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Design

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Organic and inorganic copper feed formulations have a similar impact in the selection of copper tolerant and multidrug resistant *Enterococcus faecium* from poultry farms and meat available to consumers

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Abstract

Background: Copper-Cu is used as animal feed additive. Inorganic (ITM) or organic (OTM) trace minerals feed formulations with Cu are available (OTM with lower Cu concentrations), but their effect in the selection of Cu tolerant-CuT and multidrug-resistant bacteria-MDR bacteria is unexplored. The impact of ITM/OTM-feed in the occurrence of MDR-CuT *E. faecium*-Efm in poultry production chain was studied.

Materials/methods: Broiler feces [n=34; 18 poultry-houses/flocks (10000-64000 animals each); 7 intensive-farms] were collected in each farm from 2 separated poultry-houses using ITM or OTM, at 2-3 days of broilers life (P1) and before slaughter (P2) (2019-2020/Portugal). Meat (n=18) of the same broilers was also collected after slaughter (P3). Samples were plated in Slanetz-Bartley (SL) agar with/without 1mM-CuSO₄ (37°C/48h; anaerobiosis). Efm species, tcrB gene coding for CuT were studied by PCR and antibiotics/Cu susceptibility by disk diffusion/microdilution.

Results: 256 Efm were from feces-97% and meat-67% samples. Efm-tcrB⁺ were mostly found in SL+Cu plates (46% of samples vs 15%-SL), with similar rates between OTM and ITM (54% vs 46%). Efm-tcrB⁺ had an increasing trend between P1 and P2 (56% vs 81%) samples but significantly decrease at P3 (17%). Efm-tcrB⁺ had a MIC_{Cu}>12mM (96%), with most being MDR comparing to Efm-tcrB⁻ (98% vs 57%). Efm-tcrB⁺ were more resistant to tetracycline, erythromycin, quinupristin/dalfopristin or streptomycin than Efm-tcrB⁻ (100%/100%/85%/62% vs 63%/66%/63%/21%). MDR-Efm-tcrB⁺, including to the clinical-relevant ampicillin or aminoglycosides, were mostly found in feces (P1+P2:47% vs P3:11%), both in OTM and ITM samples (31% vs 38%).

Conclusions: MDR-Efm-tcrB⁺ were similarly detected independently of OTM/ITM feed at poultry-farms but they significantly decreased in carcasses for the consumer. The Cu role in the selection and persistence of MDR-Efm deserves more studies.

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