Use of antibiotics in animals and humans (commercial poultry and poultry farmers) in rural Pakistan- A One Health quantitative study

Student: Um e Habiba

Supervisor: Dr. Muhammad Asaduzzaman

Co- Supervisor: Elia Mmbaga

Supervisor at field place: Dr. Amjad Khan

Date of submission: 29 June 2022

Department of Community Medicine and Global Health

Institute of Health and Society

Faculty of Medicine

University of Oslo

ACKNOWLEDGEMENTS

I would like to express my sincere gratitude to all those people who helped me and supported me in the completion of my master's thesis.

My earnest thanks to my supervisors, Muhammad Asaduzzaman and Elia Mmbaga at the University of Oslo, and my local mentor, Amjad Khan (Quaid-i-Azam University Islamabad, Pakistan). This thesis would have been incomplete without their guidance and supervision. Thank you Asad and Elia for your regular meetings and feedback on time. I would specially thank Amjad who helped me a lot in getting ethical clearance and in the data collection phase in Pakistan.

I am thankful to the Department of Community Health and Global Health, University of Oslo for providing me with such an excellent platform to learn and extend my knowledge in a competitive environment. Thanks to our coordinators, Terese Eriksen and Birthe Neset for their help throughout this master's program.

Thanks to my supportive family especially my husband Muhammad Faizan for his support. I sincerely thank my father Muhammad Daud for his motivation and encouragement. I am thankful to my brothers Nouman, Abdul Mateen, Abdul Mannan and Abdul Jabbar for helping me in finding the poultry farms and data collection in a very challenging setting. I would like to thank my mother for her emotional and moral support in the data collection phase. Thanks to my friend Fatou for her guidance and useful tips.

Table of Content

ACKNOWLEDGEMENTS 2

List of Tables 5

List of Figures 5

List of Abbreviations 6

Abstract 7

- **Background** 7
- **Aim** 7
- **Method** 7
- **Results** 7
- **Conclusion** 7

Introduction 8

- **One Health Approach** 8
- **Antibiotic Use in Poultry** 9
- **Antibiotic Use in Humans** 11
- **Environment as a source of AMR** 12

Country Profile: Pakistan 12

Problem Statement/Research Gap 13

Rationale/Justification for the study 13

- **General objective** 14
- **Specific objectives** 14

Methodology 14

- **Study Design** 14
- **Study Setting and Participants** 14
- **Sample Size and Sampling** 14
- **Inclusion and Exclusion criteria** 15
- **Data collection Tools and Procedures** 15

Data management and analysis 15

Ethical Approval 16

- **Ethical Considerations** 16

Funding 16

Results 17
Demographic data of farmers and characteristics of poultry farms 17
Poultry wastes disposal 22
Health care seeking behaviour and antibiotic use in poultry farmers 22
Knowledge of poultry farmers about antibiotics 24
Discussion 26
Pattern of antibiotic use and health care seeking behaviour in poultry farms 26
Pattern of antibiotic use and health care seeking behaviour in poultry farmers 27
Limitations 28
Conclusion 29
References 30
APPENDICES 35

List of Tables

Table 1. Characteristics of Poultry farms and demographic data of farmers (N=40) 17
Table 2. Pattern of Source and Use of Antibiotics in Poultry farming (N=40) 19
Table 3. Distribution of the types of Antibiotics used in small-scale commercial poultry farms..... 20
Table 4. Distributions of antibiotics misuse by demographic characteristics of poultry farmers 21
Table 5.Disposal of poultry wastes..... 22
Table 6. Pattern of antibiotic use among poultry farmers (N=40)..... 23
Table 7.Knowledge of poultry farmers about antibiotics 25

List of Figures

Figure 1: One Health system and development of Antibiotic resistance..... **9**
Figure 2: Map of Pakistan **13**
Figure 3: Clinical conditions for using antibiotics in poultry..... **18**
Figure 4: Most used antibiotics (>5) in poultry Farms **20**
Figure 5.Clinical Conditions for using antibiotics in poultry farmers **23**

List of Abbreviations

ABR: Antibiotic resistance

AGP: Animal growth promoter

AMR: Antimicrobial resistance

AMU: Antimicrobial usage

API: Active pharmaceutical ingredient

ARG: Antibiotic resistance gene

CDC: Centre of disease control

FAO: Food and Agriculture Organization of the United Nations

HPI: Human Poverty Index

LMICs: Low and middle income countries

MRSA: Methicillin-resistant Staphylococcus aureus

OECD: Organization for Economic Co-operation and Development

OIE: World Organisation for Animal Health

TSD: Tjenester(service) for Sensitive Data

WHO: World Health Organization

Abstract

Background

Antibiotic resistance (ABR) is a multifaceted problem that threatens both human and animal health, which requires an integrated One Health (OH) approach to mitigate. Misuse of antibiotics in food animals is an important driver for antibiotic resistance and food safety in developing country context. Therefore, there is a need for evidence generation of antibiotic misuse in farming practices to contain this emerging issue.

Aim

This study aims to explore the pattern and indications of antibiotic use for commercial poultry farms and farmers and its potential for contributing to the development of ABR in rural Pakistan as well as the dissemination of ABR through poultry waste to surrounding environment.

Method

In this cross-sectional study, we included 40 poultry farms and interviewed 40 farmers from 10 villages (4 farmers/village) in Tehsil Pindi gheb, district Attock, Punjab, Pakistan. We conducted descriptive analysis and differences between antibiotic use in humans and poultry with various explanatory categorical variables were examined using the Chi-square test.

Results

Use of antibiotics for growth promotion, lack of proper biosecurity in animal farming, poor healthcare-seeking behavior and knowledge favoring ABR in both human and animal were the major findings in our study. Antibiotics were used in all poultry farms while 60% (n=24) obtained antibiotics without veterinary prescription. Colistin sulphate and Amoxicillin trihydrate were the most commonly used antibiotics (60%). Farmers having no or primary education used more antibiotics for growth promotion (60% & 80% respectively) and had less knowledge about antibiotic resistance, antibiotic use and prohibited antibiotics in poultry as compared to those having secondary or more education. However, the correlation between farmers' knowledge on antibiotic resistance and their professional farm training along with number of years in farming was statistically significant ($p<0.05$). 85% of farms had no wastewater drainage system. Regarding human use, 52.5% (n=21) of the participants obtained antibiotics without consulting a physician. The major reasons for using antibiotics were flu (42%) and chest infections (32%).

Conclusion

Our study findings are important for national ABR strategy and food safety in Pakistan and similar settings. Therefore, an integrated and sustainable OH approach, ABR and food safety policy needs to be adopted with inclusion of farmers' education, mass awareness, organic farming and strict antibiotic usage guidelines.

Keywords: One Health; Poultry; antibiotic resistance; environment; Poultry farmers; health care seeking behavior, Antibiotics.

Introduction

Antimicrobial resistance (AMR) is an increasing global health problem driven by an increase in antimicrobial use (AMU) both in humans and in animals [1]. It has been considered a serious threat to modern health care system which endangers the ability to prevent and treat many infectious diseases [2-4]. AMR has been gradually increasing during the last decades and it currently accounts for over 7 million death per year which is estimated to be around 10 million in the year 2050 with 90% of these deaths in low and middle income countries (LMICs) in Africa and Asia [5, 6]. Moreover, AMR is also associated with approximately 2 million infections and 23000 deaths in the United stated and 25000 deaths in European Union per year. [7, 8]. The world bank estimated the loss of 3.8% of annual gross domestic product (GDP) in the world due to drug-resistant infections [9].

Several factors have been identified for AMR development. For example: inappropriate prescribing, easy availability of antimicrobials over the counters without professional controls, use of less effective low potency drugs as a result of poor manufacture, availability of antibiotics on roadside stalls, and hawkers with little or no knowledge about drug dosing, dosing regimens, contraindications and indications, counterfeit drugs, self-medication, lack of awareness on antibiotics resistance and unaware of global threats for our societies due to AMR [10-14]. In LMICs, few additional factors such as lack of diagnostic laboratories, poor sanitation level, less potent activity of few antibacterial agents, improper food handling and poor infection control accelerate AMR spread within population [15, 16].

In recent decades, antimicrobial consumption has increased gradually in LMICs due to which drug-resistant infections are increasing [17]. In many developing countries, antibiotic use in both animals and humans is unregulated which results in uncontrolled and overuse of antibiotics [18]. In addition, many locally produced antibiotics are counterfeit or with sub-therapeutic concentration [19]. Therefore, to overcome the AMR issue particularly in LMICs, the current pattern of AMU and the contributing factors need to be determined.

One Health Approach

According to One Health High-Level Expert Panel (OHHLEP), '***One Health (OH) is an integrated, unifying approach that aims to sustainably balance and optimize the health of people, animals and ecosystems'*** [20]. OH system recognizes that human health is closely linked to animal health and the surrounding environment. Many people live in close contact with domestic animals (livestock & pets) due to which there are more chances to transmit diseases from animals to humans. Moreover, intensive farming practices and disruption in environmental conditions result in disease transfer to humans and animals. OH

approach is an effective way to fight health issues at the Human-Animal-Environment interface among which AMR and food safety are major themes [21]. Several major organizations like the World Health Organization (WHO), the United Nations Environment Programme (UNEP), the World Organisation for Animal Health (OIE), and Food and Agriculture Organization of the United Nations (FAO) are exponents of OH [20, 22].

The following figure depicts how AMR develops through OH process.

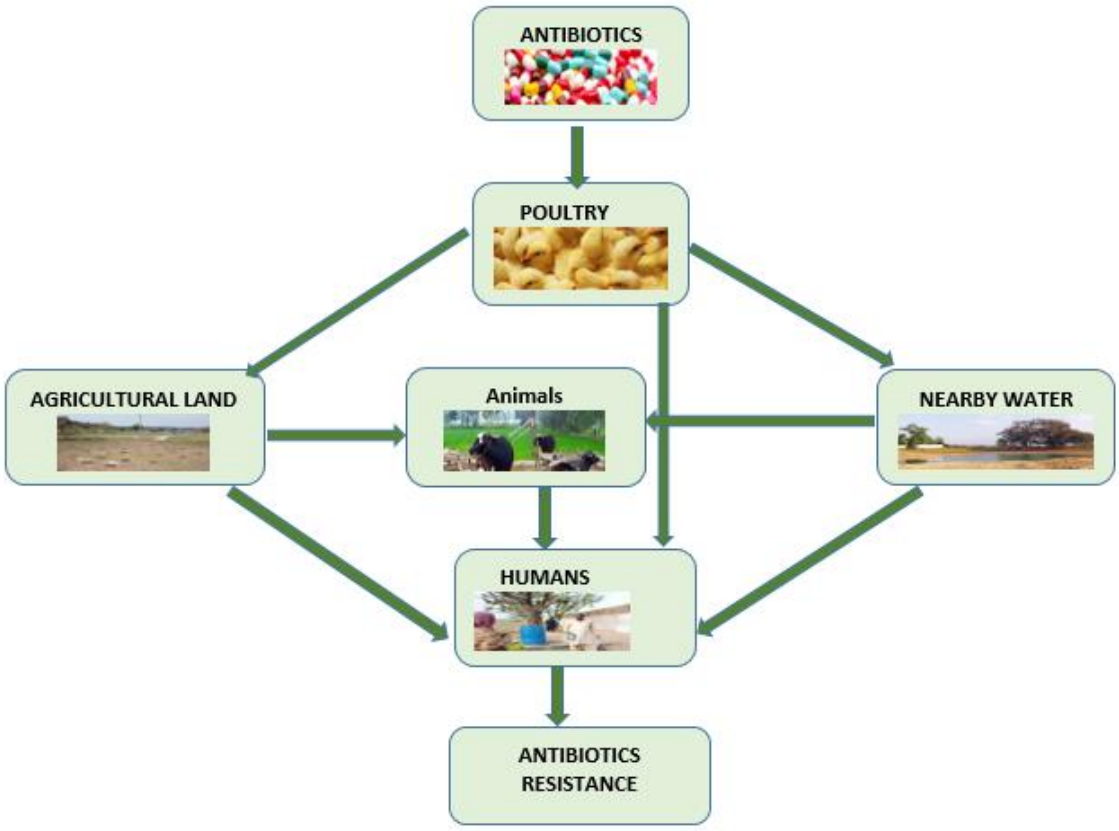

Figure 1: One Health system and development of Antibiotic resistance

Antibiotic Use in Poultry

Antibiotic use as animal growth promoters (AGPs) without veterinary prescription was started in Europe, particularly in West Germany in 1951 and gradually spread in the whole Europe, the USA and around the globe [23]. To tackle growing AMR, the WHO recommends against antibiotic use in healthy animals, adding to the growing recognition of misuse and overuse in food production [24]. While Europe initiated antibiotic use as AGPs, its rectification steps were also in place quite a bit ahead of others. Since January 2006, the EU banned antibiotics as animal growth promoters whereas the situation in Asia, Africa and Latin America is more frustrating [25].

Antibiotics used in the poultry industry are of three types such as prophylactic (to avoid disease), therapeutic (to prevent bacterial infections), and growth promoters (to increase growth rate). The duration of therapeutic antibiotics is less than the animal growth promoters (AGPs) though given at low doses. However, antibiotics administered beyond permissible limits without having any adherence to the withdrawal period, the antibiotic residues deposit continuously in the tissue of chicken and become hazardous to human health upon consumption. Antibiotic residue or resistant bacteria in chicken meat can cause very serious health problems e.g., allergic reactions, AMR, and imbalance of intestinal microbiota [26]. Resistant bacterial strains can tolerate the MIC (minimum inhibitory concentration) of antibiotics. If humans consume the food containing this strain, it will be transferred to human beings through improper processing. After entering the human body, they will transfer resistant genes to human microbiota, and it will decrease the effectiveness of antibiotics used by that individual. In this way, resistance will develop in human beings too as these microbes have resistant genes that can tolerate the action of antibiotics. As a result, this strain will be dominant in the population and can be transmitted through mutation and plasmid mediation [26].

The intensive use of antibiotics in food producing animals has increased over the last decades because of the increase in meat consumption [27]. This extensive use of antibiotics in animal production with long exposure periods provides favourable conditions to fix antibiotic resistance genes (ARGs) in food chain. These ARGs can be transmitted to human gut microbiota through contaminated food or animals. Environmental bacteria, particularly those in soil and water, are the most prevalent organisms that serve as a carrier for transferring ARGs to animals and human beings [28].

Poultry is the largest source of meat around the globe. In the past few decades, more than a 100% increase in poultry is observed globally, with a total chicken meat consumption of 13% in 1965 to 28% in 2015 [26]. As per the report presented by Food and Agriculture Organization (FAO) and Organization for Economic Co-operation and Development (OECD), the estimated poultry meat production in 2014 was 108.5 million tons, which will reach 134.5 million tons in 2023 [26]. This increasing demand puts pressure on farmers to produce in the minimum time e.g. 6 weeks instead of 9 or 10 weeks. This intensification leads to the transmission of drug-resistant pathogens and diseases [26] and misuse of antibiotics mainly due to improper rules and regulations regarding antibiotics use, lack of awareness, and low education level among the farmers [29]. Even, farmers who work in the poultry production facilities have high rates of antibiotic resistance because of occupational exposure [30].

Pakistan is among the top 10 countries that are producing animals through modern intense farming practices [31]. These farming practices rely on antibiotics on routine basis as growth promoters and disease prevention [18]. There is no estimation of annual AMU in food-producing animals in Pakistan and the choice of antibiotics is based on availability and cost, not on diagnosis. Therefore, AMR is one of the major health

and food safety challenges in Pakistan [32]. It is hard to estimate the exact antibiotic usage for treatment, prevention, or as growth promoters but practices indicate that antibiotics are mostly used for prevention and growth promotion on a routine basis in food-producing animals [18].

Usually, in small-scale poultry, workers who are involved in taking care of poultry give antibiotics without knowing the match between the antimicrobials and pathogenic organisms. As a result, many health problems are increasing because of emerging antimicrobial resistant pathogens [32].

Antibiotic Use in Humans

Antibiotics have been considered as 'magic bullet' to treat dreadful infectious diseases since its discovery and played a significant role in saving lives. Unfortunately, irrational use of antibiotics leads to the development of AMR, increase in mortality and morbidity, and later it became a major health challenge globally and a threat to human wellbeing [33]. It is estimated that by the year 2050 about 100 trillion USD of economic output and about 100 million lives a year are in danger because of increase in drug resistance infections if we don't find proactive solutions now to decrease drug resistance [34].

Major contributing factors in developing AMR in LMICs are antibiotic overuse in all health sectors, unnecessary prescription by the physicians, unsuitable selection of antibiotic, poor practices, non-existent policies for the institutional antibiotic, lack of regulations, etc. [32]. To overcome this, a positive attitude, optimal practices, adequate knowledge about antibiotics and antibiotic resistance issues can play an important role [35]. In high-income countries, interventions regarding antibiotic use and AMR in health facilities have been implemented but in LMICs, the burden of AMR is difficult to quantify. In LMICs especially in rural areas, microbiological cultures and sensitivity tests may not be performed due to lack of equipment, workers, and above all financial resources which result in misuse of available antibiotics in those settings and ultimately emergence and spread of AMR [36].

Pakistan is ranked third highest county among low-income countries where antibiotic consumption is very high [37]. It is common practice there to seek treatment from a local medical store or using antibiotic by getting advice from relatives or through previous experience. Therefore antibiotics consumption and resistance are increasing [38]. Several studies reported a high percentage of antibiotic prescriptions e.g., 51.5%,52% and 52.4% [39-41]. It is also evident that 30 to 50% of patients in hospitals receive at least one antimicrobial agent [42]. As per one report, antibiotics consumption in Pakistan increased from 0.8 to 1.3 billion Daily defined dose (DDD) (65%) between 2000 to 2015 [37]. Approx. more than 600,000 quacks are active and 50,000 unnecessary products are registered in Pakistan and the number is increasing day by day due to no proper check and balance [18].

Environment as a source of AMR

Antibiotics used in livestock transfers to the environment through different ways e.g., from animals to agricultural lands, grazing of animals and using organic fertilizer (animals waste) [43]. Several studies have shown a high percentage of transfer of antibiotics used in livestock to the environment through direct application of animal manure whereas the concentration of antibiotics varies from antibiotic to antibiotic. This results in the accumulation of residual antibiotics in the aquatic ecosystem and into soil changes soil microbial environment [43-45].

In many LMICs, poultry wastes are considered as best fertilizer for agricultural land [46]. Antibiotics present in poultry wastes are mostly bioactive and result in increased ABR in exposed bacteria in the environment [46]. This will increase the chances of transfer of antibiotic resistance genes (ARGs) from poultry to agricultural land and then to humans [47]. Moreover, a study revealed that poultry wastes are used to feed aquaculture (fishes and shellfishes) which leads to high resistance in Enterococcus spp. in fish intestines [48].

A study conducted in rural areas in India revealed that the chances of resistant bacteria and gene transmission from poultry to human beings is more because of shared living and sleeping areas and because of no proper waste disposal from poultry farms. Moreover, biosecurity measures are almost absent in small-scale farming [49]. Another study in Bangladesh reported that poultry wastes after slaughtering and feces are disposed into municipal drains or nearby open land [50]. These wastes containing ABR with ARGs can affect the immune system and metabolism and develop ABR in humans. Treating these resistant bacteria with antibiotics is difficult and it increases the mortality rate [47]. To stop this emerging health issue, effective measures like monitoring and controlling antibiotic use in poultry can play a significant role.

Country Profile: Pakistan

Pakistan (officially Islamic Republic of Pakistan) is in the South Asia region. Pakistan is bordered by India, Iran, Afghanistan, and China. It is the 5th most populated country in the world having a population of more than 228 million and the 33rd largest country. [51] The population density in Pakistan is 287 per Km2 (742 people per mi^2). Majority of the population (64.9%) live in rural areas, mostly engaged in agriculture and farming [52].

Economically Pakistan is a LMIC (low middle-income country) and according to Human Poverty Index (HPI), it ranks 65th among 102 developing countries in the world [53]. Only 27% of the population gets full health care benefits which include government employees and armed forces members while the remaining 73% depends on out-of-pocket payments&[53]. The health care system in Pakistan is facing challenges like

lack of resources, inequality, untrained human resources, gender insensitivity, and structural mismanagement [53].

Figure 2: Map of Pakistan[54]

Problem Statement/Research Gap

Antibiotic use in animals and humans has increased in the past few decades in Pakistan. This leads to the emergence of resistant microorganisms in animals, humans, and the surrounding environment. Excessive use of antibiotics in food producing animals especially for growth promotion and as prophylaxis transmits ABR genes in the surrounding environment and humans along with the development of ABR in the population. Various studies have been done in Pakistan about antibiotic use in humans and poultry, but not many studies have been conducted in the context of OH approach. There is a need to implement a collaborative approach such as OH to cope with the increasing ABR issue in developing countries like Pakistan.

Rationale/Justification for the study

ABR is a growing risk for global health concerns and is regarded as a OH issue. The spread of antibiotic resistance among the domains of the OH system (human-animals-environment) is a quickly worsening health problem worldwide with a very high rate in LMICs. Improving antibiotics stewardship interventions and following policies to restrict antibiotics use locally and internationally is of utmost importance. Studies

have been conducted in Pakistan on antibiotic use in poultry and human beings but to our knowledge, no concurrent research has been carried targeting both the ABR in poultry and poultry farmers. This study aimed to explore how health care seeking behaviour and antibiotic use contribute towards development and spread of ABR at the human-animal-environment interface. This will help to improve health care seeking behaviour and interventions to reduce spread of ABR between OH domains.

Objectives

General objective

- To explore the pattern of use of antibiotics for commercial poultry farms and farmers and its potential for contribution to the development of AMR in rural Pakistan.

Specific objectives

- To understand the pattern of health care seeking behaviour and pathway of getting antibiotics for both poultry and poultry farmers.
- To assess the purpose of using antibiotics in poultry and in human
- To describe the poultry wastes disposal system in rural Pakistan

Methodology

Study Design

A cross-sectional study design was selected to estimate the use of antibiotics in poultry farms and by poultry farmers. This study design was the best suitable way to estimate the pattern of antibiotic use in poultry farms in this selected population of farmers.

Study Setting and Participants

Pakistan has 4 provinces (Punjab, KPK (Khyber Pakhtunkhwa), Sindh and Balochistan), and capital territory (Islamabad). Each province has further subdivisions e.g., Division, district, tehsil and union councils, etc. We conducted our cross-sectional study in the Attock district. It is in the north of the Punjab province. It is one of the densely populated districts in Punjab and has about 1.89 million population [55].

This district has further 6 Tehsils, of which we selected Tehsil Pindi gheb which has a population of 271,931 as per the survey conducted in 2017 and of which 83.4% are in the rural area where most people earn through agriculture and farming. This tehsil has a total of 134 villages [56]. For our study, we selected randomly 10 villages and 40 farms (4 from each village).

Sample Size and Sampling

We did not find any similar study in Pakistan and this is a baseline surveillance study to describe the patterns of antibiotic use in humans and poultry in rural areas. We sampled all eligible farmers in the selected study

area the same as per the study in north western China [57]. We sought to capture the level of variability in antibiotic use by selecting 4 farms each village in 10 villages (n=40 farms), which will reduce the likelihood of sampling bias.

Inclusion and Exclusion criteria

Inclusion:

- Healthy adult farmers (>18 years) responsible for poultry rearing.
- Agreed to provide informed consent.

Exclusion:

- Persons who are not related to poultry farming, not engaging in day-to-day care of the poultry farm

Data collection Tools and Procedures

We collected data between January to March 2021 from poultry rearing farmers through a structured questionnaire having 5 sections (including demographics). Before data collection, we explained the aims and objectives of our study to our participants and got their consents on consent forms.

Data was collected by using a validated questionnaire from a study conducted recently in North-western China regarding the use of antibiotics in poultry [57].For the use of antibiotics by farmers themselves a validated questionnaire from a study conducted in the Northwest region of Pakistan regarding the self-medication and antibiotic use by the public was used [58]. The questionnaire was translated into Urdu (the national language of Pakistan) to make it easy for farmers to understand and all the communication with farmers was in Urdu and Punjabi (local languages).

To maximize the validity of the questionnaire, a pilot study (pretesting of the questionnaire) was done on 4 poultry farms randomly from another village other than the selected for data collection.

We also took the suggestions and information from the local livestock officer, veterinary doctor, and medical doctor regarding illness and the use of antibiotics both in humans and in poultry.

After data collection, we entered all the data from the paper questionnaires into TSD (software for collecting sensitive data information) provided by UiO.

Data management and analysis

All the responses from participants were entered in Nettskjema (Online software for data collection provided by UiO) after data collection. Later we connected this to TSD as our data had some sensitive information. Lastly, we exported data from TSD to UiO remote desktop to analyze our data.

We analyzed the data using SPSS software and performed different statistical analysis. We conducted descriptive statistics where categorical variables were summarized using proportions and continuous variables were summarized by calculating means and standard deviations. Differences between categorical variables were examined using the Chi-square test while differences in means were examined using the student t-test.

Ethical Approval

Being an international student in Norway, before the start of my study, ethical approval was obtained from Norwegian Centre for Research Data (NSD) in Norway with project reference no. 726029. In addition to this approval, ethical approval was also obtained from the Bioethics Committee of faculty of biological sciences, Quaid-i-Azam University Islamabad, Pakistan Under the reference no. #BEC-FBS-QAU2021-250 to get access to participants in the selected area for data collection.

Ethical Considerations

All the participants of my study were healthy adults (>18 years). While interviewing participants, I have ensured that the rights of my participants are protected. I ensured them that there are no detrimental effects or risks of taking part in the study and that their privacy and confidentiality will be maintained. All of them participated voluntarily in my study without any force.

The purpose of the study was explained in detail to all participants and an opportunity was given to ask questions or seek clarification. Moreover, information about the study was provided in the form of a participant information letter, which was translated into Urdu. Some participants read the information letter by themselves while in some cases, I read and explained it to them in the local language. I also informed participants about their right to withdraw from the study at any stage.

A consent form was then signed by each participant to ensure anonymity. All collected data was stored in TSD (UIO service for research data) and only I had access to it. All questionnaires were allocated a unique study identification number. Moreover, I have used codes to make the data more anonymous so that it may not reveal the identity of any participants.

Funding

Our project was funded by the Institute of Health and Society, Faculty of Medicine, University of Oslo. A masters student stipend was also provided to cover the travel and fieldwork expenses.

Results

Demographic data of farmers and characteristics of poultry farms

We enrolled 40 poultry rearing farmers in the study. All were male and nearly two-thirds of the farmers (62.5%) had less than 15 years of farming experience. Of the 40 participants, 20 (50.0%) participants reported to have completed secondary education (10 years of education), 12.5% had completed primary education level (5 or less years of education) and 12.5% completed above secondary education level (12 or more years of education). A quarter of the participants (n= 10; 25.0%) had never been to school. With regards to professional farm training, 87.5% of the farmers had no professional training or education relevant to farming while only 12.5% had professional farming training. Moreover, based on the number of chickens we divided farms into three categories i.e. small, medium and large. About 15.0% of the farms were small-scale (up to 2000 chickens), nearly half (47.5%) of the farms were medium size (2001 to 4000 chickens), and 37.5 % were large-sized poultry farms (above 4000 chicken). There was only one worker in all small-sized poultry farms except one while out of 19 medium-sized poultry farms, 7 had 2 or more workers while the rest had one worker. A majority (13 out of 15) of large-sized poultry farms had 2 or more workers.

The number of years in the poultry farming profession ranged from 2 months to 35 years but almost two-thirds of the poultry farmers (62.5%) had experience in poultry farming of less than 15 years (**Table 1**).

Table 1. Characteristics of Poultry farms and demographic data of farmers (N=40)

Characteristics	Total
	n (%)
No. of year/s in farming	
<15	25(62.5)
15-30	13(32.5)
>30	2(5.0)
Education level of farmers	
Not educated	10(25.0)
Primary	5(12.5)
Secondary	20(50.0)
Above Secondary	5(12.5)
Professional farm training	
No	35 (87.5)
Yes	5(12.5)
Size of Poultry farm*	
Small	15 (37.5)
Medium	19(47.5)
Large	6(15.0)

*Size of the poultry farm is based on the number of chickens in the poultry farm.

Health care seeking behaviour for poultry and antibiotic use in poultry

Health services seeking behaviours, source, and use of antibiotics for poultry farming are presented in Table 2. All participants reported that they use antibiotics in every flock. Out of 40 farmers, 45.0% farmers were using antibiotics as a growth promoter in poultry while 55.0% were using antibiotics for various clinical conditions e.g., Flu, infectious coryza (respiratory infections), fever, Coli disease, Infectious bursitis (Gumboro), typhoid, pneumonia, etc. Figure 3 clearly shows the clinical conditions for which antibiotics were used in poultry.

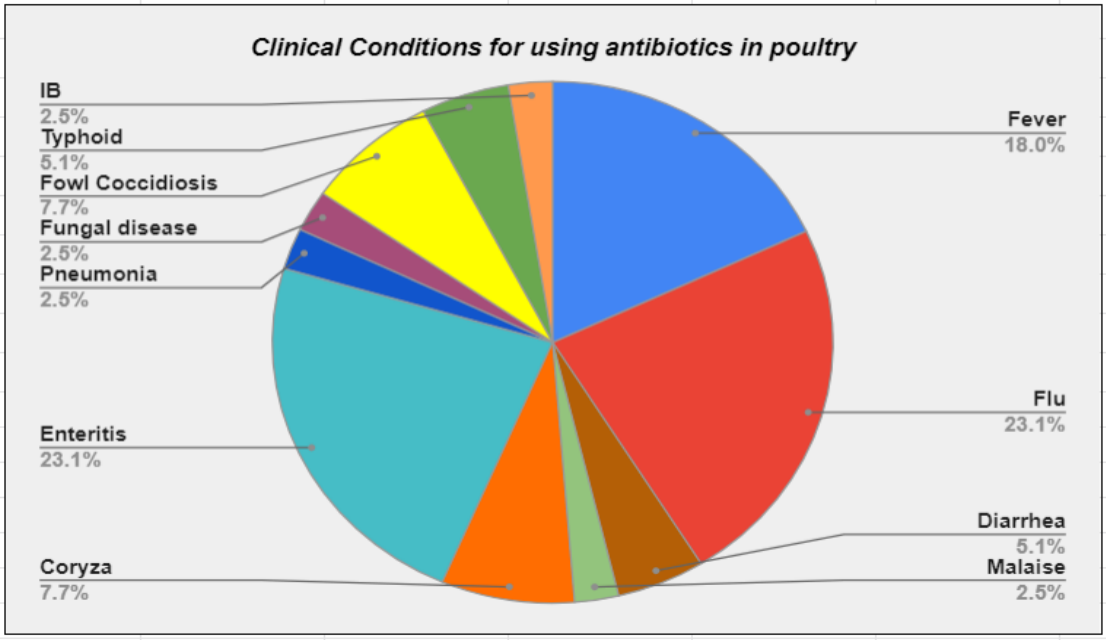

Figure 3: Clinical conditions for using antibiotics in poultry

Participants were asked about the source of veterinary services and antibiotics for poultry to investigate the health care seeking behaviour for poultry. A total of 16 out of 40 farmers (40.0%) reported that they used prescribed antibiotics (obtained antibiotics after veterinary doctor prescription) while 60.0% were using non-prescribed antibiotics. A large majority (82.5%) of the respondents reported that they obtained antibiotics from the agents while only 7 respondents (17.5%) were from the local pharmacy/drug store. Agents are those who supply feed and medicines to the poultry farms and act as a third party between the poultry farmers and feed/veterinary drug companies. Moreover, 45.0% of respondents reported that they have received veterinary services from feed companies.

We also found that about three-quarters of the participants (72.5%) used antibiotics frequently. Half of the poultry farmers (52.5%) did not follow withdrawal periods of the antibiotics used and even, those (55%)

using antibiotics for various clinical conditions, some might be viral, and antibiotics had no role such as Flu, fungal infections, or malaise **(Figure 3).**

Table 2. Pattern of Source and Use of Antibiotics in Poultry farming (N=40)

Characteristics	Total N (%)
Antibiotic/s use in poultry	
No	0(0)
Yes	40(100)
Veterinary doctor Prescription for getting antibiotic/s	
No	24(60.0)
Yes	16(40.0)
Source of veterinary services	
Local livestock officer	1(2.5)
Private veterinary doctor	14(35.0)
By Yourself	2(5.0)
Feed company	18(45.0)
Government source	5(12.5)
Source of getting antibiotic/s	
Agents	33(82.5)
Local pharmacy/ drug shop	7(17.5)
Use of antibiotic/s for clinical conditions	
No	18(45.0)
Yes	22(55.0)
Use of antibiotic/s as Growth promotion	
No	22(55.0)
Yes	18(45.0)
Frequency of antibiotic/s use	
Occasionally*	11(27.5)
Regularly**	29(72.5)
No. of days of antibiotic/s administration	
1-3 days	20(50.0)
4-7 days	12(30.0)
>7days	8(20,0)

*Occasionally: Have not used antibiotics in every flock. **Regularly: Used antibiotics in every flock

Table 3 outlines the pattern of antibiotic use including class and types. Twelve classes of antibiotics containing eighteen different types were used in poultry farming by participants in this study. These antibiotics were used both separately and in combination. Out of these antibiotics, both colistin and a combination of colistin sulphate and Amoxicillin trihydrate were most frequently (n=24; 60.0%) used. Besides these, Enrofloxacin, Tylosin and Doxycyclin were commonly used antibiotics (35.0%, 25.0% and 22.5% respectively) **(Figure 4).** It was observed that mostly farmers were using multiple antibiotics in combination, and some were using the same antibiotic more than once but under different brand names.

Apart from antibiotics other antimicrobials e.g., antivirals (Amantadine HCl) and antifungal (Nystatin) were used by 25.0% and 2.5% of poultry farmers for the treatment of viral and fungal diseases.

Table 3. Distribution of the types of Antibiotics used in small-scale commercial poultry farms

Antibiotic class	Antibiotic	No of farms using antibiotics (N=40) n (%)
Aminopenicillins	Amoxicillin trihydrate	3(7.5)
Tetracyclines	Chlortetracycline	1(2.5)
	Oxytetracycline	1(2.5)
	Doxycycline	9(22.5)
Polymyxins	Colistin	24(60.0)
Macrolides	Tylosin	10(25.0)
	Erythromycin	2(5.0)
Fluoroquinolones	Ciprofloxacin	1(2.5)
	Enrofloxacin	14(35.0)
Penicillin	Penicillin	2(5.0)
Polypeptides	Bacitracin	4(10.0)
Trimethoprim	Trimethoprim	1(2.5)
Sulfonamides	Sulfamethoxypyridazine	1(2.5)
	Sulfamethazine	1(2.5)
Aminoglycosides	Neomycin	4(10.0)
	Streptomycin	2(5.0)
	Gentamycin	2(5.0)
Nitrofurans Derivatives	Furaltadone	1(2.5)
Aminopenicillins/ Polymyxins	Amoxicillin trihydrate + colistin sulphate	24(60.0)

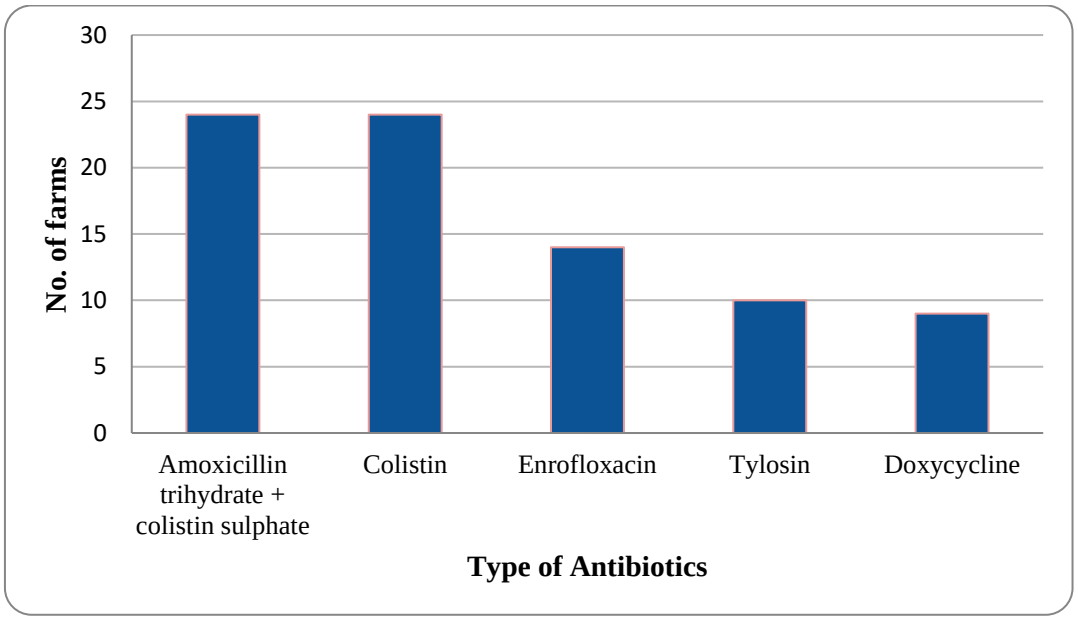

Figure 4: Most used antibiotics (>5) in poultry Farms

Table 4 illustrates the distributions of antibiotics misuse by demographic characteristics of poultry farmers. It can be seen from **Table 4** that the ratio of antibiotic misuse is more in farmers having education less than higher secondary (except 20.0% farmers having higher secondary or above education) when it comes to obtaining antibiotics without prescription, using antibiotics as growth promoters and withdrawal period follow up. In case of getting antibiotics from agents, there is no correlation between farmer's education level and professional farm training as mentioned in **Table 3**.

On the other hand, farmers having professional farm training (12.5%) have knowledge about antibiotic misuse. Results shows that only 1 out of 5 farmers were not following the withdrawal period and getting antibiotics without prescription while the ratio is comparatively high (20 out of 35 in both cases) among farmers having no professional farm training (87.5%). Professionally trained farmers were not using antibiotics as a growth promoter.

Table 4. Distributions of antibiotics misuse by demographic characteristics of poultry farmers

Characteristics	**Total N**	**Antibiotics Misuse**			
		Antibiotic/s given without Veterinary doctor's prescription n (%)	**Obtained antibiotic/s from agents n (%)**	**Antibiotic/s used as growth promotors n (%)**	**Didn't follow withdrawal period n (%)**
Education level of farmers					
Not educated	10	5(50.0)	9(90.0)	6(60.0)	7(70.0)
Primary	5	4(80.0)	5(100)	4(80.0)	3(60.0)
Secondary	20	11(55.0)	15(75.0)	7(35.0)	10(50.0)
Higher Secondary or >	5	1(20.0)	4(80.0)	1(20.0)	1(20.0)
Professional Farm training					
No	35	20(57.1)	30(85.7)	18(51.4)	20(57.1)
Yes	5	1(20.0)	3(60.0)	0(0)	1(20.0)

Poultry wastes disposal

Most of the poultry farmers (85.0%) reported that they do not have a wastewater drainage system in the farms, but they have a non-concrete and open drainage system to drain water to open sites near the farms or into the agricultural land. Only six farms (15.0%) had proper drainage systems for poultry farms. Additionally, 24 (60.0%) farmers reported that they use poultry wastes as fertilizer while 16(40.0%) reported disposing poultry wastes in an open area surrounding poultry farms and open areas outside the villages where the land is not in use. **(Table 5)**

Table 5.Disposal of poultry wastes

Characteristics	Total N(%)
Type of drainage in farm	
Concrete and covered	6(15.0)
Non-Concrete and open	34(85.0)
Poultry wastes disposing method	
Open area	16(40.0)
Use as fertilizer	24(60.0)

Health care seeking behaviour and antibiotic use in poultry farmers

Of the 40 participants, more than one-third (37.5%) used antibiotics within the last month preceding the survey, 5.0% in the last 1-3 months while 25.0% of the participants used antibiotics more than 6 months ago before the survey. A third (32.5%) of the participants did not remember when they took the antibiotic last time. Moreover, about half (n= 21; 52.5%) of respondents reported self-medicating with antibiotic without a physician's prescription while (n= 19; 47.5% obtained antibiotic after physician's prescription. Almost half of the participants (n=19; 47.5%) obtained used previously used antibiotics without consulting a physician, while 2.5% used antibiotic after getting advice from relatives. Some participants had no access to physicians, so they used antibiotic without a physician's prescription.

When participants were asked about the source of antibiotic, 75.0% reported obtaining them from local pharmacies, 20.0% from leftover antibiotics at home, and 5.0% obtained from rural medical practitioners (unqualified doctors). Moreover, 70.0% of the respondents used antibiotic for 1-3 days, 20.0% used for 4-7 days, and 10.0% used for more than 7 days. **(Table 6)**

Table 6. Pattern of antibiotic use among poultry farmers (N=40)

Characteristics	Total n (%)
Purpose of antibiotic/s use	
Flu (common cold)	17(42.5)
Stomach problems	4(10.0)
Chest infections	13(32.5)
Fever	3(7.5)
Others*	3(7.5)
Physician prescription	
No	21(52.5)
Yes	19(47.5)
Reason behind self-medication	
None	19(47.5)
Not access to physician care	1(2.5)
Previous experience	19(47.5)
Advice from relatives	1(2.5)
Source of getting antibiotic/s	
Pharmacy	30(75.0)
Leftover household antibiotics	8(20.0)
Rural practitioner (Untrained doctor)	2(5.0)
Duration of antibiotic/s use	
1-3 days	28(70.0)
4-7 days	8(20.0)
>7 days	4(10.0)

*Others include skin infections and one farmer had Inguinal hernia.

A large proportion (42.5%) of the participants used antibiotic to treat flu/common cold and about a third (30%) used them for chest infections treatment **(Figure 5).**

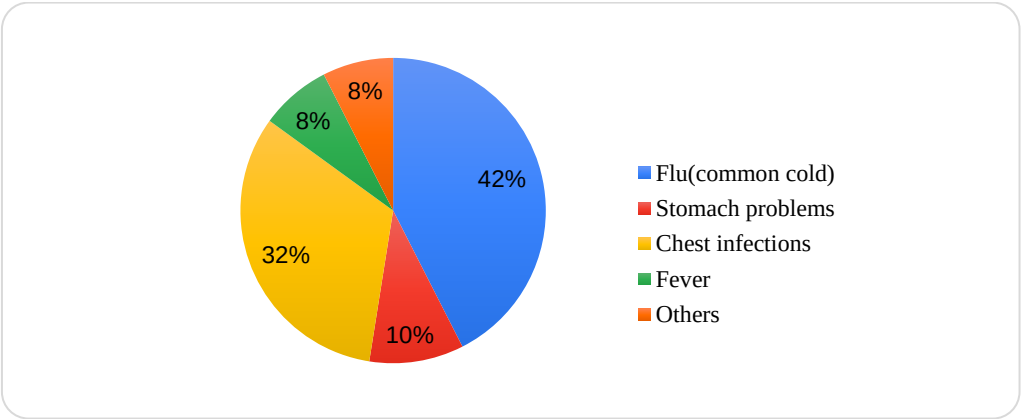

Figure 5.Clinical Conditions for using antibiotics in poultry farmers

Knowledge of poultry farmers about antibiotics

Majority (90.0%) of respondents who were not educated had no knowledge about antibiotic use, prohibited antibiotics in poultry and no farmers in this category had knowledge about antibiotic resistance. Farmers having primary level education had no knowledge about antibiotic use, resistance and prohibited antibiotics. Of the 20 farmers who had secondary level education, 25.0 % had knowledge about antibiotic use, 15.0% about prohibited antibiotics and 10.0% had about antibiotic use. A large majority (80.0%) of respondents having higher secondary level education or more had knowledge about antibiotic use while over half (60.0%) had knowledge about prohibited antibiotics and 40.0% had knowledge about antibiotic resistance. There is a significant association between the education level of farmers and knowledge about antibiotic use ($p=0.012$) and knowledge about prohibited antibiotics ($p= 0.051$). **(Table 7)**

Table 7.Knowledge of poultry farmers about antibiotics

Variables	Total	Knowledge about *AB use			Knowledge about prohibited AB			Knowledge about AB resistance		
	N (%)	No N (%)	Yes N (%)	p-value	No N (%)	Yes N (%)	p-value	No N (%)	Yes N (%)	p-value
Education level of farmers				0.012			0.051			0.083
Not educated	10(25.0)	9(90.0)	1(10.0)		9(90.0)	1(10.0)		10(100.0)	0(0)	
Primary	5(12.5)	5(100.0)	0(0)		5(100.0)	0(0)		5(100.0)	0(0)	
Secondary	20(50.0)	15(75.0)	5(25.0)		17(85.0)	3(15.0)		18(90.0)	2(10.0)	
Higher Secondary or >	5(12.5)	1(20.0)	4(80.0)		2(40,0)	3(60.0)		3(60.0)	2(40.0)	
Professional Farm Training				0.002			<0.001			0.017
No	35 (87.5)	29(82.9)	6(17.1)		32(91.4)	3(8.6)		33(94.3)	2(5.7)	
Yes	5(12.5)	1(20.0)	4(80.0)		1(20.0)	4(80.0)		3(60.0)	2(40.0)	
No. of year/s in farming				0.026			0.004			<0.001
1-15	25(62.5)	21(84.0)	4(16.0)		23(92.0)	2(8.0)		24(96.0)	1(4.0)	
15-30	13(32.5)	9(69.2)	4(30.8)		10(76.9)	3(23.1)		12(92.3)	1(7.7)	
>30	2(5)	0(0)	2(100.0)		0(0)	2(100.0)		0(0)	2(100.0)	

Discussion

Pattern of antibiotic use and health care seeking behaviour in poultry farms

In our study, we have evaluated the practice of antibiotic use and health care seeking behaviour of poultry farmers in rural areas of Punjab, Pakistan in OH approach, which means the practice for humans and for poultry. This is the first one health study done in Pakistan to evaluate the emergence of antibiotic resistance.

Our study findings confirm that use of antibiotics in poultry is not well regulated in Pakistan, and it has the potential to contribute to the emergence of antibiotics resistance pathogens and the development of antibiotic resistance. More than half of the participants used antibiotics as growth promoters without any consultation of trained veterinarians. This observation of the unregulated use of antibiotics in food producing animals in Pakistan is similar to what has been reported in other studies elsewhere [18, 59]. In contrast to this, in European Union countries, antimicrobial use for growth promotion was banned since 2006 and antimicrobials use is prescription-only [60]. In addition, we observed a significant seasonal variation in antibiotic use for poultry as prophylaxis. The poultry farmers reported that they use more antibiotics in winters than during summertime as chickens are more prone to diseases in cold weather. The results of this study also revealed the use of improper dosage, wrong combination of antibiotics, misuse, and overuse of antibiotics as also reported in other studies [61].

The majority of the participants in this study purchased antibiotics based on previous experience and from local agents which is a clear indication of the patron-client relationship and undue influence for unnecessary usage. Such resistance provoking drug purchase behaviour and practice are also evident in similar LMIC settings [18, 29, 62, 63].

Our findings show that nineteen different antibiotics were used by poultry farmers belonging to eleven different classes of antibiotics. Antibiotics were used separately and in combination where colistin sulphate and amoxicillin trihydrate combination were the most used. Twenty-four participants used this combination under different brand names e.g., Colimoxin, Almoxin-C, Colimoxin forte, Amoxi-Hi, Colistamoxyl, Neo-AC. Although colistin is considered as the last defence antibiotic and used for the treatment of multi-drug resistance (MDR) infections. Despite of the reserved nature of this antibiotic it was excessively being used by farmers to treat enteric disease and for growth promotion. Overuse and misuse of colistin lead to the development of antibiotic resistance as reported in previous studies [64-66]. It was also observed that the farmers were using the same antibiotic but under different brand names and they were unaware of this. Some used antibiotics as a supplement on daily basis. Few participants reported that they have used antibiotics on alternative days without following the duration of treatment and withdrawal time. One-fourth of the participants used antiviral (Amantadine HCl) while one farmer delineated that he used antifungal (Nystatin).

Farmers were also using antibiotics for antiviral diseases e.g., flu. Lack of education, lack of professional farm training, and not getting advice from the veterinary doctor were the common reasons behind antibiotics misuse and these findings are in lined with other studies [18, 29, 57].

Our study found that nearly half of the participants were not following the recommended withdrawal period and they were unaware of the term ‘‘Withdrawal period’’. This practice increases the possibility of high level of antibiotic residues in poultry meat with detrimental health consequences. In our studies, most used antibiotics e.g., Colistin sulphate, amoxicillin trihydrate, tylosin, and doxycycline have withdrawal period 7 days, while 4 days for enrofloxacin [63, 67]. Farmers sold poultry within the withdrawal period. These findings of violations of the withdrawal period for antibiotics have also been in other studies [62, 63, 68]. Not adhering to the withdrawal period is a public health concern as antibiotic residues in food producing animals may also lead to MDR pathogens in humans [63].

Thirty-four farms had no wastewater drainage system, and they drained wastewater into nearby agricultural land or open sites near farms. These practices increase the chances of antibiotics contamination to agricultural land as reported in various studies which have shown that antibiotics have been detected in raw and treated wastewater [43]. Moreover, poultry wastes were used as fertilizer by more than half of the participants as poultry wastes contain nutrients and are excellent fertilizers for crop growth [69]. It was observed that farmers sell poultry wastes to agricultural landowners as well. Previous studies have revealed that increased use in poultry leads to more resistant bacteria in human surroundings as a major portion of antibiotics excrete through manure and urine [70-72]. If poultry wastes are disposed into agricultural land, this changes the soil’s microbial environment and becomes a potential risk for human health [47, 73, 74].

Pattern of antibiotic use and health care seeking behaviour in poultry farmers

Our study found that one-third of the participants used antibiotics in the last month indicating the high use of antibiotics among farmers in the study area. One of the important findings was the inability of the participants to distinguish between viral and bacterial infections. Nearly half of them used antibiotics for flu (common cold) and a few used for fever which is in line with other studies done in Punjab, Pakistan [75, 76]. Similar findings have been reported in a Lebanese study where nearly three-quarters of the participants used antibiotics for all conditions, whether viral or bacterial [77].

Antibiotics are easily accessible in Pakistan and can be obtained from pharmacies without a doctor’s prescription. One-third of the participants obtained antibiotics from pharmacies. This leads to an increase in self-medication practices [78]. People have no idea about the risk of self-medication. For instance, our data shows that more than half of the farmers used antibiotics without a physician’s prescription similar to what has been reported in other studies [37, 79, 80]. Almost half of the participants reported using the

antibiotics from previous experience and leftover antibiotics at home. The main reason behind this was the financial constraints and traveling to the cities for physician consultation. This observation has also been reported in studies conducted in India, Sudan, and Nigeria [81-83]. On the other hand, the ratio of self-medication with antibiotic is comparatively less in Italy and other European countries [84, 85].

Several studies have reported that patient understanding about illness and treatment will increase the adherence to the medication [86, 87]. In our findings, the dosage regimen of antibiotics was not proper and almost three-quarters of the participants used antibiotics for 1-3 days. Participants were of the opinion that they could stop taking medicine after they feel better. Improper consumption of antibiotics results in antibiotic resistance [88]. Incomplete information about antibiotic use and getting antibiotics for a few doses because of high antibiotics prices or leftover antibiotics at home is associated with it [37]. Even from pharmacies or from rural practitioners one can get antibiotics for one-day treatment. Studies have shown that non adherence to antibiotics regimens can be improved by increasing general population knowledge and proper counselling at pharmacies and by improving pharmacist-patient interactions [88].

A significant number of participants had no knowledge about Antibiotic resistance (ABR), and antibiotics use. Knowledge of the farmers about antibiotics was associated directly with the education level of the farmers. Participants who were not educated or primary level education had no or least knowledge about antibiotic use, ABR and prohibited antibiotics in poultry as compared to participants who had secondary or more education, and these findings are in consistent with other studies [37, 58, 84]. In contrast to this, studies done in Germany, Norway and Scotland reported that a significant number of participants had knowledge about ABR [89-91].

Lack of knowledge about antibiotics results in misuse of antibiotics [92]. Therefore, interventions based on educational level can be effective to raise awareness, enhance knowledge about antibiotic use and change their health care seeking behavior. A good example is E-bug by public health England. This is an international health education source to aware and educates people about antibiotics, ABR, and infections [93, 94].

Limitations

The study included a limited number of poultry farms and farmers which cannot be generalized to the whole of Pakistan. However, these results provide a descriptive picture of the situation of antibiotic use patterns in rural Pakistan. During the conduct of this study, several participants refused to participate in the study, and this may have introduced selection bias. Moreover, the findings of this study may be affected by recall bias as participants did not remember properly the antibiotics used. Due to less medicine-related

knowledge, there is a possibility that some participants may have used antibiotics but could not be able to understand if they were antibiotics.

Conclusion

We are aware that many studies have investigated the antibiotic use in human and food producing animals along with the pattern of health care seeking behaviour, but none has investigated this issue in One health context for rural Pakistan so far. We found that there is a signification association between the education of the farmers and health care seeking behaviour. Educated and professionally trained farmers contribute less towards ABR development. Moreover, we observed that the easy accessibility of antibiotics is a growing concern for ABR development in LMICs particularly for Pakistan. Implementing strict rules to obtain antibiotics without physicians' prescription, initiating health education programs to educate mass people and farmers about their health and pattern of health care seeking can play a significant role in controlling antibiotic resistance. There should be a proper crackdown against quacks and unauthorized health practitioners. These strategies can bring positive outcomes to system.

References

1. Nayiga, S., et al., *Use of antibiotics to treat humans and animals in Uganda: a cross-sectional survey of households and farmers in rural, urban and peri-urban settings.* JAC-Antimicrobial Resistance, 2020. **2**(4): p. dlaa082.
2. O'neill, J., *Antimicrobial resistance.* Tackling a crisis for the health and wealth of nations, 2014.
3. Hay, S.I., et al., *Measuring and mapping the global burden of antimicrobial resistance.* BMC medicine, 2018. **16**(1): p. 1-3.
4. Ferri, M., et al., *Antimicrobial resistance: A global emerging threat to public health systems.* Critical reviews in food science and nutrition, 2017. **57**(13): p. 2857-2876.
5. Hofer, U., *The cost of antimicrobial resistance.* Nature Reviews Microbiology, 2019. **17**(1): p. 3-3.
6. de Kraker, M.E., A.J. Stewardson, and S. Harbarth, *Will 10 million people die a year due to antimicrobial resistance by 2050?* PLoS medicine, 2016. **13**(11): p. e1002184.
7. Uchil, R.R., et al., *Strategies to combat antimicrobial resistance.* Journal of clinical and diagnostic research: JCDR, 2014. **8**(7): p. ME01.
8. Hampton, T., *Novel programs and discoveries aim to combat antibiotic resistance.* Jama, 2015. **313**(24): p. 2411-2413.
9. Adeyi, O.O., et al., *Drug-resistant infections: a threat to our economic future.* World Bank Group, Washington, DC, 2017.
10. SMITH, R.D., J. Coast, and M.R. MILLAR, *Over-the-counter antimicrobials: the hidden costs of resistance.* Journal of Antimicrobial Chemotherapy, 1996. **37**(5): p. 1031-1032.
11. Taylor, R., O. Shakoor, and R. Behrens, *Drug quality, a contributor to drug resistance?* Lancet (London, England), 1995. **346**(8967): p. 122.
12. McGregor, A., *Counterfeit drugs flood developing world.* The Lancet, 1997. **350**(9092): p. 1690.
13. Dua, V., C.M. Kunin, and L.V. White, *The use of antimicrobial drugs in Nagpur, India. A window on medical care in a developing country.* Social science & medicine, 1994. **38**(5): p. 717-724.
14. Coenen, S., et al., *Are patient views about antibiotics related to clinician perceptions, management and outcome? A multi-country study in outpatients with acute cough.* PloS one, 2013. **8**(10): p. e76691.
15. Laxminarayan, R. and D.L. Heymann, *Challenges of drug resistance in the developing world.* Bmj, 2012. **344**.
16. Fletcher, S., *Understanding the contribution of environmental factors in the spread of antimicrobial resistance.* Environmental health and preventive medicine, 2015. **20**(4): p. 243-252.
17. Van Boeckel, T.P., et al., *Global trends in antimicrobial resistance in animals in low-and middle-income countries.* Science, 2019. **365**(6459): p. eaaw1944.
18. Ur Rahman, S. and M. Mohsin, *The under reported issue of antibiotic-resistance in food-producing animals in Pakistan.* Pak. Vet. J, 2019. **1**: p. 1-16.
19. Mamun, K.Z., P. Shears, and C. Hart, *The prevalence and genetics of resistance to commonly used antimicrobial agents in faecal Enterobacteriaceae from children in Bangladesh.* Epidemiology & Infection, 1993. **110**(3): p. 447-458.
20. *Joint tripartite and UNEP statement on definition of "One Health"*. 2021 [cited 2022; Available from: https://www.unep.org/news-and-stories/statements/joint-tripartite-and-unep-statement-definition-one-health#:~:text=One%20Health%20is%20an%20integrated,closely%20linked%20and%20inter%2Ddependent.
21. *One Health Basics*. 2022 February 7 [cited 2022 16.03.2022]; Available from: https://www.cdc.gov/onehealth/basics/index.html.

22. Lerner, H., *The philosophical roots of the" One Medicine" movement: an analysis of some relevant ideas by Rudolf Virchow and Calvin Schwabe with their modern implications.* Studia Philosophica Estonica, 2013. **6**(2): p. 97-109.
23. Kirchhelle, C., *Pharming animals: a global history of antibiotics in food production (1935–2017).* Palgrave Communications, 2018. **4**(1): p. 1-13.
24. *Stop using antibiotics in healthy animals to prevent the spread of antibiotic resistance*. 2017 [cited 2022; Available from: https://www.who.int/news/item/07-11-2017-stop-using-antibiotics-in-healthy-animals-to-prevent-the-spread-of-antibiotic-resistance.
25. Brussels. *Ban on antibiotics as growth promoters in animal feed enters into effect*. 2005 [cited 20222; Available from: https://ec.europa.eu/commission/presscorner/detail/en/IP_05_1687.
26. Muaz, K., et al., *Antibiotic residues in chicken meat: global prevalence, threats, and decontamination strategies: a review.* Journal of food protection, 2018. **81**(4): p. 619-627.
27. Collignon, P. and A. Voss, *China, what antibiotics and what volumes are used in food production animals?* 2015, BioMed Central. p. 1-4.
28. Chang, Q., et al., *Antibiotics in agriculture and the risk to human health: how worried should we be?* Evolutionary applications, 2015. **8**(3): p. 240-247.
29. Glasgow, L., et al., *Antibiotic use in poultry production in Grenada.* Veterinary medicine international, 2019. **2019**.
30. Braykov, N.P., et al., *Antibiotic resistance in animal and environmental samples associated with small-scale poultry farming in northwestern Ecuador.* Msphere, 2016. **1**(1): p. e00021-15.
31. Page, S. and P. Gautier, *Use of antimicrobial agents in livestock.* Revue Scientifique et Technique-OIE, 2012. **31**(1): p. 145.
32. Choisy, M., et al., *Assessing antimicrobial misuse in small-scale chicken farms in Vietnam from an observational study.* BMC veterinary research, 2019. **15**(1): p. 1-10.
33. Zaffiri, L., J. Gardner, and L.H. Toledo-Pereyra, *History of antibiotics. From salvarsan to cephalosporins.* Journal of Investigative Surgery, 2012. **25**(2): p. 67-77.
34. O'Neill, J., *Tackling drug-resistant infections globally: final report and recommendations.* 2016.
35. Napolitano, F., et al., *The knowledge, attitudes, and practices of community pharmacists in their approach to antibiotic use: A nationwide survey in Italy.* Antibiotics, 2019. **8**(4): p. 177.
36. Sartelli, M., et al., *Antimicrobials: a global alliance for optimizing their rational use in intra-abdominal infections (AGORA).* World journal of emergency surgery, 2016. **11**(1): p. 1-32.
37. Atif, M., et al., *What drives inappropriate use of antibiotics? A mixed methods study from Bahawalpur, Pakistan.* Infection and drug resistance, 2019. **12**: p. 687.
38. Loyola Filho, A.I.d., M.F. Lima-Costa, and E. Uchôa, *Bambuí Project: a qualitative approach to self-medication.* Cadernos de saude publica, 2004. **20**(6): p. 1661-1669.
39. Atif, M., et al., *Assessment of WHO/INRUD core drug use indicators in two tertiary care hospitals of Bahawalpur, Punjab, Pakistan.* Journal of pharmaceutical policy and practice, 2016. **9**(1): p. 1-8.
40. Hafeez, A., et al., *Prescription and dispensing practices in public sector health facilities in Pakistan-survey report.* Journal-Pakistan Medical Association, 2004. **54**(4): p. 187-191.
41. Atif, M., et al., *Assessment of core drug use indicators using WHO/INRUD methodology at primary healthcare centers in Bahawalpur, Pakistan.* BMC health services research, 2016. **16**(1): p. 1-9.
42. Vlahovic-Palcevski, V., M. Morovic, and G. Palcevski, *Antibiotic utilization at the university hospital after introducing an antibiotic policy.* European journal of clinical pharmacology, 2000. **56**(1): p. 97-101.
43. Tasho, R.P. and J.Y. Cho, *Veterinary antibiotics in animal waste, its distribution in soil and uptake by plants: A review.* Science of the Total Environment, 2016. **563**: p. 366-376.

44. Kumar, K., et al., *Antibiotic uptake by plants from soil fertilized with animal manure.* Journal of environmental quality, 2005. **34**(6): p. 2082-2085.
45. Elmund, G.K., et al., *Role of excreted chlortetracycline in modifying the decomposition process in feedlot waste.* Bulletin of Environmental Contamination and Toxicology, 1971. **6**(2): p. 129-132.
46. Barnes, K.K., et al., *A national reconnaissance of pharmaceuticals and other organic wastewater contaminants in the United States—I) Groundwater.* Science of the total environment, 2008. **402**(2-3): p. 192-200.
47. Muhammad, J., et al., *Antibiotics in poultry manure and their associated health issues: a systematic review.* Journal of Soils and Sediments, 2020. **20**(1): p. 486-497.
48. Nhung, N.T., et al., *Antimicrobial usage and antimicrobial resistance in animal production in Southeast Asia: a review.* Antibiotics, 2016. **5**(4): p. 37.
49. Stålsby Lundborg, C., et al., *Protocol: a 'One health'two year follow-up, mixed methods study on antibiotic resistance, focusing children under 5 and their environment in rural India.* BMC public health, 2015. **15**(1): p. 1-11.
50. Rousham, E.K., L. Unicomb, and M.A. Islam, *Human, animal and environmental contributors to antibiotic resistance in low-resource settings: integrating behavioural, epidemiological and One Health approaches.* Proceedings of the Royal Society B: Biological Sciences, 2018. **285**(1876): p. 20180332.
51. *Pakistan*. 2022 [cited 2022; Available from: https://www.nationsonline.org/oneworld/pakistan.htm.
52. *Pakistan Population* 2022 [cited 2022; Available from: https://www.worldometers.info/world-population/pakistan-population/.
53. *Health Systems Profile- Pakistan*. 2007 [cited 2022; Available from: https://digicollections.net/medicinedocs/documents/s17305e/s17305e.pdf.
54. *Provinces And Territories Map Of Pakistan*. 2021 [cited 2022; Available from: https://www.worldatlas.com/maps/pakistan.
55. *Population Profile Punjab*. Available from: https://pwd.punjab.gov.pk/population_profile.
56. *Pindi Gheb*. 2017; Available from: https://www.citypopulation.de/en/pakistan/distr/admin/attock/70104__pindi_gheb/.
57. Xu, J., et al., *Antibiotic use in chicken farms in northwestern China.* Antimicrobial Resistance & Infection Control, 2020. **9**(1): p. 1-9.
58. Nazir, S. and M. Azim, *Assessment of antibiotic self-medication practice among public in the northwestern region of Pakistan.* European Journal of Hospital Pharmacy, 2017. **24**(4): p. 200-203.
59. Nisar, M., et al., *Prevalence and antimicrobial resistance patterns of Campylobacter spp. isolated from retail meat in Lahore, Pakistan.* Food control, 2017. **80**: p. 327-332.
60. Agency, E.M., *Sales of veterinary antimicrobial agents in 31 European countries in 2018.* 2020.
61. Annan-Prah, A., et al., *Antibiotic use, abuse and their public health implication: the contributory role of management flaws in the poultry industry in two agro-ecological zones in Ghana.* 2012.
62. Al-Mustapha, A.I., V.O. Adetunji, and A. Heikinheimo, *Risk perceptions of antibiotic usage and resistance: a cross-sectional survey of poultry farmers in Kwara State, Nigeria.* Antibiotics, 2020. **9**(7): p. 378.
63. Guetiya Wadoum, R., et al., *Abusive use of antibiotics in poultry farming in Cameroon and the public health implications.* British poultry science, 2016. **57**(4): p. 483-493.
64. Agency, E.M., *Use of colistin products in animals within the European Union: development of resistance and possible impact on human and animal health*. 2013, European Medicines Agency London, UK.

65. Organization, W.H., *Global Antimicrobial Resistance Surveillance System (GLASS): the detection and reporting of colistin resistance*. 2018, World Health Organization.
66. Wongsuvan, G., et al., *Antibiotic use in poultry: a survey of eight farms in Thailand.* Bulletin of the World Health Organization, 2018. **96**(2): p. 94.
67. Khattab, W.O., et al., *Transmission of administered amoxicillin drug residues from laying chicken to their commercial eggs.* J Egypt Public Health Assoc, 2010. **85**(5-6): p. 297-316.
68. Turkson, P., *Use of drugs and antibiotics in poultry production in Ghana.* Ghana Journal of Agricultural Science, 2008. **41**(1).
69. Sims, J. and D. Wolf, *Poultry waste management: Agricultural and environmental issues*, in *Advances in agronomy*. 1994, Elsevier. p. 1-83.
70. Heuer, H. and K. Smalla, *Manure and sulfadiazine synergistically increased bacterial antibiotic resistance in soil over at least two months.* Environmental microbiology, 2007. **9**(3): p. 657-666.
71. Kelley, T.R., et al., *Antibiotic resistance of bacterial litter isolates.* Poultry Science, 1998. **77**(2): p. 243-247.
72. Yeom, J.-R., S.-U. Yoon, and C.-G. Kim, *Quantification of residual antibiotics in cow manure being spread over agricultural land and assessment of their behavioral effects on antibiotic resistant bacteria.* Chemosphere, 2017. **182**: p. 771-780.
73. Zhao, L., Y.H. Dong, and H. Wang, *Residues of veterinary antibiotics in manures from feedlot livestock in eight provinces of China.* Science of the Total Environment, 2010. **408**(5): p. 1069-1075.
74. Li, Y., et al., *A review on removing pharmaceutical contaminants from wastewater by constructed wetlands: design, performance and mechanism.* Science of the Total Environment, 2014. **468**: p. 908-932.
75. Arshad, M.S., M. Ijaz, and A. Hussain, *Evaluation of antibiotic use behavior in cold and flu amongst the students of Bahauddin Zakariya University Multan Pakistan.* Pak J Pharm, 2010. **23**(2): p. 15-22.
76. Anwar, M., et al., *Self-medication, home remedies, and spiritual healing: common responses to everyday symptoms in Pakistan.* Health Psychology and Behavioral Medicine, 2015. **3**(1): p. 281-295.
77. Mouhieddine, T.H., et al., *Assessing the Lebanese population for their knowledge, attitudes and practices of antibiotic usage.* Journal of infection and public health, 2015. **8**(1): p. 20-31.
78. Saleem, Z., et al., *Sale of WHO AWaRe groups antibiotics without a prescription in Pakistan: a simulated client study.* Journal of pharmaceutical policy and practice, 2020. **13**(1): p. 1-8.
79. Shet, A., S. Sundaresan, and B.C. Forsberg, *Pharmacy-based dispensing of antimicrobial agents without prescription in India: appropriateness and cost burden in the private sector.* Antimicrobial resistance and infection control, 2015. **4**(1): p. 1-7.
80. Zapata-Cachafeiro, M., et al., *Determinants of antibiotic dispensing without a medical prescription: a cross-sectional study in the north of Spain.* Journal of Antimicrobial Chemotherapy, 2014. **69**(11): p. 3156-3160.
81. Banerjee, I. and T. Bhadury, *Self-medication practice among undergraduate medical students in a tertiary care medical college, West Bengal.* Journal of postgraduate medicine, 2012. **58**(2): p. 127.
82. Omolase, C., et al., *Self medication amongst general outpatients in a Nigerian community hospital.* Annals of Ibadan postgraduate medicine, 2007. **5**(2): p. 64-67.
83. Salim, A.M. and B. Elgizoli, *Exploring the reasons why pharmacists dispense antibiotics without prescriptions in Khartoum state, Sudan.* International Journal of Pharmacy Practice, 2017. **25**(1): p. 59-65.

84. Napolitano, F., et al., *Public knowledge, attitudes, and experience regarding the use of antibiotics in Italy.* PloS one, 2013. **8**(12): p. e84177.
85. Grigoryan, L., et al., *Determinants of self-medication with antibiotics in Europe: the impact of beliefs, country wealth and the healthcare system.* Journal of Antimicrobial Chemotherapy, 2008. **61**(5): p. 1172-1179.
86. Kincey, J., P. Bradshaw, and P. Ley, *Patients' satisfaction and reported acceptance of advice in general practice.* The Journal of the Royal College of General Practitioners, 1975. **25**(157): p. 558-566.
87. Sharpe, T.R. and R.L. Mikeal, *Patient compliance with antibiotic regimens.* American Journal of Health-System Pharmacy, 1974. **31**(5): p. 479-484.
88. West, L.M. and M. Cordina, *Educational intervention to enhance adherence to short-term use of antibiotics.* Research in Social and Administrative Pharmacy, 2019. **15**(2): p. 193-201.
89. Waaseth, M., et al., *Knowledge of antibiotics and antibiotic resistance among Norwegian pharmacy customers–a cross-sectional study.* BMC Public Health, 2019. **19**(1): p. 1-12.
90. Easton, P.M., et al., *A survey on public knowledge and perceptions of methicillin-resistant Staphylococcus aureus.* Journal of Antimicrobial Chemotherapy, 2009. **63**(1): p. 209-214.
91. Raupach-Rosin, H., et al., *Knowledge on antibiotic use, self-reported adherence to antibiotic intake, and knowledge on multi-drug resistant pathogens–results of a population-based survey in Lower Saxony, Germany.* Frontiers in microbiology, 2019. **10**: p. 776.
92. Gualano, M.R., et al., *General population's knowledge and attitudes about antibiotics: a systematic review and meta-analysis.* Pharmacoepidemiology and drug safety, 2015. **24**(1): p. 2-10.
93. Eley, C.V., et al., *Young people's knowledge of antibiotics and vaccinations and increasing this knowledge through gaming: mixed-methods study using e-Bug.* JMIR Serious Games, 2019. **7**(1): p. e10915.
94. Tsuzuki, S., et al., *Factors associated with sufficient knowledge of antibiotics and antimicrobial resistance in the Japanese general population.* Scientific reports, 2020. **10**(1): p. 1-9.

APPENDICES

Are you interested in taking part in the research project?

"Use of antibiotics in animal and human (commercial poultry and poultry farmers) in rural Pakistan- A One Health quantitative study"

This is an inquiry about participation in a research project where the main purpose is to evaluate the use of antibiotics in commercial poultry farms as well as by farmers and its contribution towards the development of AMR (antimicrobial resistance). In this letter, I will give you information about the purpose of the project and what your participation will involve.

Purpose of the project

This project is my master thesis and it is a quantitative study involving the voluntary participation of the participants. Data will be collected by asking various questions involving some personal information e.g. name, address and mobile number which will keep confidential and only handled by the investigator. But if you do not want to give any personal information, you will not be forced.

The main objectives of the projects are to get information about the purpose of using antibiotics in poultry and in humans, the pathway of getting antibiotics, the pattern of health care seeking behaviour and process through which poultry wastes are being disposed off.

The purpose is to evaluate how all these processes contributing towards development of AMR in human beings.

Who is responsible for the research project?

Institute of health and society, Faculty of Medicine, University of Oslo Norway is responsible for this project.

Why are you being asked to participate?

I selected poultry farms from only District Attock, Province Punjab, Pakistan. As I am born and raised in District Attock, Pakistan. So, I selected my district as there as it is easy for me to communicate with the people in the local language and to get their trust in this study For this I selected 10 villages randomly from one tehsil i.e. Pindi gheb, and will collect data from 40 randomly selected poultry rearing farmers. So I selected your farm on a randomly basis. And all the information that I will collect from you will be confidential

What does participation involve for you?

This is a questionnaire-based study. If you chose to take part in this project, you will have to give the answers to questions on the questionnaire. It will take around 20-25 minutes. This questionnaire includes questions about

1. Farming practices and use of antibiotics in poultry.
2. Antibiotics that you used for yourself when u got sick in past few months.
3. How you dispose poultry wastes and what source of water for the poultry.

Participation is voluntary

Participation in the project is voluntary. If you chose to participate, you can withdraw your consent at any time without giving a reason. All information about you will then be made anonymous. There will be no negative consequences for you if you chose not to participate or later decide to withdraw and you will not be forced to take participate in this study if you do not want to.

Your privacy – how we will store and use your personal data

Before the start of data collection, I took ethical clearance from the ethical committee of Pakistan and from NSD (The Norwegian Centre for Research Data) and from the ethical board of my department. I ensured them that I will only use your personal data for this study and all the gathered data will be protected. I will process your personal data confidentially and in accordance with data protection legislation (the General Data Protection Regulation and Personal Data Act).

Only I will have access to your personal information and will lock all these collected information on TSD (UIO service for research data)

I will not reveal your name/contact details in my studies or anywhere and will replace with codes. The list of names, contact details and respective codes will be stored separately from the rest of the collected data in password-protected folders on TSD. Your personal information e.g. name. Poultry farm name, address, contact details will not be published anywhere.

What will happen to your personal data at the end of the research project?

The project is scheduled to end June 2021. All information will be processed and used without your name or any other information that is directly identifiable to you. Information about you will be anonymized or deleted a maximum of five years after the project has ended. The Institute of Health and Society (HELSAM), University of Oslo will be responsible for the data collected in this study.

Your rights

So long as you can be identified in the collected data, you have the right to:

- access the personal data that is being processed about you
- request that your personal data is deleted
- request that incorrect personal data about you is corrected/rectified

- receive a copy of your personal data (data portability), and
- send a complaint to the Data Protection Officer or The Norwegian Data Protection Authority regarding the processing of your personal data

What gives us the right to process your personal data? We will process your personal data based on your consent.

Based on an agreement with Institute of health and society Faculty of Medicine University of Oslo, Norway, NSD – The Norwegian Centre for Research Data AS has assessed that the processing of personal data in this project is in accordance with data protection legislation. Also ethical committee in of Pakistan allowed me to gather this information and I ensured them that I will keep all the personal information confidential.

Where can I find out more?

If you have questions about the project, or want to exercise your rights, contact:

• **Institute of Health and society Faculty of Medicine, University of Oslo Norway** via

1. Principal investigator: Um E Habiba, Master student of International Community Health Programme, Institute of Health and Society, Faculty of Medicine, University of Oslo.

   Contact Details:

   Email: u.e.habiba@studmed.uio.no

   Mobile Phone: +4793963401 & +923055134048

2. Supervisor: Dr. Muhammad Asaduzzaman, Centre for Global Health, Department of Social Medicine and Global Health, Institute of Health and Society, Faculty of Medicine, University of Oslo.

   Contact Details:

   Email: muhammad.asaduzzaman@medisin.uio.no

   Telephone: +47 96835658

3. Data Protection Officer: Data protection officer at UiO is Roger Mrkgraf-Bye.

   Contact Details:

   Email: personverrnombud@uio.no

   Telephone: +490822826

   NSD – The Norwegian Centre for Research Data AS

   Email: personverntjenester@nsd.no

   Telephone: +4755582117.

Yours sincerely,

Principal Investigator	Supervisor
Um e Habiba	Dr. Muhammad Asaduzzaman

Consent form

I have received and understood information about the project [insert project title] and have been given the opportunity to ask questions. I give consent:

- to participate in questionnaire-based study
- for my personal data to be processed
- for information to be published in the research study without revealing my personal information and my identity

I give consent for my personal data to be processed

(Signature/Thumb Impression by the participant, date)

Name of impartial witness and Signature*_____

*Required if the participant is unable to read or write

UiO : Faculty of Medicine
University of Oslo

Um E Habiba

Date: 18.09.2020

Statement from the Program Ethical Committee

The Program Ethical Committee have processed your application, number 8849626, about your project *"Use of antibiotics in animal and human (commercial poultry and poultry farmers) in rural Pakistan- A One Health study"*

The committee believe your project does not fall under the Norwegian Health Research Law (helseforskningsloven and forskningsetikkloven) and you do not need to apply to the Regional Committees for Medical and Health Research Ethic (REC). However, person sensitive information might be collected and therefore you need to apply to Norwegian Centre for Research Data (NSD) for approval.

If your project is to be conducted outside of Norway, you also need to submit the project to local authorities for approval.

Supervisor for **Um E Habiba** master project is:

- **Muhammad Asaduzzaman**-, Researcher- Institute of Health and Society at UIO

Sincerely yours

Elia John Mmbaga
Associate Professor, MD, PhD
Program leader
elia.mmbaga@medisin.uio.no

Terese Eriksen
Senior Executive Officer
terese.eriksen@medisin.uio.no
+47 22850526 or +47 22850550

Institute of Health and Society Phone: (+47) 22 85 05 50
Department of Community Medicine Telefax: (+47) 22 85 05 90
Postal addr.: PO Box 1130 Blindern, 0318
postmottak@medisin.uio.no Oslo
www.med.uio.no/helsam
Visiting addr.: Frederik Holsts hus,
Org. no.: 971 035 854
Kirkeveien 166, 0850 Oslo

NSD NORSK SENTER FOR FORSKNINGSDATA

NSD's assessment

Project title

Use of antibiotics in animal and human (commercial poultry and poultry farmers) in rural Pakistan- A One Health quantitative study

Reference number

726029

Registered

23.09.2020 av Um E Habiba - umeh@uio.no

Data controller (institution responsible for the project)

Universitetet i Oslo / Det medisinske fakultet / Institutt for helse og samfunn

Project leader (academic employee/supervisor or PhD candidate)

Muhammad Asaduzzaman, muhammad.asaduzzaman@medisin.uio.no,

tlf: 4796835658

Type of project

Student project, Master's thesis

Contact information, student

Um E Habiba, u.e.habiba@studmed.uio.no, tlf: 4793963401

Project period

01.08.2020 - 31.05.2021

Status

20.10.2020 - Assessed

Assessment (1) 20.10.2020 - Assessed

Our assessment is that the processing of personal data in this project will comply with data protection legislation, so long as it is carried out in accordance with what is documented in the

Notification Form and attachments, dated 20.10.2020. Everything is in place for the processing to begin.

NOTIFY CHANGES
If you intend to make changes to the processing of personal data in this project, it may be necessary to notify NSD. This is done by updating the Notification Form. On our website, we explain which changes must be notified. Wait until you receive an answer from us before you carry out the changes.

TYPE OF DATA AND DURATION
The project will be processing special categories of personal data about health and general categories of personal data, until 31.05.2021.

LEGAL BASIS
The project will gain consent from data subjects to process their personal data. We find that consent will meet the necessary requirements under art. 4 (11) and 7, in that it will be a freely given, specific, informed and unambiguous statement or action, which will be documented and can be withdrawn.

The legal basis for processing special categories of personal data is therefore explicit consent given by the data subject; cf. the General Data Protection Regulation art. 6.1 a), cf. art. 9.2 a), cf. the Personal Data Act § 10, cf. § 9 (2).

PRINCIPLES RELATING TO PROCESSING PERSONAL DATA
NSD finds that the planned processing of personal data will be in accordance with the principles under the General Data Protection Regulation regarding:

- lawfulness, fairness and transparency (art. 5.1 a), in that data subjects will receive sufficient information about the processing and will give their consent
- purpose limitation (art. 5.1 b), in that personal data will be collected for specified, explicit and legitimate purposes, and will not be processed for new, incompatible purposes
- data minimisation (art. 5.1 c), in that only personal data which are adequate, relevant and necessary for the purpose of the project will be processed
- storage limitation (art. 5.1 e), in that personal data will not be stored for longer than is necessary to fulfil the project's purpose

THE RIGHTS OF DATA SUBJECTS
Data subjects will have the following rights in this project: transparency (art. 12), information (art. 13), access (art. 15), rectification (art. 16), erasure (art. 17), restriction of processing (art. 18), notification (art. 19), data portability (art. 20). These rights apply so long as the data subject can be identified in the collected data.

NSD finds that the information that will be given to data subjects about the processing of their personal data will meet the legal requirements for form and content, cf. art. 12.1 and art. 13.

We remind you that if a data subject contacts you about their rights, the data controller has a duty to reply within a month.

FOLLOW YOUR INSTITUTION'S GUIDELINES

NSD presupposes that the project will meet the requirements of accuracy (art. 5.1 d), integrity and confidentiality (art. 5.1 f) and security (art. 32) when processing personal data.

To ensure that these requirements are met you must follow your institution's internal guidelines and/or consult with your institution (i.e. the institution responsible for the project).

FOLLOW-UP OF THE PROJECT

NSD will follow up the progress of the project at the planned end date in order to determine whether the processing of personal data has been concluded.

Good luck with the project!

قائداعظم یونیورسٹی

QUAID.I-AZAM UNIVERSITY

Faculty of Biological Sciences

Bioethics Committee

No. #BEC-FBS-QAU2021-250 Dated 26-01-2021

Ms, Um E Habiba

C/O Dr, Amjad Khan,

Assistant Professor,

Department of Pharmacy,

Faculty of Biological Sciences,

Quaid-i-Azam University, Islamabad

45320, Pakistan

Subject: - **"Use of Antibiotics in Animals and Ilumnn (Commercial Poultry and Poultry Farmers) in Rural Pakistan-One Ilcnlth Ouantitativc Study"**

Dear Ms. Um E Habiba,

We wish to inform you that your subject research study has been reviewed and is hereby granted approval for implementation by Bio-Ethical Committee (BEC) of Quaid-i-Azam University, Your study has been assigned protocol #BEC-FBS-QAU2020-250.

While the study is in progress, please inform us of any adverse events or new, relevant information about risks associated with the research. In case changes have to be made to the study procedure, the informed consent from and or informed consent process, the BEC must review and approve any of these changes prior to implementation.

Sincerely,

Sarwat Jahan.

Prof, Dr. Sarwat Jahan

cc:

Dean, F.B,S

Department of Animal Sciences

PROFESSOR
Department of Zoology
Quaid-i-Azam University
Islamabad

QUESTIONNAIRE

Topic: Use of antibiotics in animal and humans (commercial poultry and poultry farmers) in rural Pakistan- A One Health Quantitative Study

Respondent ID _____

1. Number of years in current occupation _____ years
2. Number of chickens in the farm? _____
3. Number of workers _____
4. What is the educational level of the respondent?
    - Non-Educated
    - Primary
    - Secondary
    - Higher Secondary or more

Questions relevant to farming practices and use of antibiotics in Poultry:

1. Do you have professional farm training?
    - Yes
    - No
2. Where do you buy poultry feed?
    - Private feed supplier
    - Local veterinary store
    - Others (give details) _____
3. In the last six months, have you bought/given any antibiotics or other treatment for your poultry?
    - Yes
    - No
4. If yes, type of treatment (tick all that apply):
    - Vaccines
    - Vitamins
    - Antibiotics
    - Growth promoters
    - Additives (give details) _____
    - Others (give details) _____

5. Give name of antibiotics provided if known (take details from package/bottle).

    _____

6. If yes, what was the treatment/ medicine taken for?
    - Diarrhea
    - Fever
    - Malaise
    - Fowl coccidiosis

- New castle disease
- Growth promotion
- Others (give details) _____

7. How is medicines/antibiotics given to poultry?
   - In water
   - In food
   - Directly fed to poultry by hand
   - Sprayed into cage/pens
   - Others (give details) _____
8. Where did you obtain the supplements/medicines/antibiotics? (tick all that apply)
   - Pharmacy/ drug shop/ drug seller
   - Agent
   - Others (give details) _____
9. Where do you get veterinary services?
   - Local livestock officer
   - Private veterinary doctor
   - By yourself
   - Feed company
   - Government source
   - Others (give details) _____
10. For how many days antibiotic/s was/were administered?
    - 1-3 day
    - 4-7 Days
    - >5 days
11. What is the frequency of antibiotics use in poultry?
    - Occasionally
    - Regularly
12. Do you get antibiotics with a veterinary prescription?
    - Yes
    - No
13. Do you follow the withdrawal period?
    - Yes
    - No

Questions relevant to antibiotics use in poultry farmers:

1. Did you use antibiotics in the last 6 weeks?
   - Yes
   - No
2. When did you use antibiotics for the last time? _____
3. What was the purpose of using antibiotics?
   - Flu (common cold)
   - Chest Infections
   - Fever

- Stomach problems
- Others (give details) _____

4. Did a physician prescribe antibiotics for you?
    - Yes
    - No
5. If no, why did you take antibiotics without a prescription?
    - Poor economic status
    - No access to physician care
    - Previous experience
    - Others (give details) _____
6. What was the source of antibiotics supply?
    - Pharmacy
    - Rural practitioner (Untrained doctor)
    - Leftover household antibiotics
    - Others (give details) _____
7. Where did you get information regarding antibiotics use?
    - Physician
    - Pharmacist
    - Relative or friend
    - Leaflet
    - Previous experience
    - Others (give details) _____
8. What was the duration of antibiotics intake?
    - 1–3 days
    - 4–7 days
    - > 7 days

Questions about the disposal of poultry wastes and water supply to farms

1. Which water supply in the farm do you use? _____
2. What is the distance of this water supply from the farm _____(meters)
3. What is the purpose of using this water?
    - Drinking water for poultry
    - Drinking water for humans
    - Washing hands (farm workers or others)
    - Washing down surfaces/floors
    - Cleaning poultry sheds
    - Others (give details) _____
4. Is there any wastewater drainage system in the farm?
    - Yes
    - No
5. What is the type of drainage?
    - Concrete and covered
    - Nonconcrete and open
    - Others (give details) _____
6. Where do you dispose the poultry waste?
    - Nearby pond

- Open area
- Use as fertilizer
- Others (give details) _____

Questions about knowledge of antibiotics use and antibiotics resistance

1. Do you have knowledge about antibiotics use in poultry?
   - Yes
   - No

2. Do you have knowledge about prohibited antibiotics in poultry?
   - Yes
   - No

3. Do you know about antibiotic resistance?
   - Yes
   - No

4. What do you understand about antibiotic resistance?

_____

_____

5. What do you think about the role of giving antibiotics for yourself and your family?

_____

_____

6. What do you think about the role of giving antibiotics to your poultry or other livestock?

_____

_____

Thank you again for taking part in this study. The findings from this research will help to reduce the health risks of infection with bacteria that are resistant to antibiot

سوال نامہ

1- آپ کتنے سال سے اس پیشے میں ہیں ؟ ----------

2- فارم میں مرغیوں کی تعداد؟ ----------

3- فارم میں کام کرنے والوں کی تعداد ؟ ----------

4- جواب دہندہ کا تعلیمی لیول؟ ----------

- کوئی نہیں
- پرائمری
- سیکنڈری
- ہائر سیکنڈری یا اس زیادہ

<u>فارمنگ کے طریقے اور پولٹری میں اینٹی بائیوٹکس کے استعمال کے بارے میں سوالات</u>

1- فارم سے متعلق پیشہ ورانہ ٹریننگ

- جی ہاں
- جی نہیں

2- آپ پولٹری کی خوراک کہاں سے خریدتے ہیں؟ ----------

3- کیا گزشتہ چھ مہینوں میں آپ نے اپنی پولٹری کے لئے سپلیمنٹ /اینٹی بائیوٹکس یا کوئی اور دوا خریدی ہے یا دی ہے؟

- جی ہاں

جی نہیں

4- اگر آپ کا جواب ہاں میں ہے تو دوا کی کونسی قسم ہے (درج ذیل میں سے نشان لگائیں)

- ایڈیٹو (تفصیل درج کریں)
- حیاتین
- اینٹی بائوٹک دوا
- اس کے علاوہ (تفصیل درج کریں) ----------

5- اگر جواب ہاں میں ہے اور معلوم ہو سکے تو اینٹی بائوٹک / دوسرے سپلیمنٹس کا نام لکھی) ----------

6- اگر آپ کا جواب ہاں میں ہے تو یہ علاج یا دوائی کس لئے لی تھی؟

- اسہال
- نیو کیسل کی بیماری
- بخار
- ملائیز
- اس کے علاوہ (تفصیل درج کریں) ----------
- صرف بڑھوتری کے لئے

7- پولٹری کو یہ دوائی یا اینٹی بائیوٹکس کیسے دی گئی؟

- پانی میں ملا کر
- پولٹری کو براہِ راست ہاتھ سے کھلائی گئی
- خوراک میں ملا کر
- پنجرے پر سپرے کر کے
- کسی اور طرح (تفصیل درج کریں) ----------

8- یہ سپلیمنٹس/دوائی یا اینٹی بائیوٹکس آپ نے کہاں سے حاصل کی؟ (درج ذیل پر نشان لگائیں)

- پولٹری کے خوراک سپلائیر سے
- فارمیسی/ دوائی کی دکان / دوا بیچنے والے سے
- سرکاری ذرائع سے
- گاؤں کے ڈاکٹر سے
- ہومیوپیتھک کے ڈاکٹر سے

9- آپ نے مویشیوں کے علاج کی سروس کہاں سے حاصل کی؟

- گورنمنٹ مویشی افسر سے
- پرائیویٹ مویشیوں کے ڈاکٹر سے
- کسی لوکل این جی او سے
- خود خریدی
- خوراک والی کمپنی سے
- کسی اور سے (تفصیل درج کریں) ----------

10- اینٹی بائیوٹکس کتنے دن استعمال کرائی گئی؟

- ایک سے تین دن
- چار سے سات دن
- سات دن سے زیادہ

11- آپ اینٹی بائیوٹکس کب استعمال کرتے ہیں؟

- کبھی کبھار
- باقاعدہ

12- کیا آپ اینٹی بائیوٹکس مویشیوں کے ڈاکٹر کے نسخے پر لیتے ہیں؟

- جی ہاں
- جی نہیں

13- کیا آپ دوا کے چھوڑنےوالا وقت پورا کرتے ہیں؟

- جی ہاں
- جی نہیں

14- کیا آپ پولٹری میں اینٹی بائیوٹکس استعمال کرنے کا حساب رکھتے ہیں؟

- جی ہاں
- جی نہیں

15- کیا آپ پولٹری میں اینٹی بائیوٹکس استعمال کرنے کا علم رکھتے ہیں؟

- جی ہاں
- جی نہیں

16- کیا آپ پولٹری میں ممنوعہ اینٹی بائیوٹکس کا علم رکھتے ہیں؟

- جی ہاں
- جی نہیں

17- کیا آپ پولٹری میں اینٹی بائیوٹکس کی مزاحمت کا علم رکھتے ہیں؟

- جی ہاں

- جی نہیں

<u>پولٹری فارم میں اینٹی بائیوٹکس استعمال کرنے کے بارے میں سوالات</u>

1- کیا آپ نے گزشتہ چھ ہفتوں کے دوران کوئی اینٹی بائیوٹکس استعمال کی؟
- جی ہاں
- جی نہیں

2- کیا آپ نے کبھی اینٹی بائیوٹکس استعمال کی؟
- جی ہاں
- جی نہیں

3- اینٹی بائیوٹکس استعمال کا مقصد کیا تھا؟
- زکام
- گلا پکنا
- اسہال
- کوئی اور (واضح کریں) ----------

4- کیا کسی معالج نے یہ دوائی تجویز کی؟
- جی ہاں
- جی نہیں

5- اگر نہیں تو آپ نے بغیر نسخے کے اینٹی بائیوٹکس کیوں دی؟
- ابتر معاشی حالات کی وجہ سے
- معالج تک رسائی نہ ہونے کی وجہ سے
- پچھلے تجربات کی وجہ سے
- کسی اور وجہ سے

6- اینٹی بائیوٹکس حاصل کا ذریعہ کیا تھا؟
- فارمیسی
- رشتہ دار یا دوست
- گھرمیں موجود تھی
- کوئی اور ----------

7- اینٹی بائیوٹکس کے استعمال کی معلومات کہاں سے حاصل کیں؟
- معالج
- ماہر ادویات
- رشتہ دار یا دوست
- دوا کے ساتھ دیا گیا کتابچہ
- پچھلے تجربات
- کوئی اور ----------

8- اینٹی بائیوٹکس کتنی مدت کے لئے استعمال کیں؟
- ایک سے تین دن
- پانچ سے سات دن
- سات دن سے زیادہ

9- مائکروبئیل مزاحمت سے آگاہی کے بارے میں کیا آپکو کوئی اندازہ ہے؟

- جی ہاں
- جی نہیں

<u>پولٹری فارم میں پولٹری فضلہ کو ٹھکانے لگانے اور نکاسیِ آب کے بارے میں سوالات</u>

1- آپ فارم میں پانی کی کونسی سپلائی استعمال کرتے ہیں؟ ----------

2- اس پانی کی سپلائی کا فارم سے فاصلہ کتنا ہے ؟ ---------- (میٹرز)

3- یہ پانی کس لئے استعمال ہوتا ہے؟ (درج زیل پر نشان لگائیں)

- پولٹری کے پینے کے لئے
- انسانوں کے پینے کے لئے
- ہاتھ دھونے کے لئے (فارم کارکن اور دوسروں کے لئے)
- نچلی سطحیں یا فرش دھونے کے لئے
- پولٹری شیڈ کی صفائی کے لئے

4- کیا فارم میں پانی کی نکاسی کا کوئی بندوبست ہے؟

- جی ہاں
- جی نہیں

5- نکاسی کی کونسی قسم ہے؟

- کنکریٹ کی اور ڈھانپی ہوئی
- کنکریٹ کی کھلی ہوی
- نیم کنکریٹ کی
- کنکریٹ کے بغیر زمینی نکاسی
- فارم کے پاس کھلی جگہ
- کوئی اور ----------

6- پولٹری کے فضلہ کو کہاں ٹھکانے لگاتے ہیں؟

- قریبی تالاب میں
- کھلی جگہ پر
- زرعی زمین میں

7- آپ خود کو اور اپنے اہل خانہ کو اینٹی بائیوٹکس دینے کے بارے میں کیا سوچتے ہیں؟

8- آپ اپنی پولٹری اور دوسرے مویشیوں کو اینٹی بائیوٹکس دینے کے بارے میں کیا سوچتے ہیں؟

اس سٹڈی میں آپ کی شمولیت کا شکریہ۔ اس ریسرچ کے حاصل کردہ نتائج سے بیکٹیریا سے ہونے والے انفکشن جو صحت کے لئے مضر ہیں کو کم کرنے میں مدد ملے گی یہ وہ بیکٹیریا ہیں جو اینٹی بائیوٹکس کے خلاف مزاحمت کرتے ہیں۔