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Does field application of manure from sulfadiazine treated pigs affect the abundance and transferability of resistance genes?

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The agricultural use of manure containing antibiotics for fertilization is assumed to promote the formation and spreading of resistance genes by mobile genetic elements such as plasmids which poses a risk for human health, e.g. by increasing resistance problems in human antibiotic therapy. The DFG research unit FOR566 aims at identifying key processes that control the fate and effects of veterinary medicines in soil. Sulfadiazine (SDZ), used as a model compound in this

project and belonging to the class of sulfonamides, is among the most widely used veterinary antibiotics in the EU. It is excreted largely unchanged by the animals and enters agricultural soils through the use of manure and slurry as fertilizer.

In the field study presented here, the effect of manure from pigs treated with sulfadiazine (SDZ-manure) on abundance and transferability of sulfonamide resistance genes *sul1* and *sul2* in the rhizosphere of maize and grass were compared to the effects in bulk soil. A significantly higher abundance of both *sul* genes was detected in the plots after application of SDZ-manure compared to manure from untreated pigs. The abundance

of *sul* genes relative to bacterial ribosomal genes was significantly lower in the rhizosphere than in bulk soil. However, only in SDZ-manure treated rhizosphere the sul gene abundance constantly deviated from control treatments with time after manuring, suggesting an ongoing antibiotic selection. Transferability of sulfonamide resistance was analysed by capturing resistance plasmids from soil communities into E. coli. Increased rates of plasmid capture were observed in samples from SDZ-manure treated bulk soil and rhizosphere of maize and grass. More than 97% of the captured plasmids belonged to the LowGC-type, giving further evidence for their important contribution to the environmental spread of antibiotic resistance.

In summary, SDZ-manure applied to the field significantly increased the abundance and transferability of antibiotic resistance. However, the effect in bulk soil and rhizosphere differed which needs to be considered in further risk assessments.