Antibiotic resistance – the impact of intensive farming on human health

A report for the Alliance to Save Our Antibiotics

Summary

Scientists and leading figures are increasingly warning of a serious health crisis in future, where some infectious diseases will no longer be treatable, if we do not urgently start using antibiotics more sparingly and intelligently. Antibiotic resistance is developing faster than new antibiotics are being developed, as finding new antibiotics is becoming increasingly difficult and expensive.

Despite previous attempts to reduce use, surveys have shown that many doctors still prescribe antibiotics far more often than necessary, a high proportion of patients still believe that antibiotics are effective against viruses, and a significant number of patients do not even complete a full course of antibiotics, sometimes saving tablets for later self-medication.

On farms, many antibiotics are used routinely for disease prevention or for the treatment of avoidable outbreaks of disease. Increasing use of antibiotics that are critically important in human medicine is also a serious concern. Some government officials and the intensive livestock industries are reluctant to reduce antibiotic use significantly, because this might increase production costs. As a result, those representing the interests of intensive livestock farmers and drug companies generally argue that the use of antibiotics does not contribute to the problem of antibiotic resistance in humans to any appreciable extent. Some governments, including the British government, accept industry arguments and claim there is no conclusive evidence that farm antibiotic use contributes to the human resistance problem.

In reality, increasing resistance levels are driven by antibiotic use in all sectors: in humans in the community and in hospitals, on farms and in companion animals. Although resistance in human infections is mainly caused by human antibiotic use, for a range of bacteria, farm-animal use contributes significantly and for some infections is the main source of resistance. This fact has been established by decades of research and is now fully accepted by organisations like the World Health Organisation and the European Food Safety Authority. This briefing sheet summarises some of the most important findings.

Antibiotic-resistant bacteria pass between humans, between animals and between humans and animals in both directions much more frequently than once realised. Copies of antibiotic-resistance genes can also move between bacteria, and this exchange can occur in the human gut, so in some cases the bacteria causing a human infection will not be of farm-animal origin, but the resistance will be.

This complexity means there are few completely conclusive results in antibiotic-resistance science. Nevertheless, the overall weight of scientific research has led to a consensus that:

- for some bacterial infections, such as *Campylobacter* and *Salmonella*, farm antibiotic use is the principal cause of resistance in human infections.
- for other infections, like *E. coli* and enterococcal infections, farm antibiotic use contributes, or has contributed, significantly to the human resistance problem.

- the emergence of resistance to critically important antibiotics, in particular of ESBL resistance in *E. coli and Salmonella*, is a major development which has occurred in recent years, which has been driven by inappropriate use of these antibiotics in both human and veterinary medicine.
- livestock-associated strains of MRSA infecting humans are also a developing problem, which results from the high use of certain antibiotics in farm animals.
- some other emerging antibiotic resistant infections in humans may in part be due to farm antibiotic use, but while research is ongoing, there is currently insufficient evidence to draw clear conclusions.

The lack of major success over past decades in developing new antibiotics means that it has become ever more important that we preserve the antibiotics that we have by using them only when they are genuinely needed in order to reduce overall use.

On many highly intensive pig and poultry farms, the approach is to increase hygiene and 'biosecurity' to reduce the spread of disease. However, the widespread use of some disinfectants, can also select for antibiotic-resistant bacteria. A more effective method for reducing disease and the need for antibiotic use in farm animals is to reduce livestock density. This is already a component of both Danish and Belgian attempts to reduce farm antibiotic use. Keeping animals in healthier conditions, where possible with greater access to the outdoors, can reduce disease still further. Selecting appropriate animal breeds, with a much greater focus on their resistance to infection and less on maximum productivity, will also contribute to a healthier animal population, needing fewer antibiotics.

Introduction

This review seeks to provide a balanced view of the relative contribution of human and veterinary use of antibiotics to the growing problem of antibiotic resistance in order to support calls for further action.

The Chief Medical Officer in the United Kingdom, Dame Sally Davies, recently warned Parliament that the rise of antibiotic resistance could cause a national emergency comparable to a catastrophic terrorist attack, pandemic flu or major coastal flooding. She told MPs of an 'apocalyptic scenario' where people going for simple operations in 20 years' time die of routine infections 'because we have run out of antibiotics'. She said that she would ask the Cabinet Office to add antibiotic resistance to the national risk register in the light of an annual report on infectious disease she will publish in March, and that there would also be a new cross-government strategy and action plan to tackle this issue published in early spring [1].

This is a welcome, if somewhat overdue, acknowledgement from the UK Government of an issue which has been highlighted many times in recent years by scientists and leading figures around the globe.

Finding new antibiotics is becoming increasingly difficult and more expensive. Only three genuinely new antibiotic classes have been developed over the last 30 years. As a result, antibiotic resistance is developing faster than new antibiotics are being developed.

At present, most antibiotic resistant infections can still be treated, but where infections are resistant to the antibiotics of choice, length of hospital stay is increased, patient recovery is slower, and costs to health services and taxpayers increase dramatically. In addition, hospital doctors are increasingly having to turn to a small number of more toxic antibiotics which have been little used in the past because they can cause serious side effects.

For some serious live-threatening infections such as *E. coli*, no new antibiotics are currently close to development, and treatment failures are already occurring [2]. It has been estimated that in the European Union alone 25,000 people die each year in from antibiotic-resistant infections and this figure could rise significantly in future [3].

In veterinary medicine the issue is currently less critical, but serious treatment problems are developing here too due to rising levels of resistance in many bacteria. Outbreaks of swine dysentery have already occurred which were resistant to all veterinary [4].

Understandably, attention tends to focus on the continuing overuse of antibiotics in human medicine, where considerable improvements could still be made in many countries. Research in the UK shows that almost half the people who visit their GPs with coughs and colds still expect to be given antibiotics, and that GPs can be concerned that a refusal to prescribe antibiotics will harm the doctor-patient relationship [5][6]. A Health Protection Agency survey found that a quarter of people who are prescribed antibiotics don't finish them, and a Welsh study concluded that approximately 1.6 million unnecessary prescriptions were made each year in the UK [5][7]. Infections in people who have taken antibiotics in the last six months are twice as likely to be resistant, so reducing unnecessary antibiotic use would have major benefits [5].

The situation in some developing countries is even more alarming. Antibiotics, including those which should be reserved for second- or third-line treatment of serious infections are on general sale, sometimes produced illegally and not full strength. Often the poor cannot afford to pay for a complete course, so they buy just a few tablets which are insufficient to kill off all the infectious bacteria, leaving the more resistant ones to proliferate. Then, due to the high level of international travel today, new types of antibiotic resistance in one country can spread worldwide within just a few years [8].

For a wide range of human diseases it is clear that the use and overuse of antibiotics in human medicine, and not farm animals or companion animals, is the cause of increasing resistance. This includes, for

example, the spread of multi-resistant tuberculosis and the emergence of resistance to the antibiotics of last resort, the carbapenems (as these are not licensed for use in farm animals).

Because of this, however, there is a tendency amongst some sections of the intensive livestock industry and even some governments, to dismiss the contribution from veterinary use almost entirely. In the UK, a Defra advisory committee has considered how cross-departmental government action could be used to counter media stories suggesting that farm antibiotic use could cause problems for people [9].

In a recent Parliamentary debate in the UK on the link between farm antibiotic use and resistance in human medicine, the Parliamentary Undersecretary for Health, Anna Soubry MP, correctly said that:
'There is scientific consensus that the use of antimicrobials[1] in human medicine is the main driving force for antimicrobial-resistant human infections' [10].

However, this is not the whole story. While antibiotic use in animals may not be the *main* driver of resistance in humans, use in farm animals (and to a lesser extent use in companion animals) is a very important contributor. For some human diseases it is actually the *main* cause of resistance. Despite this, the Minister continued by saying:
'There is no conclusive scientific evidence that food-producing animals form a reservoir of infection in the UK. Food is not considered to be a major source of infections resistant to antibiotics.'

The Minister made it clear that her notes were in part provided by Defra, where the Veterinary Medicines Directorate (VMD), a largely industry-funded executive agency of Defra, is responsible for antimicrobial resistance. The statement was clearly a reflection of VMD's position statement on antibiotic resistance which fails to recognise explicitly that farm antibiotic use contributes to resistance problems in humans [11]. In this respect, Defra's position is increasingly out of step with a broader European perspective as reflected by reports from the European Food Safety Authority (EFSA) and the World health Organisation (WHO) [3][12].

Although absolute proof of cause and effect in this field can be extremely difficult to establish because so many of the same antibiotics are used in both veterinary and human medicine, scientists have established a clear link between antibiotic use in farm animals and resistance in humans.

In particular, the scientific evidence shows that:

1. for some major human bacterial infections, such as *Salmonella* and *Campylobacter*, farm animals are the most important source of antimicrobial resistance.
2. for certain other human infections, such as *E. coli* and enterococci, there is strong evidence that farm animals are an important source of antibiotic resistance.
3. for some infections, like MRSA, there is evidence that in the UK the farm use of antibiotics currently makes a small contribution to treatment problems in human medicine. But based on the experiences in some other countries, this contribution may increase significantly unless we take decisive action very quickly.
4. for a further small number of antimicrobial-resistant infections, such as *Neisseria gonorrhoeae*, there is as yet no evidence of any link with farm antimicrobial use at all, yet there is a solid theoretical case that the horizontal transmission of resistance genes of farm-animal origin could contribute to the rise of potentially untreatable cases in humans. This would be such a serious and

[1]Antimicrobials are substances which kill or inhibit the growth of micro-organisms such as bacteria, fungi or protists. Antimicrobials can be synthetic or naturally produced by other micro-organisms. Antibiotics were originally defined as antimicrobials which are naturally produced, although the two terms are often now used interchangeably.

quite possibly irreversible development that precautionary action without waiting for evidence would be wise, even if the probability of the worst-case scenario is only moderate.

5. for many other infections, such as multi-drug resistant tuberculosis and the wide range of infections caused by antibiotic-resistant strains of *Streptococcus pneumonia*, the use of antibiotics on farms plays no part in the resistance problem in human medicine.

Antibiotic-resistant bacteria of farm-animal origin can pass to humans in a number of ways, principally on food, but also by direct contact and through the environment. Resistant bacteria can and also pass from humans to farm animals. Here they can multiply and acquire additional resistance genes, then pass back to humans.

In each case, the resistant farm-animal bacteria can contribute to higher levels of resistance in human infections in two main ways:

- they can directly cause an infection in humans, and this infection will be antibiotic-resistant.
- they can colonise the human gut (and potentially other sites such as the nares) without causing an infection, and pass on copies of their resistance genes ('horizontally') to bacteria already living in the human gut. The human-adapted bacteria receiving the resistance genes may subsequently, possibly at a much later date, cause an infection, if they get into the wrong part of the body (e.g. a urinary-tract infection). In this case, the pathogen will be of human origin, but its resistance will originate (either wholly or partly) from the farm use of antibiotics.

Tracing the origin of the resistance tends to be much easier in the first scenario, as when foodborne bacteria cause immediate outbreaks of infection in a significant number of people at once, scientists can frequently trace the source of the infection to a particular food, often meat.

In the second scenario, establishing the origin of the resistance tends to be more difficult and can involve molecular studies examining resistance genes and associated genes such as plasmids[2], rather than just comparing bacterial strains. The Minister said in the Parliamentary debate that 'The majority of resistant strains affecting humans are different from those affecting animals', but differences in strains does not always mean that the resistance is not of farm-animal origin, due horizontal gene transmission.

A factor complicating the detective work is that human antibiotic use can add to the resistance profile[3] of some bacteria, which may already be resistant to certain antibiotics due to earlier farm antibiotic use. The fact that some of the resistance in this case will be due to human use does not detract from the fact that a large amount of resistance to vitally important medical antimicrobials in these bacteria is initially coming from the use of similar, or identical, drugs in livestock production.

Companion animals can also be a source of resistant bacteria which infect humans, and there is a significant amount of resistance which can be transmitted from humans to animals as well. Transmission is often by direct contact, and this is a particular problem with MRSA which can be easily passed on in this way [13]. Although overall antibiotic use is much lower in companion animals than in the high-consuming farm species like pigs and poultry [14], efforts should nevertheless be made to ensure that unnecessary use is avoided.

[2]A plasmid is a small loop of DNA, which is separate from the bacterium's chromosome, and which can carry antibiotic-resistance genes. Copies of resistance plasmids, sometimes with more than one resistance gene can be transferred between bacteria, making the recipient bacteria resistant to all the corresponding antibiotics.
[3]The resistance profile is the list of antibiotics to which a bacterium is resistant.

The WHO has summarised the situation by saying:
'*Since this resistance has no ecological, sectoral or geographical borders, its appearance in one sector or country affects resistance in other sectors and countries. National authorities, veterinarians, physicians, patients and farmers all have key roles in preserving the power of antibiotics. The prevention and containment of antibiotic resistance therefore requires addressing all risk factors for the development and spread of antibiotic resistance across the full spectrum of conditions, sectors, settings (from health care to use in food-animal production) and countries'* [3].

In addition to the genuine scientific difficulties in establishing certainty on the origin of some antibiotic resistance, it is important to recognise that commercial interests may influence the debate. In the UK, the Responsible Use of Medicines in Agriculture Alliance (RUMA), an alliance representing the interests of the pharmaceutical and intensive-farming industries which is opposing attempts to ban the routine preventative use of antibiotics in farming, has dismissed the claim that the overuse of antibiotics in intensive farming adds to the serious public-health threat from antibiotic resistance as a 'myth', despite the wealth of evidence to the contrary [15].

Furthermore, although government scientists have produced many high-quality studies over the past decades examining the farm resistance issue, government officials recognise that implementing significant restrictions on antibiotic use in farming could increase costs. A report published last year by Defra and Department of Health scientific advisors and officials argued against taking too many measures at an EU level, saying this could put EU farmers at a commercial disadvantage leading to more imports [16].

They warned that costs might increase because, they claimed, fewer animals might survive, but also because 'livestock have to be kept more extensively or in better buildings to minimise risks of becoming infected, such as avoiding pneumonia by building better designed, well-ventilated buildings'. The report concluded that 'Unless consumers are prepared to pay a premium for food produced by means designed to lower the risk of transmitting antimicrobial resistance (while not compromising animal welfare) the potential for unintended consequences of certain measures that may be used to control antimicrobial resistance is high'.

As a result, despite accepting that improving the conditions in which animals are reared can result in significant improvements in antibiotic use, Defra officials largely continue to support the status quo. It is not difficult to imagine, then, how these considerations may also have influenced the partly incorrect scientific advice Defra gave to the Health Minister, which downplayed the contribution of farm antibiotic use to the human resistance problem.

1. Human infections where farm antibiotic use is the main source of resistance: *Salmonella* and *Campylobacter*

Salmonella is a food-poisoning infection which often causes outbreaks where a number of people are infected at the same time. Because of this it is generally easy to establish the cause, and there is now a large scientific consensus that most antibiotic resistance in human infections is of farm-animal origin.

Campylobacter infections tend to be more sporadic, making it more difficult to trace their origin precisely. Nevertheless meat, particularly poultry, is known to be a major source of infection, and the emergence of resistance in human infections to certain particularly important antibiotics, such as the fluoroquinolones, followed the introduction of these antibiotics to farming, providing strong evidence of a link [12].

EFSA concluded in its 2008 report, which reviewed the evidence on foodborne antimicrobial resistance, that:
'Resistant Salmonella *and* Campylobacter *involved in human disease are mostly spread through foods'* [12].

The WHO similarly said in its 2011 report on foodborne antibiotic resistance:
'Resistance in the foodborne zoonotic bacteria Salmonella *and* Campylobacter *is clearly linked to antibiotic use in food animals, and foodborne diseases caused by such resistant bacteria are well documented in people. Of special concern is resistance to so-called critically important antibiotics for human medicine. For example, the use of fluoroquinolones in food animals has led to a corresponding antibiotic resistance in* Salmonella *and* Campylobacter *species, thus causing infections in people. Also, antibiotic resistance in* Salmonella *has been associated with more frequent and longer hospitalization, longer illness, a higher risk of invasive infection and a twofold increase in the risk of death in the two years after infection'* [3].

2. Human infections where farm antibiotic use is an important source of resistance: *E. coli* and enterococci

With infections caused by *E. coli* which cause extra-intestinal infections (such as urinary-tract and blood-poisoning infections) and by enterococci which cause kidney and wound infections, the situation is more complicated than for *Salmonella* and *Campylobacter* because farm-animal strains can frequently contribute to resistance in human infections by transferring copies of resistance genes to human-adapted *E. coli* and enterococci in the human gut, rather than by directly causing infections.

In the case of *E. coli*, farm-animal strains, especially those originating with poultry, can also cause infections directly. This may also occur for enterococci, but there is less evidence of this happening.

However, despite the greater scientific difficulties in establishing the source of resistance for these two types of bacteria, in both cases, there is now clear evidence that farm-animal antibiotic use does contribute significantly to levels of antibiotic resistance in human infections.

2.1 *E. coli*

Headline coverage of antibiotic resistance in *E. coli* infections inevitably focuses on the most serious emerging forms resistance: extended-spectrum beta lactamase (ESBL) resistance which renders the bacteria resistant to modern cephalosporins, which have for many years been the most important antibiotics for treating cases of blood poisoning, and resistance to carbapenems, the drugs of last resort which doctors had until recently managed to hold in reserve, but are now often forced to use in life-threatening cases, because of the growing risk that modern cephalosporins will not be effective.

Resistance to other antibiotics may sometimes be seen as less important because these resistances have been around for longer, however, we have only reached the current critical situation where we are already seeing some untreatable *E. coli* infections because we have allowed a large number of antibiotics over the years to be used far too freely in both human and veterinary medicine, in the latter case most frequently on a routine basis at sub-therapeutic levels over prolonged periods, the very conditions most likely to cause resistance to become a problem. It is important, therefore, to examine the evidence that a significant proportion of the resistance to these earlier antibiotics in human infections has been caused by the farm use of antibiotics, before we look at the particular case of ESBL resistance.

Certain scientists, including some funded by the pharmaceutical industry, have used the fact that some studies have found differences in the strains of *E. coli* which colonise the intestines of farm animals and those which infect humans, to argue that resistance in farm-animal *E. coli* is largely irrelevant to human health [17].

However, this view is now disputed by many scientists, as evidence mounts that a very significant proportion of the resistance in *E. coli* causing urinary-tract and blood-poisoning infections in humans is of farm-animal origin [18][19][20]. There is now compelling evidence that food animals are a reservoir for antibiotic-resistant *E. coli*, colonising or infecting humans, and also a reservoir for resistance genes which

can transfer to *E. coli* which can cause infections in humans. This accumulating evidence has led one leading Australian scientist to warn that, with resistant *E. coli*, 'We are what we eat' [18].

The transfer of antibiotic resistance genes from farm-animal *E. coli* to human *E. coli* in the human gut was shown to occur in a study published in 1969 [21]. Since then a number of studies have confirmed the finding, including a study carried out on a Danish pig farm which found that while the *E. coli* from the pigs, the pig farmers and the environment were all genetically different they carried the same resistance plasmid [22]. Danish government scientists believe that taken together, these studies show that:
'The transfer of resistance genes between E. coli *of animal and human origin in the intestine of humans is very likely'* [19].

Some of the strongest evidence that resistance genes in human *E. coli* can originate in farm animals comes from occasions when an antibiotic has been used in veterinary medicine but not in human medicine. The antibiotic nourseothricin was used in pigs in the former East Germany in the 1980s, but no equivalent antibiotic was used in humans during the same period.

Resistance to the antibiotic was first detected in porcine *E. coli*, two years after the introduction of the antibiotic, and later resistance was found in *E. coli* from pig farmers. In subsequent years, resistance was found in *E. coli* and other pathogens, such as *Salmonella* and *Shigella* (the cause of dysentery in humans), from people in the wider community. One scientist from the UK's Veterinary Laboratories Agency commented that:
'These observations strongly support the premise that resistance genes present in the commensal flora of animals can spread to bacteria which can colonize or infect humans' [23].

EFSA has come to a similar conclusion, saying:
'Some categories of food may often be contaminated with E. coli*, including resistant isolates, and these bacteria reside long enough in the intestines of humans to be able to transfer resistance genes to the residential flora. It is therefore highly probable that food is a vehicle for spread of resistance genes between different ecosystems'* [12].

While most scientists have refrained from claiming their research provides conclusive proof, this is largely because it is not possible to observe under experimental conditions all components of resistance gene transfer and subsequent infection at one time. As such, conclusions have to be based on deduction. The small element of uncertainty introduced by words like 'highly probable' should not be taken to indicate that the incidence of such gene transfer is not frequent or that the implications of this are not significant.

Although there are differences from country to country it is important to realise that many of the findings from research in other countries are relevant to the situation in the UK. Some British research has also found evidence of resistance to important antibiotics in human *E. coli* infections originating in farm animals.

In the 1980s and 1990s, for example, government scientists working for the Public Health Laboratory Service (PHLS) produced some strong evidence that resistance to aminoglycoside antibiotics, which have been important for treating *E. coli* infections, was passing from farm animals to humans. By 1994, they said that their findings:
'support the view that resistance to gentamicin and apramycin in clinical isolates of E. coli *results from the spread of resistant organisms from animals to man, with subsequent inter-strain or inter-species spread, or both, of resistance genes on transferable plasmids'* [24].

Given this, and other similar statements by scientists from the Health Protection Agency and the Veterinary Laboratories Agency over the years, it is disappointing that the British Government appears to be so reluctant to acknowledge the significance of farm-animal-to-human transmission of antimicrobial resistance, or to take effective action to limit it.

In addition to the mounting evidence that resistance genes from farm animal *E. coli* can spread to human-adapted *E. coli*, there is also evidence that farm-animal *E. coli*, particularly *E. coli* from poultry, can directly cause infection in humans [18][19]. Two studies carried out in Spain and the United States, for example, have found strong evidence that poultry are a source of antibiotic-resistant human *E. coli* infections [25][26]. The scientists used genetic methods to compare human and poultry *E. coli* and found that human resistant *E. coli* were genetically similar to resistant poultry *E. coli* and that resistant poultry *E. coli* were also genetically similar to sensitive poultry *E. coli*. However, the human resistant *E. coli* were genetically unrelated to the human sensitive *E. coli*.

Both sets of scientists concluded that it appeared that the *E. coli* had evolved to become resistant in poultry, before being transferred to humans. The scientists working in the United States said:
'Many drug-resistant human fecal E. coli *isolates may originate from poultry, whereas drug-resistant poultry-source* E. coli *isolates likely originate from susceptible poultry-source precursors'* [26].

The WHO, commenting on the evidence, has concluded:
'Resistant E. coli *can spread from animals to people through the food-chain'* [3].

Direct evidence of the effect of food on levels of antibiotic-resistant *E. coli* in humans has been provided by a French study. This involved feeding six volunteers a near-sterile diet for an average of 17 days, after an earlier control period of 21 days. During the control period, they were fed their usual diet, and for the sterile-diet period their food was heated to 105°C for one hour, which was shown to be sufficient to destroy any *E. coli* bacteria on the food.

The day after the sterile diet began, the number of *E. coli* in the volunteers which were resistant to ampicillin, streptomycin, and tetracycline fell significantly, and it reached a minimum in just three days. For the antibiotic tetracycline, for example, the number of resistant bacteria fell by an average factor of 500. On the other hand, there was a much smaller fall in the number of sensitive *E. coli* (average factor of 3), which was not statistically significant. The scientist concluded that most resistant *E. coli* in the human gut come from food [27].

Although estimating the size of the precise contribution of farm-animal antibiotic use to resistance in human *E. coli* infections has proved more difficult than for *Salmonella* and *Campylobacter* for the reasons detailed above, evidence gathered in recent years has suggested that it is likely to be very significant.

An international study by Australian, Canadian and Danish scientists used data from 11 European countries on levels of antibiotic resistance in human and farm-animal *E. coli*, and antibiotic use in humans. They found strong and statistically significant correlations between resistance to several antibiotics, including critically important antibiotics such as fluoroquinolones and modern cephalosporins, in human *E. coli*, and resistance to the same antibiotics in poultry and pig *E. coli*. They also found that antibiotic use in humans was only correlated with antibiotic resistance in humans for two of the four classes of antibiotics examined.

They concluded that:
'In addition to the contribution of antimicrobial usage in people, a large proportion of resistant E. coli *isolates causing blood stream infections in people are likely derived from food animal sources'* [20].

ESBL E. coli

A major cause for concern regarding resistance in *E. coli* has been the emergence over the past decade, in both humans and farm animals, of extended-spectrum beta-lactamases (ESBL) resistance. This is caused by a large family of enzymes which render the bacteria resistant to modern cephalosporins, which are very important treatments for patients who have to be hospitalised with resistant *E. coli* infections. Defra and HPA scientists, and many others, believe that the emergence of these bacteria in farm animals, in the UK and throughout Europe, is partly linked to the increasing use of modern cephalosporins and

fluoroquinolones in farming, two antibiotic classes classified as critically important in human medicine by the WHO [28].

A review of the scientific evidence by Defra and Department of Health scientists in the UK, which was published last year recognised only a minor role for farm animals in the emergence of this, but did conclude:
'It is thought that emergence of ESBL bacteria in food producing animals may present a risk of resistant strains being transmitted to humans through the food chain' [28].

In view of the potential importance to human health of the emergence of ESBL *E. coli* in farm animals, it is welcome news that a three-year collaborative research project is to be undertaken by four universities, the AHVLA, Health Protection Scotland and the HPA to provide further evidence of 'the risk to public health posed by ESBLs in bacteria from non-human sources, including the food chain' [29]. However, based on the existing evidence, it would not be justifiable to delay taking effective action to limit the rise of ESBLs in farm animals until this project has been completed.

Many of the ESBL resistant infections in humans are nowadays acquired in the community. According to the Defra/DH ESBL report published last year:
'Whilst initially confined to enterobacteriaceae causing hospital acquired infection, the emergence and spread particularly in the community of Escherichia coli *(*E. coli*) strains producing CTX-M ESBLs is a very serious challenge to effective therapy of infections caused by all Gram negative bacteria[4]'* [28].

Not only is the emergence of these bacteria in the community a serious development regarding therapy, it may also be important evidence of a farming connection. In 2005, the HPA published a report on the spread of ESBL *E. coli*, and the author of the report, Dr Georgina Duckworth said:
'The findings in our report show evidence of people carrying these bacteria in their gut. If this is found to be commonplace in the general population this may point towards the food chain being a potential source' [30][31].

At the time of the publication of the HPA report, one study had found that 1.4% (8 of 565) of community-based patients had ESBL *E. coli* bacteria in their faeces, whereas just 0.25% (1 of 394) of hospital-based patients had the bacteria [32]. More recent research, carried out in Birmingham, found that 11.3% of community patients (GP patients or outpatients) had ESBL *E. coli* in their faecal matter [33]. This is a very large increase over earlier findings, and perhaps pointing towards the food chain as a possible source, as Dr Duckworth had suggested. However, it has to be recognized that the Birmingham study may not accurately reflect the national situation and that further more comprehensive surveys are needed.

A key point to note here is that modern cephalosporins are not normally prescribed by GPs, not least because, with only one exception, they are not available in tablet form. They are also not prescribed by veterinary surgeons for companion animals, so where ESBL carriage is found in members of the public who have not been treated with these antibiotics in hospitals, food or farm animals are likely to be the source of the resistance in a significant proportion of cases.

While the evidence for ESBL resistance coming from animals is a major cause for concern, it should also be pointed out that the main epidemic strain of ESBL *E. coli* in humans in the UK, called ST131, is not thought to have a significant farm-animal link. There is, nevertheless, evidence that farm animals may be a source for some of the other ESBL E. coli strains causing infection in humans, as well as of various ESBL plasmids.

In the Netherlands, where more research has been carried into ESBL *E. coli* in farm animals than has been the case in the UK (and where levels of ESBL *E. coli* in farm animals are also higher than in the UK), the evidence is even stronger. Scientists there, including Dutch government scientists, found that a very high

[4] All bacteria are classified as Gram-positive or Gram-negative. *E. coli*, like *Salmonella* and *Campylobacter*, are Gram-negative, whereas *Staphylococcus aureus* and enterococci are Gram-positive.

proportion (94%) of retail poultry was contaminated with ESBL *E. coli*, and that 39% of these bacteria belonged to strains causing human infections. They said that:
'These findings are suggestive for transmission of ESBL producing E. coli *from poultry to humans, most likely through the food chain'* [34].

Other Dutch scientists also found a proportion (80%) of retail poultry had ESBL bacteria and that the predominant ESBL genes in poultry meat and in human rectal samples were identical. They said:
'These findings suggest that the abundant presence of ESBL genes in the food chain may have a profound effect on future treatment options for a wide range of infections caused by gram-negative bacteria' [35].

Further Dutch research published this month confirmed that 40% of human ESBL E. coli were 'chicken-meat isolates'. The scientists said that:
'Therefore, chicken meat is a likely contributor to the recent emergence of ESBL E. coli *in human infections in the study region. This raises serious food safety questions regarding the abundant presence of ESBL* E. coli *in chicken meat'* [36].

EFSA has also concluded that genetic similarities between certain ESBL plasmids found in farm animals and in humans 'strongly suggests an animal reservoir for this ESBL gene variant' [45]. These particular ESBL plasmids are very common in human infections in some countries, but less common in the UK.

2.2 Enterococci

For enterococci, there is less evidence that farm-animal strains cause infection directly in humans, but they can transfer their resistance genes to human enterococci. EFSA says:
'While the direct clinical infection in humans by VRE [vancomycin-resistant enterococci*] from food sources apparently is rare although not totally excluded as a possibility, the reservoir of VRE in food-producing animals presents a definite risk of resistance genes being transferred to virulent human strains through food and other routes'* [12].

Avoparcin was an antibiotic growth promoter, widely used in pigs, poultry and cattle in the UK and throughout Europe. It is chemically very closely related to vancomycin, an extremely important hospital antibiotic for treating MRSA and enterococcal infections.

The first evidence that the widespread use of this antibiotic on farms was leading to resistance problems was produced by British scientists working at the University of Oxford: they isolated vancomycin-resistant enterococci (VRE) from pig herds and from uncooked chickens [37]. Soon after, German scientists found that VRE could be isolated from pigs, poultry and from humans in the community [38][39]. In contrast, in the United States, where avoparcin had never been licensed as a growth promoter, VRE was not found in people in the community, nor in farm animals [39][40][41].

Concerns about VRE being transmitted from farm animals to humans were a major reason for the EU ban on the growth promoters. Avoparcin was the first growth promoter to be banned throughout Europe in 1997, after first having been banned in Sweden in the 1980s, in Denmark in 1995 and then in Germany in 1996.

In hindsight, the ban appears to have had the desired effect, according to data collected in some countries (the UK and many other countries did not collect the data which would have enabled the ban's effect to be evaluated). In Germany, the incidence of VRE on poultry meat fell from 100% in 1994 to 25% in 1997, and in faecal samples taken from people in the community it fell from 12% in 1994 to just 3% in 1997 [39][42]. In Denmark, VRE prevalence in poultry fell from 82% in 1995 to less than 5% in 1998 [43], and in the Netherlands VRE prevalence fell sharply between 1997 and 1999: from 80% to 31% for broilers, from 34% to 17% for pigs and from 12% to 6% for humans [44].

Referring to some of these findings, the WHO said in their 2011 report that:
'Data have shown that this intervention resulted in reduction of vancomycin-resistant enterococci in food animals and the general population'.[3]

3. Human infections where there is compelling evidence that farm antimicrobial use in the UK contributes to a small, but likely increasing, proportion of resistance

MRSA are strains of *Staphylococcus aureus* with resistance to beta-lactam antibiotics, and often resistance to other antibiotics as well. Strains of MRSA have emerged in farm animals in recent years, and unlike many strains of *Staphylococcus aureus* of farm-animal origin, these livestock-associated MRSA strains can colonise and multiply on most species, including humans. The most common of these, MRSA ST398, was first detected in the Netherlands in 2005 [46].

The spread of MRSA ST398 throughout Europe's pig population in particular (it is also present in poultry and cattle), is recognised to have led to a growing number of these infections in humans. This strain now accounts for approximately 39% of human cases of MRSA in the Netherlands [47]. Although farmers and those in direct contact with livestock are those most at risk, MRSA ST398 can also sometimes pass from human to human. Consumers of meat contaminated with MRSA are not thought to be at great risk, but further research is needed to clarify this.

Most MRSA infections in humans in the UK currently have nothing to do with agriculture, but the recent detection of a small number of cases of MRSA ST398 and other types of MRSA in British cattle is cause for concern [48]. Livestock-associated MRSA have already caused infections in humans in the UK, and experience from abroad suggests that, for MRSA ST398 in particular, there is a real danger that it will spread widely in livestock unless changes in farm antimicrobial use are introduced urgently.

Several further types of MRSA are now emerging in pigs in Europe, North and South America, and Asia, and some of these are epidemic human strains which are thought to have transferred initially from humans to animals [49][50][51]. If these strains become widespread on farms, there is a real danger that livestock will become a very important reservoir of human MRSA infections.

It is worth noting how quickly the livestock-associated MRSA problem has emerged. Less than ten years ago, MRSA had never been detected in pigs, and the very small number of cases found in other farm animals were believed to have been incidental transfers from humans. More recently, MRSA has been found in abattoir studies in 61% of Spanish pigs, in 60% of Germany pigs and 39% of Dutch pigs [52][53][54]. The emergence of this problem, like the emergence of the highly resistant ESBL E. coli, is believed by scientists to be particularly linked to the increased use in farming of modern cephalosporins, which are classified by the WHO as critically important antibiotics in human medicine [55][56].

4. Human infections for which there is currently no evidence that the farm use of antibiotics is contributing to resistance, but where theoretical considerations suggest this could happen

Modern cephalosporins are first-line treatments for gonorrhoea, and Health Protection Agency scientists have warned that any emergence of resistance to these antibiotics would be a 'catastrophic development' [57].

As mentioned above, resistance to modern cephalosporins, in the form of ESBL resistance, already occurs in *E. coli* from humans and farm animals, and HPA scientists are worried that this resistance could transfer in the genitourinary tract from *E. coli* to *Neisseria gonorrhoeae*. They say that at the moment, in the UK, most cases of ESBL E. coli occur in older patients in the community. However, they point out that studies from abroad, in Canada and Hong Kong, are finding significant levels of ESBL *E. coli* in women of all ages. The scientists say:

'Rising rates of E. coli *with CTX-M ESBLs* [a type of ESBL resistance]*in the genitourinary tracts of sexually active women raise the alarming possibility that these enzymes might "escape" into sexually transmitted bacterial pathogens, specifically* Neisseria gonorrhoeae' [57].

Since it is known that the presence of ESBL *E. coli* in farm animals and on food is contributing to the presence of these bacteria in the human gut, the use of these antibiotics on farms is increasing the risk that ESBL genes will eventually spread from *E. coli* to *Neisseria gonorrhoeae*.

5. Increasing farm use of critically important antibiotics in human medicine

Modern cephalosporins (3rd and 4th generation cephalosporins) and fluoroquinolones are two of the most important classes of antibiotics used in human medicine, and have been classified by the WHO as critically important in human medicine.

The increasing use of these antibiotics in agriculture over the past decade is widely recognised to have contributed to the emergence of a range of highly resistant bacteria in farm animals, such as ESBL *E. coli*, ESBL *Salmonella*, fluoroquinolone-resistant *Campylobacter* and MRSA.

Statistics in the UK from the Veterinary Medicines Directorate show that, after fluoroquinolone use was cut significantly in 2000 following warnings in a report by the House of Lords Committee on Science and Technology in 1998 and a report by the Advisory Committee on the Microbiological Safety of Food in 1999, the use of both fluoroquinolones and modern cephalosporins has increased in most years since then. See the table below in kgs of active ingredient compiled by the Soil Association from data provided by the Veterinary Medicines Directorate.

	1999	2000	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011
Modern cephalosporins	220	220	310	410	410	500	672	739	854	887	976	1,463	1166
Fluoroquinolones	2290	1230	1320	1370	1360	1410	1460	1620	1951	1928	1849	2232	2085

While these are relatively small amounts in comparison to the quantities of some antibiotics used in farming, it is important to realise that these are very potent antibiotics, and the weight of the active ingredient of one dose is very low in comparison to most other antibiotics (the European Medicines Agency says that one dose of fluoroquinolones, for example, typically weighs 30 to 70 times less than a dose of tetracyclines [58]).

The increase in the use of these antibiotics is therefore a significant concern, particularly as it appears to be happening throughout most of Europe (in fact the situation in some other European countries is significantly worse in this respect compared to the UK) [58].

6. The need for action

In addition to action on inappropriate antibiotic use, overall farm antibiotic use must be reduced. This is widely recognised as the most likely strategy which will slow, or even reverse, the growth of antibiotic resistance.

The European Union has already taken some welcome action aimed at reducing the veterinary use of critically important antibiotics, particularly the modern cephalosporins. However, much more remains to be done [59]. Industry initiatives tend to promote 'biosecurity' and hygiene, which can have some benefits. However, British research has shown that disinfectants can also 'co-select' for antibiotic resistance [60].

Some of the most important elements of a truly effective strategy would be:

1. **A legally binding timetable for the phased ending of all routine prophylactic, non-therapeutic use of antibiotics.**
Although the European Medicines Agency has clarified that, for products licensed through Europe's centralised procedure, routine prophylactic use is not permitted if no signs of disease are present [61], it is clear that many within the industry continue to believe this practice is both acceptable, and at times desirable [62].

2. The ban on the use of modern cephalosporins in poultry must be fully implemented, and a ban should be also be introduced on their use in pigs and for dry-cow therapy in cattle.
The European Commission's has taken an important decision that Member States must add a statement to the Summary of Product Characteristics for modern cephalosporins to make it clear that the products must not be used in poultry. Based on past experiences the Commission will need to verify that statements are in fact added to SPCs throughout Europe, so that the ban comes into force as soon as possible.
Danish, Dutch and French pig producers have already introduced voluntary bans on the use of modern cephalosporins [63]. Very recently published research found that the occurrence of ESBL *E. coli* in Danish pigs at slaughter fell from 11.8% in 2010 to 3.6% in 2011 after the voluntary ban was introduced in July 2010. The decline in resistance for pigs tested on farms was even larger, from 11% in 2010 to 0% in 2011 [64].
Dutch dairy farmers have also already introduced a voluntary ban on the use of modern cephalosporins for dry-cow therapy. According to the Dutch Chief Veterinary Officer, these voluntary bans have contributed to a 92% reduction in the Dutch farm use of these antibiotics between 2009 and 2012 [65].

3. **Modern cephalosporins should no longer be permitted to be used off-label**
The risk of ESBL resistance and MRSA transferring from farm animals to humans is too great to permit use of these antibiotics in animals which is not fully regulated.

4. **A ban on the use of fluoroquinolones in poultry**
Fluoroquinolones are critically important antibiotics in human medicine because of their importance for treating infections such as Campylobacter, Salmonella and E. coli. Poultry are recognised as an important source of these infections in humans, and in the case of Campylobacter are by far the most important source. The United States banned the use of these antibiotics in poultry for that reason [66] and an advisory committee to the French Prime Minister has added its voice to calls for such a ban in Europe [63].

5. **New legislation aimed at ensuring that farm animals are kept in healthier, less intensive conditions, wherever possible with access to the outdoors.**
It is essential that a farm-animal health and welfare strategy should be recognised as a key tool in helping to address the rise of antibiotic resistance. Improving animal health through increased animal welfare, better system design and the selection of breeds that are less susceptible to disease can dramatically reduce the need for antibiotics. There are a number of studies finding significantly lower use of antibiotics and correspondingly lower levels of antibiotic-resistant bacteria in organically farmed animals, and both the Belgian [67] and Danish [63] Governments are beginning to require reductions in livestock stocking density in order to reduce the use of antibiotics.

6. **Improved surveillance of antibiotic use and antibiotic-resistant bacteria in farm animals**
The European Commission has already taken some important initiatives in this regard, both in relation to antibiotic sales data and to the surveillance of antibiotic resistance . However, it is crucial that sales data be provided by antibiotic class in each animal species. Without this it has little value in monitoring the relationship between antibiotic use and resistance levels. It is also vital that the Commission uses its powers to ensure that all Member States routinely monitor levels of antibiotic resistance in farm animals and on retail meat for *E. coli*, *Campylobacter*, *Enterococci*, *Salmonella* and *Staphylococcus aureus*.

Cóilín Nunan and Richard Young, March 2013.

References

[1] Ian Sample, 2013. Antibiotic-resistant diseases pose 'apocalyptic' threat, top expert says, *The Guardian*, http://www.guardian.co.uk/society/2013/jan/23/antibiotic-resistant-diseases-apocalyptic-threat

[2] Livermore D.M., 2008. New antibiotics – what we'll get & what we need, Powerpoint presentation on European Antibiotics Awareness Day, http://www.online-web-presentations.com/EAA08/PDF/DavidLivermore.zip

[3] WHO, 2011. Tackling antibiotic resistance from a food safety perspective in Europe, http://www.euro.who.int/__data/assets/pdf_file/0005/136454/e94889.pdf

[4] Defra Antimicrobial Resistance Co-ordination Group, 2011. Report of 41st meeting 3 May 2011, http://www.vmd.defra.gov.uk/pdf/darc_MinsMay11.pdf

[5] Health Protection Agency, 2012. Antibiotics and you, http://www.hpa.org.uk/NewsCentre/NationalPressReleases/2012PressReleases/121114AntibioticsandYou/

[6] Butler CC, Rollnick S, Pill R, Maggs-Rapport F, Stott N., 1998. Understanding the culture of prescribing: qualitative study of general practitioners' and patients' perceptions of antibiotics for sore throats, *British Medical Journal*, **317**: 637-42, http://www.ncbi.nlm.nih.gov/pmc/articles/PMC28658/pdf/637.pdf

[7] Butler CC, Simpson SA, Dunstan F, Rollnick S, Cohen D, Gillespie D, Evans MR, Alam MF, Bekkers MJ, Evans J, Moore L, Howe R, Hayes J, Hare M, Hood K., 2012. Effectiveness of multifaceted educational programme to reduce antibiotic dispensing in primary care: practice based randomised controlled trial, *British Medical Journal*, **344**, http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3270575/pdf/bmj.d8173.pdf

[8] Aymes, Sebastian, 2001. Magic Bullets, Lost Horizons. Taylor and Francis, London and New York

[9] DARC, 2011. Consideration of independent evaluation of research to augment communication of research finding, Report of the Forty Second meeting of the Defra Antimicrobial Resistance Co-ordination Group, item 7, 2 August 2011 http://www.vmd.defra.gov.uk/public/antimicrobial_darc.aspx#summary

[10] Hansard, 2013. http://www.publications.parliament.uk/pa/cm201213/cmhansrd/cm130109/halltext/130109h0002.htm#13010947000001

[11] VMD, 2009. Defra Antimicrobial Resistance in Animals policy Statement, http://www.vmd.defra.gov.uk/public/antimicrobial_statement.aspx

[12] European Food Safety Authority, 2008. Foodborne antimicrobial resistance as a biological hazard, Scientific Opinion of the Panel on Biological Hazards, Adopted on 9 July 2008, http://www.efsa.europa.eu/de/scdocs/doc/765.pdf

[13] Lloyd D.H., 2007. Reservoirs of Antimicrobial Resistance in Pet Animals, *Clinical Infectious Diseases*, **45** S2: S148-S152, http://cid.oxfordjournals.org/content/45/Supplement_2/S148.full.pdf+html

[14] Veterinary Medicines Directorate, 2012. Sales of antimicrobial products authorized as veterinary medicines in the UK in 2011, http://www.vmd.defra.gov.uk/pdf/salesanti11.pdf

[15] Responsible Use of Medicines in Agriculture, 2012. RUMA comment on the Early Day Motion in the House of Commons on the use of antibiotics in intensive farming, http://www.ruma.org.uk/news/20121105.htm

[16] DARC and ARHAI, 2012. ESBLs – A threat to human and animal health? http://www.vmd.defra.gov.uk/pdf/ESBL_report.pdf

[17] Phillips I., Casewell M., Cox T., De Groot B., Friis C., Jones R., Nightingale C., Preston R. and Waddell J., 2004. Does the use of antibiotics in food animals pose a risk to human health? A critical review of

published data, Journal of Antimicrobial Chemotherapy, 53: 28-52, http://jac.oxfordjournals.org/content/53/1/28.full.pdf+html

[18] Collignon P., 2009. Resistant Escherichia coli – We Are What We Eat, Clinical Infectious Diseases, 49: 202-4, http://cid.oxfordjournals.org/content/49/2/202.full.pdf+html

[19] Hammerum A.M. and Heuer O.E., 2009. Human health hazards from antimicrobial-resistant Escherichia coli of animal origin, Clinical Infectious Diseases, 48: 916-921, http://cid.oxfordjournals.org/content/48/7/916.full.pdf+html

[20] Vieira A.R., Collignon P., Aarestrup F.M., McEwen S.A., Hendriksen R.S., Hald T. and Wegener H.C., 2011. Association Between Antimicrobial Resistance in Escherichia coli Isolates from Food Animals and Blood Stream Isolates from Humans in Europe: An Ecological Study, Foodborne Pathogens and Disease, 8: 1295-301

[21] Smith H.W., 1969. Transfer of antibiotic resistance from animal and human strains of Escherichia coli to resident E. coli in the alimentary tract of man, Lancet, 1: 1174-6

[22] Moodley A. and Guardabassi L., 2009. Transmission of IncN plasmids carrying blaCTX-M-1 between commensal Escherichia coli in pigs and farm workers, Antimicrobial Agents and Chemotherapy, http://aac.asm.org/content/53/4/1709.full.pdf+html

[23] Teale C.J., 2010. ESBLs in animals, *Journal of Antimicrobial Chemotherapy*, **65** Suppl. 1, i8, http://jac.oxfordjournals.org/content/65/suppl_1/i3.full.pdf+html

[24] Johnson A.P., Burns L., Woodford N., Threlfall E.J., Naidoo J., Cooke E.M. and George R.C., 1994. Gentamicin resistance in clinical isolates of Escherichia coli encoded by genes of veterinary origin, Journal of Medical Microbiology, 40: 221-6, http://jmm.sgmjournals.org/content/40/3/221.long

[25] Johnson J.R., Kuskowski M.A., Menard M., Gajewski A., Xercavins M. and Garau J., 2006. Similarity between human and chicken Escherichia coli isolates in relation to ciprofloxacin resistance status, The Journal of Infectious Diseases, 194: 71-8, http://jid.oxfordjournals.org/content/194/1/71.full.pdf+html

[26] Johnson J.R., Sannes M.R., Croy C., Johnston B., Clabots C., Kuskowski M.A., Bender J., Smith K.E., Winokur P.L. and Belongia E.A., 2007. Antimicrobial drug-resistant *Escherichia coli* from humans and poultry products, Minnesota and Wisconsin, 2002-2004, *Emerging Infectious Diseases*, **13**: 828-46, http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2792839/pdf/06-1576_finalR.pdf

[27] Corpet D.E., 1988. The New England Journal of Medicine, 318: 1206-7, http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2814222/pdf/halms430932.pdf

[28] Defra Antimicrobial Resistance Co-ordination Group and Advisory Committee on Antimicrobial Resistance and Healthcare Associated Infection, 2012. ESBLs – A threat to humanand animal health?, http://www.vmd.defra.gov.uk/pdf/ESBL_report.pdf

[29] Woodford Neil, 2013. New funding secured for ESBL research, AMRHAI news Winter 2013 Issue 02, Health Protection Agency

[30] Health Protection Agency, 2005a. HPA publishes report on infections caused by ESBL-producing E. coli, HPA press release 12 September 2005, http://www.hpa.org.uk/webc/HPAwebFile/HPAweb_C/1253205583156

[31] Health Protection Agency, 2005b. Investigations into multi-drug resistant ESBL-producing Escherichia coli strains causing Infections in England: 2005, http://www.hpa.org.uk/webc/HPAwebFile/HPAweb_C/1274090495083

[32] Munday C.J., Whitehead G.M., Todd N.J., Campbell M. and Hawkey P.M., 2004. Predominance and genetic diversity of community- and hospital-acquired CTX-M extended-spectrum beta-lactamases in York, UK, Journal of Antimicrobial Chemotherapy, 54: 628-33, http://jac.oxfordjournals.org/content/54/3/628.full.pdf+html

[33] Eustace A., Wickramasinghe N., Xu L., Saluja T., Shabir S. and Hawkey P., 2011. High faecal carriage rates of CTX-M ESBL-producing Escherichia coli in the Birmingham area: implications of global origin, Abstract presented at the 21st European Congress of Clinical Microbiology and Infectious Diseases (ECCMID), http://registration.akm.ch/einsicht.php?XNABSTRACT_ID=125298&XNSPRACHE_ID=2&XNKONGRESS_ID=136&XNMASKEN_ID=900

[34] Leverstein-van Hall M.A., Dierikx C.M., Cohen Stuart J., Voets G.M., van den Munckhof M.P., van Essen-Zandbergen A., Platteel T., Fluit A.C., van de Sande-Bruinsma N., Scharinga J., Bonten M.J., Mevius D.J. and the National ESBL surveillance group., 2011. Dutch patients, retail chicken meat and poultry share the same ESBL genes, plasmids and strains, *Clinical Microbiology and Infection*, **17**: 873-80

[35] Overdevest I., Willemsen I., Rijnsburger M., Eustace A., Xu L., Hawkey P., Heck M., Savelkoul P., Vandenbroucke-Grauls C., van der Zwaluw K., Huijsdens X. and Kluytmans J., 2011. Extended-spectrum β-lactamase genes of Escherichia coli in chicken meat and humans, The Netherlands, Emerging Infectious Diseases, 17: 1216-22, http://wwwnc.cdc.gov/eid/article/17/7/pdfs/11-0209.pdf

[36] Kluytmans JA, Overdevest IT, Willemsen I, Kluytmans-van den Bergh MF, van der Zwaluw K, Heck M, Rijnsburger M, Vandenbroucke-Grauls CM, Savelkoul PH, Johnston BD, Gordon D, Johnson JR., 2013. Extended-Spectrum β-Lactamase-Producing Escherichia coli From Retail Chicken Meat and Humans: Comparison of Strains, Plasmids, Resistance Genes, and Virulence Factors, *Clinical Infectious Diseases*, **56**: 478-87

[37] Bates J, Jordens JZ, Griffiths DT., 1994. Farm animals as a putative reservoir for vancomycin-resistant enterococcal infection in man, *Journal of Antimicrobial Chemotherapy*, **34**: 507-14

[38] Klare I, Heier H, Claus H, Reissbrodt R, Witte W., 1995. vanA-mediated high-level glycopeptide resistance in *Enterococcus faecium* from animal husbandry, *FEMS Microbiology Letters*, **125**: 165-71

[39] Wegener HC, Aarestrup FM, Jensen LB, Hammerum AM, Bager F., 1999. Use of antimicrobial growth promoters in food animals and Enterococcus faecium resistance to therapeutic antimicrobial drugs in Europe, *Emerging Infectious Diseases*, **5**: 329-335, http://wwwnc.cdc.gov/eid/article/5/3/pdfs/99-0303.pdf

[40] Coque TM, Tomayko JF, Ricke SC, Okhyusen PC, Murray BE., 1996. Vancomycin-resistant enterococci from nosocomial, community, and animal sources in the United States, *Antimicrobial Agents and Chemotherapy*, **40**: 2605-9, http://www.ncbi.nlm.nih.gov/pmc/articles/PMC163584/pdf/402605.pdf

[41] Silverman J, Thal LA, Perri MB, Bostic G, ZervosMJ., 1998. Epidemiologic evaluation of antimicrobial resistance in community-acquired enterococci, *Journal of Clinical Microbiology*, **36**: 830-2, http://www.ncbi.nlm.nih.gov/pmc/articles/PMC104638/pdf/jm000830.pdf

[42] Klare I, Badstübner D, Konstabel C, Böhme G, Claus H, Witte W., 1999. Decreased incidence of VanA-type vancomycin-resistant enterococci isolated from poultry meat and from fecal samples of humans in the community after discontinuation of avoparcin usage in animal husbandry, *Microbial Drug Resistance*, **5**: 45-52

[43] Bager F, Aarestrup FM, Madsen M, Wegener HC., 1999. Glycopeptide resistance in Enterococcus faecium from broilers and pigs following discontinued use of avoparcin, *Microbial Drug Resistance*, **5**: 53-6

[44] van den Bogaard AE, Bruinsma N, Stobberingh EE., 2000. The effect of banning avoparcin on VRE carriage in The Netherlands, *Journal of Antimicrobial Chemotherapy*, **46**: 146-8, http://jac.oxfordjournals.org/content/46/1/146.full.pdf

[45] EFSA Panel on Biological Hazards (BIOHAZ), 2011. Scientific Opinion on the public health risks of bacterial strains producing extended-spectrum β-lactamases and/or AmpC β-lactamases in food and food-producing animals, http://www.efsa.europa.eu/en/efsajournal/doc/2322.pdf

[46] Voss A., Loeffen F., Bakker J.,Klaassen C. and Wulf M., 2005. Methicillin-resistant Staphylococcusaureus in pig farming, EmergingInfectious Diseases, 11: 1965–1966, http://wwwnc.cdc.gov/eid/article/11/12/pdfs/05-0428.pdf

[47] NethmapMaran 2012, http://www.uu.nl/SiteCollectionImages/Fac_DGK/Nieuwsplaatjes/Nieuws/2012/NethmapMaran_Web.pdf

[48] Paterson et al., 2012. First detection of livestock-associated meticillin-resistant Staphylococcus aureus CC398 in bulk tank milk in the United Kingdom, January to July 2012, Eurosurveillance,17, http://eurosurveillance.org/ViewArticle.aspx?ArticleId=20337

[49] Khanna T., Friendship R., Dewey C. and Weese J.S., 2007. Methicillin resistant Staphylococcus aureus colonization in pigs and pig farmers, *Veterinary Microbiology*, **128**: 298-303

[50] Arriola C.S., Güere M.E., Larsen J., Skov R.L., Gilman R.H., Gonzalez A.E. and Silbergeld E.K., 2012. Presence of methicillin-resistant Staphylococcus aureus in pigs in Peru, *PlosOne*, **6**, http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3234269/pdf/pone.0028529.pdf

[51] Pantosti A., 2012. Methicillin-Resistant Staphylococcus aureus Associated with Animals and Its Relevance to Human Health, *Frontiers in Microbiology*, **3**: 127,http://www.ncbi.nlm.nih.gov/pmc/articles/pmid/22509176/

[52] Porrero MC, Wassenaar TM, Gómez-Barrero S, García M, Bárcena C, Alvarez J, Sáez-Llorente JL, Fernández-Garayzábal JF, Moreno MA, Domínguez L., 2012. Detection of methicillin-resistant Staphylococcus aureus in Iberian pigs, *Letters in Applied Microbiology*, **54**: 280-5

[53] Tenhagen BA, Fetsch A, Stührenberg B, Schleuter G, Guerra B, Hammerl JA, Hertwig S, Kowall J, Kämpe U, Schroeter A, Bräunig J, Käsbohrer A, Appel B., 2009. Prevalence of MRSA types in slaughter pigs in different German abattoirs, *Veterinary Record*, **165**: 589-93

[54] de Neeling AJ, van den Broek MJ, Spalburg EC, van Santen-Verheuvel MG, Dam-Deisz WD, Boshuizen HC, van de Giessen AW, van Duijkeren E, Huijsdens XW., 2007. High prevalence of methicillin resistant Staphylococcus aureus in pigs, *Veterinary Microbiology*, **122**: 366-72

[55] Aarestrup FM, Wegener HC, Collignon P., 2008. Resistance in bacteria of the food chain: epidemiology and control strategies, *Expert Review of Anti-Infective Therapy*, **6**: 733-50

[56] Burch D., 2010. MRSA in pigs: a link with cephalosporins?, *Pig Progress*, **26**, http://www.octagon-services.co.uk/articles/cephalosporins.pdf

[57] Woodford N., Carattoli A., Karisik E., Underwood A., Ellington M.J. and Livermore D.M., 2009. Complete nucleotide sequences of plasmids pEK204, pEK499, and pEK516, encoding CTX-M enzymes in three major Escherichia coli lineages from the United Kingdom, all belonging to the international O25:H4-ST131 clone, Antimicrobial Agents and Chemotherapy, 53: 4472-82, http://aac.asm.org/content/53/10/4472.full.pdf+html

[58] European Medicines Agency, 2011. Trends in the sales of veterinary antimicrobial agents in nine European countries, Reporting period: 2005-2009, http://www.ema.europa.eu/docs/en_GB/document_library/Report/2011/09/WC500112309.pdf

[59] Commission Implementation Decision concerning, in the framework of Article 35 of Directive 2001/82/EC of the European Parliament and of the Council; the marketing authorisations for veterinary medicinal products which contain the active substances "Cefquinome and Ceftiofur"

[60] Gaze WH, Zhang L, Abdouslam NA, Hawkey PM, Calvo-Bado L, Royle J, Brown H, Davis S, Kay P, Boxall AB, Wellington EM., 2011. Impacts of anthropogenic activity on the ecology of class 1

integrons and integron-associated genes in the environment, *The ISME Journal*, **5**: 1253-61, http://www.nature.com/ismej/journal/v5/n8/pdf/ismej201115a.pdf

[61] European Medicines Agency, 2011. Question and answer on the CVMP guideline on the SPC for antimicrobial products (EMEA/CVMP/SAGAM/383441/2005), http://www.ema.europa.eu/docs/en_GB/document_library/Other/2011/07/WC500109155.pdf

[62] Responsible Use of Medicines in Agriculture, 2012. RUMA comment on the Early Day Motion in the House of Commons on the use of antibiotics in intensive farming, http://www.ruma.org.uk/news/20121105.htm
This press release by the alliance representing the pharmaceutical and farming industries argues against a Parliamentary motion calling for a ban on 'routine prophylactic use'. RUMA also explicitly says that it 'does not consider the delay of treatment until the development of clinical signs of disease is always appropriate', apparently in breach of the EMA's 2011 clarification.

[63] Centre d'Analyse Stratégique, 2012. Les bactéries résistantes aux antibiotiques, http://www.strategie.gouv.fr/system/files/2012-11-15-antibiotiques-na299_0.pdf

[64] Agersø Y. and Aarestrup F.M., 2013. Voluntary ban on cephalosporin use in Danish pig production has effectively reduced extended-spectrum cephalosporinase-producing Escherichia coli in slaughter pigs, *Journal of Antimicrobial Chemotherapy*, **68**: 569-72

[65] Bruschke CLM, 2013. Letter from the Dutch Chief Veterinary Officer to Peter Stevenson of Compassion in World Farming

[66] http://www.fda.gov/AnimalVeterinary/SafetyHealth/RecallsWithdrawals/ucm042004.htm

[67] DARC, 2011. ESBL in livestock, Report of the Forty Second meeting of the Defra Antimicrobial Resistance Co-ordination Group, item 6.3, 2 August 2011

[68] European Medicines Agency, 2012. Sales of veterinary antimicrobial agents in 19 EU/EEA countries in 2010, http://www.ema.europa.eu/docs/en_GB/document_library/Report/2012/10/WC500133532.pdf

[69] EFSA and ECDC, 2012. The European Union Summary Report on Trends and Sources of Zoonoses, Zoonotic Agents and Food-borne Outbreaks in 2010, http://www.efsa.europa.eu/en/efsajournal/doc/2597.pdf