**Distribution of the *pco* operon among swine *Escherichia coli* from a controlled feeding trial**

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As one component of a study on alternatives to antibiotics, copper, zinc, and oregano oil (with negative controls and low- and high-dose tetracycline positive controls) were administered in feed to 400 pigs over a 49-day period. *Escherichia coli* were isolated from fecal samples on Days 0 and 28. The *pco* operon, a copper transport system, has been previously shown to increase tolerance to copper by active efflux. The prevalence of the operon was determined by PCR among 403 *E. coli* isolates, as well as the extended spectrum beta-lactamase genes $bla_{CMY}$ and $bla_{CTX\text{-}M}$, and tetracycline resistance genes *tet*(A) and *tet*(B). Thirty-four isolates (8%) were *pco*-positive, while 15% carried $bla_{CMY}$ and 3% carried $bla_{CTX\text{-}M}$. Isolates with and without the *pco* operon had virtually identical susceptibility to copper in the form of copper(II) sulfate, with MICs around 20mM; these MICs were unaffected by induction with lower concentrations of copper (1mM and 5mM). The expression of the *pco* operon was compared between several isolates, induced and uninduced, using reverse transcription of total RNA, and no significant differences were observed. There was no statistically significant association between the presence of the *pco* operon and any other treatment factor, including addition of 125 ppm copper in feed, though *pco* was negatively associated with the presence of $bla_{CMY}$. Targeted sequencing revealed the operon to be closely associated with a Tn7-like transposon, and was flanked by both chromosomal and plasmid-like DNA in different isolates. The *pco* operon also appears to be randomly distributed among various multi-locus sequence types, independently of genetic relationships. Whole genome analysis using next-generation sequencing of a subset of isolates is ongoing to further characterize the local genetic environment of the operon. In the absence of an associated copper resistance phenotype and of any selective effect of copper in feed, the significance of *pco* for co-selection of other antimicrobial resistance factors may be questionable. However, the association with a transposable element, as well as documented co-location with other antimicrobial resistance genes warrants further study.