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**Tetracycline-resistant *Escherichia coli* from American crocodiles (*Crocodylus acutus*) captured in aquaculture facilities in Costa Rica**

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**Abstract:**

**Background/Objective:** The use of antimicrobial substances influences the natural bacterial communities in the surrounding environment. Apex predators are exposed to both antimicrobial agents and the resistant microbes that have accumulated across the different trophic levels of related ecosystems. The aim of this study was to characterize tetracycline-resistant *Escherichia coli* isolated from American crocodiles (*Crocodilus acutus*) captured in a Costa Rican aquaculture farm held in captivity before release.

**Methods:** During release activities, cloacal swab samples were taken from 53 wild individuals and streaked on McConkey agar containing 16 µg/ml tetracycline. The presence of genetic resistance traits in *Escherichia coli* isolates were determined by whole-genome sequencing and bioinformatics analysis.

**Results:** A total of 24 swab samples contained tetracycline-resistant *E. coli* (45.3%). The isolates carried either the *tet(A)* (15/20), *tet(B)* (4/20), or *tet(C)* (1/20) genes. Furthermore, genetic sequences were detected among the isolates that demonstrate resistance mechanisms against ß-lactam antibiotics (*blaEC*: 20/20; *blaTEM1*: 1/20), sulfonamides (*sul2*: 3/20), fosfomycin (*glpT\_E448K*: 3/20), aminoglycosides (*aph(6)-Id,aph(3'')-Ib*: 2/20), phenicol (*floR*: 1/20), quinolones (*qnrS1*: 1/20) or trimethoprim (*dfrA14*: 1/20). Genome sequencing further revealed that a longer abode in the aquaculture premises may influence the occurrence of specific sequence types and the prevalence of antibiotic resistance carriage. Moreover, some isolates harbored virulence factors.

**Conclusion:** Considering the regional human-wildlife conflicts (e.g. bites) and the potential transmission whether from or into aquaculture facilities, the detection of mono- and multiresistant *E. coli* strains needs consideration by producers, local communities, and the national public health authorities.

**Keywords:** human-animal-environment interface, plasmid carriage, biosecurity, captivity