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J Antimicrob Chemother 2016 doi:10.1093/jac/dkw245 Advance Access publication 20 June 2016

# Detection of the mcr-1 gene in Escherichia coli prevalent in the migratory bird species Larus argentatus

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Sir,

To the best of our knowledge, we describe the first known occurrence of the mcr-1 gene in Escherichia coli carried by a wild migratory bird—the European herring gull (Larus argentatus). Recently, the first plasmid-mediated colistin resistance determinant, mcr-1, has been identified in Enterobacteriaceae prevalent in humans, domestic animals and food. Colistin is a polymyxin antibiotic widely used in animal production and currently increasingly prescribed for therapeutic usage in human medicine, as a consequence of the spread of MDR Gram-negatives. The mcr-1 gene, encoding a phosphoethanolamine transferase, was first identified among Chinese enterobacterial isolates<sup>2</sup> and later on it was detected in some European countries including Denmark,<sup>3</sup> the UK,<sup>4,5</sup> Germany,<sup>6</sup> Belgium<sup>7</sup> and France.<sup>8</sup>

In the present case, the mcr-1 gene was detected in non-ESBLproducing E. coli. The study involved 22 ESBL-positive and 95 ESBL-negative E. coli isolates obtained from 160 European herring gulls tested for the presence of MDR. The study protocol was approved by the Lithuanian Environmental Protection Agency (protocol number 15.10-A4-8844). Among 117 isolates tested, 8 of them had elevated colistin MICs of 4-8 mg/L and a single isolate harboured the mcr-1 gene. The gene was detected by PCR following the protocol described previously. Faeces of birds were collected on Kaunas (Lithuania) city dump in January 2016. Rectal swabs were taken individually and inoculated on to MacConkey Agar (Thermo Scientific, UK) and Brilliance ESBL Agar (Thermo Scientific). Isolates were identified using biochemical testing followed by sequencing of 16S rRNA genes in the case of uncertain identification. MICs of antimicrobials were determined using Sensititre plates and automated system ARIS 2X (Thermo Scientific). Interpretation of results was performed according to EUCAST clinical breakpoints. 10 Sequencing of the mcr-1 gene was performed using a 3730 Series DNA Analyzer (Thermo Scientific). Sequences were analysed using Molecular Evolutionary Genetic Analysis software (version 6). The basic local alignment search tool was used for comparison of obtained sequences with the sequences presented in the database of the National Centre of Biotechnology Information. Migration routes of European herring gulls were assessed using data from the EU for Bird Ringing database and the Lithuanian Bird Ringing Centre.

The obtained results confirmed the identity of the mcr-1 gene sequence from E. coli isolated from the migratory bird with that of the mcr-1 gene from plasmid SHP45 described by Liu et al.<sup>2</sup> (GenBank accession no. KU341381.1). The isolate carrying the mcr-1 gene was resistant to ampicillin, ampicillin/sulbactam and colistin, and susceptible to all other antimicrobial agents

To the best of our knowledge, this is the first case of detection of the mcr-1 gene in bacteria from a migratory bird. European herring gulls according to their biology are considered as seabirds, although the birds more often are found on dumps where they have access to food waste. Non-appropriate management of medical, biological and food waste may influence spread of infectious agents including resistant microorganisms though the wild birds, particularly gulls. The natural habitat of European herring gulls includes northern, central, western and eastern Europe and the Baltic States. Some European herring gulls, particularly those resident in colder areas, migrate further south in winter. During the winter, these European herring gulls may be considered as vagrant birds as they periodically change their living places, which can be several hundred miles apart from each other. Figure 1 demonstrates the migration routes of European herring gulls after they had been ringed in Lithuania. According to data from the Lithuanian Bird Ringing Centre, juvenile European herring gulls ringed in the Baltic States were later observed in almost all European countries. The lifestyle of gulls allows them to carry and disseminate pathogenic and resistant microorganisms despite country borders. ESBL-producing E. coli detected in this study (22 of 160) also indicates a high prevalence of resistant bacteria within European herring gulls. Water contaminated by faeces of birds should be foreseen as an important risk factor for transmission of resistant bacteria.

#### **Acknowledgements**

We would like to acknowledge Ricardas Patapavicius (Head of the Lithuanian Bird Ringing Centre) for providing data on the migration routes of European herring gulls.

## Funding

The study was funded by a grant (SIT-06/2015) from the Research Council of Lithuania.



**Figure 1.** Main migration routes of European herring gulls from Lithuania to other European countries. European herring gulls ringed as juveniles in Lithuania were later observed in many European countries.

### **Transparency declarations**

None to declare.

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