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Comparison of different approaches to antibiotic restriction in food-producing animals: stratified results from a systematic review and meta-analysis

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ABSTRACT

Background We have previously reported, in a systematic review of 181 studies, that restriction of antibiotic use in food-producing animals is associated with a reduction in antibiotic-resistant bacterial isolates. While informative, that report did not concretely specify whether different types of restriction are associated with differential effectiveness in reducing resistance. We undertook a sub-analysis of the systematic review to address this question.

Methods We created a classification scheme of different approaches to antibiotic restriction: (1) complete restriction; (2) single antibiotic-class restriction; (3) single antibiotic restriction; (4) all non-therapeutic use restriction; (5) growth promoter and prophylaxis restriction; (6) growth promoter restriction and (7) other/undetermined. All studies in the original systematic review that were amenable to meta-analysis were included into this substudy and coded by intervention type. Meta-analyses were conducted using random effects models, stratified by intervention type.

Results A total of 127 studies were included. The most frequently studied intervention type was complete restriction (n=51), followed by restriction of non-therapeutic (n=33) and growth promoter (n=19) indications. None examined growth promoter and prophylaxis restrictions together. Three and seven studies examined single antibiotic-class and single antibiotic restrictions, respectively; these two intervention types were not significantly associated with reductions in antibiotic resistance. Though complete restrictions were associated with a 15% reduction in antibiotic resistance, less prohibitive approaches also demonstrated reduction in antibiotic resistance of 9%–30%.

Conclusion Broad interventions that restrict global antibiotic use appear to be more effective in reducing antibiotic resistance compared with restrictions that narrowly target one specific antibiotic or antibiotic class. Importantly, interventions that allow for therapeutic antibiotic use appear similarly effective compared with those that restrict all uses of antibiotics, suggesting that complete bans are not necessary. These findings directly inform the creation of specific policies to restrict antibiotic use in food-producing animals.

Key questions

What is already known?

- Antimicrobial resistance (AMR) is a threat to public health, with the Tripartite Collaboration (WHO, the Food and Agriculture Organisation of the United Nations and the World Organisation for Animal Health) calling for a One Health approach to address this crisis.
- A recent systematic review and meta-analysis suggested that, in general, interventions that restrict antibiotic use in food-producing animals are effective in reducing AMR in these animals and in certain subgroups of human population, though whether certain types of interventions are more effective than others remains unknown.

What are the new findings?

- A wide spectrum of interventions, from limiting antibiotics for growth promoter or feed additive purposes only to limiting all uses of antibiotics (including for therapy), were associated with a 9%–30% absolute reduction in antibiotic resistance.
- Interventions that restrict the use of only one antibiotic or antibiotic class were not significantly associated with a reduction in antibiotic resistance.

What do the new findings imply?

- Highly targeted interventions that limit the use of only a single antibiotic or antibiotic class may have limited effectiveness in reducing antibiotic resistance.
- Interventions that broadly target overall antibiotic use or that restrict the use of multiple antibiotic classes are recommended as these appear to be associated with reductions in antibiotic resistance, though a complete restriction of antibiotics does not appear to be necessary.

INTRODUCTION

Antimicrobial resistance (AMR) has been recognised as a threat to public health worldwide, being associated with increased morbidity, mortality and societal costs.[1–4] It is

estimated that by 2050, AMR will contribute to 10 million deaths per year, a 2%–3.5% reduction in gross domestic product, and cost $100 trillion US$ worldwide.[5] Over-prescription and unnecessary non-prescription antibiotic use are the main contributors to increase AMR in humans.[6] Widespread antibiotic use in agriculture and aquaculture also likely plays a role,[7–9] especially as many of the antibiotics used in animals are the same, or are in the same class, as antibiotics used in humans.[8 10–12] The WHO, the Food and Agriculture Organisation of the United Nations and the World Organisation for Animal Health, known as the Tripartite Collaboration, have called for a One Health approach, with recognition that animal, human and environmental health are linked, to address the problem of AMR.[13]

A systematic review conducted by our group showed that interventions that aimed to reduce antibiotic use in food-producing animals are associated with a reduction in AMR in these animals, as well as in certain subgroups of the human population (particularly those with direct contact with animals).[14] These findings were critical in demonstrating that reducing antibiotic use in agriculture is an effective avenue by which to combat the growing problem of AMR worldwide. However, the studies included in the systematic review used many different approaches to reduce and/or to restrict antibiotic use. Our report did not address whether different types and extent of antibiotic restriction lead to different levels of reduction in antibiotic resistance. That is, though antibiotic restrictions appear, in a broad sense, to be effective in reducing resistance, it is unclear whether specific types of restrictions are more effective than others.

Antibiotics can be used in food-producing animals for therapeutic purposes (ie, to treat existing infectious disease), for disease control within a herd or flock, and for non-therapeutic purposes.[15] This results in a wide spectrum of possible approaches to antibiotic restriction. The least restrictive approaches would include those that prohibit the use of only one antibiotic or antibiotic class, and those that restrict the use of antibiotics for specific non-therapeutic indications only such as for growth promotion. On the opposite end of the spectrum is the complete prohibition of the use of all antibiotics, for any indication. With the least restrictive approaches, there is risk of increased use of other antibiotics in the place of the restricted drug(s), thereby raising the question of whether such measures actually reduce AMR.[16 17] On the other hand, while antibiotic-free strategies may be effective in reducing AMR, the inability to use antibiotics, even to treat diagnosed clinical infectious diseases, is detrimental for animal production and economics as well as to animal welfare.[18 19]

The development of national and international guidelines and policies requires greater detail about the effectiveness of different interventions so that specific recommendations can be made as to what type of antibiotic restrictions should be implemented. We were commissioned by the WHO to undertake a subanalysis of the original systematic review and meta-analysis to explore the associations between different interventions that restrict antibiotic use in food-producing animals and antibiotic resistance in these animals, to inform the WHO Guidelines on the use of antibiotics in food-producing animals.[20] Our findings provide crucial insights into the type and extent of antibiotic restriction that optimises desired effects of reducing AMR.

METHODS

The methods for the broader systematic review and meta-analysis, of which this is a substudy, have been described in detail in a prior publication.[14] The systematic review and meta-analysis was conducted following a predetermined protocol and in accordance with Preferred Reporting Items for Systematic Reviews and Meta-Analyses reporting standards.[21] Ethics approval was not required, as the study is based on a review of published literature.

Search strategy

The search strategy consisted of controlled vocabulary and keywords, under three themes: animal populations of interest (theme 1), resistance to antibiotics (theme 2)[22 23] and interventions to restrict antibiotic use (theme 3). These three themes were combined with the Boolean operator 'AND'. Electronic databases were searched in initially searched in July 2016, and again in January 2017. Databases included Agricola (1970–present), AGRIS (http://agris.fao.org), BIOSIS Previews (1980–present), CAB Abstracts (1910–present), MEDLINE (1946–present), EMBASE (1974–present), Global Index Medicus (http://www.globalhealthlibrary.net; non-MEDLINE indices included AIM (AFRO), LILACS (AMRO/PAHO), IMEMR (EMRO), IMSEAR (SEARO), WPRIM (WPRO), WHOLIS (KMS) and SciELO), ProQuest Dissertations and Science Citation Index (1899–present). No limits were placed based on publication date or language. An update to the search was conducted on 8 July 2019, focusing on the electronic databases MEDLINE, EMBASE, CAB Abstracts, and AGRIS.

Reference lists of included articles (published 2010 onward) were manually searched. Grey literature searching included websites of relevant health agencies, professional associations and other specialised databases. The WHO Guideline Development Group as well as experts in antimicrobial use and resistance, veterinary medicine and animal health policy were contacted to identify potential missed, ongoing or unpublished studies.

Abstract screening and full-text review

Two authors independently reviewed all identified titles and abstracts for eligibility. Only articles reporting original research that described an intervention aimed to reduce antibiotic use in animals and described antibiotic resistance in animals or humans were selected for full-text review. At the full-text review stage, articles were

retained and ultimately included into this substudy if they were original research meeting the following inclusion criteria: (1) population studied included food-producing animals (within the classifications of avian, swine, bovine, caprine, camel, equine, rabbit, ovine, fish, bees, molluscs and crustaceans); (2) interventions restricted the use of antibiotics in food-producing animals; (3) presence of a comparator group without antibiotic use restrictions (historical comparators were considered eligible); (4) outcomes reported phenotypic antibiotic resistance in bacteria in food-producing animals and (5) sufficient data reported to calculate risk differences (RDs) in proportion of isolates with antibiotic resistance in the intervention versus the comparator group (to allow for meta-analysis).

Data extraction and assessment of individual study quality

Two authors (KT and NC) extracted data from each included study using a predesigned form. Data extracted included study design, country, animal characteristics, sampling characteristics, description of intervention, description of comparator, bacteria investigated, and prevalence of antibiotic resistance in intervention and comparator groups. The same authors independently assessed the methodological quality of each study based on pre-specified study quality indicators adapted from the Downs and Black checklist.[24] The results of the quality assessment are described in a prior publication.[14]

Patient and public involvement

Due to the nature of the research question, which was defined by the WHO and which used data from our prior review of published literature, patients were not involved in this study.

Creation of an intervention classification scheme

The WHO commissioned this study to inform the development of Guidelines on this topic. The initial request for a classification scheme therefore originated from the WHO Advisory Group on Integrated Surveillance of Antimicrobial Resistance (WHO AGISAR) Guideline Development Group committee members. Because there is no widely accepted classification scheme to categorise interventions that restrict and/or reduce antibiotic use, we developed one from the ground-up, based on the types of interventions found in the literature. The preliminary categories that were developed were then presented to WHO AGISAR for input and feedback, and then iteratively refined.

We began by establishing standard terminology to be used in this classification scheme, as different jurisdictions may use terminology differently. For example, the definition for metaphylaxis provided by the US Department of Agriculture includes the prophylactic use of antibiotics in healthy animals to prevent disease (even when there are no clinically affected animals present),[25] whereas the definition from the European Medicines Agency does not.[26] Furthermore, some consider metaphylaxis to be a therapeutic indication of antibiotic use (ie, it is considered to be 'group treatment' of animals)[26] while others note that antibiotic use is only therapeutic if administered in clinically infected animals.[27] The latter definition would therefore consider metaphylaxis not to be therapy, but rather disease prevention. We consulted the veterinary experts on the study team along with the WHO Guideline Development Group for definitions for the terms 'antibiotic growth promotor', 'metaphylaxis', 'prophylaxis', 'non-therapeutic antibiotic use' and 'therapeutic antibiotic use'. Consensus was reached for the definitions provided in table 1, which were then used in our classification scheme.

In total, we created seven categories of interventions (table 2): (1) complete restriction; (2) restriction of use of a single antibiotic class; (3) restriction of use of a single antibiotic; (4) all non-therapeutic use restriction; (5) growth promoter and prophylaxis restriction; (6) growth promoter restriction and (7) other/undetermined. Each intervention was assigned only one category. If a study included more than one intervention, then each intervention was classified separately based on

Table 1 Definitions for terms used in the classification scheme for interventions

Terminology	Definition
Antibiotic growth promoter	Administration of subtherapeutic doses of antibiotics to stimulate growth in animals or to increase feed efficiency.[27 167]
Non-therapeutic antibiotic use	Administration of antibiotics to animals without identifiable infectious disease.[167] This includes antibiotic use for growth promotion, disease prophylaxis and metaphylaxis.
Metaphylaxis	Treatment of a group of animals without evidence of disease, but which are likely in an incubation phase, due to being in close contact with clinically diseased animals.[26]
Prophylaxis	Administration of antibiotics to animals at high risk of infectious disease (but without current disease and where there is no known disease in the herd or flock).[167] Prophylaxis is commonly used when environmental conditions or changes portend increased risk for infection. Examples of such conditions include transport of animals and confining animals to small, crowded spaces.[167]
Therapeutic antibiotic use	Administration of antibiotics to treat animals with clinical evidence of infectious disease only.[27 167]

Table 2 Classification of interventions that restrict antibiotic use in food-producing animals

Category	Description
Complete restriction	Restriction on the use of all antibiotics
Single antibiotic-class restriction	Restriction on the use of one class of antibiotics, for all indications of use
Single antibiotic restriction	Restriction on the use of a single individual antibiotic, for all indications of use
All non-therapeutic use restriction	Restriction on the use of antibiotics for all non-therapeutic indications including growth promotion, prophylaxis and metaphylaxis (treatment of diseased animals permitted only)
Growth promoter and prophylaxis restriction	Restriction on the use of antibiotics for the non-therapeutic indications of growth promotion and prophylaxis (treatment and metaphylaxis permitted)
Growth promoter restriction	Restriction on the use of antibiotics for purposes of growth promotion only (treatment, metaphylaxis and prophylaxis permitted)
Other/undetermined	Inability to classify the intervention type into one of the above categories, or where the indication for antibiotic use that is targeted by the intervention is not specified

the above approach. The 'growth promoter restriction' category did not require the restriction of all available antibiotic growth promoters. That is, interventions that restricted one or more growth promoters were eligible to be included in this category, even if there was residual use of other non-restricted growth promoters (eg, ionophores and flavophospholipols). The 'other/undetermined' category captures studies that did not specify the type of antibiotic use or indication that was targeted in the antibiotic restriction strategy. This includes studies, for example, that compare regions or farms using 'more' versus 'less' antibiotics with no indication of what is specifically targeted or described, or studies that assess the impact of reducing antibiotic use in a jurisdiction without delineating how this is achieved. An algorithm was created to ensure reproducibility in how interventions are classified into the different categories (figure 1).

We anticipated that some studies may use labels to define the intervention, without further description. Such labels might include 'organic' or 'antibiotic-free' production. We established a set of decision rules a priori. These included the following:

a. Interventions involving organic production in the USA were classified as 'complete restriction', as organic certification in the USA specifies that animals are raised without any exposure to antibiotics.[28]
b. Interventions involving organic production in Europe were classified as 'all non-therapeutic use restriction' as the European Commission on organic production specifies that animals are allowed limited antibiotics for therapeutic purposes.[29 30]
c. We referred to organic certification standards, if cited, for interventions involving organic production in countries outside of the USA and Europe.
d. Interventions where no such certifications exist (eg, 'antibiotic-free', 'pasture' or 'free range') were classified as 'undetermined/other' unless sufficient detail was provided for classification into any other category.

Outcome measure

Antibiotic resistance was considered a dichotomous outcome, as classified by the individual primary studies. Intermediate susceptibility was considered susceptible. Absolute RDs were calculated for each individual antibiotic in each study by subtracting the proportion of resistant isolates in the control group from the proportion in the intervention group.

Meta-analysis

All meta-analyses were stratified by intervention type. To allow for meaningful and adequately powered analysis within each intervention stratum, all included studies were pooled, regardless of the animal populations, sample types or bacterial species studied. A single effect estimate (absolute RD) was generated for each study by conducting within-study meta-analysis using random effects models.

Absolute RDs across all studies were then pooled using DerSimonian and Laird random-effects models. This method was chosen due to the known clinical heterogeneity across studies, with studies from different regions examining different animal populations, sample types and bacteria.[31] A lower prevalence of antibiotic resistance in the intervention group compared with the control group would result in a negative pooled absolute RD. Recognising that RDs must be interpreted in the context of baseline prevalence of antibiotic resistance, we conducted additional meta-analysis, pooling the prevalence of antibiotic resistance in the comparator groups, stratified by intervention type, using random-effects models. Heterogeneity across studies was evaluated using the I^2 statistic.[32 33] Meta-regression was conducted, with each intervention type being a covariate. A joint test for all covariates was conducted, to test whether intervention type was associated with the size of the outcome effect (ie, antibiotic resistance).[34]

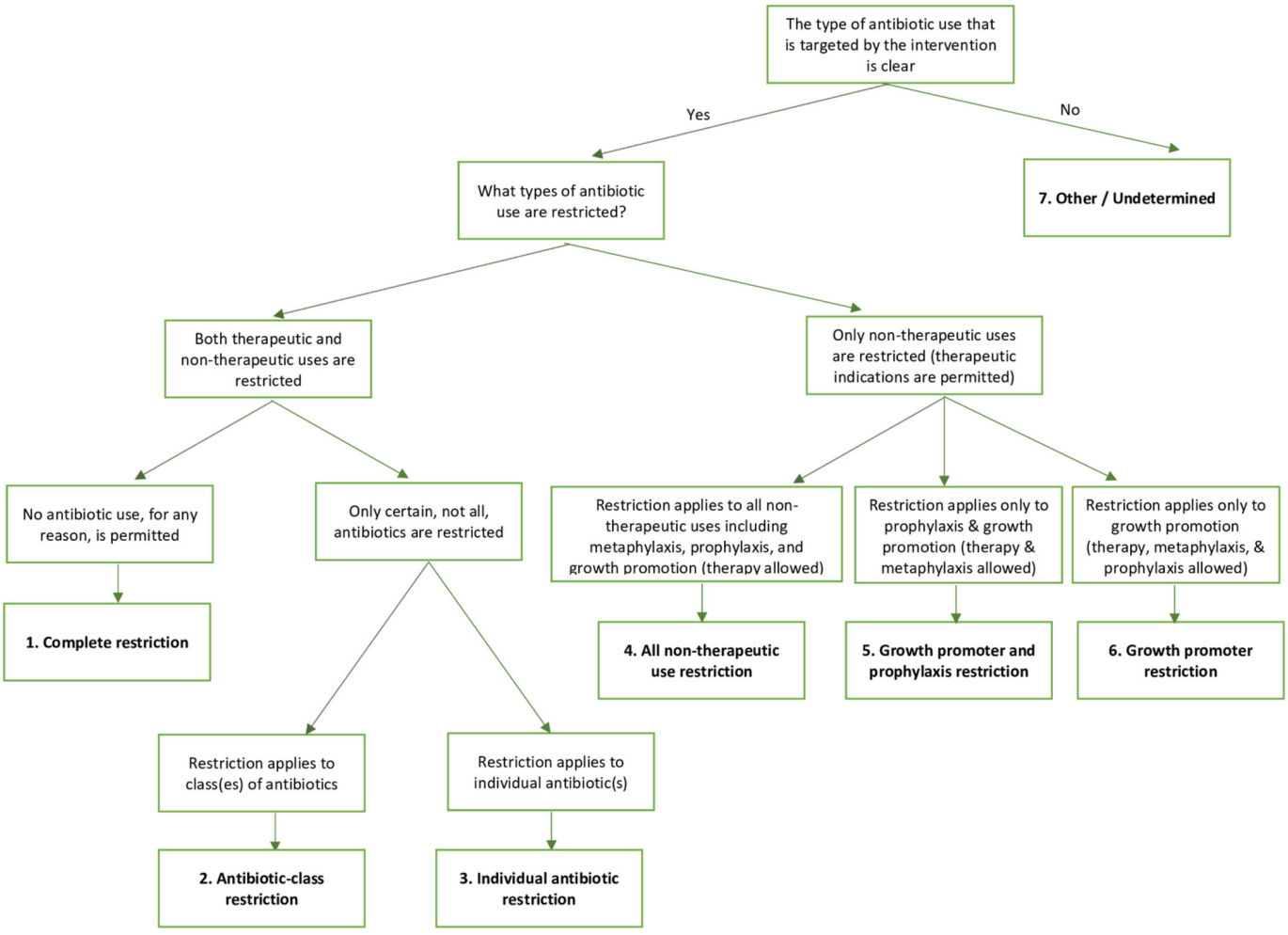

Figure 1 Algorithm for the classification of interventions to restrict antibiotic use in food-producing animals.

Role of the funding source

The WHO was involved in both the original systematic review and meta-analysis, as well as this substudy. They were involved in developing the research question, the study design and the study protocol. They had no involvement in data extraction or interpretation of findings. The authors have been given permission by the WHO to publish this article.

RESULTS

Identification of studies

The initial search strategy identified 9008 citations from electronic databases. An additional 56 studies were identified by contacting experts, and another 82 by searching reference lists. After removal of duplicates, 5945 records underwent title and abstract review. Of these, 5559 records were not relevant to the research objective, and 386 full-text articles were reviewed. A total of 181 studies were included in the larger original systematic review. Of these, two were excluded as they examined AMR outcomes in humans but not animals, 17 were excluded as they reported presence of resistant genetic elements with no phenotypic resistance outcomes, and 48 were excluded as there were insufficient data to allow for meta-analysis. Therefore, 114 studies from the original systematic review were included in this substudy. In addition, an update to the search was conducted July 2019, at which time a total of 1208 new records were identified. After duplicates were removed, 703 underwent title and abstract review. Of these, 659 were excluded as were not relevant to the research objective, and 44 full-text articles were reviewed, of which 13 ultimately met criteria to be included into this study. In total, 127 studies were included into this systematic review and meta-analysis (figure 2).

Study characteristics

Of the 127 studies, 51 restricted all use of antibiotics (complete restriction),[35–85] three restricted use of a single antibiotic class[86–88] and seven restricted use of a single specific antibiotic.[89–95] In all, 33 studies restricted use of antibiotics for all non-therapeutic purposes,[48 96–127] and 19 restricted antibiotic growth promoters only.[128–146] A total of 21 studies were classified into the 'other/undetermined' category.[52 53 83 100 137 147–162] Of note, seven studies consisted of two different interventions and were therefore included into two separate categories.[48 52 53 83 100 122 137]

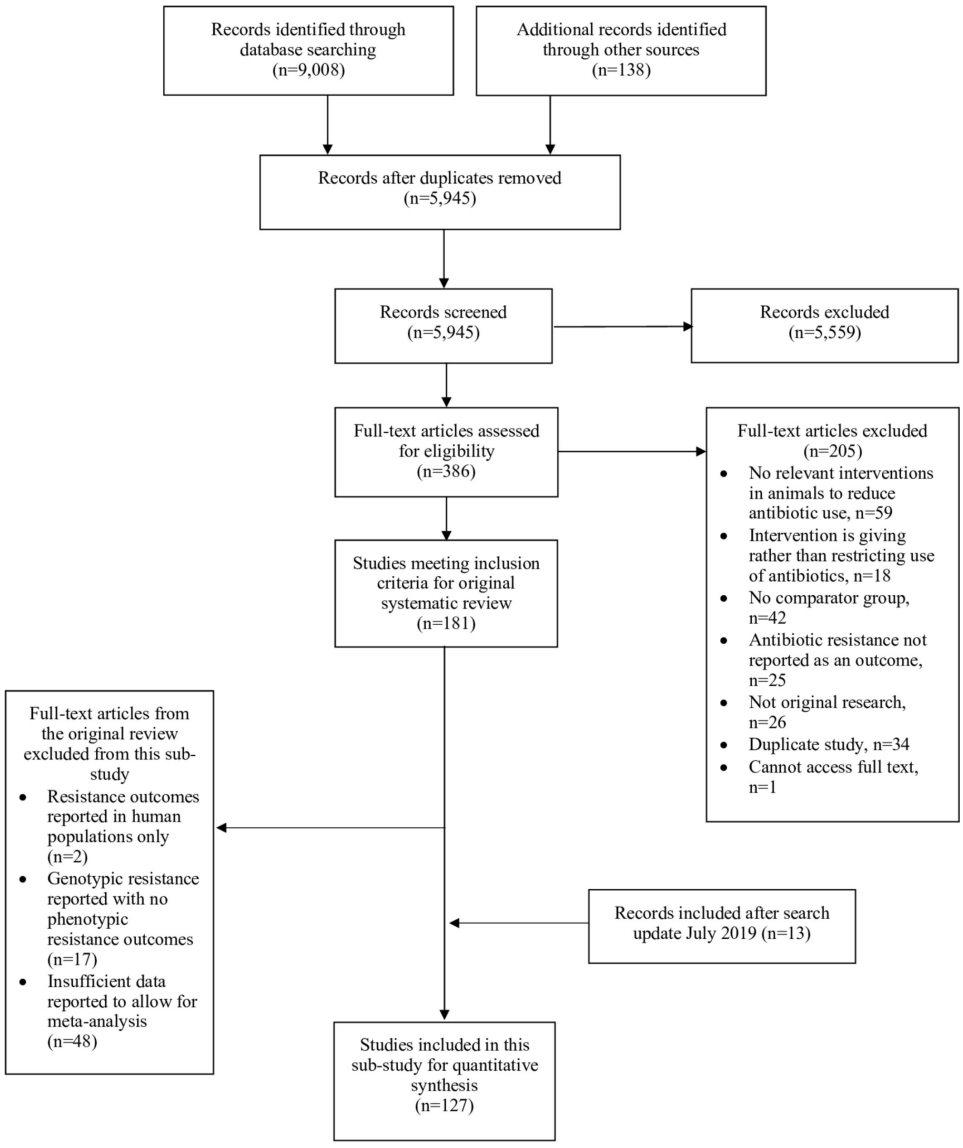

Figure 2 PreferredReporting Items for Systematic Reviews and Meta-Analyses flow diagram of the study selection process.

A summary of study characteristics is found in table 3. In total, 114 of the 127 studies were journal articles. There were eight dissertations and six meeting abstracts/conference proceedings. The majority had a cross-sectional design. Poultry (n=69) was the most commonly studied animal population, followed by swine (n=42) and dairy cattle (n=19). Antibiotic resistance was most commonly assessed in the bacterial group Enterobacteriaceae. In all, 65 studies were from North America and 53 were from Europe. Few study populations were from Asia (n=6), Africa (n=1), Australasia (n=2) and South America (n=1). Detailed study characteristics for individual studies can be found in a prior publication,[14] as well as in online supplementary appendix 1 table S1.

Meta-analysis by intervention category

All intervention types were associated with a significantly lower pooled risk of antibiotic resistance in the intervention group compared with the comparator group except for single antibiotic-class and single antibiotic restrictions (RD −0.02, 95% CI −0.10, 0.05 and RD −0.11, 95% CI −0.21, 0.01 respectively, see table 4). The pooled risk reduction of antibiotic resistance was greatest for growth promoter restrictions (RD −0.30, 95% CI −0.42 to -0.17). That is, for interventions that restricted the use of antibiotic growth promoters, there was a 30% reduction in the proportion of isolates that were antibiotic resistant in the intervention group compared with the comparator group. Similarly, there was a 10% and 15% reduction in the proportion of antibiotic-resistant isolates for interventions that restricted all non-therapeutic uses of antibiotics and interventions that completely restricted all (non-therapeutic and therapeutic) uses of antibiotics, respectively. The I^2 for each intervention stratum ranged between 89.0% and 98.5%, suggesting the presence of considerable heterogeneity. The meta-regression joint p value was 0.046, suggesting that the type of intervention significantly affected the magnitude of reduction in antibiotic resistance.

Table 3 Summary of study characteristics

Study characteristic (n)	Complete restriction (n=51) n (%)	Antibiotic-class restriction (n=3) n (%)	Individual antibiotic restriction (n=7) n(%)	All non-therapeutic use restriction (n=33) n (%)	Growth promoter restriction (n=19) n (%)	Other/ undetermined (n=21) n(%)	Total number of studies n=127
Type of article							
Journal article	43 (84.3)	3 (100.0)	7 (87.5)	31 (93.9)	19 (100.0)	16 (76.2)	114
Abstract only	4 (7.8)	–	–	1 (3.0)	–	3 (14.3)	6
Dissertation	4 (7.8)	–	1 (12.5)	1 (3.0)	–	2 (9.5)	8
Study design							
Non-randomised controlled trial	–	–	1 (14.3)	–	–	–	1
Cross-sectional	45 (88.2)	1 (33.3)	2 (28.6)	28 (84.8)	8 (42.1)	16 (76.2)	95
Longitudinal	6 (11.8)	2 (66.7)	4 (57.1)	5 (15.2)	11 (57.9)	5 (23.8)	31
Geographical region where intervention was implemented*							
North America	46 (90.2)	1 (33.3)	6 (85.7)	3 (9.1)	–	13 (61.9)	65
Europe	4 (7.8)	2 (66.7)	–	28 (84.8)	16 (84.2)	7 (33.3)	53
Asia	–	–	1 (14.3)	3 (9.1)	2 (10.5)	–	6
Australasia	1 (2.0)	–	–	–	–	1 (4.8)	2
Africa	–	–	–	–	1 (5.3)	–	1
South America	1 (2.0)	–	–	–	–	–	1
Population studied†							
Beef cattle	4 (7.8)	–	–	3 (9.1)	1 (5.3)	6 (28.6)	14
Dairy cattle	10 (19.6)	–	1 (14.3)	9 (27.3)	–	–	19
Poultry	24 (47.1)	2 (66.7)	5 (71.4)	13 (39.4)	16 (84.2)	13 (61.9)	69
Swine	17 (33.3)	2 (66.7)	1 (14.3)	9 (27.3)	10 (52.6)	5 (23.8)	42
Goats	2 (3.9)	–	–	1	–	–	3
Sample studied†							
Faeces/cloaca/caeca	33 (64.7)	2 (66.7)	6 (85.7)	12 (36.4)	18 (94.7)	11 (52.4)	77
Meat or carcass	16 (31.4)	1 (33.3)	2 (28.6)	10 (30.3)	4 (21.1)	11 (52.4)	42
Milk	7 (13.7)	–	–	10 (30.3)	–	–	16
Eggs	2 (3.9)	–	–	3 (9.1)	–	–	5
Nasal swabs	2 (3.9)	–	–	1 (3.0)	–	3 (14.3)	6
Bacteria studied†							
Campylobacter spp.	12 (23.5)	2 (66.7)	1 (14.3)	4 (12.1)	2 (10.5)	3 (14.3)	23
Enterococcus spp.	7 (13.7)	–	–	4 (12.1)	14 (73.7)	5 (23.8)	29
Staphylococcus spp.	8 (15.7)		–	14 (42.4)	–	8 (38.1)	29
Enterobacteriaceae	25 (49.0)	1 (33.3)	7 (100.0)	20 (60.6)	3 (15.8)	10 (47.6)	63
Other	4 (7.8)	–	–	6 (18.2)	–	2 (9.5)	11

*One study included intervention group samples from Denmark and the USA and was therefore counted twice.
†Categories are not mutually exclusive and studies can be included in more than one category.

Pooled proportions of antibiotic resistance in comparator groups

The pooled proportion of bacterial isolates with antibiotic resistance in comparator groups was lowest for studies that single antibiotic-class restrictions (pooled proportion 0.163, 95% CI 0.075 to 0.252, see table 4), and highest for studies examining interventions that restricted growth promoter use only (pooled proportion 0.492, 95% CI 0.261 to 0.723). The pooled proportions for complete restriction, all non-therapeutic use restriction and other/undetermined restriction were similar, between 0.32 and 0.34.

DISCUSSION

Though our broader systematic review and meta-analysis was important in bringing to light the effectiveness of antibiotic use restrictions on decreasing antibiotic resistance in food-producing animals, what has remained unknown until now is how to best implement this broad

Table 4 Meta-analysis stratified by intervention category

Intervention category*	Number of studies	Baseline prevalence of AMR (95% CI)†	Pooled absolute risk difference (95% CI)
Complete restriction	51	0.320 (0.165 to 0.468)	−0.15 (−0.18 to −0.12)
Single antibiotic-class restriction	3	0.163 (0.075 to 0.252)	−0.02 (−0.10 to 0.05)
Single antibiotic restriction	7	0.405 (0.027 to 0.784)	−0.11 (−0.21 to 0.01)
All non-therapeutic use restriction	33	0.322 (0.076 to 0.568)	−0.10 (−0.13 to −0.08)
Growth promoter restriction	19	0.492 (0.261 to 0.723)	−0.30 (−0.42 to −0.17)
Other/undetermined	21	0.338 (0.082 to 0.593)	−0.09 (−0.13 to −0.06)

*Meta-regression joint p-value=0.046.
†Pooled proportion of resistance in the comparator group.
AMR, antimicrobial resistance.

principle into practice and policy. This subanalysis plays a critical role in providing answers that can guide antibiotic use strategies in food-producing animals.

We demonstrate that highly targeted interventions limiting the use of single antibiotics or a single class of antibiotics are unlikely to be effective in reducing overall AMR. One reason for this finding may be that the use of the restricted antibiotic(s) is simply replaced by other antibiotics, such that there is no overall reduction in antibiotic use. This phenomenon was seen in Denmark. After the ban on the antibiotic growth promoter avoparcin, there was increased use of other growth promoters, including tylosin and virginiamycin, in its place.[131] Furthermore, there may be continued resistance to certain antibiotic classes even after selected classes have been banned or restricted because of co-selection. Because genes that encode resistance to different antibiotics may be linked (ie, carried on the same mobile genetic element), the continued use of just one of these antibiotics is sufficient to select for all of the linked resistance mechanisms to the different antibiotics.[163] This phenomenon was described in pigs where macrolide and glycopeptide resistance genes were linked. In this case, the ban of avoparcin did not result in reduced glycopeptide resistance, due to continued macrolide use.[164 165]

Conversely, a complete ban on the use of all antibiotics in food-producing animals does not appear to be necessary. Though antibiotic-free practices were associated with a 15% reduction in antibiotic resistance, less prohibitive practices are associated with similar reductions. Given that complete restrictions do not appear superior in this regard, and with the added economic, production and ethical challenges of such practices, complete bans are not recommended. Beyond this, it is more difficult to ascertain whether certain less-restrictive types of interventions are superior to others.

At first glance, interventions that restrict antibiotic growth promoters appear to be most effective at reducing AMR in food-producing animals (RD −0.30, 95% CI −0.42 to 0.17). However, growth promoter bans are often the first types of restrictions implemented; other interventions such as those limiting other non-therapeutic uses of antibiotics or all uses of antibiotics tend to be later interventions that are implemented after growth promoter bans or after other efforts to reduce antibiotic use are already in place. The large effect of antibiotic growth promoter bans relative to those of other interventions may therefore be due to the different comparator groups across the different interventions. Lending support to this hypothesis is that growth promoter ban studies tended to be published earlier (median year of publication 2001, IQR 2000–2004) compared with studies examining all other types of interventions (median 2010, IQR 2006–2015). Further support is provided through stratified meta-analysis of baseline proportions of isolates demonstrating antibiotic resistance. As predicted, the pooled baseline proportion of antibiotic resistance for growth promoter ban studies was higher compared with non-therapeutic antibiotic restriction and complete restriction studies (49% vs 32%). The smaller effect size for non-therapeutic restriction studies may therefore be explained, at least in part, by the lower baseline risk of antibiotic resistance (resulting in smaller RDs even if relative effects of the intervention are as large as the ones seen with growth promoter ban studies) and/or the smaller incremental benefit to antibiotic restriction once strategies to ban growth promotion claims on medically important antibiotics are already in place. We therefore cannot conclude that restrictions that target antibiotic growth promoters alone are more effective in reducing AMR compared with restrictions that target non-therapeutic indications more broadly. On the other hand, we have demonstrated that antibiotic growth promoter bans are effective in reducing AMR and therefore recommend that these be implemented on a global scale.

There are limitations to this systematic review. First, the comparison among intervention types through stratified analysis is inferior to comparison through head-to-head randomised controlled trials. However, such head-to-head randomised comparisons of different antibiotic restriction strategies do not exist in the primary literature. Furthermore, our stratified analysis findings are powerful especially as the differences in outcome effect across intervention types are consistent with prior experience and are biologically plausible (particularly the finding that very narrow restrictions are ineffective in

reducing AMR while broader restrictions are). Second, there is known clinical heterogeneity across studies, with different countries, livestock production sectors, animal groups and resistance to different bacterial species included. Despite this, our original systematic review and meta-analysis demonstrated consistency in findings across many different layers of stratification, suggesting the presence of an overall effect. Third, we were limited in our classification of interventions by the lack of detailed description of interventions within primary studies. Similarly, because the majority of studies did not provide any description of the implementation process, we were not able to assess how the quality of implementation may affect the effectiveness of interventions in reducing AMR. Our analysis suggests that well-implemented interventions that have national certification standards (eg, for organic production) may be more effective than interventions that have similar claims but no such standard (eg, 'antibiotic-free' products). The former was categorised as 'complete restriction' (if undertaken in the USA), which was associated with a 15% reduction in antibiotic resistance, while the latter was classified in the 'other/undetermined' category, which was associated with a 9% reduction. A more in-depth analysis, though, could not be completed without more information and description about implementation of each intervention in the primary studies. Lastly, the vast majority of studies originated from either North America or Europe. Generalisability of these findings to other jurisdictions may be limited, particularly in low-income countries where there may be limited access to veterinarians, less investment in biosecurity[166] and different antimicrobial use patterns.

Though we previously found that interventions that restrict antibiotic use in food-producing animals in general are effective in reducing AMR,[14] the practical applications were limited due to the broad nature of the research question and analyses. It has been unclear until now which specific interventions should or should not be recommended to achieve the goal of reducing AMR. This substudy provides insight to these policy-relevant questions. We show that broad interventions that restrict the use of a full spectrum of antibiotic classes are needed. We also show, however, that complete bans on all antibiotic use are not necessary, as judicious use of antibiotics (such as for the treatment of clinical disease in affected animals) does not appear to hinder efforts to reduce AMR. These findings have directly informed WHO Guidelines on use of medically important antimicrobials in food-producing animals,[20] and are directly relevant to public health policy globally.

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REFERENCES

1. de Kraker MEA, Davey PG, Grundmann H, *et al*. Mortality and hospital stay associated with resistant Staphylococcus aureus and Escherichia coli bacteremia: estimating the burden of antibiotic resistance in Europe. *PLoS Med* 2011;8:e1001104.
2. de Kraker MEA, Wolkewitz M, Davey PG, *et al*. Burden of antimicrobial resistance in European hospitals: excess mortality and length of hospital stay associated with bloodstream infections due to Escherichia coli resistant to third-generation cephalosporins. *J Antimicrob Chemother* 2011;66:398–407.
3. Cosgrove SE. The relationship between antimicrobial resistance and patient outcomes: mortality, length of hospital stay, and health care costs. *Clin Infect Dis* 2006;42:S82–9.
4. Cosgrove SE, Carmeli Y. The impact of antimicrobial resistance on health and economic outcomes. *Clin Infect Dis* 2003;36:1433–7.
5. O'Neill J. *The Review on Antimicrobial Resistance: Tackling drug-resistant infections globally - Final report and Recommendations*. UK, 2016.
6. Laxminarayan R, Duse A, Wattal C, *et al*. Antibiotic resistance-the need for global solutions. *Lancet Infect Dis* 2013;13:1057–98.
7. World Health Organization. *Global principles for the containment of antimicrobial resistance in animals intended for food: report of a*

who consultation with the participation of the food and agriculture organization of the United nations and the. Geneva, Switzerland: Office International des Epizooties, 2000.
8. World Health Organization. *Joint FAO/OIE/WHO expert workshop on non-human antimicrobial usage and antimicrobial resistance: scientific asssessment*. Geneva, Switzerland, 2003.
9. World Health Organization. *Critically important antibacterial agents for human medicine for risk management strategies of non-human use: report of a who Working group consultation*. Canberra, Australia, 2005.
10. World Health Organization. *Antimicrobial use in aquaculture and antimicrobial resistance. Report of a joint FAO/OIE/WHO expert consultation on antimicrobial use in aquaculture and antimicrobial resistance*. Seoul, Republic of Korea, 2006.
11. Tuševljak N, Dutil L, Rajić A, *et al*. Antimicrobial use and resistance in aquaculture: findings of a globally administered survey of aquaculture-allied professionals. *Zoonoses Public Health* 2013;60:426–36.
12. Food and Agriculture Organization of the United Nations. *Joint FAO/WHO/OIE expert meeting on critically important antimicrobials*. Rome, 2008: 60.
13. World Health Organization, Food and Agriculture Organization of the United Nations, World Organisation for Animal Health. *A manual for developing national action plans*. Geneva, 2016.
14. Tang KL, Caffrey NP, Nóbrega DB, *et al*. Restricting the use of antibiotics in food-producing animals and its associations with antibiotic resistance in food-producing animals and human beings: a systematic review and meta-analysis. *Lancet Planet Health* 2017;1:e316–27.
15. Landers TF, Cohen B, Wittum TE, *et al*. A review of antibiotic use in food animals: perspective, policy, and potential. *Public Health Rep* 2012;127:4–22.
16. Casewell M, Friis C, Marco E, *et al*. The European ban on growth-promoting antibiotics and emerging consequences for human and animal health. *J Antimicrob Chemother* 2003;52:159–61.
17. Cogliani C, Goossens H, Greko C. Restricting antimicrobial use in food animals: lessons from Europe. *Microbe* 2011;6:274–9.
18. Karavolias J, Salois MJ, Baker KT, *et al*. Raised without antibiotics: impact on animal welfare and implications for food policy. *Transl Anim Sci* 2018;2:337–48.
19. Phillips I, Casewell M, Cox T, *et al*. Does the use of antibiotics in food animals pose a risk to human health? A critical review of published data. *J Antimicrob Chemother* 2004;53:28–52.
20. World Health Organization. Who guidelines on use of medically important antimicrobials in food-producing animals, Geneva, 2017. Available: http://apps.who.int/iris/bitstream/handle/10665/258970/9789241550130-eng.pdf;jsessionid=FC6969336920B3D0DA150C31EF36D25B?sequence=1 [Accessed 20 Dec 2018].
21. Moher D, Liberati A, Tetzlaff J, *et al*. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. *Ann Intern Med* 2009;151:264–9.
22. WHO Advisory Group on Integrated Surveillance of Antimicrobial Resistance (AGISAR). Critically important antimicrobials for human medicine, 2011. Available: http://apps.who.int/iris/bitstream/10665/77376/1/9789241504485_eng.pdf [Accessed 11 Dec 2018].
23. World Organisation for Animal Health (OIE).. OIE list of antimicrobial agents of veterinary importance, 2015. Available: http://www.oie.int/fileadmin/Home/eng/Our_scientific_expertise/docs/pdf/Eng_OIE_List_antimicrobials_May2015.pdf [Accessed 30 Jun 2016].
24. Downs SH, Black N. The feasibility of creating a checklist for the assessment of the methodological quality both of randomised and non-randomised studies of health care interventions. *J Epidemiol Community Health* 1998;52:377–84.
25. U.S. Department of Agriculture. Feedlot 2011, part IV: health and health management on U.S. feedlots with a capacity of 1,000 or more head Fort Collins, Colorado, 2013. Available: https://www.aphis.usda.gov/animal_health/nahms/feedlot/downloads/feedlot2011/Feed11_dr_PartIV.pdf [Accessed 18 Sep 2018].
26. European Medicines Agency. Guideline for the demonstration of efficacy for veterinary medicinal products containing antimicrobial substances London, United Kingdom, 2016. Available: https://www.ema.europa.eu/documents/scientific-guideline/final-guideline-demonstration-efficacy-veterinary-medicinal-products-containing-antimicrobial_en.pdf [Accessed 18 Sep 2018].
27. Codex Alimentarius. Code of practice to minimize and contain antimicrobial resistance, CAC/RCP 61-2005: food and agriculture organization of the United nations / World Health organization, 2005. Available: http://www.fao.org/fao-who-codexalimentarius/sh-proxy/fr/?lnk=1&url=https%253A%252F%252Fworkspace.fao.org%252Fsites%252Fcodex%252FStandards%252FCAC%2BRCP%2B61-2005%252FCXP_061e.pdf [Accessed 7 Feb 2018].
28. U.S. Department of Agriculture. Organic livestock requirements, 2013. Available: https://www.ams.usda.gov/sites/default/files/media/Organic%20Livestock%20Requirements.pdf
29. European Union Commission. Commission regulation (EC) NO 889/2008: laying down detailed rules for the implementation of Council regulation (EC) NO 834/2007 on organic production and labelling of organic products with regard to organic production, labelling and control 2008.
30. European Union Commission. Agriculture and rural development: organic farming, 2017. Available: http://ec.europa.eu/agriculture/organic/eu-policy/eu-rules-on-production/livestock_en [Accessed 27 Jan 2017].
31. DerSimonian R, Kacker R. Random-effects model for meta-analysis of clinical trials: an update. *Contemp Clin Trials* 2007;28:105–14.
32. Higgins JPT, Thompson SG. Quantifying heterogeneity in a meta-analysis. *Stat Med* 2002;21:1539–58.
33. Higgins JPT, Thompson SG, Deeks JJ, *et al*. Measuring inconsistency in meta-analyses. *BMJ* 2003;327:557–60.
34. Harbord RM, Higgins JPT. Meta-Regression in Stata. *Stata J* 2008;8:493–519.
35. Aarestrup FM. Occurrence of glycopeptide resistance among *Enterococcus faecium* isolates from conventional and ecological poultry farms. *Microb Drug Resist* 1995;1:255–7.
36. Abdalrahman L, Stanley A, Wells H, *et al*. Isolation, virulence, and antimicrobial resistance of methicillin-resistant Staphylococcus aureus (MRSA) and methicillin sensitive Staphylococcus aureus (MSSA) strains from Oklahoma retail poultry meats. *Int J Environ Res Public Health* 2015;12:6148–61.
37. Antimicrobial resistance of enteric bacteria among ceftiofur treated and non-antimicrobial treated co-mingled pasture beef cows. *4Th ASM conference on antimicrobial resistance in zoonotic bacteria and foodborne pathogens*. Washington, United States, 2015.
38. Alali WQ, Thakur S, Berghaus RD, *et al*. Prevalence and distribution of *Salmonella* in organic and conventional broiler poultry farms. *Foodborne Pathog Dis* 2010;7:1363–71.
39. Bailey MA, Taylor RM, Brar JS, *et al*. Prevalence and antimicrobial resistance of *Campylobacter* from antibiotic-free broilers during organic and conventional processing. *Poult Sci* 2019;98:1447–54.
40. Bombyk RAM, Bykowski AL, Draper CE, *et al*. Comparison of types and antimicrobial susceptibility of Staphylococcus from conventional and organic dairies in west-central Minnesota, USA. *J Appl Microbiol* 2008;104:1726–31.
41. Bunner CA, Norby B, Bartlett PC, *et al*. Prevalence and pattern of antimicrobial susceptibility in *Escherichia coli* isolated from pigs reared under antimicrobial-free and conventional production methods. *J Am Vet Med Assoc* 2007;231:275–83.
42. Cho S, Fossler CP, Diez-Gonzalez F, *et al*. Antimicrobial Susceptibility of Shiga Toxin-Producing *Escherichia coli* Isolated from Organic Dairy Farms, Conventional Dairy Farms, and County Fairs in Minnesota. *Foodborne Pathog Dis* 2007;4:178–86.
43. Cicconi-Hogan KM, Belomestnykh N, Gamroth M, *et al*. Short communication: prevalence of methicillin resistance in coagulase-negative staphylococci and Staphylococcus aureus isolated from bulk milk on organic and conventional dairy farms in the United States. *J Dairy Sci* 2014;97:2959–64.
44. Cui S. Detection and characterization of Escherichia coli O157:H7 and Salmonella in food [Ph. *D.]. University of Maryland, College Park* 2004.
45. Cui S, Ge B, Zheng J, *et al*. Prevalence and antimicrobial resistance of Campylobacter spp. and Salmonella serovars in organic chickens from Maryland retail stores. *Appl Environ Microbiol* 2005;71:4108–11.
46. Cuny C, Friedrich AW, Witte W. Absence of livestock-associated methicillin-resistant Staphylococcus aureus clonal complex CC398 as a nasal colonizer of pigs raised in an alternative system. *Appl Environ Microbiol* 2012;78:1296–7.
47. Gebreyes WA, Thakur S, Morrow WEM. Comparison of prevalence, antimicrobial resistance, and occurrence of multidrug-resistant Salmonella in antimicrobial-free and conventional pig production. *J Food Prot* 2006;69:743–8.
48. Gellin G, Langlois BE, Dawson KA, *et al*. Antibiotic resistance of gram-negative enteric bacteria from pigs in three herds with different histories of antibiotic exposure. *Appl Environ Microbiol* 1989;55.
49. Halbert LW, Kaneene JB, Linz J, *et al*. Genetic mechanisms contributing to reduced tetracycline susceptibility of Campylobacter isolated from organic and conventional dairy farms in the midwestern and northeastern United States. *J Food Prot* 2006;69:482–8.
50. Halbert LW, Kaneene JB, Ruegg PL, *et al*. Evaluation of antimicrobial susceptibility patterns in *Campylobacter* spp isolated

from dairy cattle and farms managed organically and conventionally in the midwestern and northeastern United States. *J Am Vet Med Assoc* 2006;228:1074–81.
51. Han F, Lestari SI, Pu S, *et al*. Prevalence and Antimicrobial Resistance Among *Campylobacter* spp. in Louisiana Retail Chickens After the Enrofloxacin Ban. *Foodborne Pathog Dis* 2009;6:163–71.
52. Joseph S, Sapkota A, Cullen P, *et al*. Reduced resistance to antibiotics among Salmonella spp. recovered from U.S. organic poultry farms. antimicrobial resistance in zoonotic bacteria and foodborne pathogens in animals, humans and the environment. *American Society for Microbiology* 2008;17.
53. Joseph SW, Paramadhas R, Cullen P, *et al*. Reduced resistance to antibiotics among Enterococcus faecium of organic poultry farm origin. Abstracts of the Interscience conference on antimicrobial agents and chemotherapy 2007;47:95–6.
54. Kassem II, Kehinde O, Kumar A, *et al*. Antimicrobial-Resistant *Campylobacter* in Organically and Conventionally Raised Layer Chickens. *Foodborne Pathog Dis* 2017;14:29–34.
55. Keelara S, Scott HM, Morrow WM, *et al*. Longitudinal study of distributions of similar antimicrobial-resistant Salmonella serovars in pigs and their environment in two distinct swine production systems. *Appl Environ Microbiol* 2013;79:5167–78.
56. Kieke AL, Borchardt MA, Kieke BA, *et al*. Use of Streptogramin Growth Promoters in Poultry and Isolation of Streptogramin-Resistant *Enterococcus faecium* from Humans. *J Infect Dis* 2006;194:1200–8.
57. Langlois BE, Cromwell GL, Stahly TS, *et al*. Antibiotic resistance of fecal coliforms after long-term withdrawal of therapeutic and subtherapeutic antibiotic use in a swine herd. *Appl Environ Microbiol* 1983;46:1433–4.
58. Langlois BE, Dawson K, Cromwell G, *et al*. Antibiotic resistance in pigs following a 13 year ban. *J Anim Sci* 1986;62(Suppl. 3):18–32.
59. Lestari SIKA, Han F, Wang FEI, *et al*. Prevalence and antimicrobial resistance of Salmonella serovars in conventional and organic chickens from Louisiana retail stores. *J Food Prot* 2009;72:1165–72.
60. Lou R. *Dietary mannan-oligosaccharide as an approach for altering prevalence of antibiotic resistance and distribution of tetracycline resistance determinants in fecal bacteria from swine [Ph.D]*. University of Kentucky, 1995.
61. Luangtongkum T, Morishita TY, Ison AJ, *et al*. Effect of conventional and organic production practices on the prevalence and antimicrobial resistance of Campylobacter spp. in poultry. *Appl Environ Microbiol* 2006;72:3600–7.
62. Nulsen MF, Mor MB, Lawton DEB. Antibiotic resistance among indicator bacteria isolated from healthy pigs in New Zealand. *N Z Vet J* 2008;56:29–35.
63. Peng M, Salaheen S, Almario JA, *et al*. Prevalence and antibiotic resistance pattern of *Salmonella* serovars in integrated crop-livestock farms and their products sold in local markets. *Environ Microbiol* 2016;18:1654–65.
64. Pettey EA. *Comparison of antibiotic susceptibility characteristics of fecal lactobacilli and the distribution of tetracycline resistance genes on two swine farms with different histories of antibiotic use [Ph.D]*. University of Kentucky, 2008.
65. Pol M, Ruegg PL. Relationship between antimicrobial drug usage and antimicrobial susceptibility of gram-positive mastitis pathogens. *J Dairy Sci* 2007;90:262–73.
66. Price LB, Johnson E, Vailes R, *et al*. Fluoroquinolone-Resistant *Campylobacter* Isolates from Conventional and Antibiotic-Free Chicken Products. *Environ Health Perspect* 2005;113:557–60.
67. Price LB, Lackey LG, Vailes R, *et al*. The Persistence of Fluoroquinolone-Resistant *Campylobacter* in Poultry Production. *Environ Health Perspect* 2007;115:1035–9.
68. Ray KA, Warnick LD, Mitchell RM, *et al*. Antimicrobial susceptibility of Salmonella from organic and conventional dairy farms. *J Dairy Sci* 2006;89:2038–50.
69. Rollo SN, Norby B, Bartlett PC, *et al*. Prevalence and patterns of antimicrobial resistance in *Campylobacter* spp isolated from pigs reared under antimicrobial-free and conventional production methods in eight states in the Midwestern United States. *J Am Vet Med Assoc* 2010;236:201–10.
70. Rossa LS, EvR S, Diez DC, *et al*. Antimicrobial resistance and occurrence of indicator and pathogenic bacteria in organic and conventional chicken meat: comparative study. *Biotemas* 2013;26:211–20.
71. Salaheen S, Peng M, Biswas D. Ecological dynamics of Campylobacter in integrated mixed crop-livestock farms and its prevalence and survival ability in post-harvest products. *Zoonoses Public Health* 2016;13.
72. Sanchez HM. *Antibiotic resistance in bacteria isolated from commercial meat samples and air samples near agricultural sites [Ph.D*. Los Angeles: University of California, 2015.
73. Sapkota AR, Hulet RM, Zhang G, *et al*. Lower prevalence of antibiotic-resistant enterococci on U.S. conventional poultry farms that transitioned to organic practices. *Environ Health Perspect* 2011;119:1622–8.
74. Sapkota AR, Kim A, Hulet RM, *et al*. Trends in the prevalence and antibiotic-resistance of Salmonella after conventional poultry farms transition to organic practices. *Abstr Gen Meet Am Soc Microbiol* 2010;110.
75. Sapkota AR, Kinney EL, George A, *et al*. Lower prevalence of antibiotic-resistant Salmonella on large-scale U.S. conventional poultry farms that transitioned to organic practices. *Sci Total Environ* 2014;476-477:387–92.
76. Sato K, Bartlett PC, Saeed MA. Antimicrobial susceptibility of Escherichia coli isolates from dairy farms using organic versus conventional production methods. *J Am Vet Med Assoc* 2005;226:589–94.
77. Sato K, Bennedsgaard TW, Bartlett PC, *et al*. Comparison of antimicrobial susceptibility of Staphylococcus aureus isolated from bulk tank milk in organic and conventional dairy herds in the midwestern United States and Denmark. *J Food Prot* 2004;67:1104–10.
78. Siemon CE, Bahnson PB, Gebreyes WA. Comparative investigation of prevalence and antimicrobial resistance of Salmonella between pasture and conventionally reared poultry. *Avian Dis* 2007;51:112–7.
79. Smith TC, Gebreyes WA, Abley MJ, *et al*. Methicillin-Resistant Staphylococcus aureus in pigs and farm workers on conventional and antibiotic-free swine farms in the USA. *PLoS One* 2013;8:e63704.
80. Thakur S, Gebreyes WA. Prevalence and antimicrobial resistance of Campylobacter in antimicrobial-free and conventional pig production systems. *J Food Prot* 2005;68:2402–10.
81. Tikofsky LL, Barlow JW, Santisteban C, *et al*. A Comparison of antimicrobial susceptibility patterns for *Staphylococcus aureus* in organic and conventional dairy herds. *Microbial Drug Resistance* 2003;9(supplement 1):39–45.
82. Vikram A, Rovira P, Agga GE, *et al*. Impact of “Raised without Antibiotics” beef cattle production practices on occurrences of antimicrobial resistance. *Appl Environ Microbiol* 2017;83.
83. Wanninger S, Donati M, Di Francesco A, *et al*. Selective pressure promotes tetracycline resistance of Chlamydia suis in fattening pigs. *PLoS One* 2016;11:e0166917.
84. Zhang J, Massow A, Stanley M, *et al*. Contamination rates and antimicrobial resistance in *Enterococcus* spp., *Escherichia coli*, and *Salmonella* isolated from "no antibiotics added"-labeled chicken products. *Foodborne Pathog Dis* 2011;8:1147–52.
85. Zwonitzer MR, Soupir ML, Jarboe LR, *et al*. Quantifying attachment and antibiotic resistance of from conventional and organic swine manure. *J Environ Qual* 2016;45:609–17.
86. Agersø Y, Aarestrup FM. Voluntary ban on cephalosporin use in Danish pig production has effectively reduced extended-spectrum cephalosporinase-producing Escherichia coli in slaughter pigs. *J Antimicrob Chemother* 2013;68:569–72.
87. Gallay A, Prouzet-Mauléon V, Kempf I, *et al*. Campylobacter antimicrobial drug resistance among humans, broiler chickens, and pigs, France. *Emerg Infect Dis* 2007;13:259–66.
88. Nannapaneni R, Hanning I, Wiggins KC, *et al*. Ciprofloxacin-resistant *Campylobacter* persists in raw retail chicken after the fluoroquinolone ban. *Food Addit Contam Part A* 2009;26:1348–53.
89. Agunos A, Arsenault RK, Avery BP, *et al*. Changes in antimicrobial resistance levels among *Escherichia coli, Salmonella,* and *Campylobacter* in Ontario broiler chickens between 2003 and 2015. *Can J Vet Res* 2018;82:163–77.
90. Bauer-Garland J, Frye JG, Gray JT, *et al*. Transmission of Salmonella enterica serotype typhimurium in poultry with and without antimicrobial selective pressure. *J Appl Microbiol* 2006;101:1301–8.
91. Dutil L, Irwin R, Finley R, *et al*. Ceftiofur Resistance in *Salmonella enterica* Serovar Heidelberg from Chicken Meat and Humans, Canada. *Emerg Infect Dis* 2010;16:48–54.
92. Hiki M, Kawanishi M, Abo H, *et al*. Decreased Resistance to Broad-Spectrum Cephalosporin in *Escherichia coli* from Healthy Broilers at Farms in Japan After Voluntary Withdrawal of Ceftiofur. *Foodborne Pathog Dis* 2015;12:639–43.
93. Patchanee P. *Epidemiology of Salmonella enterica related to swine production system and food safety [Ph.D]*. The Ohio State University, 2008.

94. Tragesser LA, Wittum TE, Funk JA, *et al*. Association between ceftiofur use and isolation of *Escherichia coli* with reduced susceptibility to ceftriaxone from fecal samples of dairy cows. *Am J Vet Res* 2006;67:1696–700.
95. Verrette L, Fairbrother JM, Boulianne M. Effect of Cessation of Ceftiofur and Substitution with Lincomycin-Spectinomycin on Extended-Spectrum-β-Lactamase/AmpC Genes and Multidrug Resistance in *Escherichia coli* from a Canadian Broiler Production Pyramid. *Appl Environ Microbiol* 2019;85. doi:10.1128/AEM.00037-19. [Epub ahead of print: 01 Jul 2019].
96. Álvarez-Fernández E, Cancelo A, Díaz-Vega C, *et al*. Antimicrobial resistance in E. coli isolates from conventionally and organically reared poultry: A comparison of agar disc diffusion and Sensi Test Gram-negative methods. *Food Control* 2013;30:227–34.
97. Alvarez-Fernandez E, Dominguez-Rodriguez J, Capita R, *et al*. Influence of housing systems on microbial load and antimicrobial resistance patterns of Escherichia coli isolates from eggs produced for human consumption. *J Food Prot* 2012;75:847–53.
98. Bennedsgaard TW, Thamsborg SM, Aarestrup FM, *et al*. Resistance to penicillin of Staphylococcus aureus isolates from cows with high somatic cell counts in organic and conventional dairy herds in Denmark. *Acta Vet Scand* 2006;48:24.
99. Boutet P, Detilleux J, Motkin M, *et al*. A comparison of somatic cell count and antimicrobial susceptibility of subclinical mastitis pathogens in organic and conventional dairy herds. *Ann Med Vet* 2005;149:173–82.
100. Cohen Stuart J, van den Munckhof T, Voets G, *et al*. Comparison of ESBL contamination in organic and conventional retail chicken meat. *Int J Food Microbiol* 2012;154:212–4.
101. El-Shibiny A, Connerton PL, Connerton IF. Enumeration and diversity of campylobacters and bacteriophages isolated during the rearing cycles of free-range and organic chickens. *Appl Environ Microbiol* 2005;71:1259–66.
102. Fraqueza MJ, Martins A, Borges AC, *et al*. Antimicrobial resistance among Campylobacter spp. strains isolated from different poultry production systems at slaughterhouse level. *Poult Sci* 2014;93:1578–86.
103. Garmo RT, Waage S, Sviland S, *et al*. Reproductive performance, udder health, and antibiotic resistance in mastitis bacteria isolated from Norwegian red cows in conventional and organic farming. *Acta Vet Scand* 2010;52:11.
104. Guarddon M, Miranda JM, Rodríguez JA, *et al*. Quantitative detection of tetracycline-resistant microorganisms in conventional and organic beef, pork and chicken meat. *CyTA - Journal of Food* 2014;12:383–8.
105. Huijbers PMC, van Hoek AHAM, Graat EAM, *et al*. Methicillin-Resistant Staphylococcus aureus and extended-spectrum and AmpC β-lactamase-producing Escherichia coli in broilers and in people living and/or working on organic broiler farms. *Vet Microbiol* 2015;176:120–5.
106. Kempf I, Kerouanton A, Bougeard S, *et al*. Campylobacter coli in organic and conventional pig production in France and Sweden: prevalence and antimicrobial resistance. *Front Microbiol* 2017;8:955.
107. Kerouanton A, Rose V, Chidaine B, *et al*. Comparison of organic and conventional pig productions on prevalence, antibiotic resistance and genetic diversity of Escherichia coli Journées de la Recherche porcine en France 2014;46:179–80.
108. Kim Y-J, Park J-H, Seo K-H. Comparison of the loads and antibiotic-resistance profiles of Enterococcus species from conventional and organic chicken carcasses in South Korea. *Poult Sci* 2018;97:271–8.
109. Larsen JL, Nielsen NC. Influence of restrictive use of antibiotics on the development of drug resistance in intestinal Escherichia coli from pigs. *Nord Vet Med* 1975;27:353–64.
110. Malissiova E, Papadopoulos T, Kyriazi A, *et al*. Differences in sheep and goats milk microbiological profile between conventional and organic farming systems in Greece. *Journal of Dairy Research* 2017;84:206–13.
111. Meemken D, Blaha T. Research on the occurrence of methicillin-resistant Staphylococcus aureus (MRSA) in domestic pigs and wild boars in Germany. *Dtsch Tierarztl Wochenschr* 2009;116:297–301.
112. Miranda JM, Guarddon M, Mondragon A, *et al*. Antimicrobial resistance in Enterococcus spp. strains isolated from organic chicken, conventional chicken, and turkey meat: a comparative survey. *J Food Prot* 2007;70:1021–4.
113. Miranda JM, Guarddon M, Vázquez BI, *et al*. Antimicrobial resistance in Enterobacteriaceae strains isolated from organic chicken, conventional chicken and conventional turkey meat: a comparative survey. *Food Control* 2008;19:412–6.
114. Miranda JM, Mondragón A, Vázquez BI, *et al*. Influence of farming methods on microbiological contamination and prevalence of resistance to antimicrobial drugs in isolates from beef. *Meat Sci* 2009;82:284–8.
115. Miranda JM, Mondragón A, Vázquez BI, *et al*. Microbiological quality and antimicrobial resistance of *Escherichia coli* and *Staphylococcus aureus* isolated from conventional and organic "Arzúa-Ulloa" cheese. *CyTA - Journal of Food* 2009;7:103–10.
116. Miranda JM, Vázquez BI, Fente CA, *et al*. Antimicrobial resistance in Escherichia coli strains isolated from organic and conventional pork meat: a comparative survey. *Eur Food Res Technol* 2008;226:371–5.
117. Miranda JM, Vazquez BI, Fente CA, *et al*. Comparison of antimicrobial resistance in Escherichia coli, Staphylococcus aureus, and Listeria monocytogenes strains isolated from organic and conventional poultry meat. *J Food Prot* 2008;71:2537–42.
118. Morley PS, Dargatz DA, Hyatt DR, *et al*. Effects of restricted antimicrobial exposure on antimicrobial resistance in fecal *Escherichia coli* from feedlot cattle. *Foodborne Pathog Dis* 2011;8:87–98.
119. O'Neill C. *Antibiotic-resistant staphylococci in the agricultural environment: reservoirs of resistance and infection [Ph.D*. University of Warwick, 2010.
120. Österberg J, Wingstrand A, Nygaard Jensen A, *et al*. Antibiotic resistance in Escherichia coli from pigs in organic and conventional farming in four European countries. *PLoS One* 2016;11:e0157049.
121. Roesch M, Perreten V, Doherr MG, *et al*. Comparison of antibiotic resistance of udder pathogens in dairy cows kept on organic and on conventional farms. *J Dairy Sci* 2006;89:989–97.
122. Sato K, Bartlett PC, Kaneene JB, *et al*. Comparison of prevalence and antimicrobial susceptibilities of Campylobacter spp. isolates from organic and conventional dairy herds in Wisconsin. *Appl Environ Microbiol* 2004;70:1442–7.
123. Schwaiger K, Schmied E-MV, Bauer J. Comparative analysis of antibiotic resistance characteristics of gram-negative bacteria isolated from laying hens and eggs in conventional and organic keeping systems in Bavaria, Germany. *Zoonoses Public Health* 2008;55:331–41.
124. Schwaiger K, Schmied E-MV, Bauer J. Comparative analysis on antibiotic resistance characteristics of *Listeria* spp. and *Enterococcus* spp. isolated from laying hens and eggs in conventional and organic keeping systems in Bavaria, Germany. *Zoonoses Public Health* 2010;57:171–80.
125. Suriyasathaporn W. Milk quality and antimicrobial resistance against mastitis pathogens after changing from a conventional to an experimentally organic dairy farm. *Asian Australas. J. Anim. Sci* 2010;23:659–64.
126. Tamang MD, Gurung M, Nam H-M, *et al*. Prevalence and characterization of Salmonella in pigs from conventional and organic farms and first report of S. serovar 1,4,[5],12:i:- from Korea. *Vet Microbiol* 2015;178:119–24.
127. Tenhagen B-A, Alt K, Pfefferkorn B, *et al*. Short communication: methicillin-resistant Staphylococcus aureus in conventional and organic dairy herds in Germany. *J Dairy Sci* 2018;101:3380–6.
128. Aarestrup FM, Bager F, Andersen JS. Association between the use of avilamycin for growth promotion and the occurrence of resistance among *Enterococcus faecium* from broilers: epidemiological study and changes over time. *Microb Drug Resist* 2000;6:71–5.
129. Aarestrup FM, Hasman H, Jensen LB, *et al*. Antimicrobial resistance among enterococci from pigs in three European countries. *Appl Environ Microbiol* 2002;68:4127–9.
130. Aarestrup FM, Kruse H, Tast E, *et al*. Associations Between the Use of Antimicrobial Agents for Growth Promotion and the Occurrence of Resistance among *Enterococcus faecium* from Broilers and Pigs in Denmark, Finland, and Norway. *Microbial Drug Resistance* 2000;6:63–70.
131. Aarestrup FM, Seyfarth AM, Emborg H-D, *et al*. Effect of abolishment of the use of antimicrobial agents for growth promotion on occurrence of antimicrobial resistance in fecal enterococci from food animals in Denmark. *Antimicrob Agents Chemother* 2001;45:2054–9.
132. Avrain L, Humbert F, L'Hospitalier R, *et al*. Antimicrobial resistance in Campylobacter from broilers: association with production type and antimicrobial use. *Vet Microbiol* 2003;96:267–76.
133. Boerlin P, Wissing A, Aarestrup FM, *et al*. Antimicrobial growth promoter ban and resistance to macrolides and vancomycin in enterococci from pigs. *J Clin Microbiol* 2001;39:4193–5.
134. Borgen K, Simonsen GS, Sundsfjord A, *et al*. Continuing high prevalence of VanA-type vancomycin-resistant enterococci on

Norwegian poultry farms three years after avoparcin was banned. *J Appl Microbiol* 2000;89:478–85.
135. Borgen K, Sørum M, Wasteson Y, *et al*. VanA-type vancomycin-resistant enterococci (VRE) remain prevalent in poultry carcasses 3 years after avoparcin was banned. *Int J Food Microbiol* 2001;64:89–94.
136. Del Grosso M, Caprioli A, Chinzari P, *et al*. Detection and characterization of vancomycin-resistant enterococci in farm animals and raw meat products in Italy. *Microbial Drug Resistance* 2000;6:313–8.
137. Desmonts M-H, Dufour-Gesbert F, Avrain L, *et al*. Antimicrobial resistance in Campylobacter strains isolated from French broilers before and after antimicrobial growth promoter bans. *J Antimicrob Chemother* 2004;54:1025–30.
138. Heuer OE, Pedersen K, Andersen JS, *et al*. Vancomycin-Resistant enterococci (VRE) in broiler flocks 5 years after the avoparcin ban. *Microbial Drug Resistance* 2002;8:133–8.
139. Kruse H, Johansen BK, Rørvik LM, *et al*. The use of avoparcin as a growth promoter and the occurrence of vancomycin-resistant *Enterococcus* species in Norwegian poultry and swine production. *Microb Drug Resist* 1999;5:135–9.
140. Kuhn I, Iversen A, Finn M, *et al*. Occurrence and relatedness of vancomycin-resistant enterococci in animals, humans, and the environment in different European regions. *Appl Environ Microbiol* 2005;71:5383–90.
141. Lauderdale T-L, Shiau Y-R, Wang H-Y, *et al*. Effect of banning vancomycin analogue avoparcin on vancomycin-resistant enterococci in chicken farms in Taiwan. *Environ Microbiol* 2007;9:819–23.
142. Li W, Hou M, Liu C, *et al*. Dramatic decrease in colistin resistance in Escherichia coli from a typical pig farm following restriction of colistin use in China. *Int J Antimicrob Agents* 2019;53:707–8.
143. Nwankwo C, Ayogu T, Iroha I, *et al*. Cloacal feacal carriage and occurrence of antibiotic resistant Escherichia coli in chicken grown with and without antibiotic supplemented feed. *J. Vet. Med. Anim. Health* 2014;6:91–4.
144. Smith HW, Lovell MA. *Escherichia coli* resistant to tetracyclines and to other antibiotics in the faeces of U.K. chickens and pigs in 1980. *J Hyg* 1981;87:477–83.
145. Sørum M, Holstad G, Lillehaug A, *et al*. Prevalence of vancomycin resistant enterococci on poultry farms established after the ban of avoparcin. *Avian Dis* 2004;48:823–8.
146. van den Bogaard AE, Bruinsma N, Stobberingh EE. The effect of banning avoparcin on VRE carriage in the Netherlands. *J Antimicrob Chemother* 2000;46:146–8.
147. Agunos A, Gow SP, Léger DF, *et al*. Antimicrobial use and antimicrobial resistance Indicators—Integration of Farm-Level surveillance data from broiler chickens and turkeys in British Columbia, Canada. *Frontiers in Veterinary Science* 2019;6.
148. Dorado-García A, Graveland H, Bos MEH, *et al*. Effects of reducing antimicrobial use and applying a cleaning and disinfection program in veal calf farming: experiences from an intervention study to control livestock-associated MRSA. *PLoS One* 2015;10:e0135826.
149. ESCMID/ASM Conference. *Prevalence of methicillin-resistant Staphylococcus aureus (MRSA) in organic and confinement swine operations in the midwestern United States*. London, UK, 2009.
150. Haskell KJ, Schriever SR, Fonoimoana KD, *et al*. Antibiotic resistance is lower in Staphylococcus aureus isolated from antibiotic-free RAW meat as compared to conventional RAW meat. *PLoS One* 2018;13:e0206712.
151. Hässig M, Eugster S, Lewis FI. Herd level antimicrobial resistance in beef calves in Switzerland 1986 through 2011. *OJVM* 2014;04:247–54.
152. Kilonzo-Nthenge A, Brown A, Nahashon SN, *et al*. Occurrence and antimicrobial resistance of enterococci isolated from organic and conventional retail chicken. *J Food Prot* 2015;78:760–6.
153. LeJEUNE JT, Christie NP. Microbiological Quality of Ground Beef from Conventionally-Reared Cattle and “Raised without Antibiotics” Label Claims. *J Food Prot* 2004;67:1433–7.
154. Lenart-Boron A, Augustyniak K, Boron P. Screening of antimicrobial resistance and molecular detection of fluoroquinolone resistance mechanisms in chicken faeces-derived Escherichia coli. *Veterinarni Medicina* 2016;61:80–9.
155. O'Brien AM, Hanson BM, Farina SA, *et al*. Mrsa in conventional and alternative retail pork products. *PLoS One* 2012;7:e30092.
156. Obeng AS, Rickard H, Ndi O, *et al*. Antibiotic resistance, phylogenetic grouping and virulence potential of Escherichia coli isolated from the faeces of intensively farmed and free range poultry. *Vet Microbiol* 2012;154:305–15.
157. Tadesse DA. *Molecular epidemiology of Campylobacter and Yersinia enterocolitica isolates from pigs reared in conventional and antibiotic free farms from different geographic regions [Ph.D]*. The Ohio State University, 2009.
158. Teramoto H, Salaheen S, Biswas D. Contamination of post-harvest poultry products with multidrug resistant Staphylococcus aureus in Maryland-Washington DC Metro area. *Food Control* 2016;65:132–5.
159. Thapaliya D, Forshey BM, Kadariya J, *et al*. Prevalence and molecular characterization of Staphylococcus aureus in commercially available meat over a one-year period in Iowa, USA. *Food Microbiol* 2017;65:122–9.
160. van den Bogaard AE*et al*. Antibiotic resistance of faecal Escherichia coli in poultry, poultry farmers and poultry slaughterers. *J Antimicrob Chemother* 2001;47:763–71.
161. Vikram A, Miller E, Arthur TM, *et al*. Similar Levels of Antimicrobial Resistance in U.S. Food Service Ground Beef Products with and without a "Raised without Antibiotics" Claim. *J Food Prot* 2018;81:2007–18.
162. Zhang Y. *Antimicrobial resistance of Listeria monocytogenes and Enterococcus faecium from food and animal sources [Ph.D]*. University of Maryland, College Park, 2005.
163. Wales A, Davies R. Co-selection of resistance to antibiotics, biocides and heavy metals, and its relevance to foodborne pathogens. *Antibiotics* 2015;4:567–604.
164. Aarestrup FM. Characterization of glycopeptide-resistant Enterococcus faecium (GRE) from broilers and pigs in Denmark: genetic evidence that persistence of GRE in pig herds is associated with coselection by resistance to macrolides. *J Clin Microbiol* 2000;38:2774–7.
165. Bager F, Aarestrup FM, Madsen M, *et al*. Glycopeptide resistance in *Enterococcus faecium* from broilers and pigs following discontinued use of avoparcin. *Microb Drug Resist* 1999;5:53–6.
166. Coker RJ, Hunter BM, Rudge JW, *et al*. Emerging infectious diseases in Southeast Asia: regional challenges to control. *The Lancet* 2011;377:599–609.
167. Paulson JA, Zaoutis TE. Nontherapeutic use of antimicrobial agents in animal agriculture: implications for pediatrics. *Pediatrics* 2015;136:e1670–7.