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Goulas, A; Belhadi, D; Descamps, A; Andremont, A; Benoit, P; Courtois, S; Dagot, C; Grall, N; Makowski, D; Nazaret, S; Nelieu, S; Patureau, D; Petit, F; Roose-Amsaleg, C; Vittecoq, M; Livoreil, B; Laouenan, C 2020 How effective are strategies to control the dissemination of antibiotic resistance in the environment? A systematic review Environmental Evidence 9, 1–32 10.1186/s13750-020-0187-x

Background and objective

Antibiotic resistance is a major concern for public and environmental health. The role played by the environment in disseminating resistance is increasingly considered, as well as its capacity for mitigation. The authors reviewed the literature on strategies to control dissemination of antibiotic-resistant bacteria (ARB), antibiotic resistance genes (ARG) and mobile genetic elements (MGE) in the environment. This systematic review focused on three main strategies: (i) restriction of antibiotic use (S1), (ii) treatments of liquid/solid matrices (S2) and (iii) management of natural environment (S3). Here we report only results regarding treatment of liquid/solid matrices.

Search strategy and selection criteria

The search for articles was conducted in English language with no restriction in date. The first search was conducted in nine publication databases in July 2017 (17th–21st, depending on the database): Web of Science, Pubmed, Scopus, DOAJ, JSTOR, Agricola, Ingenta Connect, AGRIS FAO and BioOne. Access to Web of Science, PubMed and Ingenta Connect has been allowed through INSERM/INIST institutional subscription, while that of Scopus thanks to a temporary account as a reviewer. Contrary to statements in protocol, the search in Wiley Online Library was not performed due to time constraints. he search in Drug Resistance Updates was not performed because our systematic review question did not correspond to the aims and scope of this journal (publication on drug resistance in infectious disease and cancer, novel drugs and strategies to overcome clinical drug resistance). A second search was made in Google Scholar (July 2017) and references obtained in the two first pages of results were checked for eligibility. Grey literature was searched both in English and French on 23 websites listed in the protocol. Researchers and private companies were contacted to ask for unpublished data. Grey literature was assessed using the same criteria as for articles. The authors' rationale was to focus on studies in which antibiotic resistance was measured in samples directly in contact with natural habitats, as this review is interested at minimizing risk of contamination. The authors excluded studies in which antibiotic resistance was measured in biological samples from humans, cultivated plants or domestic animals. Studies on ATBR in the hospital indoor environment, on infectious diseases, molecular or genetic characterization, and occurrence of pharmaceuticals, bacteria or virulence genes, without ATBR measurement were also excluded. To assess the internal validity of each study, sources of bias were determined through discussion with the review team. Only studies reporting measurements of antibiotic-resistant bacteria (ARB), antibiotic resistance genes (ARG) and mobile genetic elements (MGE) in environmental samples were included.

Data and analysis

Several random-effects models were fitted to the dataset by restricted maximum likelihood with R (version 3.5.1, package lme4). In each model, the study identifier was included as random effect and the empirical variances were used to weight the individual log ratio. Mean effect sizes and their associated 95% confidence intervals (CI) were estimated for the different types of organic treatments defined above.

Number of papers	Population	Intervention	Comparator	Outcome	Quality score
98	Livestock waste and sewage sludge	Six types of treatments were considered: aerobic and anaerobic digestion, aerobic and anaerobic lagoon storage, composting, drying, pasteurization and pile storage.	No treatment	Metric: Relative abundance of antibiotic resistance markers (e.g., number of antibiotic resistance genes copies in total microbial biomass estimated by number of 16S rRNA copies in environmental sample).; Effect size: Logarithm of ratio of the considered metrics in the intervention to the considered metrics in the control	100

Results

- The overall effect size for composting was not significant since high heterogeneity was obtained with only three studies.Heterogeneity between the three studies could be explained by differences of study scale, temperature and initial abundance of ARB.Heterogeneity between the three studies could be explained by differences of study scale, temperature and initial abundance of ARB.
- Concerning anaerobic digestion, although overall effect was not significant, all studies reported a decrease of ARB whatever the antibiotic used in susceptibility tests or experimental conditions including temperature. It could be a trend for higher ARB reduction with thermophilic temperatures, but more studies are needed to confirm this hypothesis.
- In spite of the heterogeneity in published results, the meta-analysis showed that composting and drying were efficient treatments to reduce the relative abundance of ARG and MGE in organic waste, by 84% [65%; 93%] and 98% [80%; 100%], respectively. The relative abundance of ARG/MGE was significantly reduced when composting was applied to livestock effluents, contrary to sludge composting.
- The overall effect of anaerobic digestion (on the complete dataset) was not statistically significant (51% reduction [− 2%; 77%]) ($p = 0.068$). However, the treatment was efficient on for livestock effluents.
- Some of these studies also showed a decrease of resistance as the distance from the wastewater discharge point increases, related to a natural resilience capacity of aquatic environments. Concerning wildlife, nine medium/high validity studies showed that animals exposed to anthropogenic activities carried more ARB.

Factors influencing effect sizes

- Temperature : For composting and aerobic treatments, the maximum temperature ranged between 34 and 86 °C and its effect on the reduction of ARG/MGE relative abundance was not significant ($p = 0.54$). For anaerobic digestion, the maximum temperature (mostly fixed during treatment) ranged between 10 and 63 °C and its effect was significant ($p = 0.012$).
- Type of manure : The composting was significantly efficient for chicken and pig manure as well as for other manure mixtures. Variability observed for cow manure could be caused by the presence of co-substrates like wheat straw in variable proportions because a highest straw proportion may lead to manure dilution and so a lower initial ARG/MGE abundance level.

Conclusion

The authors obtained significant results for composting, drying and a (non-significant) trend for anaerobic digestion in reducing ARG/MGE relative abundance, when organic waste treatments were compared together in the same model. Thermophilic treatments showed greater reductions in ARG/MGE relative abundance than mesophilic ones after anaerobic digestion. Consequently, treatments with thermophilic phases should be implemented before the application of organic waste products on agricultural soils. Pasteurization resulted in non-significant effect, due to a large variability and low number of observations (N=4). Anaerobic or aerobic lagoon storage and solid manure pile storage have no significant effect on antibiotic resistance genes.