



Antimicrobial Resistance: A One Health Concept Perspective Analysis

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Introduction

Since the 1940s, antimicrobial drugs have substantially reduced morbidity and mortality associated with infectious diseases. The widespread availability of effective antimicrobials has enabled advancements in modern medical and veterinary practice that would have been unachievable without them. Antimicrobial drugs are a precious resource used every day all over the world to effectively treat infections in humans and animals.

This paper aims to review under “One Health” concept in particular antimicrobial resistance and the unnecessary use of antibiotics as well as strategies and alternatives to keep antimicrobial drugs useful as long as possible.

About Antimicrobial Resistance

Antimicrobial resistance represents the ability of a microorganism (like a bacterium, fungi, virus or parasite) to stop an antimicrobial drug from working against it. As a result, standard medical treatments could become ineffective, infection persists and may spread to others. Resistance to current available antimicrobials is increasing faster than the development of new drugs, and so effective treatments cannot keep pace.

The ability of bacteria to develop resistance was recognised almost as soon as antibiotics were discovered, and the microorganisms were challenged with them [1]. The evolution of resistant strains, to some extent, is a natural phenomenon that occurs when bacteria replicate themselves erroneously or when antimicrobial resistance traits are exchanged among bacteria. Each time antibiotics are used bacteria either die or adapt by acquiring resistance.

Antimicrobial resistance is recognised as a significant global health priority that threatens to take modern medical and veterinary practice back into the pre-antibiotic era, when infection risks prevented procedures that are today considered routine, and simple infections caused significant harm. It is a shared concern of both developed and developing countries.

Implications of Antimicrobial Resistance

Antimicrobial resistant infections implicate additional laboratorial investigations, more complex and expensive treatments, longer hospital stays, and lead to higher mortality. On the other hand, extended recovery time means that patients remain infectious for longer periods, shading the infectious agent to the environment and contact persons, increasing the risk of resistant organisms spreading to others. Treating resistant infections is also extremely expensive, as more costly medicines and resources need to be used. Longer hospital stays result in increased healthcare costs for patients and society.

In animals, antimicrobial resistant infections result in reduced animal health, reduced welfare, poor bio-security and underperforming production outcomes. Antimicrobial resistant infections in animals can also result in the transfer of resistant organisms to humans who come into direct contact with them, or by vectors or following animal consumption (foodborne transmission), which underscores the importance of the bond between human and veterinary medicines, as observed by the “One Health” approach [2].

In an attempt to conduct a global assessment of this topic, WHO reviewed the resistance data of selected bacteria from 114 countries and the findings were alarming. Antimicrobial resistance associated with common infections (e.g., urinary tract infections, pneumonia, and bloodstream infections) has reached a level where standard treatment most readily available in many parts of the world has been rendered ineffective. In addition, antibiotic usage in Animal Health adds further complexity to this process due to the spread of resistance genes across species, and which should be carefully monitored [3]. Resistance to last-line therapy, such as carbapenems, and a lack of novel antibiotic drugs adds to problematic current global situation [3]. Moreover, systematic reviews show that patients infected with antibiotic resistant bacteria are not only at risk of poorer health outcomes but also consume significantly more health resources (e.g., lengthier hospital stays, more frequent ICU admission or use of long-term care facilities) [3].

The Increase of Antimicrobial Resistance

There are several concomitant factors contributing to the increasing antimicrobial resistance while it is clear though that certain anthropogenic activities accelerate this process of increasing resistance, the single most powerful contributor to this finding is the global unrestrained use of antibiotic drugs. This includes underuse, overuse and misuse and applies both to the use of such drugs in human and animal health.

Non-prescription Access to Antimicrobials

Many individuals can buy antimicrobials and other drugs without a prescription, either because they cannot access formal clinical advice, or because they decide to self-medicate as a matter of personal choice. However, governments and health authorities should seek to reduce this kind of self-medication in shortest possible term. This goal can be achieved by improving access to proper clinical advice, restraining the free or non-prescription access to antimicrobials, controlling the medicine distribution chain, as well as by implementing educational campaigns about understanding the personal and public risks of self-medicating (whilst recognizing that in some very particular settings, informal or over-the-counter access to antimicrobials may be the only route to accessing urgent medications). The same problem can be tackled in animal health; many livestock producers can buy antimicrobials without veterinary prescription leading to overuse, underuse and misuse, which in time, leads to drug's resistance. This occurs even in countries in which the access to antimicrobials is highly regulated but poorly controlled.

Medicines sales over the internet is today a common phenomenon that only began in the 1990s [4] but that aggravates this scenario of antimicrobial resistance. Online sales of medicines are convenient for patients who are unable to reach a pharmacy, either for reasons of mobility or simple convenience. Properly regulated online pharmacies, servicing legitimate prescriptions, are a thus natural and indeed welcome evolution of the pharmaceutical retail sector. However, there are nonetheless significant risks associated with the growth of online non-regulated pharmaceutical sales, especially with respect to the sale of antimicrobials and the development of antimicrobial resistance.

Poor Quality Forged Antimicrobial Drugs

The risks associated with the internet sales of antimicrobials go far beyond the problem of excessive consumption or the sale of antimicrobial drugs without a prescription. The problem of falsified and poor quality products arises daily. Regardless of how antimicrobials are accessed, doctors and patients need to be sure that the drugs that they use are effectively what they are supposed to be, and of good quality - something that, sadly, cannot always be taken for granted. Poor quality antimicrobials can represent a significant public health concern, as they deliver a sub-therapeutic dosage of the active ingredient. This provides a selective advantage to drug-resistant microbes, encouraging such organisms to develop and spread. Illegal internet pharmacies that take advantage of regulatory gaps or blind spots will often be operating beyond

normal arrangements for oversight of the quality of the products on sale. This increases the risk that the drugs sold by spurious websites could be falsified or of very poor standards. For instance, a fake version of the antiviral drug Tamiflu was available on fraudulent internet pharmacy sites within weeks of the 2009 H1N1 pandemic being declared by the World Health Organization [5]. Even when online vendors sell antimicrobials "by prescription", there may be no mechanism to verify the authenticity and accuracy of a script. There is no doubt that such phenomenon has the potential to have a significant impact on antimicrobial drug sales [6] and that the emergence of unregulated and unscrupulous online retailers represents huge regulatory challenge. Demonstrating the potential scale of the problem, a study carried out in 2009 showed that a wide range of antibiotics were available online, including penicillin's, macrolides, fluoroquinolones and cephalosporin's, identifying 136 unique vendors who would accept to ship them to the United States without a prescription [7].

The availability of antibiotics on line represents an international problem, and requires global answer from pharmaceutical regulators, customs authorities, transport companies and internet companies. At present, we are confronted with significant gaps in the international regulations, which prevent the governments and authorities to control the movement of antibiotics and other medicines from one country to another. Concerted international efforts are needed to ensure effective antimicrobial access control and consistent standards in internet sales. The international coordination, led by Interpol, targeting illegal online pharmacies was a notable success story in 2015. Operation Pangea VIII, counted with 115 countries involved, targeted criminal networks responsible for the sale of forged medicines by illegal online pharmacies. This operation resulted in the seizure of 81 million USD worth of potentially dangerous medicines, 156 arrests across the world and the shutdown of two internet domain names selling these drugs. It represented the largest ever internet-based operation and involved multiple international agencies from government agencies, to private sector companies such as Google, MasterCard, Visa and PayPal [8]. This shows how relevant the organizations from across the world coming together to address this issue can achieve.

Unnecessary Use of Antimicrobials

There is a global vast overuse of antibiotic drugs, either in rich or poorer countries. There are no reliable data available to document the unnecessary use of antibiotics globally, but the scale of the problem is no doubt enormous. Globally, the increase in antibiotic resistance has been driven by the unrestrained use of antibiotics both in human and animal health.

An academic study of medical prescriptions in United States primary and outpatient care considered adult patients visiting their doctor with a respiratory problem [9] described the following pattern of antibiotic use: for a total of 106 million visits in one year, 86 million patients were considered to have respiratory problems for which antibiotics would be of no help, for example bronchitis or asthma. Of those 86 million persons, however, 27

million patients were prescribed an antibiotic unnecessarily.

Use of Antimicrobials in Animal Health

Antimicrobials are used in Animal Health for exactly the same purposes and reasons that they are used in humans - to prevent and treat diseases. Many consumers express concerns that in modern agricultural units, pigs and poultry are kept indoors in high densities than in traditional systems. Closely packed and stressed animals will succumb easily to infectious diseases and need more antibiotics. However, modern agricultural units can only flourish because the animals are productive; but they are only productive if they are healthy and not stressed.

The growth-promoting antibiotic products and the use of medically important antibiotics to improve weight gain and feed efficiency were banned throughout Europe in 2006. Antibiotics were sometimes used at lower concentrations in feed given to cattle, pigs and poultry to control the bacteria in the gut, which caused low-level diseases, which prevented the animals from developing at their optimum rate. This was quite evident when Denmark became the first country to ban such products; Danish farmers suffered higher mortalities and morbidities in young piglets and initially had to double the use of antibiotics to treat them [10].

A poor welfare can be a primary predisposition to specific diseases and may affect animal health by altering the susceptibility to several diseases. Changes in the livestock industry have had positive consequences like greater hygiene and bio-security that reduce the risk of infectious disease. Keeping pigs and poultry indoors allows farmers to maintain conditions that will protect the health and welfare of their animals - for example, keeping out predators, such as foxes, wild rodents and birds that can introduce infectious diseases. But whichever farming system is used, once a new disease is introduced into a social group it will spread rapidly because animals are in close contact with each other and use the same water and food troughs.

Some strains of antibiotic-resistant bacteria are found in farm animals and there is a theoretical risk that they could cause health problems in humans, either by direct transmission from animal to human, bacterial contamination of food or by resistance gene exchange between animal and human bacterial strains.

Despite recognition by producers and veterinarians that these risks do exist, it is clear that they don't play a clear major role in the development of multi-drug resistant bacterial infections in humans. Scientific evidence shows that hospital patients colonized/infected with much resistant organisms contracted organisms from another patient health personal or from contaminated surfaces of the hospital environment. Evidences supporting such assumption arise from a Center for Disease Control report [11].

In 2013, that listed 18 strains of antibiotic-resistant bacteria which pose a threat to human health; in only two cases were able to identify that livestock could be a potential source for resistant strains of *Salmonella* and *Campylobacter*. Both these agents are omnipresent in the environment and can cause unpleasant

gastrointestinal infections whether the strain is resistant or not. Risks can be effectively eliminated by good kitchen, hygiene and proper cooking of all meat and dairy products [12].

The Veterinary Antibiotic Resistance and Sales Surveillance Report - Veterinary Medicines Directorate in 2015, documented that "isolates of *E. coli* from the caeca of healthy pigs randomly selected at slaughter were tested for resistance; of the 150 isolates of *E. coli* tested, 1% were resistant to ciprofloxacin; none were resistant to cefotaxime, ceftazidime or colistin and no carbapenemase or OXA-48 producing *E. coli* were detected in 294 caecal samples cultured on selective agar". Furthermore, a total of 1594 *Salmonella* isolates from cattle, sheep, pigs, chickens and turkeys were tested and resistance to the highest priority critically important antibiotics was found to be very low, with 1.3% (20/1594) of all *Salmonella* resistant to ciprofloxacin and 0.1% were resistant to cephalosporins [13]. However, *E. coli* isolates from a combination of all livestock species were most frequently resistant to streptomycin, tetracycline and ampicillin. Resistance to the highest priority critically important antibiotics tested was generally low, with 9.3% resistant to cefotaxime, 7.2% resistant to cefpodoxime, and 10.7% resistant to enrofloxacin. This report concludes that the overall the level of resistance was low in bacteria associated with respiratory disease in sheep and cattle; nevertheless 0.6% of 313 samples from randomly sampled pigs and 1.2% of 163 isolates from clinical surveillance were positive for the *mcr-1* gene [13].

The tetracycline group makes up 40% of the total veterinary market. These were one of the first antibiotics to be developed and in many countries at present its use is limited to human patients only. But despite its sustained use for years in animals, veterinarians have found little evidence of resistant strains causing hard to treat infections [14]. However, there is a wide overlap in the use of medically valuable antibiotic groups such as macrolides, fluoroquinolones and cephalosporins; the latter is only used for individual animal treatment. Veterinary fluoroquinolones, for example, make up less than 2% of all antibiotic use in Europe; in United States is just over 1% [15]. These compounds are effective a treatment for cattle and pig respiratory diseases but are only available for use through a veterinarian's prescription. When used adequately, the likelihood of fluoroquinolone-use in cattle giving rise to an untreatable bacterial disease in a human patient is quite small - one recent study calculated that such finding would occur at a rate of a single case of *Salmonella* every 293 years in the United States [16].

The European Union summary report on antimicrobial resistance in zoonotic and indicator bacteria from humans, animals and food in 2015 describes two serovars in particular, *S. typhimurium* and monophasic *S. typhimurium*, contributing significantly to the overall numbers of multidrug-resistant *Salmonella* in Europe, but only one single *S. typhimurium* isolate from a calf under one year of age displayed high-level resistance to ciprofloxacin, while microbiological resistance was low in *Salmonella* spp. from pig meat (4.3%), from bovine meat (2.5%) and from fattening pigs (4.7%). The importance of these facts, from a public health

perspective, is relevant because ciprofloxacin is a common first-line treatment for invasive salmonellosis in humans [17]. However, despite resistance to azithromycin in *Salmonella* spp. isolates from pig meat being generally low or not detected, Portugal and Cyprus reported high levels of resistance, namely 37.5% and 25% prevalence respectively. Regarding resistance to carbapenems in *Salmonella* from fattening pigs and calves less than one year of age and meat resistance was not observed in any of the 28 reporting countries [17]. Regarding *Campylobacter* the resistance values are high both in human as in animal health for ciprofloxacin. The report clearly states that for human health “the level of acquired resistance to fluoroquinolones is so high in some Member States that these drugs can no longer be considered appropriate for routine empirical treatment of human *Campylobacter* infection” [17].

It is not clear that the resistances observed in human be the same found in animal isolates. The figures don't match and the data geographically does not match either [17]. The majority of bacteria are adapted to living on a particular host; thus, a bacterial strain found in cattle or sheep is unlikely to survive in humans. Surprisingly, a series of studies from different countries found bacteria resistant to the same range of antibiotics in humans and animals [12]. However, recent studies using accurate analytical techniques in Scotland and in the Netherlands have found that the genes causing resistance among different species are actually quite distinct, meaning that such resistant strains must have emerged independently [18]. Even the emerging antimicrobial resistances detected in humans due to antimicrobial residues present in the food chain are hard to explain. National survey results regarding antimicrobial residues in US and Europe typically report a rate of positive samples of substantially less than 1% [19]. This arises from the fact that any authorized medicine used in livestock will have a statutory withdrawal period, that is, there is a minimal amount of time that must be observed after drug treatment before meat, milk or eggs from that animal can enter the food chain. If contamination does occur, it is usually due to a mistake or oversight and not de liberate, since the penalties can be quite high.

Antimicrobial Resistance: A Comprehensive Perspective

Anti-microbial resistance constitutes a global threat and concerns several sectors, such as human and veterinary medicine, animal husbandry, farming, the environment and trade [20]. Thus, such a multifactorial problem demands comprehensive policy and concrete attitudes to be tasked with success.

Short Term Measures

The measures that should be implemented in an immediate way are condensed in the European Medicines Agency's Committee for Medicinal Products for Veterinary Use (CVMP) strategy on antimicrobials 2016-2020 (October 6th, 2016, EMA/CVMP/209189/2015):

“Aim 1: To provide opinions for the authorization of effective antimicrobial veterinary medicinal products ensuring that the

necessary risk management measures are applied so that products can be used safely and sustainably.

Aim 2: To consider and advise on the risk to public health that could arise from the use of antimicrobials in animals, and to balance this against the need to protect animal health. To provide advice in a One Health context, considering the interaction between humans, animals and the environment as sources of antimicrobial resistance genes.

Aim 3: To maintain the effectiveness of antimicrobial substances that are already authorized in veterinary medicinal products by monitoring and analysing their sales and usage, encouraging surveillance for changes in susceptibility of target pathogens and zoonotic bacteria, and subsequently reviewing the authorization of substances and/or products, especially when there is evidence that there may be a related change in the benefit-risk of the authorization.

Aim 5: To support the responsible use of antimicrobials both in accordance with Marketing Authorizations and under the cascade.”

Aim 5 is particularly important as the responsible use of antimicrobials involves, among other things, under veterinary control, with Antimicrobial Susceptibility Tests (AST), supporting the rational use, avoiding empiric use or misuse such as sub-therapeutical doses, fighting forgeries and smuggle of antimicrobials and controlling quality of antimicrobials and treatments. Controlling the easy access to antimicrobials can be a very effective tool to achieve all the previous points. Countries like Sweden and The Netherlands have shown how it is possible to keep antibiotic use low with current technology, both in human and animal health. More recently, countries like China and Brazil have made progress in reducing over-the-counter sales of antibiotics in large urban centres.

Strategically, all the stakeholders should look to the antimicrobials use with responsibility, ranging from the industry that produce and sells them, through physicians, veterinary surgeons, pharmacists, regulating authorities, customs, researchers, and finally to those who benefit from their use (patients either humans or animals).

Considering the public health, the CVMP supports the categorization of human critically important antimicrobials provided by the European Medicines Agency's Antimicrobial Advice Ad Hoc Expert Group (AMEG) and the establishment of a list of specific substances which represent a last resource for treatment of life-threatening diseases in humans and should be excluded from veterinary use. However, as it is also recognized by the CVMP, “the greatest producer of antimicrobial resistance among humans is the use of antimicrobials in human medicine” and CVMP also considers that risk management measures applied to Veterinary medical products should be proportionate and scientifically evidence-based. As the CVMP points out: “It is hoped that the new guidance will provide greater transparency

for pharmaceutical companies considering antimicrobial product development and address the “regulatory uncertainty” that has been identified as a contributor to the recent limited development of antimicrobial veterinary medicines”.

Long Term Measures

The need to develop new antimicrobial agents and novel strategies to control microbial pathogens arose, and considerable efforts have been made - although not always rewarded by satisfactory results - to find new functional compounds that can be used as an alternative to antibiotics when the latter are no longer strictly required, as pointed out by the European Medicines Agency’s Committee for Medicinal Products for Veterinary Use (CVMP) statement on strategy on antimicrobials 2016-2020 (October 6th, 2016, EMA/CVMP/209189/2015):

“**Aim 4:** To encourage the development of new and existing antimicrobial veterinary medicinal products (particularly those in the AMEG’s category 1) and alternatives to antimicrobials. To encourage the development of these products specially to fill therapeutic gaps and for minor uses and minor species.”

The origin of antimicrobial resistance in the environment is relevant to human health because of the increasing importance of zoonotic diseases as well as the need for predicting emerging resistant pathogens. The presence of mobile antimicrobial resistance elements in pathogenic bacteria made this topic more complex because of the prevalence of horizontal gene transfer, the process by which bacteria acquire genes from the environment. Many of the known antibiotic resistance genes are found in transposons, integrons or plasmids, which can be mobilized and transferred to other bacteria of the same or distinct species. There are evidences of the transfer of resistance elements to known human commensal bacteria from pathogens; gene transfer among members of the human intestinal microbiome is known to be extensive [21].

Several alternatives to antimicrobial use or adjuvant supporting techniques aiming to improve their use have so far been proposed such as:

Cooperating Commensals in Microbiota or Probiotics

One of the possibilities is to address the commensal microbiota. Administration of specific consortia of commensal bacteria can re-establish colonization resistance against highly antibiotic-resistant pathogens - for example: cooperating commensals restore colonization resistance to Vancomycin-Resistant *Enterococcus faecium*. Vancomycin-Resistant *Enterococcus* (VRE) can densely colonize the intestine and cause bloodstream infections. The intestinal microbiota provides resistance against VRE colonization. Caballero and colleagues demonstrated in mice that *Blautia producta* and *Clostridium boltea* can restore mucosal resistance against VRE. These findings suggest that therapeutic or prophylactic administration of defined bacterial consortia to individuals with compromised microbiota composition may reduce inter-patient transmission and intra-patient dissemination of highly antibiotic-resistant pathogens [22].

Biocontrol using Bacteriophage or Phage Therapy

Bacteriophages are viruses that only infect bacterial cells. Bacteriophage treatment of food-producing animals reduces the probability of bacterial contamination of the resulting food products during processing. Risk assessment models indicate that a 1 and 2-log reduction in the number of pathogens shed in feces of the slaughtered animal could reduce the risks to the consumers by 45 and 75 %, respectively. Scientific trials estimate that a reduction of 2 log *Campylobacter* loads in poultry intestines is sufficient to diminish 30-fold the incidence of campylobacteriosis associated with consumption of chicken meals [23]. *Campylobacter* phage administered to poultry by oral gavage associated to a reduction of 2 log CFU/g of the level of *C. coli* and *C. jejuni* in feces [24]. Similar studies have also been conducted with cattle showing reduction of fecal shedding of *E. coli* O157:H7 [25]. Many other trials have been conducted with success for reducing intestinal colonization and fecal shedding of *E. coli*, *Salmonella* and *Campylobacter* [26].

Bacteriophages are also effective in biocontrol of foodborne pathogens: *Salmonella* phage F01-E2 when added to turkey deli meats and chocolate milk resulted in 5 log reduction of CFU and a 3-log reduction when applied to hot dogs [27]. Another use was biocontrol of *Listeria monocytogenes* on Ready to Eat (RTE) poultry, where a reduction of CFU by 2.5 log at 30°C in RTE chicken; at 5°C, regrowth was prevented over 21 days [28]. Even in processed food the use of bacteriophages was interesting: a phage cocktail added to pasteurized milk challenged with *S. aureus*, led to reduction of *S. aureus* to undetectable levels after 6 h in fresh cheese and continuous reductions in hard cheese. In curd a reduction of 4.64 log CFU per g was obtained in comparison with control [29].

Unconventional Antimicrobials

Researching effective and safe novel compounds could also become a reliable alternative to conventional antibiotics. Antimicrobial peptides are a primitive component of the innate immune system of animals. There are already good examples of the antimicrobial effect of these compounds and their application in clinical practice; the combination of the antimicrobial peptide AMP2041, Chlorhexidine digluconate and Tris-EDTA. The peptide AMP2041, thanks to its particular amino acid sequence and high degree of hydro-solubility, makes a unique synergy possible with other components of the combination. This allowed building a formula with broad spectrum activity, fast effect, at low doses; safe in use and a low environmental impact, resulting in a new gel otologic product for the treatment of acute external otitis or recurrent bacterial and/or yeasts external otitis in dogs. The results have been confirmed by studies conducted by the laboratory of the Department of Medical and Veterinary Sciences, University of Parma [30-33].

The two antimicrobial peptides predominant families in mammalian species are cathelicidins and defensins. [34]. Cathelicidins are a family of small, cationic antimicrobial peptides,

which function as natural antibiotics through antimicrobial and immunomodulatory activities [34]. Cathelicidins directly kill a broad spectrum of bacteria [35], fungi [36] and parasites [37] through electrostatic interaction of positively charged peptides with the negative pathogen cell membrane, resulting in the formation of trans-membrane pores [38]. In addition, cathelicidins indirectly target pathogens by modulating cells of the innate and adaptive immune systems to alter the local cellular and chemical environment. This includes the release and suppression of pro-inflammatory mediators [39-41], modulating the formation of chemotactic gradients [42], the immune cell development and angiogenesis [43,44].

Not only peptides have antimicrobial effect; oligosaccharides, many of them present in milk, including the human milk, may establish those properties which help to explain why, among other things like passive antibodies transfer, breastfeeding is so important for the neonate. It has been demonstrated that human milk exhibits modulatory effects upon streptococcal biofilm formation. For instance, lactoferrin and IgA inhibit biofilm formation while lactose and casein enhance biofilm formation by *Streptococcus mutans* [45]. Pooled human milk oligosaccharides from distinct donors modulate biofilm formation by *Streptococcus agalactiae*. Furthermore, this particularly studies also reveal that pooled human milk oligosaccharides can inhibit bacterial growth [46].

Responsible use of unconventional antimicrobials could help to control any bacterial disease that does appear and could be particularly valuable in protecting healthy animals during weaning steps or transportation, when their normal immune defences are known to be diminished.

Vaccines

Vaccines help to prevent infections; consequently, they reduce the need to use antibiotics. This is true for vaccines that prevent bacterial infections, and it is also true for vaccines that prevent viral infections, such as the flu, a condition that should not be treated with antibiotics, but often is, unfortunately. This may result from the lack of rapid diagnostic tests or because patients buy drugs over the counter. Vaccine development is a high-risk endeavour, with high chances of failure, and often takes 10 years or more to complete, meaning we are a long way away from having them on the market. Vaccines already play an important role in preventing many diseases in farm animals, but they are likely to have a greater role in the future, since the economic pressure increases in farm animal production (it has been shown, along the years that is cheaper to vaccinate than to treat). The same scenario can be drawn for pets; it is not only cheaper to vaccinate than treat but also the surviving rates are much higher, and the sequelae are lower or inexistent. Many studies have shown the large gains for human health, as well, as costs avoided by vaccination [47]. Vaccines are considered among the most cost-effective ways to prevent morbidity and mortality from infectious diseases [48]. A recent study estimated that global coverage with a universal pneumococcal conjugate vaccine could potentially prevent 11.4 million days of antibiotic use per year in children younger

than five; these children would otherwise have been treated for *Streptococcus pneumoniae* [49].

Efforts must be developed in the short term to increase the use of the existing vaccines and to improve delivery in both the community and hospitals, as well as in farming systems. This will involve providing financial support in some cases. On the other hand, in long term, it is needed a renewed impetus in the science of vaccines and alternative approaches to make sure researchers from a wide range of fields and countries are looking for solutions that will reduce our dependence on antibiotics and will help tackle drug resistance. There are no licensed vaccines for any of the bacteria that are considered by the United States Centres for Disease Control and Prevention (CDC) to represent our most urgent antimicrobial resistant threats [50]. In Veterinary Medicine we can produce auto-vaccines from a bacterium isolate and test it in the flock or herd and apply it in a farm or in an outbreak area, preventing threat from spreading. However, for universal vaccines and more particularly the ones regarding viral diseases we come across large obstacles. The huge cost of developing vaccines has also led to the stagnation of many vaccine candidates in late stages of the pipeline. These often do not advance further due to the lack of funding of the regional large field or clinical trials. Clinical trials for vaccines require higher safety issues than those for drug trials. They are used on healthy subjects rather than people who are already ill. It may also be difficult to recruit into these kinds of trials due to the challenges of identifying 'at risk' patients. There are also large upfront capital costs for building and certifying production facilities, which often needs to take place before the results of clinical trials are available. If a vaccine candidate is not successful during phase III trials, then the company loses their research and development costs, along with the cost of their manufacturing facilities which were constructed in parallel with the phase III trials. It will require a long term sustained funding from several stakeholders, not relying solely on the pharmaceutical industry, which at the present moment, is keener on producing generic drugs less risky than researching vaccines. It may be needed the involvement of the public sector, universities and even philanthropic organizations. What better example than the malaria vaccine? It has been a challenge for so many years and finally we could witness the approval in 2015 by the European Medicines Agency [51], and is the first vaccine ever licensed for use against a parasite in Medicine (In Veterinary Medicine already existed a vaccine against Leishmaniosis). The point is: the vaccine was developed through a partnership between industry, academia and non-profit organizations. Though the efficacy of the vaccine is relatively low, it still represents a milestone achievement in the fight against malaria.

Rapid Diagnostics Tests

Since the development and market availability of new antimicrobials is insufficient to keep up the face with the increase in drug resistance, older drugs and adjuvants are used more widely, but microbes also evolve to resist them. Rapid point-of-care diagnostic tests are a central part of the solution to this demand problem, which results currently in enormous unnecessary antibiotic use.

Take, for example, a modern health care system such as the one in the United States: a study found adult patients visiting the doctor to treat respiratory problems, more than two-thirds of courses of antibiotics were likely to have been inappropriately prescribed for conditions that were not infections at all, or were caused by viruses - for which an antibiotic would do nothing [9]. To solve the problem of antibiotic unnecessary use, and to get the right drug to the right patient at the right time, neither regulation or stewardship programmes will not be enough; we need urgently new rapid diagnostic tests. The world needs a change on the paradigm in the way that technology is incorporated into the decision-making process around antibiotic use - whether that is in at home, the pharmacy, a doctor's office, hospital ward or farm.

Rapid diagnostic tests can play an important role in improving how we can use antimicrobials to improve infection treatment; help to slow down the rise of drug resistance by reducing the unnecessary use of antimicrobials, in particular antibiotics; and change our approach to treating bacterial infections through supporting targeted and more precise therapies. Improvement in rapid diagnostic tests, particularly AST, along with incentives to encourage their use, may well have a significant role in improving the accuracy of prescription use in the medium term.

Genomic and proteomic tests were especially important on the identification of the microorganisms but, regarding susceptibility only in some cases (facing well characterized mechanisms of resistance) it could help. New methods are in evaluation taking particular relevance the development of flow cytometric tests [52]. Phenotypically fast results can be obtained for the main antimicrobials from isolated colonies but also from positive blood cultures (patented).

Conclusion

Antimicrobials cure bacterial diseases in people and they also cure bacterial diseases in animals. They help to the compliance of our moral duty regarding the animals in our care. Antimicrobials play an important role in sustainable livestock production by preventing waste and inefficiencies caused by disease, and helping provide a safe supply of food. They keep our pets healthy, preventing the zoonotic diseases transmission to humans, thereby allowing a continued strong and healthy human-animal relationship.

Antimicrobials should always be used with high responsibility, under physician or veterinary prescription, and only when strictly required. They are powerful medical tools and their benefits need to be preserved for the generations to come. Consequently, they should always be handled in such a way that limits their potential for stimulating the development of resistant clones. This requires concerted policies and strategies by all stakeholders, governments, medical and veterinary authorities, borders and customs authorities, physicians, veterinary surgeons, farmers, pet owners and general public. Informed decision-making should base upon reliable and sound facts and must be able to distinguish these from myths and tactics, which misinform and mislead policy-makers and the general public. By themselves, antimicrobials are not a panacea

for dealing with health problems in man or animal.

Regarding animal health in particularly, veterinary surgeons, farmers and pet owners must appreciate that effective disease control does not rely on one single class of drugs. It requires a balanced approach involving careful attention to good hygiene, bio-security measures, nutrition, rapid and accurate diagnostics and the use of other preventive measures such as vaccines. We have always shared our environment and lived with bacteria and always will; paradoxically we can't survive without them. We need to study them and seek the way to control them and their dissemination, while at the same time manage the adequate use of antibiotics, vaccines and all veterinary medicines in order to maintain their efficacy.

Rapid diagnostic methods for AST could help the stewardship programmes and reduce the institution of broad-spectrum therapy when there is no need.

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