Original Article

IMPACT OF LEGISLATION ON ANTIBIOTIC USE AND AWARENESS OF BEEKEEPERS

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Abstract

Legislation plays a key role in regulating beekeeping practices and biosecurity measures, but different countries present conflicting legislative frameworks in terms of allowing antibiotic use. Currently the EU and the UK only permit antibiotics under the cascade principle, but while the UK considers the application in cases of European Foulbrood, the EU strongly discourages antibiotic use. In the US, antibiotics are registered for honeybees and their treatments are regularly authorised. This study relied on an international survey to assess the current use of antibiotics and general awareness among beekeepers, within the three legislative contexts. The results showed a similar use of antibiotics in all three geographical areas, regardless of the legislative context. General use and awareness indicated that the EU and US would likely benefit from national inspectors. Furthermore, even though beekeepers were somewhat aware of the various risks associated with improper antibiotic use, they would likely benefit from targeted training on the matter.

Keywords: antibiotic awareness, antibiotic use, AMR, bee management, legislation

INTRODUCTION

Wild and domestic pollinators contribute to local ecosystems and agricultural production by improving approximately 75% of global crops and increasing global crop value by nearly £153 billion (Gallai et al., 2009). Over recent decades pollinators have been subjected to anthropogenic disturbances and exponential disease spread due to a considerable increase in international honeybee and honeybee product trade (Mutinelli, 2011). The Terrestrial Animal Health Code established by the World Organisation for Animal Health (OIE) covers such infectious honeybee diseases as American foulbrood (*Paenibacillus larvae*) and European foulbrood (*Melissococcus plutonius*), which present significant challenges to honeybee health. Treatment of these diseases is currently impeded by a lack of approved pharmaceuticals applicable to honeybees. Countries typically prefer to encourage the improvement of good beekeeping practices and biosecurity measures that enable disease prevention and reduce the need for veterinary medicines. In recent years, interest in preventative biosecurity measures has been furthered by the need to reduce residues of veterinary medicines within honeybee products and prevent the development of antimicrobial resistance (FAO Factsheet on AMR in food, 2020). The World Health Organization (WHO) developed guidelines that present evidence-

based recommendations and best practice statements on the use of medically important antimicrobials in food-producing animals (WHO, 2017).

Legislation plays a key role in the responsible use of antibiotics (AB) and education of AB related risks (Kirchhelle, 2018). In the European Union (EU), there are no antibiotics currently approved for use in honeybees. A cascade principle has been put in place for antibiotic prescriptions, whereby MRLs are defined according to the Regulation (EU) 2018/470 (EC, 2018). However, many EU member states strongly discourage the use of antibiotics for American Foulbrood (AFB) and European Foulbrood (EFB) and enforce this recommendation with a 'zero residue' policy applied to internationally traded and imported honey ("Residues of Veterinary Medicinal Products - Food Safety - European Commission", 2020; Ministero della Salute, 2012; Tierarzneimittelkontrollgesetz Nr. 28/2002). Regardless of national policies, studies continue to report antibiotic residues in honey samples, suggesting regular and widespread use (Al-Waili et al., 2012; Reybroek et al., 2012; Galarini et al., 2015; Reybroek, 2017).

The United Kingdom (UK), while compliant with EU legislation regarding the residue limits, actively considers protocols that permit the use of antibiotics under the prescribing cascade in cases of European Foulbrood (The cascade: prescribing unauthorised medicines, 2020). UK legislation defines the prescribing cascade as a risk-based decision tree that permits veterinarians to prescribe medicines that have not been authorised for that specific species or condition ("Guidance on the use of cascade", 2013). Veterinarians therefore take on the responsibility of recommending the most appropriate formulation and defining the correct dose, administration and withdrawal period. For honeybees, the traditional role of veterinarians is instead replaced by the figure of the bee inspector. National bee inspectors work as part of the National Bee Unit in order to carry out regular inspections and testing to monitor honeybee disease outbreaks.

The official protocol for bee inspectors outlines that, subject to a laboratory diagnosis, lightly infected colonies can be treated between April and September with a single dose of antibiotics, prescribed by the Veterinary Medicine Directorate (Wilkins et al., 2007; Animal and Plant Health Agency, 2015). Antibiotic treatment can be conducted to hold back EFB until the following spring, when the shook swarm technique is applied to remove potentially infected and AB contaminated matrices (Animal and Plant Health Agency, 2015; Beebase - Beekeeping information resource for Beekeepers, 2020). On the contrary, use of antibiotics against American Foulbrood is strictly prohibited in the UK, and all cases of the disease must be reported to a local bee inspector for further action (Honeybees: protecting them from pests and diseases, 2018).

In the United States of America (US), the FDA has approved eleven applications for use in honeybees including oxytetracycline, tylosin and lincomycin, with added measures instituted in 2017 that require veterinary prescription for these ABs because of relevant concern in human medicine (Using Medically Important Antimicrobials in Bees, 2020). Because ABs in the US are approved and advised for treatment against such diseases as AFB and EFB, more specific guidelines define recommended use, including appropriate formulation, dosage and administration in honeybees as well as legally defined withdrawal periods before honey can be harvested (Databank, 2020). However, the recommended 42-day withdrawal period for the soluble powder concentrate form of oxytetracycline registered for AFB and EFB appears incompatible with reports from studies showing persistence of active metabolites within hive matrices for up to 290 days (Reybroek et al., 2012). Currently, legally defined MRLs in the US indicate that veterinary drug residues are permitted in the market's regulation. These have recently been defined as 750 ppb (µg/L) for Oxytetracycline and Lincomycin, and 500 ppb (µg/L) for Tylosin (Maximum Residue Limits (MRL) Database, 2020).

This study relied on an international survey to assess how legislation impacts the use of antibiotics and general antibiotic awareness among

beekeepers. The three geographical areas of the EU, UK and US presenting three different legislative frameworks were compared and evaluated.

MATERIAL AND METHODS

Data acquisition took place through an online survey designed in collaboration with the BPRACTICES project, the AMR Inter-departmental Working Group (AMRWG) and TECA beekeeping of the Food and Agriculture Organization of the United Nations (FAO), and Appalachian State University. The aim of the survey was to gather information regarding antibiotic use across the global beekeeping landscape as well as beekeepers' awareness surrounding antibiotic resistance. In order to gather an international overview of current AB use, the survey was offered on the FAO website and promoted by relevant partners involved in reaching out to a number of beekeeping associations. The survey was made available for a period of twelve months and offered in ten languages for the purpose of inclusivity and further outreach (Harzing et al., 2013).

Respondents voluntarily completed the anonymous survey, providing demographic information such as geographical region of practice, country of origin and years of experience in beekeeping. For the purpose of the current study, only responses from European Union countries, the United Kingdom and the United States of America were included, in order to facilitate an accurate analysis of AB use and awareness under the three chosen legislative frameworks. Responses within the EU were primarily from Italy, Denmark, Belgium and France, all presenting a similar approach in discouraging antibiotic use. The number of respondents vary across countries, so we aggregated the data. The survey asked a variety of questions (see Appendix) pertaining to practices engaged by beekeepers to address diseases. For each statement, respondents were asked to indicate whether they agreed or disagreed. Variables were coded 0="no" or "disagree", 1="yes" or "somewhat agree", and increased incrementally for each category. A primary statistical analysis focused on mean comparisons between the outlined areas of the EU, UK and US. This approach identified statistically significant differences among key regions allowing for insight into discrepancies under the different legislative frameworks. One-way ANOVA testing was carried out with the aim of comparing apiary practices in the EU (control group) to those in the UK and US. A minimum significance threshold of $p<0.05$ was set to determine statistical significance.

RESULTS

Of the 298 responses, 48.9% (n=146) were from EU countries, 32.2% (n=96) were from the UK and 18.8% (n=56) were from the US. Hobbyist beekeepers were the most common beekeeper type in all regions, with the UK presenting the highest percentage (93%), and US and EU reporting respectively 89% and 78%.

Antibiotic use

Antibiotic use ranged from two to four percent in the three geographical areas, with the EU and US reporting the highest percentages (4%), however with no statistical difference between the EU and US or UK (2%). Respondents in the UK were less likely to use ABs without following label instructions, but the difference was not significant. Reports pertaining to the diseases treated with ABs showed that use against *Nosema* was significantly higher among US beekeepers (p=.0095) and European Foulbrood (p=.0045) compared to that of the EU (Fig. 1 & 2).

EU beekeepers primarily purchased antibiotics from pharmacies compared to the US where beekeepers acquired ABs from agro-chemical supply shops or the internet, and the UK where beekeepers resorted to national bee inspectors. All three were likely to purchase from veterinarians with no statistical difference between areas (5-11%).

A lack of significance showed accordance among all three geographical areas, indicating ABs were primarily used to treat infections instead

of prevention. Moreover, in all areas, between 60-65% of respondents stated the necessity of a prescription to purchase antibiotics.

Antibiotic awareness

In a self-identifying knowledge score, all three regions reported having little or some knowledge pertaining to AB issues, with UK respondents reporting significantly less knowledge (p=.013) (Fig. 4). When investigating the function of ABs, the US beekeepers reported believing ABs are medicines that prevent diseases (p=.003) (Fig. 3). The UK beekeepers were more aware of ABs being used to target bacteria compared to the EU, which presented the lowest average response rate. When asked where information regarding ABs was found, EU respondents were statistically more likely to obtain information from veterinarians or other beekeepers compared to both the US and the UK (p=.031; p=.0022), but also the least likely to obtain the information from a beekeeper association (p=.0017; p=.0003). US respondents were more likely to obtain information from books, the internet (p=.0009) and extension services (p=.00001). UK respondents reported using national bee inspectors (p=.0035).

Experience in recognizing bee resistance to medicines was significantly higher for the EU compared to the UK (p=.03) and US (p=.016) (Fig. 5). US responders indicated being more likely to witness AB failure in treating honeybees, although the difference was not significant. UK respondents were also less informed regarding AB residues compared to EU respondents. Responders from all three regions agreed that products deriving from AB treated hives should not be consumed. Beekeepers also believed antibiotic resistance would have a strong impact on their personal lives and hives. Both questions yielded no statistical difference among geographical areas.

DISCUSSION

The aim of the current study was to determine whether differing legislative frameworks have an impact on biosecurity measures and beekeeper behaviour. Specifically, the study sought to discern whether more restrictive policies resulted in better use and understanding of ABs. Hence, the current study surveyed beekeepers from countries presenting three different legal contexts to define current beekeeping practices and antibiotic use, while also assessing knowledge and awareness with regards to antibiotic related risks. The results suggested that beekeepers under all three frameworks have a general understanding of legislative oversight and are for the most part compliant with legal requirements. The findings however also indicated that AB use and knowledge do not appear to be significantly affected by legislation and that other contributing factors may be involved. Two tables were produced to summarize outputs obtained by the application of this survey as a risk assessment tool: the first outlining the major shortcomings of antibiotic use and awareness, and the second suggesting corrective actions (Tab. 1 and 2).

Antibiotic use

Compliance was assessed by asking beekeepers if antibiotics had been used over the past two years, and respondents in all three regions indicated a use ranging from 2 to 4%, with no statistical difference across regions. These findings are likely a conservative estimate, given that beekeepers would be unlikely to admit to unsanctioned AB use. This is further supported by the numerous studies that have exposed frequently detected residues in honey samples, indicating antibiotic use among EU member states (Al-Waili et al., 2012; Reybroek et al., 2012; Galarini et al., 2015; Reybroek, 2017). Al-Waili et al. (2012) reported that 1.7% out of 3855 tested honey samples were noncompliant with current EU standards based on the detection of seven different antibiotic residues. It is not always possible to discern the source of contamination, whether from explicit AB use, honey admixture or environmental pollutants, which makes the current findings so significant (Lodesani & Costa, 2005).

Misuse of ABs is facilitated if beekeepers know where to purchase them and how to avoid the

Table 1.

Identified shortcomings of antibiotic use and awareness

	EU	UK	US
Antibiotic use	Low persistent use of ABs. Highest reported use of ABs (5%). ABs purchased from other beekeepers.	Low but persistent use of ABs.	Low persistent use of ABs. ABs mainly used for *Nosema* (no AB registered for this). ABs purchased online. Most likely to not follow label instructions.
Antibiotic awareness	Little knowledge on AB function.	Little knowledge on AB function.	Little knowledge on AB function (e.g. indicated a preventative function). Most likely to see ABs fail to treat. Acquire AB information online.

Table 2.

Suggested corrective actions

	EU	UK	US
Antibiotic use	Minor antibiotic use and illegal treatments (official controls). Verify traceability of Abs. Verify preparedness and training of officers and veterinarians.	Continue to monitor antibiotic use and respect of protocol (official controls). Continue to verify preparedness and training of bee inspectors and veterinarians.	Monitor antibiotic use and illegal treatments (official controls). Verify traceability of Abs. Verify preparedness and training of officers and veterinarians.
Antibiotic awareness	Training of beekeepers concerning antimicrobials.	Training of beekeepers concerning antimicrobials.	Training of beekeepers concerning antimicrobials, especially ABs use following label instructions.

necessary legal route. AB acquisition was investigated and suggested some level of veterinary engagement, but beekeepers were primarily purchasing ABs from pharmacies (EU), the internet (US) or national bee inspectors (UK). The locations of purchase can often be indicative of illegal intent, especially when considering that 55% of US based websites selling antibiotics do not require a prescription according to Garcia et al. (2020). Undocumented trade and use of antimicrobial treatments could result in imprudent use of the product, leading to the development of superbugs, health issues within the hive and AB residues contaminating bee products (Bártíková et al., 2016).

Respondents in the US reported a significant use of ABs against EFB and less frequently against AFB. This finding is suggestive of general awareness regarding the long-lived spores of *P. larvae* which cannot be killed, but only temporarily controlled with antibiotics (Krongdang et al., 2017). Studies have in fact shown that while AB treatments can suppress symptoms of the disease, the spores remain in the environment surviving the treatment and eventually causing the hive to relapse (Krongdang et al., 2017).

The US, however, also indicated a higher use of ABs against *Nosema* for which no antibiotics are currently approved. In 2013 the FDA withdrew the application of approval for fumagillin (New Animal Drug Applications; Alfaprostol; Bicyclohexylammonium Fumagillin; N, 2013). Furthermore, a number of studies have reported a substantial increase in resistance and possible toxic effects of fumagillin suggested in both honeybees and humans (Rodriguez-Garcia et al., 2018; Burnham, 2019). A current lack of approved active ingredients for *Nosema* presents a significant demand for new medications that safely and effectively treat honeybee colonies against this pathogen (Burnham, 2019). Non-significant findings showed a general consensus regarding the function of antibiotics used for treating diseases as opposed to preventing them, suggesting appropriate use of ABs. Studies in fact show that prophylactic and metaphylactic use of antibiotics in production species have resulted in the development of resistant bacteria within the animals and their surrounding environments (Zhu et al., 2013; Larsson, 2014; Gonzalez Ronquillo & Angeles Hernandez, 2017). Similarly, all three geographic areas reported no statistical differences concerning the need for a prescription for antibiotics. However, the average response rate reached 60%, a lower rate compared to the small animal veterinary industry, where owners are more conscious of the need for a prescription when purchasing antibiotics. This usually occurs because pet owners are in direct contact with veterinarians that must provide prescriptions before dispensing medications (Redding & Cole, 2019). In beekeeping, veterinarians are often foregone either due to their lack of knowledge on honeybee pathologies or because veterinary consults may be deemed too expensive for beekeepers.

Antibiotic awareness

All three geographical areas reported having little to some knowledge of AB issues, with the UK self-reporting the lowest average knowledge. A general lack of knowledge was further supported by the results of a subsequent question investigating beekeeper understanding of AB function. The US indicated that ABs are mainly used to prevent diseases, a practice that is strongly discouraged by veterinary and health professionals worldwide, as prophylactic use often results in the development of resistance (Gonzalaz Ronquillo & Angeles Hernandez, 2017). The EU, instead, underreported AB use to target bacteria, indicating a lack of awareness regarding the correct function of antibiotics as medicines that target bacteria specifically. Misuse and overuse have been described as the major contributors to antibiotic resistance, suggesting the importance of beekeepers understanding AB function and correct use (Sifri et al., 2019). For this reason, it is necessary to ensure beekeepers have access to accurate information, ideally provided by reliable sources and trained experts.

The survey identified that only EU respondents sought veterinary advice on treatment, while the UK and US relied heavily on books and the

internet. These findings show a stark contrast to other animal-related fields, where owners are more likely to consult a veterinarian prior to treatment (De Briyne et al., 2013). A persistent lack of beekeeper-veterinary rapport not only establishes a knowledge gap within the industry but further results in beekeepers seeking advice elsewhere, as seen in these results, often from other beekeepers that may be similarly deficient in antibiotic related knowledge (Williams, 2000). The UK, however, reported the involvement of national bee inspectors, who are trained to diagnose honeybee diseases, and are the only entity allowed to issue and administer ABs in the UK (Wilkins et al., 2007). Inspectors are also tasked with inspecting hives annually alongside randomly testing hive products and matrices for antibiotic residues, ensuring regular compliance of all beekeepers. The current findings would suggest that while AB use is generally dictated by legislation and mostly compliant, knowledge regarding AB issues is somewhat lacking. Awareness campaigns have already been adopted in multiple countries in an attempt to educate beekeepers as to the risks of antibiotic resistance, mostly through the National Bee Unit in the UK and extension services in the US, but efforts are still underway to increase their impact (Huttner et al., 2019).

When asked to report how often ABs fail to treat honeybees, the US reportedly witnessed this the most. Such a response is supported by the multiple studies that have indicated widespread resistance to the ABs commonly used in apiaries (Alippi et al., 1999; Krongdang et al., 2017). This finding also indicates beekeepers may be using ABs imprudently or adopting practices that increase the chance of resistance developing. Furthermore, the response rate from all three areas (40% EU, 76% US, 42% UK) would suggest that imprudent use of ABs is occurring more frequently than declared in previous questions.

Beekeepers in the UK appeared consistently more aware and compliant throughout, further supported by an increased awareness of drug resistance infections, but respondents in the UK were also less aware of AB residues compared to those in the EU and US. This finding was perhaps less surprising since those in the UK also reported the lowest number of professional beekeepers (7%). A study by Jones et al. (2015), in fact, suggested that individuals that are economically affected by product contamination are generally more aware of residues, compared to hobbyists producing for themselves.

Most respondents agreed that contaminated products should not be consumed, with no statistical differences appearing between areas. This belief reflects the findings of a study assessing consumer perception regarding health and safety when consuming animal-derived products, suggesting that consumer demand for safer products highly influenced marketing and sale of such products (Kos Skubic et al., 2018). General consensus was similarly met regarding the belief that AB resistance would have a significant impact on various spheres of life. AB resistance has been shown to be an increasingly significant threat to medicine, veterinary medicine and the environment. Studies are reporting more frequent and severe cases of resistance in most medical fields, as well as high levels of residues in many bodies of water and soil samples (Martens & Demain, 2017; Jia et al., 2017).

Further considerations

Some limitations were considered alongside the need for further research. A more active distribution of the survey, with the involvement of additional beekeepers' associations and social platforms would have prompted a larger population sample, providing more insight and more robust tests. However, the sample was large enough to produce valid results. The survey included both hobbyists and professional beekeepers, combining different practices since professionals often require more stringent biosecurity and resort more easily to ABs to maintain hive health throughout the year (Reybroek, 2012). The current study set out to assess the impact that legislation has on antibiotic use and awareness. It was not possible to discern significant differences caused by legislation, but the study was able to ascertain that ABs are apparently used by a small percentage of

beekeepers in all three areas, regardless of the legislative context. Moreover, the analysis of the survey data identified a lack of in-depth AB awareness, suggesting that stronger campaigns are needed to increase widespread understanding of AB principles and related risks. The use of a globally distributed survey could continue to be employed as a risk-assessment tool, outlining shortcomings within the beekeeping community and allowing for a targeted approach in advancing sustainable beekeeping practices.

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Supplementary material

Fig. 1

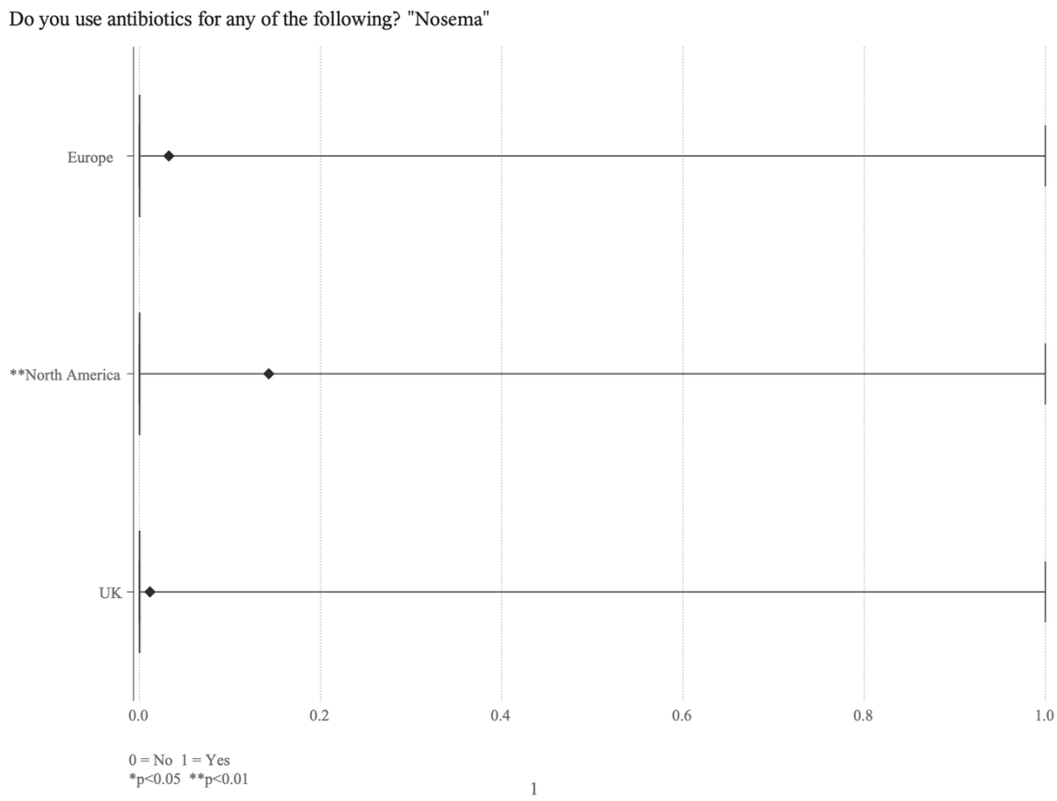


Fig. 2

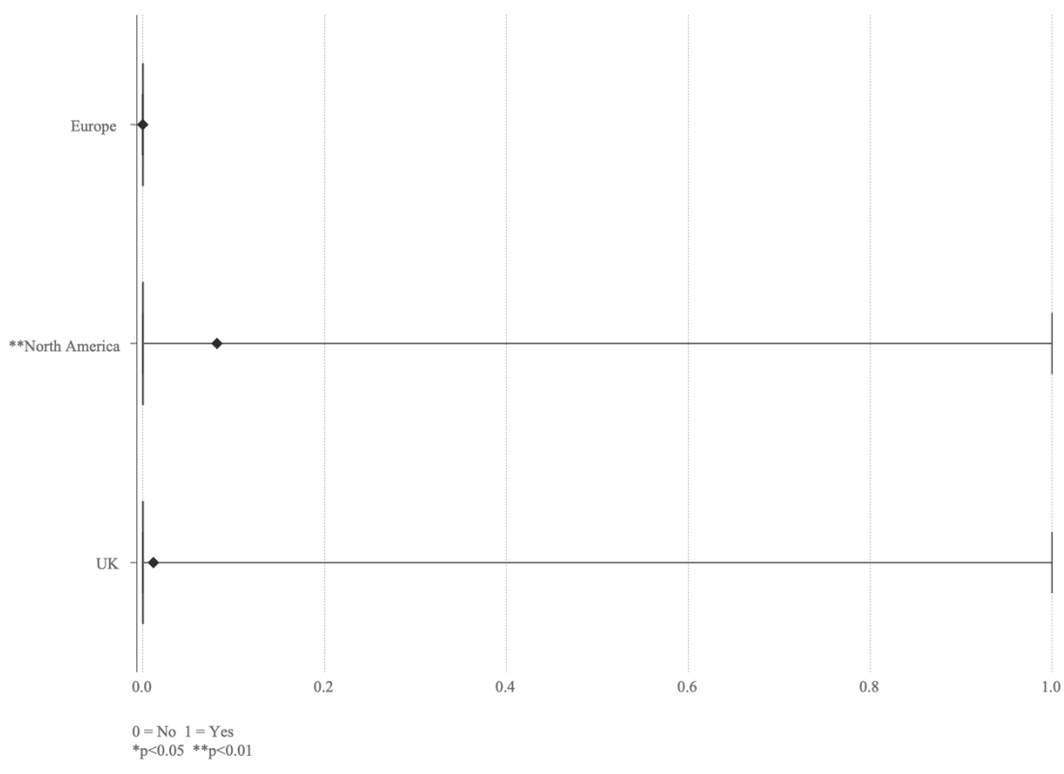
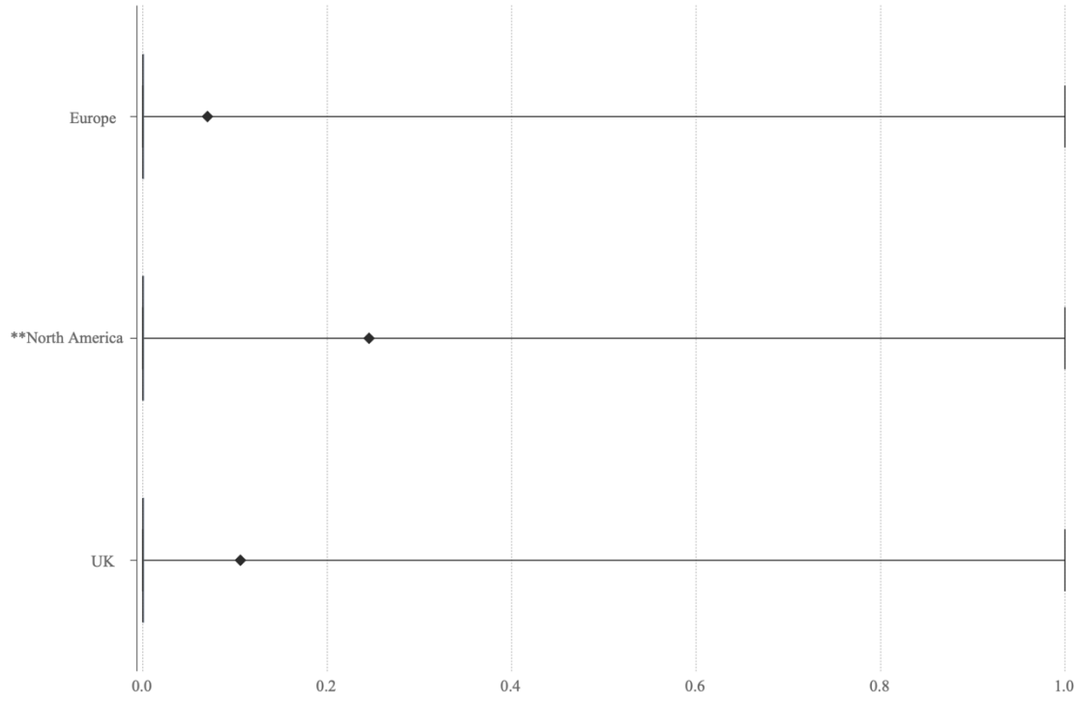


Fig. 3

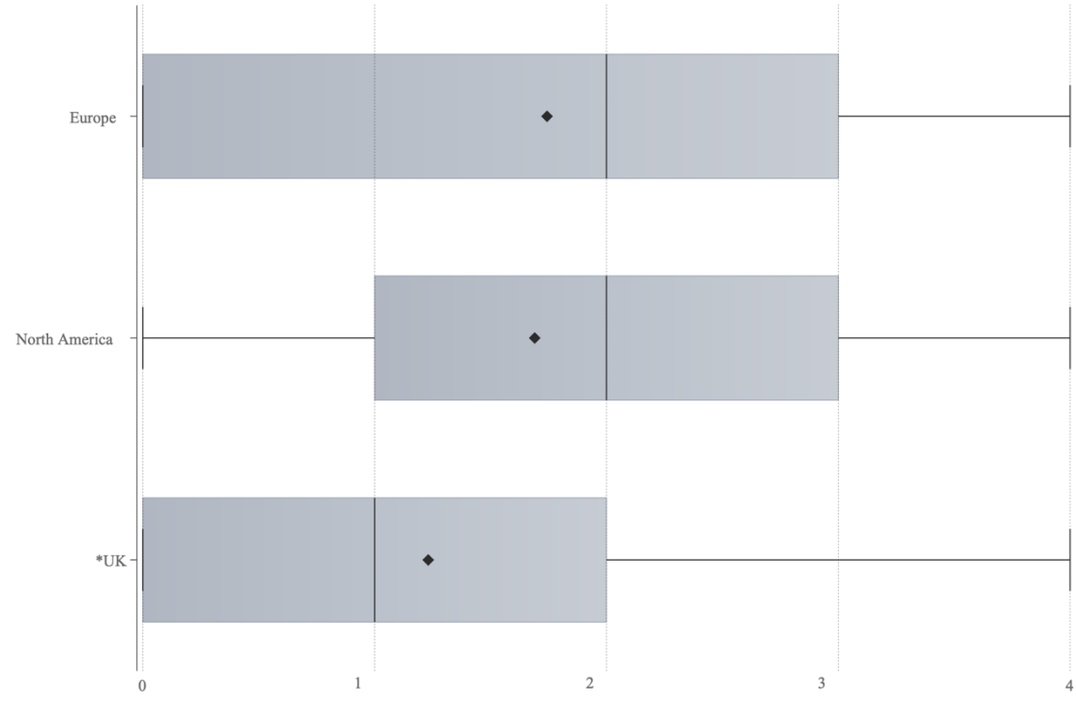
What are antibiotics intended to do? "Medicines that prevent diseases"



0 = No 1 = Yes
*p<0.05 **p<0.01

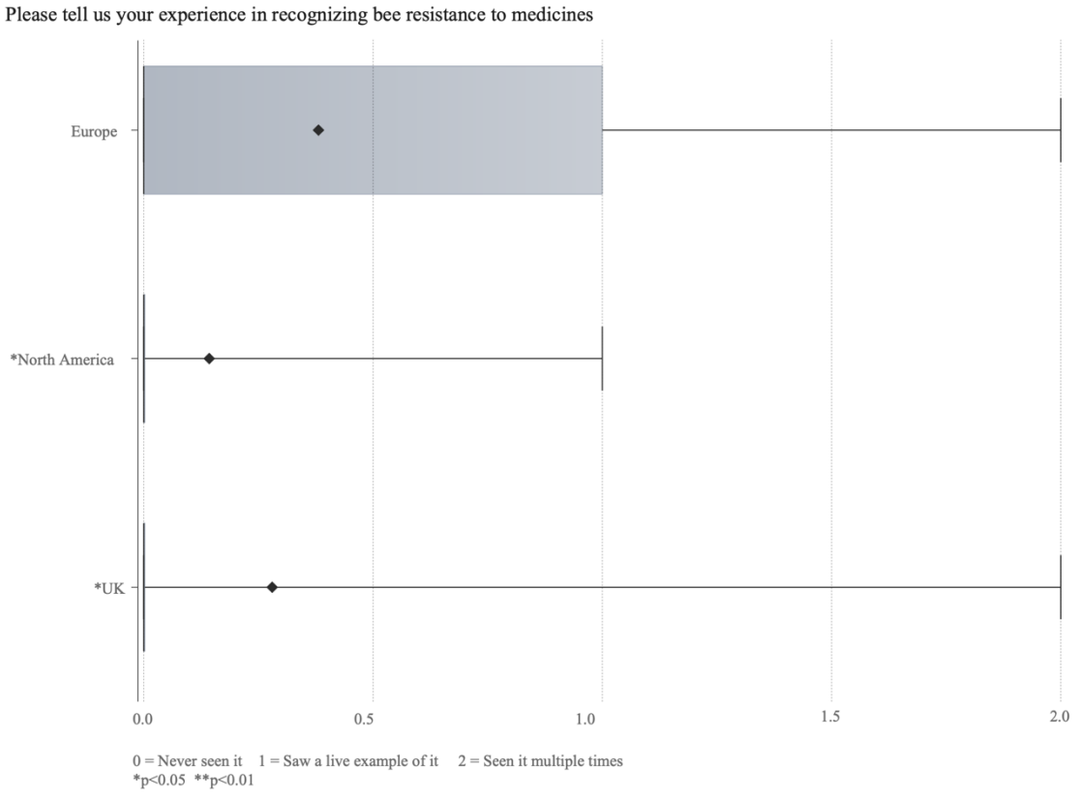
Fig. 4

How knowledgeable are you in issues of antibiotics intended to be used on bees?



0 = No knowledge 1 = Little knowledge 2 = Somewhat knowledgeable 3 = Moderately knowledgeable 4 = Extremely knowledgeable
*p<0.05 **p<0.01

Fig. 5



Croppi_Appendix