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https://dx.doi.org/10.4314/nvj.v43i3.2 **ORIGINAL ARTICLE**

HYGIENE AND ENVIRONMENTAL SANITATION CONSEQUENCES ON THE SPREAD OF *Klebsiella species* AND ITS ANTIBIOTIC RESISTANCE IN A RUMINANT ANIMAL FARM.

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SUMMARY

This study assessed the sanitation and hygiene practices in an animal farm through walkthrough observation and one-on-one interviews, monitored the levels of selected air parameters in animal houses using portable air samplers, identified the *Klebsiella species* from the bacterial population in the nose and skin of selected ruminant animals, and skin of consented animal handlers; and examined the antibiotic sensitivity test (AST) on the confirmed *Klebsiella* (K.) isolates following standard procedures. Most handlers (77.78%) wash their hands before starting work without soap (55.56 %), with well (77.78 %) or spent water (22.22 %). A total of 181 and 132 distinct bacterial colonies and gram-negative bacteria species, respectively, were obtained. The highest bacteria count (28cfu/ml) was from Cattle nasal swab samples, while the lowest (17cfu/ml each) was from the skin samples of Goats and Sheep. Also, the highest gram-negative bacteria (24cfu/ml) was from sheep skin swab samples, while the least was from goat nasal swab samples. Eighteen of thirty-two presumed *K. species* were confirmed with colony characteristics and biochemical tests. Multidrug resistance (MDR) was prevalent in 77.7% of the confirmed *K. species*. Two-thirds of *Klebsiella* isolates in goats and cattle showed multidrug resistance, compared with 60% in Sheep and 100 % in handlers. *Klebsiella's* estimated resistance percentages against Penicillin, Cefuroxime, Ceftazidime, Nitrofurantoin, and Gentamicin were 100, 67.7, 50, 38.9, and 22.2, respectively. This study found a significant gram-negative bacteria load in the skin swabs of animal handlers, which will only worsen with inadequate personal hygiene and poor sanitation.

Keywords: Animal handlers, Poor hygiene, Zoonoses.

Running title: Sanitation, Hygiene, and Antibiotic Resistance Menace

INTRODUCTION

Environmental health concerns are critical when considering infectious disease emergence and spread. In small-scale animal farms, the availability of sustainable Water, Sanitation and Hygiene (WaSH) systems is questionable. Humans can be exposed to pathogens from poorly managed animal faeces, transmitted via WaSH-related pathways where household livestock, small-scale animal operations, and free-roaming animals are common (Penakalapati *et al.,* 2017); handlers may infect animals. According to the Food and Agricultural Organization (FAO), "domestic animals such as poultry, cattle, sheep, and pigs generate 85% of the world's animal faecal waste, proportionally a far more significant amount than the contribution by the human population (FAO, 2018). Many bacteria with high virulence have been isolated from livestock and their air environment, especially *Staphylococcus aureus* (Anika Friese *et al.,* 2012); alpha-haemolytic cocci (Murphy, 2012); *Klebsiella species* (Wareth & Neubauer, 2021) and others. Air pollutants in indoor environments of animal enterprises have been generally recognised as a threat to animals and farm workers (Donham. 2010). For instance, chickens produce a large amount of dust from epithelial desquamation, including feed, manure, faeces and litter (Matkovic *et al*., 2009). One of the bacteria which have been named as a threat to global health because the handling, interaction, and consumption of *K. species*-infected animals and their products could be a significant source of antibiotic resistance with potential human health implications and dire consequences on animal welfare is *K. species* (Effah *et al*., 2020). High morbidity and mortality of ruminant animals have been traced to infectious and non-infectious species of *Klebsiella* (Munoz *et al*., 2007). The factors responsible for *Klebsiella species* in animal production or enterprise can be grouped into environmental, animal, and handling factors (Studdert *et al*., 2012). Environmental factors include relative humidity, ambient temperature, sun intensity, wind speed, geographical region, disasters, insurgence, vegetation and ecological factors (Mercks, 2010); animal factors include genetic (species, age, allergy, animal behaviour), animal's nutritional status, and physiological adaptation to internal and external stress (Scott, 2007); and others are poor management systems, farm practices, sanitation and hygiene in animal houses (Biobaku & Shamaki, 2010). *Klebsiella (K.) species* is a zoonotic pathogen found in the digestive, urinary, and respiratory tracts, mostly of ruminant animals. This pathogen is of One health importance because it can also be found in various places, including air, food, water, sewage, soil, plants, and mammalian mucosal surfaces. It belongs to a group of bacteria frequently associated with antimicrobial resistance (AMR) known as ESKAPE (*Enterococcus. faecium, Staphylococcus aureus, K. species, Acinetobacter baumannii*, *Pseudomonas aeroginosa*, and *Enterobacter species*) (Rice, 2010), now ESKAPEE with the addition of *Escherichia coli* (Ngoi *et al.,* 2021). It can cause septic infections like abdominal, digestive system, intra-abdominal and bloodstream infections (BSIs) and meningitis (Bradley and Scoular, 2019). It is an essential cause of pneumonia, and epidemic metritis,

leading to high losses in milk production, decreased milk quality, weight loss, reduced animal performance and even high mortalities among affected animals (Hertl, 2014), which can result in noticeable economic losses in the dairy industry (Paulin-Curlee, 2008). *Klebsiella (K.)* spp is second to *E. coli*, with an incidence of 9.1 per 100,000/year in British Columbia (Reid *et al.*, 2019). *Klebsiella*-related infections can affect humans and animals by causing severe infections. Isolates of *Klebsiella species* often display drug-resistance traits, making it challenging to choose sensitive antibiotics for treatment. Increasing resistance to Augmentin (AUG) among *Klebsiella species* has also been reported (Schlievert *et al.*, 2010; Nirwati *et al.*, 2019; Awoke *et al.*, 2021). In addition, handling, interaction and consumption of products of these animals affected by *Klebsiella* may be an essential source of antibiotic resistance of possible human health significance (Shaban *et al*., 2013). The emergence of Carbapenem-resistant *K. species* has been confirmed in various hospital-related samples (Akinyemi *et al*., 2021). There is a need to ascertain the presence of different strains of *K. species* in animal enterprise since it has already been confirmed to be the primary cause of infections in neonates, infants and children (Akindolire *et al*., 2016; Uzoamaka *et al*., 2017). *Klebsiella* might be opportunistic, hyper-virulent or multidrug-resistant (Martin & Bachman, 2018) and pathogenic, with livestock acting as a reservoir. This organism is poorly understood and highly important in veterinary and environmental health. The risk of human infection via animal contact is understudied, especially in developing countries like Nigeria, where most studies on *K. species* have focused on infections in hospital settings (Navon-Venezia *et al.*, 2017). Hence, this study assessed sanitation and hygiene practices, the air quality within the animal, houses, and isolated *K. species* from the bacterial population in nasal lavage swabs, skin swabs of selected ruminant animals, and skin swabs of consented animal handlers while also carrying out antibiotic susceptibility tests (AST) on all the confirmed *K. species* subspecies.

METHODS

Study area and population

The study was conducted at a farm in the Odeda local government, Southwest Nigeria. Goats and sheep are kept in pens and enclosed by planks and nets for easy ventilation, and they are allowed to graze during the day. The goat, Sheep, and cattle pens have an area of 64680 cm^2, 65680 cm^2, and 18740 cm^2, respectively. The total population of the ruminant animals is 27 goats (10 males, 10 females, 3 kids, and 4 growers), 18 sheep (12 male and 8 female), and 55 cattle (37 female and 18 male). Also, there were 12 animal handlers across the three animal pens.

Sources of data

Walkthrough observation

A walkthrough on-site physical observation of the farm to assess the environmental conditions and hygiene practices was carried out to ascertain the responses provided by the animal handlers.

Interview

The interview was done using copies of the questionnaire written in English and then

translated into Fulfude and Yoruba, which are the native languages of the respondents.

Air quality monitoring

In-situ air quality monitoring was done in the windward direction in the sampled animal buildings for SO_2, VOC, $PM_{2.5}$, PM_{10}, H_2S, CH_4, CO_2 and CO using hand-held Aeroqual GasSensing active gas monitor (300 Series) portable air quality monitoring equipment. The Aeroqual Gas Monitor is a digital meter that takes time-weighted average values. For each monitored parameter, specific gas sensors are inserted and run for about 3 mins, and then results are displayed on the screen for recording. The measurements were obtained at the chest level to avoid interference from fugitive dust. The equipment was calibrated before and after each sampling batch according to the manufacturer's recommendations.

Sample collection for microbial analysis

Thirty nasal lavage and skin swab samples - 10 each for cattle (CN), goats (GN), and Sheep (SN) and nine skin swab samples of consented animal handlers (AH) (cattle – 5, goats – 2, and Sheep – 2) were collected purposively from selected ruminant animals to represent gender, age and breed using sterile wooden swab sticks soaked in peptone broth after sample collection. The sampling was done aseptically following standard procedures. Moreover, the consent of the handlers and management of the farm was sought and gotten. The swab sticks were transported to the laboratory on ice for culturing. The descriptions of the animal handlers and animals are presented in Tables III and IV, respectively. Indoor air microbial population was done by using a modified settled-plate method. Two plates (one for Nutrient agar and the other for MacConkey Agar) were opened and placed on each palm of the researcher. The researcher walked around the animal pens for about 3 minutes to settle viable bacteria on the plates. The researcher used hand gloves and nose masks to avoid media contamination. The description of pens are Sheep pen (SP), Goat pen (GP), and Cattle pen (CP). All plates were labelled and immediately transported to the laboratory for overnight incubation. It is worth noting that all samples were collected from healthy handlers and animals.

Bacterial isolation and identification of *Klebsiella species*

The moistened swab sticks were streaked on the nutrient and MacConkey agars for total aerobic and coliform counts in cfu/ml of suspension (peptone water) between 18 - 24 hours. *Klebsiella* spp. were identified based on the following criteria: gram-negative rods, lactose fermentation on MacConkey agar, negative oxidase test, non-motility, production of urease, utilisation of citrate, and usually indole negativity (Pondei *et al*., 2013; Mir *et al*., 2021; Zaidi *et al.*, 2022).

Antibiotic sensitivity testing (AST)

All confirmed *K. species* bacteria were tested for their sensitivity against eight antibiotics using the Kirby-Bauer disc diffusion method on Mueller-Hinton agar (MHA). The following antibiotics and their concentrations were used: Ceftazidime (CAZ) (30 µg), Cefuroxime (CXM) (30 µg), Gentamicin (GEN) (10 µg), Ciprofloxacin (CPR) (5 µg), Ofloxacin (OFL) (5 µg), Amoxycillin/Clavulanate (AUG) (30 µg), Nitrofurantoin (NIT) (300 µg), and

Ampicillin (AMP) (10 µg) (ABTEK) as prepared by the manufacturer. The inoculating loop was sterilised using the spirit lamp. The sterile loop was allowed to cool and then used to pick a colony of the confirmed *K. pneumonia* isolate and spread it evenly over the surface of the MHA (Geta *et al.*, 2019). The antibiotic discs were placed on the surface of the inoculated plates and incubated for 24 hours at 37ºC. The inhibition zone diameters were measured at millimetres and interpreted as susceptible, intermediate and resistant (CLSI, 2017).

RESULTS

Walkthrough observation and interview

The results of the on-the-spot walkthrough observation by the researcher and responses by consented animal handlers are presented in Table I.

Most respondents claimed that they wash their hands before starting the day's work (77.78%), while all of them agreed that they do not wash their hands in-between activities but at the end of the day's work. Meanwhile, our observation revealed that less than 50% wash their hands before commencing the day's work. Similarly, provision was not made for tap water and veronica buckets, but rather water used for hygiene activities is fetched from the well (77.78 %) or spent water (22.22 %) (already used water). Most (55.56 %) handlers always wash their hands with soap. However, our observation revealed that none of them practices handwashing with soap. Most (66.67 %) of the animal handlers believed that their contact with animals is the primary source of hand contamination, and 77.78 % claimed to have received on-the-job training on animal handling, none of them goes for medical check-ups except when they feel sick (55. 56 %).

TABLE I: Interview responses and researcher's observation

Variables	Interview responses	Frequency (%) (n = 9)	Observation
Washing of hands before the day's work	Yes	7 (77.78)	4
	No	2 (22.22)	5
Handwashing between activities during work	Yes	0 (0)	0
	No	9(100)	9 (100)
Washing of hands after the day's work	Yes	9 (100)	7 (77.78)
	No	0 (0)	2 (22.22)
Source of water for handwashing	Tap	0 (0)	
	Veronica Buckets	0 (0)	
	Well-fetched in bucket	7 (77.78)	Yes
	Spent water	2 (22.22)	
Use of soap to wash hands	Never	1 (11.11)	Never
	Often	3 (33.33)	
	Always	5 (55.56)	
The perceived major source of hand contamination	Environment	3 (33.33)	
	Animal	6 (66.67)	
Received on-the-job training in animal handling	Yes	7 (77.78)	
	No	2 (22.22)	
Frequency of medical check-ups	Never	1 (11.11)	
	When sick	5 (55.56)	
	Often	3 (33.33)	
	Always	0 (0)	

Air quality parameters

Good air quality leads to healthy animals and productive facilities. The levels of air quality parameters monitored are presented in Table II. The ranges of SO_2, VOC, H_2S, CH_4, CO_2 and CO (ppm) were 0.0 – 0.07±0.115, 155.33±34.29 – 414.67±13.051, 0.20±0.058 – 0.27±0.00, 144±0.298 – 201.67±21.362, 707±27.495 – 1072±314.98 and 34.67±2.517 – 81±14.107, respectively. Also, $PM_{2.5}$ and PM_{10} (mg/m^3) ranged between 0.01±0.00 – 0.07±0.036b and 0.03±0.00 – 0.04±0.005, respectively. Sheep houses have the highest VOC and CO_2, goat houses have the lowest CO level, and cattle houses have the highest H_2S, CH_4, $PM_{2.5,}$ and PM_{10}.

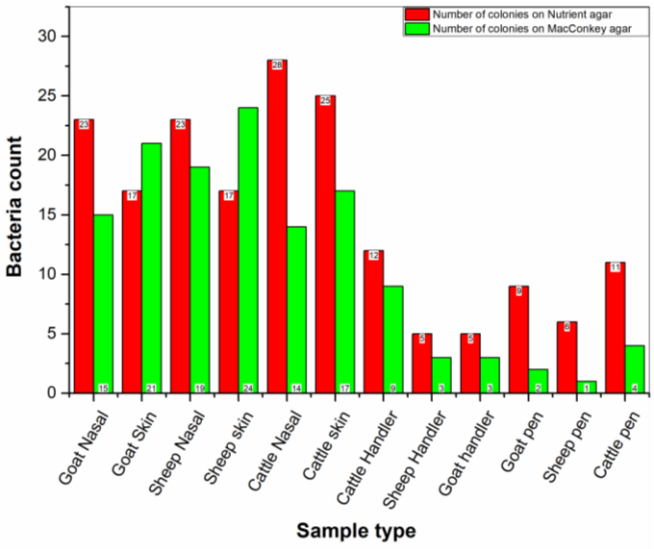
TABLE II: Mean values of air quality parameters monitored and analysis of variance (ppm)

Animal	SO_2	VOC	H_2S	CH_4	CO_2	CO	$PM_{2.5}$	PM_{10}
Sheep	0.07±0.115	414.67±13.051b	0.20±0.058	144±0.298	1072±314.98	81±14.107a	0.01±0.00a	0.03±0.001
Goat	0	155.33±34.294a	0.23±0.115	155.67±34.239	811±33.045	34.67±2.517a	0.01±0.00a	0.03±0.00
Cattle	0	187±98.727a	0.27±0.00	201.67±21.362	707±27.495	47.33±22.368b	0.07±0.036b	0.04±0.005

Bacteria counts and identification.

Figure 1 shows the proportion (counts) of the culturable bacteria on nutrient agar (NA) and McConkey agar plates from the 69 samples from ruminant animals and consented animal handlers. On the NA, there was a total bacterial count of 181 with the distribution CN>CS>GN=SN>GS=SS>CH>CP>GP>SP>SH=GH. Similarly, on the MacConkey agar plates, with the distribution SS>GS>SN>CS>GN>CN>CH>CP>SH=GH>GP>SP.

Fig1. Bacteria count on Nutrient and MacConkey Agar



A total of 6 different gram-negative organisms were isolated from the samples (Tables III and IV). The microorganisms are *Salmonella, Klebsiella, Proteus mirabilis, Escherichia coli, Shigella,* and *Pseudomonas*. Of all the organisms isolated, *Klebsiella* was the highest, followed by *E. coli*, with *Proteus* being the least isolated organism. *Proteus mirabilis* was isolated only in the nasal swab of cattle; meanwhile, *Shigella* species were isolated only from skin swabs of animals. Also of note is that the occurrence of the isolated organisms was more from the skin swabs than the nasal lavages, which could be because the skin is an outer covering and is prone to infestation by different agents which might be present in the ambience.

TABLE III: Description of handlers and Gram-negative organisms isolated

Sample ID	Sex	Age range	Tribe	Organisms
CH1	Male	40-50	Fulani	*Klebsiella*
CH2	Male	40-50	Fulani	*Proteus, Klebsiella*
CH3	Male	40-50	Fulani	-
CH4	Male	40-50	Yoruba	-
CH5	Male	45-55	Yoruba	*Salmonella spp*
SH1	Male	40-50	Yoruba	-
SH2	Female	45-55	Yoruba	-
GH1	Male	30-40	Yoruba	*Salmonella spp*
GH2	Male	35-45	Yoruba	*Salmonella spp*

CH (Cattle handler); SH (sheep handler); GH (Goat handler)

TABLE IV: Description of sampled animals and Gram-negative organisms isolated from animals.

Sample ID	Age	Gender	Breed	Nasal swabs	Skin swabs
G1	4 years	Male	WAD	*Salmonella*	*Klebsiella*
G2	1 year	Male	WAD	*Klebsiella*	*Pseudomonas*
G3	3 years	Male	Red Sokoto	*Salmonella*	*Salmonella, Shigella, E. coli*
G4	1 year	Male	WAD	-	*Klebsiella*
G5	1 month	Male	WAD	*E. coli*	*E. coli*
G6	3 years	Female	WAD	-	*Klebsiella spp*
G7	3 years	Female	WAD	*Klebsiella*	*E. coli*
G8	3 years 5 months	Female	CB	*Klebsiella*	*E. coli*
G9	1 month	Female	WAD	-	-
G10	1 year	Female	WAD	-	-
SI	2 years	Female	CB	-	-
S2	2 years 6 months	Female	CB	-	*Salmonella, Shigella*
S3	3 years	Male	WAD	*Klebsiella, Salmonella*	*Pseudomonas spp*
S4	3 years	Male	WAD	*Klebsiella*	*E. coli*
S5	3 years	Male	WAD	*E. coli*	*Salmonella, Shigella*
S6	2 months	Male	CB	-	*Klebsiella*
S7	2 months	Female	WAD	*E. coli*	*Pseudomonas spp*
S8	3 years	Female	WAD	*Klebsiella*	-
S9	2 years	Female	CB	-	-
S10	2 years	Female	CB	-	-
C1	3 years	Male	White Fulani	*Klebsiella spp*	*Salmonella, Shigella, Pseudomonas*
C2	5 years	Male	White Fulani	-	*Pseudomonas*
C3	3 years	Male	Red Bororo	-	*Klebsiella, Salmonella, Shigella, E. coli*
C4	3 years	Male	Muturu	*Klebsiella spp*	-
C5	3 months	Male	White Fulani	*Proteus mirabilis*	*E. coli*
C6	4 years	Female	Ndama	*Proteus mirabilis*	*Klebsiella*
C7	5 years	Female	Ndama	-	*E. coli*
C8	5 years	Female	White Fulani	*E. coli*	*Klebsiella*
C9	3 years	Female	White Fulani	-	-
C10	3 months	Female	Ndama	*Proteus mirabilis*	-

G (Goat), S (Sheep); C (Cattle): WAD (West African Dwarf), CB (Crossbreed)

TABLE V: Multidrug-resistant pattern of *Klebsiella* isolates

Sample	Cephalosporin		Aminoglycoside	Penicillin		Nitrofuran	Fluoroquinolones		MDR
ID	CAZ	CXM	GEN	AMP	AUG	NIT	OFL	CPR	
SN3	R	R	S	S	R	S	S	S	NO
SN4	S	S	R	R	R	S	S	S	YES
SN8	S	S	S	S	R	S	S	S	NO
SS6	R	R	S	R	R	S	S	S	YES
SS8	R	R	S	R	R	R	S	S	YES
GN7	R	R	S	R	R	S	S	S	YES
GN8	S	R	S	R	R	S	S	S	YES
GN10	S	R	S	R	R	S	S	S	YES
GS1	R	R	S	R	R	S	S	S	YES
GS4	S	S	R	R	R	R	S	S	YES
GS6	S	S	S	R	R	S	S	S	NO
CH1	R	R	S	R	R	R	S	S	YES
CH2	R	R	R	R	R	R	S	S	YES
CN1	S	R	S	R	R	S	S	S	YES
CN4	S	R	S	R	R	R	S	S	YES
CS3	R	R	S	R	R	S	S	S	YES
CS6	S	S	R	R	R	R	S	S	YES
CS8	S	S	S	R	R	S	S	S	NO

CAZ - Ceftazidime, CXM - Cefuroxime, GEN - Gentamicin, AUG -Amoxicillin/clavulanate, NIT- Nitrofurantoin; CPR - Ciprofloxacin; OFL – Ofloxacin; AMP - Ampicillin
R – Resistant, S - Sensitive

After the confirmatory tests, *Klebsiella* bacteria were isolated from eighteen out of the 69 samples (26.09%), comprising 6 from goats (GN7, GN8, GN10, GS1, GS4, GS6), 5 from Sheep (SN3, SN4, SN8, SS6, SS8), 5 from cattle (CN1, CN4, CS3, CS6, CS8) and 2 from cattle handlers (CH1, CH2). More *Klebsiella* was isolated from goats (33%), Sheep (27.7%), cattle (27.7%) and 11.1% from handlers. A closer look at the distribution of confirmed *Klebsiella* bacteria shows that in goats, they were all from the nasal samples, while in sheep, 60% were from nasal samples, and 40% were from skin swab samples. *Klebsiella* was isolated from 2 of the 9 animal handlers, and the two were cattle handlers. The results showed that *K. species* was confirmed in 7 females and 8 males of 15 animals; only a particular female sheep had *K. species* in both her skin and nasal swab. The ages of the animals that had confirmed *K. species* ranged between 2 months and 5 years. Most goats and sheep are pure (as identified and reported by the keepers and handlers) West African Dwarf breeds; 5 of the 8 WAD goats and 3 of the 5 WAD sheep harbour *K. species* in their nasal passage or skin swab.

Antibiotic sensitivity testing

The antibiotic sensitivity results are presented in Table III, showing the antibiotic resistance profiles of *Klebsiella* isolates from different host origins. Seventy-seven per cent of the total confirmed *Klebsiella spp* were MDR.

Although, for goats and cattle, about 83% of the confirmed *Klebsiella* were MDR, while it was 60% for Sheep and 100% for handlers. The isolated *Klebsiella* had 100% resistance to Augmentin, 67.7% resistance to Cefuroxime, 50% to Ceftazidime, 38.9% to Nitrofurantoin, and 22.2% to Gentamicin. On the other hand, there was a 100% sensitivity to the Fluoroquinolone class of antibiotics.

DISCUSSION

The possibility of the growth of any non-fastidious bacteria is evidenced in the bacterial count reported in this study, almost twice the count of the gram-negative bacteria on the MacConkey media (Quinlan, 2022), with the implication that there might be other bacteria with a high level of public health significance (Neal, 2022). This study has shown that many bacteria colonise the respiratory tract of apparently healthy ruminant animals, as seen in Sheep (Yimer & Asseged, 2007). *Escherichia coli* and *Klebsiella species* are two of the most common bacteria that cause animal diseases and are resistant to multiple antibiotics (Friese *et al*., 2013; Harada *et al*., 2016; Haulisah *et al*., 2021). The predominant species among the Gram-negative isolates were Klebsiella, unlike the study of Yimer & Asseged (2007), where *E. coli* was predominant., Gram-positive bacteria are the dominant species inhabiting the respiratory tract of seemingly healthy ruminants (Megra *et al.*, 2006; Yimer & Asseged, 2007; Addisu *et al.*, 2017). Bacteria found in animal confinement buildings increase the risk of spreading diseases among livestock and pose health hazards to farm workers and nearby residents (Roque *et al*., 2016). According to Ghanem *et al*. (2015), pneumonia infection caused by *Klebsiella species* isolated from the respiratory tract of pneumonic Boer goats causes pulmonary dysfunction. The results in this study align with the fact reviewed by Biobaku & Amid (2018) that animal factor plays a vital role in disease emergence and transmission. Studies have isolated *K. species* in hospitalised people (Akinyemi *et al.*, 2021) and abattoirs (Ayoade & Olayioye, 2016) but failed to establish a link to the animal-man transfer, especially during contact (Marques *et al*., 2019). This study found a high gram-negative bacterial load in the skin swabs of animal handlers, which will only get worse with the non-usage of personal protective equipment (Odo *et al*., 2015), poor waste management practices (Sobsey *et al*., 2006) and environmental conditions (Biobaku & Amid, 2018). Air quality in animal houses is a significant factor in animal health and disease protection (Quintana *et al.*, 2020). This study found that the monitored gaseous and particulate matter levels are within the threshold limits. Microbial air pollution from the livestock industry has raised concerns about potential public health risks and environmental impact. The current data suggest that bacterial pathogens are typically too sparse and short-lived in the air; they are most easily settled on surfaces such as the skin of animals and handlers and possibly breathed in through the nostril of humans and animals. Dust particles emitted may contain some pathogens that can pose a risk of airborne infection to humans in the vicinity and animals on other farms (Zhao *et al*., 2014), as indicated by the mortality of dairy cattle (Cox *et al*., 2016). Regarding the multidrug resistance pattern of the isolates, the isolates from humans were all multidrug-resistant (100%); this is higher than the 93% reported by Iramiot *et al*. (2020) in Uganda and the 90.4% MDR rate reported by Yang *et al*. (2019) in China. The cow had the

second-highest MDR of 80%, similar to what Iramiot *et al*. (2020) but higher than the 52% MDR rate reported by Yang *et al*. (2019). The high MDR observed in this study raises a concern: Cheng *et al*. (2018) confirmed hypervirulent *K. species* strains from cattle nasal swabs samples. Hence, the milk from such cattle might not be free of MDR-*Klebsiella* bacteria. This claim corroborates the report of Salauddin *et al.* (2019), which also found MDR-*Klebsiella* in milk samples of mastitic cattle. In addition, Yang *et al*. (2019) reported that sheep had the lowest MDR, similar to this research. The higher rate of MDR by both Cattle handlers is noteworthy.

Antibiotic resistance is one of the world's three major problems (Munita & Arias, 2016); about 94% resistant-*K. Species* to Augmentin (AUG), there is a threat to the effectiveness of AUG in treating broad-spectrum bacterial infections (Abubakar, 2020). Increasing resistance to Amoxicillin/clavulanate (AUG) among *Klebsiella species* has also been reported (Schlievert *et al.*, 2010). Meanwhile, the low resistance rate to Gentamicin which is below 30%, is similar to what was reported by Cheng *et al*. (2018). The total (100%) resistance of the isolates to Penicillin is similar to the report of Josy *et al*. (2018) and Wu *et al.* (2022). Results of our study indicated the prevalence rate of *Klebsiella* spp. in nasal swabs of animal handlers to be 22.2% which is lower than the 43.3% reported by Josy *et al*. (2018). *Klebsiella* found in cattle handlers could be due to the nomadic lifestyle of cattle handlers (Modupe *et al*., 2019) and poor hand hygiene. However, there is no limit to where K. *species* can be found, e.g. oral cavity of smokeless tobacco consumption (Harshal *et al*., 2020, fitness centres surfaces (Mukherjee *et al*., 2014), street foods and drinks **(**Budiarso *et al*., 2021).

The resistance rate to Cephalosporins, which was above 50%, is similar to the Cephalosporins reported by Yang *et al*. (2019). The highest resistance rate was found in humans, followed by Cattle.

CONCLUSION

Cross-infection potential due to *Klebsiella* and other gram-negative bacteria through the human, animal, and environmental medium is established. Similarly, absolute antibiotic resistance of the identified *Klebsiella* species to Penicillin (Augmentin) and the exhibition of MDR characteristics raise a public health concern. We recommend the efficient use of personal protective devices during operation in animal houses, routine maintenance of hygiene and sanitation practices, and responsible use of antibiotics.

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