. Appl. Ecol. 55 (3), 1276–1287.
Howison, R.A., Belting, H., Smart, J., Smart, M., Schuckard, R., Thorup, O., Piersma, T., International Wader Study Group, 2019. Een horizon vol weidevogels in Zuidwest-Friesland/Meadowbirds on the horizon of southwest Friesland. In: Internationale ecologische kennis die de landbouw in Zuidwest-Friesland met biodiversiteit verbindt/Harvesting international ecological knowledge to connect farming practices in the southwest of Friesland with biodiversity. International Wader Study Group, Workum.
Kannan, K., Corsolini, S., Falandysz, J., Oehme, G., Focardi, S., Giesy, J.P., 2002. Perlfluorooctanesulfonate and related fluorinated hydrocarbons in marine mammals, fishes, and birds from coasts of the Baltic and the Mediterranean Seas. Environ. Sci. Technol. 36, 3210–3216.
Kentie, R., Hooijmeijer, J.C.E.W., Trimbos, K.B., Groen, N.M., Piersma, T., 2013. Intensified agricultural use of grasslands reduces growth and survival of precocial shorebird chicks. J. Appl. Ecol. 50 (1), 243–251.
Kentie, R., Senner, N.R., Hooijmeijer, J.C.E.W., Márquez-Ferrando, R., Figuerola, J., Masero, J.A., Verhoeven, M.A., Piersma, T., 2016. Estimating the size of the Dutch breeding population of Continental Black-tailed Godwits from 2007–2015 using resighting data from spring staging sites. Ardea 114, 213–225. https://doi.org/10.5253/arde.v104i3.a7.
Kentie, R., Marquez-Ferrando, R., Figuerola, J., Gangoso, L., Hooijmeijer, J.C.E.W., Loonstra, A.H.J., Robin, F., Sarasa, M., Senner, N., Valkema, H., Verhoeven, M.A., Piersma, T., 2017. Does wintering north or south of the Sahara correlate with timing and breeding performance in black- tailed godwits? Ecol. Evol. 7, 2812–2820.
Kentie, R., Coulson, T., Hooijmeijer, J.C.E.W., Howison, R.A., Loonstra, A.H.J., Verhoeven, M.A., Both, C., Piersma, T., 2018. Warming springs and habitat alteration interact to impact timing of breeding and population dynamics in a migratory bird. Global Change Biol. 24, 5292–5303. https://doi.org/10.1111/gcb.14406.
Kim, D., Cui, R., Moon, J., Kwak, J.I., Kim, S.W., Kim, D., An, Y.J., 2018. Estimation of the soil hazardous concentration of methylparaben using a species sensitivity approach. Environ. Pollut 242, 1002–1009.
Kleijn, D., Berendse, F., Smit, R., Gilissen, N., 2001. Agri-environment schemes do not effectively protect biodiversity in Dutch agricultural landscapes. Nature 413 (6857), 723–725.
Kleijn, D., van Zuijlen, G.J., 2004. The conservation effects of meadow bird agreements on farmland in Zeeland, The Netherlands, in the period 1989–1995. Biol. Conserv. 117 (4), 443–451.
Kushlan, J.A., 1997. The conservation of wading birds. Colonial Waterbirds 129–137.
Laue, H., Kern, S., Badertscher, R.P., Ellis, G., Natsch, A., 2017. *p*-Alkyl-Benzoyl-CoA conjugates as relevant metabolites of aromatic aldehydes with rat testicular toxicity—studies leading to the design of a safer new fragrance chemical. Toxicol. Sci. 160 (2), 244–255. https://doi-org.naturalis.idm.oclc.org/10.1093/toxsci/kfx178.
Li, B., Li, H., Pang, X., Cui, K., Lin, J., Liu, F., Mu, W., 2018. Quaternary ammonium cationic surfactants increase bioactivity of indoxacarb on pests and toxicological risk to Daphnia magna. Ecotoxicol. Environ. Saf. 149, 190–196.

Lourenço, P.M., Piersma, T., 2008. Stopover ecology of Black-tailed Godwits Limosa limosa limosa in Portuguese rice fields: a guide on where to feed in winter. Hous. Theor. Soc. 55 (2), 194–202.
Lourenço, P.M., Kentie, R., Schroeder, J., Alves, J.A., Groen, N.M., Hooijmeijer, J.C., Piersma, T., 2010. Phenology, stopover dynamics and population size of migrating Black-tailed Godwits Limosa in Portuguese rice plantations. Ardea 98 (1), 35–42.
Lucia, M., Bocher, P., Cosson, R.P., Churlaud, C., Robin, F., Bustamante, P., 2012. Insight on trace element detoxification in the Black-tailed Godwit (Limosa limosa) through genetic, enzymatic and metallothionein analyses. Sci. Total Environ. 423, 73–83.
Lv, Z., Li, X., Wang, Y., Hu, X., An, J., 2021. Responses of soil microbial community to combination pollution of galaxolide and cadmium. Environ. Sci. Pollut. Res. 28 (40), 56247–56256.
Matsinos, Y.G., Wolff, W.F., 2003. An individual-oriented model for ecological risk assessment of wading birds. Ecol. Model. 170 (2–3), 471–478.
Merola, C., Perugini, M., Conte, A., Angelozzi, G., Bozzelli, M., Amorena, M., 2020. Embryotoxicity of methylparaben to zebrafish (Danio rerio) early-life stages. Comparative Biochem Physiol. C: Toxicol Pharmacol 236, 108792.
Meyer, J., Jaspers, V.L., Eens, M., de Coen, W., 2009. The relationship between perfluorinated chemical levels in the feathers and livers of birds from different trophic levels. Sci. Total Environ. 407 (22), 5894–5900.
Minomo, K., Ohtsuka, N., Nojiri, K., Hosono, S., Kawamura, K., 2011. Polychlorinated dibenzo-p-dioxins, dibenzofurans, and dioxin-like polychlorinated biphenyls in rice straw smoke and their origins in Japan. Chemosphere 84 (7), 950–956.
Mizukami-Murata, S., Suzuki, Y., Sakurai, K., Yamashita, H., 2021. Freshwater alga Raphidocelis subcapitata undergoes metabolomic changes in response to electrostatic adhesion by micrometer-sized nylon 6 particles. Environ. Sci. Pollut. Res. 28 (47), 66901–66913.
Monteleone, A., Schary, W., Fath, A., Wenzel, F., 2019. Validation of an extraction method for microplastics from human materials. Clin. Hemorheol. Microcirc. 73 (1), 203–217.
Movalli, P., Duke, G., Osborn, D., 2008. Introduction to monitoring for and with raptors. Ambio 37 (6), 395–396. https://doi.org/10.1579/0044-7447(2008)37[395:ITMFAW]2.0.CO;2.
Movalli, P., Dekker, R., Koschorreck, J., Treu, G., 2017. Bringing together raptor collections in Europe for contaminant research and monitoring in relation to chemicals regulations. Environ. Sci. Pollut. Res. 24 (31), 24057–24060. https://doi.org/10.1007/s11356-017-0096-x.
Movalli, P., Duke, G., Ramello, G., Dekker, R., Vrezec, A., Shore, R.F., García-Fernández, A., Wernham, C., Krone, O., Alygizakis, N., Badry, A., Barbagli, F., Biesmeijer, K., Boano, G., Bond, A.I., Choresh, Y., Christensen, J.B., Cincinelli, A., Danielsson, S., Dias, A., Dietz, R., Eens, M., Espín, S., Eulaers, I., Frahnert, S., Fuiz, T. I., Gkotsis, G., Glowacka, N., Gómez-Ramírez, P., Grotti, M., Guiraud, M., Hosner, P., Johansson, U., Jaspers, V.L.B., Kamminga, P., Koschorreck, J., Knopf, B., Kubin, E., LoBrutto, S., Lourenço, R., Martellini, T., Martínez-López, E., Mateo, R., Nika, M.-C., Nikolopoulou, V., Osborn, D., Pauwels, O., Pavia, M., Glória Pereira, M., Rüdel, H., Sanchez-Virosta, P., Slobodnik, J., Sonne, C., Thomaidis, N., Töpfer, T., Treu, G., Väinölä, R., Valkama, J., van der Mije, S., Vangeluwe, D., Warren, B.H., Woog, F., 2019. Progress on bringing together raptor collections in Europe for contaminant research and monitoring in relation to chemicals regulation. Environ. Sci. Pollut. Res. 26 (20), 20132–20136. https://doi.org/10.1007/s11356-017-0096-x.
Nikolopoulou, V., Alygizakis, N.A., Nika, M.C., Oswaldova, M., Oswald, P., Kostakis, M., et al., 2022. Screening of legacy and emerging substances in surface water, sediment, biota and groundwater samples collected in the Siverskyi Donets River Basin employing wide-scope target and suspect screening. Sci. Total Environ. 805, 150253.
Park, K., Barghi, M., Lim, J.E., Ko, H.M., Nam, H.Y., Lee, S.I., Moon, H.B., 2021. Assessment of regional and temporal trends in per-and polyfluoroalkyl substances using the Oriental Magpie (Pica serica) in Korea. Sci. Total Environ. 793, 148513.
Previšić, A., Vilenica, M., Vučković, N., Petrović, M., Rožman, M., 2021. Aquatic insects transfer pharmaceuticals and endocrine disruptors from aquatic to terrestrial ecosystems. Environ. Sci. Technol. 55 (6), 3736–3746.
Rattner, B.A., Hoffman, D.J., Melancon, M.J., Olsen, G.H., Schmidt, S.R., Parsons, K.C., 2000. Organochlorine and metal contaminant exposure and effects in hatching black-crowned night herons (Nycticorax nycticorax) in Delaware Bay. Archives of Environmental Contamination and Toxicology 39, 38–45.
Robuck, A.R., McCord, J.P., Strynar, M.J., Cantwell, M.G., Wiley, D.N., Lohmann, R., 2021. Tissue-specific distribution of legacy and novel per-and polyfluoroalkyl substances in juvenile seabirds. Environ. Sci. Technol. 8 (6), 457–462.
Roodbergen, M., Klok, C., van der Hout, A., 2008. Transfer of heavy metals in the food chain earthworm Black-tailed godwit (Limosa limosa): comparison of a polluted and a reference site in The Netherlands. Sci. Total Environ. 406 (3), 407–412.
Sakellarides, T.M., Konstantinou, I.K., Hela, D.G., Lambropoulou, D., Dimou, A., Albanis, T.A., 2006. Accumulation profiles of persistent organochlorines in liver and fat tissues of various waterbird species from Greece. Chemosphere 63 (8), 1392–1409.
Sampaio, F.G., Carra, M.L., Jonsson, C.M., Gonçalves, V.T., Dal'Bo, G., Nunes, K.S.D., Reyes, F.G.R., 2016. Effects of dietary exposure to sulfamethazine on the hematological parameters and hepatic oxidative stress biomarkers in Nile tilapia (Oreochromis niloticus). Bulletin of environmental contamination and toxicology 97, 528–535.Chicago.
Schroeder, J., Lourenço, P.M., van der Velde, M., Hooijmeijer, J.C., Both, C., Piersma, T., 2008. Sexual dimorphism in plumage and size in Black-tailed Godwits *Limosa limosa limosa*. Ardea 96 (1), 25–37.
Shi, Y.F., Zhang, L.L., Wu, Z.I., 2012. Inhibitory effects of pyrazoles on soil nitrification: effects of chemical structure. Fresenius Environ. Bull. 21 (6), 1385–1389.
Shore, R.F., Pereira, E., Walker, L.A., Thompson, D.R., 2011. Mercury in nonmarine birds and mammals. In: Beyer, W.N., Meador, J.P. (Eds.), *Environmental Contaminants In Biota: Interpreting Tissue Concentrations*. 2. CRC Press, Boca Raton, pp. 609–626.
Shore, R.F., Taggart, M.A., 2019. Population-level impacts of chemical contaminants on apex avian species. Curr. Opin. Environ. Sci. Health 11, 65–70.
Spyridakis, I., Tzanakakis, V.A., Pashalidis, I., Kalderis, D., Anastopoulos, I., 2022. Polyamide nylon 6 as a potential carrier of nitrate anions in aqueous environments. J. Mol. Liq. 352, 118706.
Strupp, C., Quesnot, N., Richert, L., Moore, J., Bomann, W.H., Singh, P., 2020. Weight of evidence and human relevance evaluation of the benfluralin mode of action in rodents (Part I): liver carcinogenesis. Regul. Toxicol. Pharmacol. 117, 104758.
Taggart, M.A., Richards, N., Kinney, C.A., 2016. Impacts of pharmaceuticals on terrestrial wildlife. In: Hester, R.E., Harrison, R.M. (Eds.), Issues Environ. Sci. Technol, vol. 41, pp. 216–254.
Tang, Y., Fan, Z., Yang, M., Zhang, S., Li, M., Fang, Y., Lie, J., Feng, X., 2022. Low concentrations of the antidepressant venlafaxine affect courtship behaviour and alter serotonin and dopamine systems in zebrafish (Danio rerio). Aquay. Toxicol. 204, 106082.
Timm, K., Myant, C., Spikes, H.A., Grunze, M., 2011. Particulate lubricants in cosmetic applications. Tribol. Int. 44 (12), 1695–1703.
Uemura, H., Arisawa, K., Hiyoshi, M., Satoh, H., Sumiyoshi, Y., Morinaga, K., Kodama, K., Suzuki, T., Nagai, M., Suzuki, T., 2008. PCDDs/PCDFs and dioxin-like PCBs: recent body burden levels and their determinants among general inhabitants in Japan. Chemosphere 73 (1), 30–37.
Verhoeven, M.A., Loonstra, A.H.J., McBride, A.D., Both, C., Senner, N.R., Piersma, T., 2020. Migration route, stopping sites, and non-breeding destinations of adult Black-tailed Godwits breeding in southwest Fryslân, The Netherlands. J. Ornithol. 162, 61–76.
Verreault, J., Letcher, R.J., Gentes, M.L., Braune, B.M., 2018. Unusually high Deca-BDE concentrations and new flame retardants in a Canadian Arctic top predator, the glaucous gull. Sci. Total Environ. 639, 977–987.
Xue, J., Kannan, K., 2016. Accumulation profiles of parabens and their metabolites in fish, black bear, and birds, including bald eagles and albatrosses. Environment international 94, 546–553.Chicago.
Zaccaroni, A., Niccoli, C., Andreani, G., Scaravelli, D., Ferrante, M., Lucisano, A., Isani, G., 2011. Trace metal concentration in wild avian species from Campania. Italy. Open Chem. 9 (1), 86–93.
Zhang, C.-X., Xi, J., Zhao, T.-P., Ma, Y.-X., Wang, X.-D., 2020. β-carbolines norharman and harman in vegetable oils in China. Food Addit. Contam.: B Surveill. 13 (3), 193–199.
Zhu, B.-R., Verkuil, Y.I., Conklin, J.R., Yang, A., Lei, W., Alves, J.A., Hassell, C.J., Dorofeev, D., Zhang, Z., Piersma, T., 2021. Discovery of a morphologically and genetically distinct population of black-tailed godwits in the East asian-australasian flyway. Ibis 163, 448–462.