Journal of Consumer Protection and Food Safety
https://doi.org/10.1007/s00003-018-1187-4

Journal of Consumer Protection and Food Safety
Journal für Verbraucherschutz und Lebensmittelsicherheit

RESEARCH ARTICLE

Prevalence of β-haemolytic multi-drug resistant *E. coli* in cow and camel milk in Kenya

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Received: 19 April 2018 / Revised: 18 September 2018 / Accepted: 20 September 2018

Abstract
The aim of this study was to find the prevalence of β-haemolytic *Escherichia coli* in milk, as well as their sources, and their sensitivity to antibiotics. *E. coli* was isolated from samples of cow and camel milk, cow and camel udder surfaces, and milking persons' hands. The organisms were identified using API20E biochemical kit. Haemolytic activity was tested on 7% defibrinated sheep blood agar while antibiotic sensitivity was tested using the Kirby-Bauer disc diffusion method. The prevalence of β-haemolytic isolates from cow and camel milk was 25% and 32% respectively. None of the isolates from the udder swabs, the milking persons' hands, or water was β-haemolytic. In cow milk, the prevalence of isolates resistant to Ampicillin, Cefotaxime, and Cefepime was 25, 37.5 and 12.5% respectively, while in camel milk it was 52.4, 23.8 and 28.6% respectively. Prevalence of β-haemolytic and multidrug resistant isolates to the three antibiotics was 12.5% for cow milk and 19% for camel milk. None of the isolates was resistant to Ciprofloxacin, Piperacillin/Tozobactam, Amikacin, and Imipenem. The prevalence of *E. coli* resistant to Cefotaxime and Cefepime indicates growing resistance of the microorganisms to drugs that are supposed to be effective against them. The presence of β-haemolytic isolates in milk and their absence on the animals' udder surface and hand swabs could indicate their better survival in the udder from which they are shed into the milk. This is a public health concern especially for pastoral communities who have a preference for consumption of raw milk.

Keywords *E. coli* · Milk · Antibiotics · Haemolysis · Multidrug resistance

1 Introduction

Kenya's dairy sector is estimated to be worth USD 1 billion with annual milk production of 4.5 billion litres of which 86% is from cows, 10% is from camels and the rest from goats (GOK 2010). The country's milk production from cows is dominated by Smallholder farmers while camel milk production is dominated by pastoralists (FAO 2011). Smallholder and pastoral milk producers are faced with many challenges in delivering good quality milk to the market. These include lack of proper hygiene during milking due to lack of water in the arid and semi-arid pastoral production areas. Water quality for udder preparation and cleaning of milk containers has been reported to be either scarce or of questionable quality (Noor et al. 2013). The level of hygiene in milk is indicated by the presence of coliforms in milk, which *Escherichia coli* is a dominant member. Presence of *E. coli* also indicates the presence of other enteric pathogenic microorganisms in the milk (Baylis et al. 2011) since it is associated with the guts of warm blooded animals and is frequently shed in the stools of these animals (Fernandes 2008). The microbes would then contaminate the udder and move up the teat canal not only contaminating the milk but also causing mastitis (Jay et al. 2005). Due to this relationship, the type of mastitis caused by *E. coli* has been demonstrated to be

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Published online: 10 October 2018

environmental mastitis rather than contagious mastitis (Khan and Khan 2006; Ahmed and Shimamoto 2011; Iyer et al. 2014). The lactating animals suffering from mastitis then shed culprit microbes with the milk (El-Ziney and Al-Turki 2008; Fernandes 2008). It has also been shown that cow and camels suffer from udder mastitis in the same way and the implicated microbes are the same (Iyer et al. 2014). Since mastitis is a disease condition in lactating animals, the *E. coli* that would be implicated are the pathogenic types with haemolysin production being one way in which the microbes express pathogenicity (Naveen and Mathai 2005). Haemolysin production in *E. coli* has also been associated with strains that cause enteric diseases in some species of animals and extra-intestinal diseases in man (Short and Kurtz 1971).

In Kenya, the bacteria that contribute most to human infections are those in which drug resistance is most evident, and these include pathogenic *E. coli* (GARP 2011). However, like in most of the African continent, there is no formal system for surveillance of antibiotic resistance in agricultural bacterial isolates (GARP 2011). *E. coli* has been demonstrated to be resistant to a number of antibiotics including ampicillin, ciprofloxacin, Cefepime, amikacin, and imipenem in human patients (Neto et al. 2003; Brinas et al. 2005; Chong et al. 2009) and dairy cows (Adefurin et al. 2011; Davies et al. 2015). Resistance of *E. coli* to third generation cephalosporins such as cefotaxime is due to the presence of plasmid mediated Extended Spectrum Beta Lactamase (ESBL) enzymes that are capable of hydrolyzing all beta lactams (Ahmed and Shimamoto 2011). The resistance gene can be acquired by a pathogen via horizontal gene transfer from one organism to another or by mutation though some organisms are inherently resistant (Vincent et al. 2010).

Previous studies indicate increasing resistance of *E. coli* to prescribed antimicrobials, especially, beta-lactams which makes empirical treatment of infections difficult (Neto et al. 2003; Vincent et al. 2010). Many of these studies have been done on patients with urinary tract infections, a few in mastitis animals and even fewer in foods. However, we are not aware of any research that has associated β-haemolytic *E. coli* to antibiotic resistance despite the compelling evidence that food could be a reservoir for *E. coli* that causes community acquired urinary tract infections (Vincent et al. 2010) and also a source of resistant bacteria (CDC 2013). Presence of pathogenic *E. coli* in milk is thus a public health concern especially for smallholder and pastoral communities where there is a lot of human contact with milk during milking, transportation and bulking. Pastoral communities too, have a preference for consumption of raw milk that increases risk of infection (Kaindi et al. 2011). The aim of this study was therefore to find the prevalence of β-haemolytic *E.coli* in milk, establish their sources and the sensitivity of these pathotypes to selected antibiotics.

2 Materials and methods

2.1 Sample collection, study area

The study was carried out between February and June 2016. Milk samples were taken directly from the udder during milking for both cow and camel milk. For cow milk, milk samples were also taken from bulking centres and milk bars while for camel milk, samples were taken from the secondary collection centre in Isiolo town and from the Nairobi market (Nato et al. 2018). For the purpose of this study, samples collected during milking were labelled as udder milk while those collected along the value chain were labelled as post-udder milk. The value chain in this context includes milk collection and bulking centres where milk may be cooled while either awaiting transportation to the processing plant or sale to consumers (Nato et al. 2018). For cow milk, samples were collected from 47 cows on 16 farms with 2–4 lactating cows per farm. Two farms had two milking persons while the rest had one each. Samples of water used for handwashing, udder and receptacle cleaning were taken from 10 farms since some farms shared water wells. Udder swabs from the lactating animals and hand swabs from the milking persons were also taken, as well as water for washing milking persons' hands, cow udders and milk receptacles. For camel milk, samples were collected from 3 kraals, each with a population of 9, 12 and 13 lactating camels. In each kraal, at least 2 camel udders were swabbed, and each of the kraals had at least two milking persons, both milking one camel at a time. However, for camel milk, only water for receptacle cleaning was sampled since udder cleaning and handwashing before milking was not practiced. For cow milk, 21 milk samples were collected from each of the 3 bulking centres within Nakuru County. Two of the bulking centres were owned by farmers' co-operative societies while one bulking centre was owned by a commercial milk processor. The rest of the milk samples were collected from 10 milk bars within Nakuru town, taking 3 milk samples from each milk bar. The milk samples from the bulking centres and milk bars constituted post-udder milk. Along the camel milk value chain, 46 milk samples were taken from the secondary collection centre in Isiolo town and 55 milk samples were collected from the Nairobi market. Milk collected from both the collection centre and Nairobi market constituted post-udder milk.

The milk and water samples were collected in 60 ml sterile bottles while hand and udder swabs were taken using sterile cotton swabs and put in 10 ml peptone water

in diluent bottles. Samples from the pastoral camel milk producers from Northern Kenya (Isiolo) were taken to the Isiolo County referral hospital laboratories for culturing, while samples taken from Nairobi market were cultured at the University of Nairobi laboratories. Samples from the Smallholder cow milk producers within Nakuru County were cultured at the Egerton University laboratories where isolation and biochemical identification was done for all isolates. All the samples were transported at 4–7 °C in a cool box, and refrigerated before culturing.

2.2 Isolation and identification of *E. coli*

The isolates were cultured on Eosin Methylene Blue agar (Oxoid, Hampshire, England) at 37 °C for 24 h. This was repeated until discrete dark colonies with or without a metallic sheen were isolated. Single colonies were picked and transferred to nutrient agar slants for morphological examination. Colonies that were Gram negative short rods were further identified using the API20E system (bioMereux SA, Lyon, France) according to manufacturer's instructions. The isolates were diluted to 0.5 McFarland standards before inoculation onto the ampules. The strips were incubated at 35 ± 2 °C for 24 h and the colour change in the ampules recorded as positive or negative. The microbial identity was done using an API20E software version 4.1 (bioMereux SA, Lyon, France). The remaining colonies were kept in 1 ml of 1 M sucrose solution in 2 ml cryo-vials and stored at − 18 °C to await further analysis. When required, the stored culture was re-cultivated in nutrient broth at 37 °C for 24 h.

2.3 Test for Beta haemolysis

Test for haemolytic activity was done by plating the isolates on Blood agar (Oxoid, Hampshire, England) with 7% defibrinated sheep blood at 37 °C for 24 h. In summary, the blood was obtained from ewes on the Egerton University farm using sterile hypodermic syringes. The blood was defibrinated and mixed by swirling with the blood agar base at a temperature of 45 °C then pouring about 20 ml onto sterile plates and allowed to set. The isolates were then streaked onto the solid plated and incubated to at 37 °C for 24 h after which haemolysis was observed (Fig. 1).

2.4 Antibiotic sensitivity testing

The β-haemolytic isolates were tested for antibiotic sensitivity based on the principle of the Kirby-Bauer disc diffusion method (Bauer et al. 1966). Ampicillin was chosen for this study because it was reported to be the drug of choice by the Veterinary officers in the study areas. This

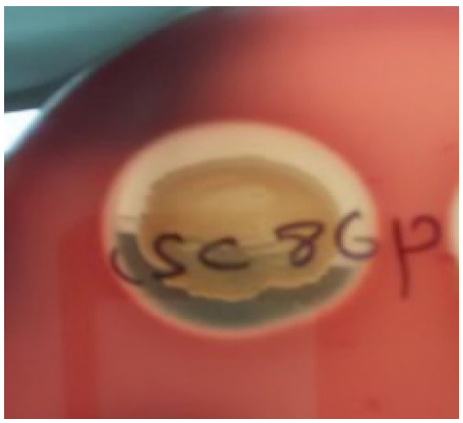

Fig. 1 A section of a Blood agar plate with 7% sheep blood inoculated with beta-hemolytic *E. coli* showing a clear zone

was corroborated by a study by Lamuka et al. (2017) which found Ampicillin administration at 27% for treatment of camel infections in Isiolo County. To expand the study, antibiotics recommended for treatment of Gram negative infections in human populations were also included in the study. These are Beta-lactams, aminoglycosides, and fluoroquinolones (Tamma et al. 2012). The Beta-lactams included cephalosporins, with Cefotaxime and Cefepime representing the third and fourth generation cephalosporins respectively; and carbepenem represented by Imipenem. Ciprofloxacin and Amikacin represented Fluoroquinolone and Aminoglycosides respectively while Piperacillin/Tozobactam (Penicillin/beta-lactamase inhibitor) was used to test susceptibility in the event the test organisms were resistant to Beta-lactams. All the antibiotic susceptibility discs were sourced from Liofilchem (Italy). Briefly, sterile solid plates of Muller-Hinton agar (Oxoid, Hampshire, England) were prepared and a broth of *E. coli* adjusted to 0.5 McFarland turbidity standards spread on its surface. Antibiotic discs were then carefully put on the solid plate, the plate inverted, and incubated at 35 ± 2 °C for 24 h. The inhibition zone was measured as diameter across the

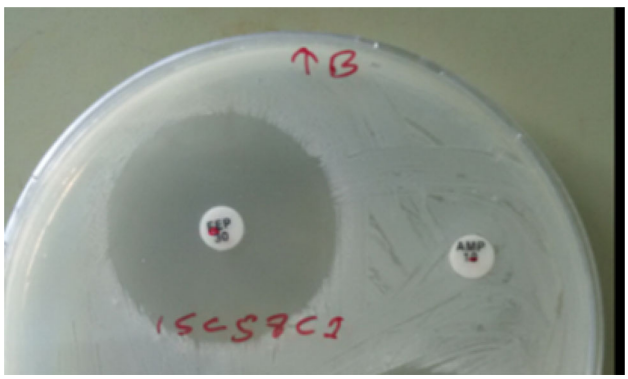

Fig. 2 A section of a Muller-Hinton agar plate showing susceptibility of beta-hemolytic *E. coli* to Cefepime (clear zone) and Ampicillin

antibiotic disc with a clear zone (Fig. 2). This was recorded in mm and interpreted according to the Clinical & Laboratory Standards Institute (CLSI) recommendations (CLSI 2016). *E. coli* ATCC 25922 was used as a standard.

2.5 Data analysis

Chi square test of equality of proportions for independent samples was used to compare prevalence of *E. coli* isolates and their antibiotic sensitivity, between cow and camel milk, and between udder milk and post-udder milk. The null hypothesis of equal proportions was rejected and that proportions were unequal if $p \leq 0.05$. SAS version 9.3 (SAS institute Inc, Cary, NC, USA) was used for all computations.

3 Results and discussion

3.1 Haemolytic activity

The sample type, number of samples taken, number of *E. coli* isolates and those tested for haemolytic activity and antibiotic sensitivity are shown in Table 1. Plates which had a clear zone around the colony were β-haemolytic, while those without a clear zone were considered non-haemolytic. The proportion of *E. coli* isolates that tested positive for beta-haemolysis were 25% and 33.3% for cow milk and camel milk respectively. None of the isolates from udder swab, milking persons hand and water were positive for beta-haemolysis.

Presence of *E. coli* from 142 cow milk samples was 90.1% which was significantly lower than 96.2% in camel milk from 136 milk samples, with $p \leq 0.05$. Prevalence for β-haemolytic *E. coli* as a proportion of *E. coli* isolates was not significantly different between cow and camel milk. The prevalence was 25.0% of 128 *E. coli* isolates for cow milk and 32.1% of 131 *E. coli* isolates for camel milk, with $p = 0.1439$. Along the chain, the prevalence of β-haemolytic *E. coli* in milk as a proportion of total *E. coli* in both cow and camel milk collected during milking (udder milk) was not significantly different from that isolated from milk from the rest of the value chain (post-udder milk). Similarly, prevalence of β-haemolytic *E. coli* as a proportion of total *E. coli* isolates between cow and camel udder milk, and between cow and camel post-udder milk was not significantly different as shown in Table 2. In both cow and camel milk, the proportion of β-haemolytic *E. coli* was significantly higher in post-udder milk compared to that of the udder milk. This is shown in Table 3.

It has since been established that *E. coli* causes infections due to the presence of virulent factors which include beta-haemolysins, ESBL and biofilms which endow it ability to survive, multiply and cause disease (Kukanur et al. 2015). The presence of *E. coli* with these virulent factors in milk and not on the udder surface, milking persons' hands and water therefore indicate the adaptation and colonization of these microbes in the lactating animals' udder. It has been shown that *E. coli* is one of the major microorganisms isolated from camel mastitis milk (Abdelgadir 2014), and in Kenya, Wanjohi et al. (2013) reported that the prevalence of *E. coli* isolated from camel milk was 60%. Indeed, a previous study by the authors indicated high levels of coliforms, which *E. coli* is a member, in camel milk in Kenya (Nato et al. 2018).

In this study, it is probable that the β-haemolytic *E. coli* were from infected udder tissues. This is because β-haemolytic *E. coli* are known to be mostly associated with diseased tissues because they have selective advantage by releasing iron from erythrocytes and enhances pathogenicity by destroying phagocytic and epithelial cells (Kukanur et al. 2015). The predominance of β-haemolytic *E. coli* in the udder milk could be because the protein haemolysin, which is the virulence factor, is only produced upon receiving certain signals from the host (China and Goffaux 1999), which in this study points at the udder of

Table 1 Types of samples collected along the smallholder and pastoral value chain for cow and camel milk respectively

Dairy value chain	Type of sample	Number of samples	Samples positive for *E. coli*	Number of β-haemolytic isolates
Smallholder dairy value chain for cow milk	Cow milk	142	128	32
	Udder swab	22	16	0
	Milking person hand swab	20	8	0
	Water	10	4	0
Pastoral dairy value chain for camel milk	Camel milk	136	131	42
	Udder swab	16	6	0
	Milking person hand swab	14	4	0
	Water	8	2	0

The number of samples collected during milking were 47 for cow milk and 34 for camel milk. The rest of the milk samples were collected along the value chain

Table 2 Prevalence of β-haemolytic *E. coli* as a proportion of *E. coli* isolates (n) between cow and camel milk; and between udder and post-udder milk in both cows and camels

Milk type	Udder milk (%, n)	Post-udder milk (%, n)	p value
Cow milk	20%, 35	26.9%, 93	0.4429
Camel milk	36.7%, 30	30.7%, 101	0.5382
p value	0.1344	0.5584	

p values in the rows and columns are for proportions in the rows and columns respectively

Table 3 Proportion of β-haemolytic *E. coli* isolated along the chain for cow and camel milk

Milk type	Udder milk	Post-udder milk	p value
Cow milk, n = 32	21.9%	78.1%	< 0.0001
Camel milk, n = 42	26.2%	73.8%	< 0.0001

n is the number of β-haemolytic *E. coli* isolates in both udder milk and post-udder milk. p values are for proportions in the rows

the lactating animal. In the environmental isolates, the genes of virulence may be present in the bacterial genome or the plasmid, even though the virulence factor is not expressed (Harel and Martin 1999).

3.2 Haemolytic activity and antibiotic sensitivity

Prevalence of ampicillin (AMP) resistant β-haemolytic *E. coli* was 52.4% in camel milk which was significantly higher than 25.0% in cow milk at $p < 0.05$. Resistance to cefotaxime (CTX) was at 37.5% for isolates from cow milk which was not significantly different compared to 23.8% for isolates from camel milk. Similarly, resistance to cefepime (FEP) for isolates from cow milk was at 12.5% and was not significantly different from that of camel milk which was 28.6%. Multidrug resistance to AMP/CTX/FEP too was not significantly different with 12.5% for isolates from cow milk and 19.0% for those from camel milk (Table 4). None of the β-haemolytic *E. coli* isolates was resistant to Piperacillin/Tozobactam, Ciprofloxacin, Amikacin, and Imipenem.

The emergence and spread of resistance of pathogens to antimicrobials is a worldwide phenomenon which may arise from their overuse, inadequate dosing, poor adherence, and substandard antimicrobials (GARP 2011; Patel and Levitin 2014). These antimicrobials are used in the control of diseases in human, animals and crops. In Kenya, penicillins are the most prescribed antibiotics in human subjects (GARP 2011) despite the fact that it is rarely recommended for treatment of serious Gram negative infections (CDC 2013). In this study, although the

Table 4 Antibiotic resistance of β-haemolytic *E.coli* isolated from cow and camel milk in Kenya

Antibiotic type	Source of microbe (milk)	% Resistance
Ampicillin (AMP)	Cow	52.4
	Camel	25.0
Cefotaxime (CTX)	Cow	37.5
	Camel	23.8
Cefepime (FEP)	Cow	12.5
	Camel	28.6
AMP/CTX/FEP	Cow	12.5
	Camel	19.0

resistance of β-haemolytic *E. coli* from cow and camel milk to AMP was 25% and 52.4% respectively, it was lower than 89% for *E. coli* isolates from healthy human subjects in Kenya (GARP 2011). The higher resistance in isolates from human sources is likely due to overuse of the antibiotic in human populations with contribution of consumption of animal products with high antibiotic levels. Indeed, Lamuka et al. (2017) reported that 46% of the pastoralists administer veterinary drugs to sick camels without seeking services of veterinary officers. This results to either underdosing, overdosing, or administering the wrong drug to the animal and lack of observation of withdrawal periods. The significantly higher resistance of isolates from camel milk compared to cow milk can therefore be attributed to the lack of antibiotic stewardship in administration of antibiotics to camels in the study area. The resistance to AMP can also be used to predict resistance to amoxyllin (CLSI 2016) which was also found by Lamuka et al. (2017) to be in common use among the pastoralists for treatment of camels, and also commonly prescribed to human populations in Kenya (GOK 2016). From our data however, it was not possible to directly link the cause for resistance of the pathogens to CTX and FEP because the drugs are not used in the study area for animal disease management, but are listed for use in human treatment in Kenya (GOK 2016). There is therefore a possibility of resistant strains from human subjects being passed on to animals. It could also be due to inherent ESBL's in the microbes. This is because resistance to CTX is a good marker of ESBL prevalence (Wiedemann et al. 2014), and demonstrates the ability of the *E. coli* strains to be resistant to other third generation cephalosporins (Paterson and Bonomo 2005). The level of resistance to Cefepime is of great concern being that it was only introduced into clinical practice in the 1990s and recommended for treatment of enterobacteriaceae infections. More so, in previous studies, FEP has been demonstrated to have 'in vitro' activity against ESBL-producing

enterobacteriaceae (Nogueria et al. 2011). This finding therefore demonstrates the prevalence of ESBL producing *E. coli* in the study area reducing treatment options for both infected persons and animals. This is because some ESBL's are known to destroy AMP, second, and third generation oxyimino-cephalosporins (Shaikh et al. 2015). It is however positive that none of the isolates was resistant to the second-line and last line agent, ciprofloxacin and Imipenem respectively. Ciprofloxacin is however restricted to high-level facilities, in this case, district hospitals and above (GARP 2011) which implies that individuals infected with these microbes cannot get treatment at lower medical facilities such as dispensaries. In addition, the use of ciprofloxacin in paediatric patients has been limited due to the possibility of arthropathy (Adefurin et al. 2011) and therefore children remain at great risk if infected. The use of last line carbepenem (Imipenem) would result in selection of carbepenem resistant enterobacteriaceae which is an emerging global time-bomb.

In conclusion, this study established that the source of β-haemolytic *E. coli* in milk was inside the lactating animals' udder. There's therefore need for antibiotic stewardship so that the right antibiotic is administered in the right way for the right infection. In this case, there should be a shift from blanket administration of penicillins to treat mastitis infections in the study areas to pathogen-specific antibiotic prescription based on microbial culture results. Meanwhile, the veterinary officers should change from penicillin drugs to *E. coli* (Gram negative) sensitive antibiotics for the treatment of mastitis in case the infected animals do not respond to antibiotics targeting Gram-positive microbes. However there is still controversy on whether treatment regimens for mastitis caused by Gram-negative microbes is effective or not. Prevention of mastitis infection should be primary in improvement of animal health. Interventions for microbiological contamination of milk should start from udder health instead of focusing only on nodes along the value chain. It is also important for pastoral communities to desist from tasting and consuming raw milk due to the presence of β-haemolytic *E. coli* strains in the milk. More concerted efforts should be deployed towards periodical tracking of resistant patterns of *E. coli* in milk.

Acknowledgements The study was facilitated by the RELOAD project with funding from the German Ministry of Education and Research (BMBF) within the framework of GlobE initiative, Grant no. 031A247A-D. The authors wish to thank Isiolo referral hospital and University of Nairobi for allowing them to use their microbiology laboratories. The authors wish to thank Ms Bernadette Misiko, Ms Ruth Imbahale, and Mr James Mwangi for their assistance in laboratory work. Special recognition is accorded to Mr Peter Lamuka (deceased), formally of University of Nairobi for the inspiration, encouragement and advice during the course of the study.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

References

Abdelgadir AE (2014) Mastitis in camels (*Camelus dromedaries*): past and recent research in pastoral production system of both East Africa and Middle East. J Vet Med Anim Health 6(7):208–216

Adefurin A, Jacqz-Aigrain E, Choonara I (2011) Ciprofloxacin safety in paediatrics: a systematic review. Arch Dis Child 96(9):874–880

Ahmed AM, Shimamoto T (2011) Molecular characterization of antimicrobial resistance in Gram-negative bacteria isolated from bovine mastitis in Egypt. Microbiol Immunol 55:318–327

Bauer AW, Kirby WM, Sherris JC, Turk M (1966) Antibiotic susceptibility testing by a standardized single disk method. Am J Clin Pathol 45(4):493–496

Baylis C, Uyttendaele M, Joosten H, Davies A (2011) The Enterobacteriaceae and their significance to the food industry. A report commissioned by the ILSI Europe emerging microbiological issues task force. International Life Sciences Institute. http://ilsi.eu/wp-content/uploads/sites/3/2016/06/EP-Enterobacteriaceae.pdf. Accessed 20 Apr 2014

Brinas L, Lantero M, Diego I, Alvarez M, Zarazaga M, Torres C (2005) Mechanisms of resistance to expanded-spectrum cephalosporins in *Escherichia coli* isolates recovered in a Spanish hospital. J Antimicrob Chemother 56:1107–1110

CDC (Center for Disease Control of the United States of America) (2013) Antibiotic resistance threats in the United States, 2013. US Department of Health and Human Services. www.cdc.gov. Accessed 10 July 2017

China B, Goffaux F (1999) Secretion of virulence factors by *Escherichia coli*. BMC Vet Res 30(2–3):181–202

Chong Y, Yakushiji H, Ito Y, Kamimura T (2009) Cefepime-resistant gram-negative bacteremia in febrile neutropenic patients with hematological malignancies. Int J Infect Dis 14s:e171–e175

CLSI (2016) Performance standards for antimicrobial susceptibility testing. 26th edition. CLSI supplement M100S. Clinical Laboratory Standards Institute, Wayne, PA. http://ljzx.cqrmhospital.com/upfiles/201601/20160112155335884.pdf. Accessed 13 Dec 2017

Davies MA, Sischo WM, Jones LP, Moore DA, Ahmed S, Short DM, Besser TE (2015) Recent emergence of *Escherichia coli* with cephalosporin resistance conferred by bla_{CTX-M} on Washington State dairy farms. Appl Environ Microbial 81:4403–4410

El-Ziney MG, Al-Turki AI (2008) Microbiological quality and safety assessment of camel milk (*Camelus dromedaries*) in Saudi Arabia (Qassim region). Appl Ecol Env Res 5(2):112–115

FAO (2011) Global food losses and food waste. Extent, causes and prevention. International Congress "Save Food", Düsseldorf, Germany. http://www.fao.org/docrep/014/mb060e/mb060e00.pdf. Accessed 21 Jun 2017

Fernandes R (2008) Microbiology handbook: dairy products. Leatherhead Publishing, Leatherhead

GARP (Global Antibiotic Resistance Working Partnership)—Kenya Working Group (2011) Situation analysis and recommendations: antibiotic use and resistance in Kenya: Washington DC, and New Delhi: Center for Disease Dynamics, Economics and Policy. https://cddep.org/wp-content/uploads/2017/08/garp-kenya_es.pdf. Accessed 21 Apr 2014

GOK (Government of Kenya) (2010) Kenya dairy master plan. Ministry of livestock development, Nairobi, Kenya. https://kdb.co.ke/press/publications/reports/5-kenya-national-dairy-master-plan/file. Accessed 12 Apr 2014

GOK (Government of Kenya) (2016) Kenya essentials medicines list 2016. Ministry of health http://apps.who.int/medicinedocs/documents/s23035en/s23035en.pdf. Accessed 23 Aug 2018

Harel J, Martin C (1999) Virulence gene regulation in pathogenic *E. coli*. BMC Vet Res 30(2–3):131–151

Iyer PA, Albaik M, Baghallab I (2014) Mastitis in camels in African and Middle East countries. J Bacteriol Parasitol 5:3

Jay JM, Losssner MJ, Golden AG (2005) Modern food microbiology. Springer Science + Business Media Inc, New York

Kaindi DW, Schelling E, Wangoh JM, Imungi JK, Farah Z, Meille L (2011) Risk factors for symptoms of gastrointestinal illness in rural town Isiolo, Kenya. Zoonoses Public Health 59(2):118–125

Khan MZ, Khan A (2006) Basic facts of mastitis in dairy animals: a review. Pakistan Vet J 26(4):204–208

Kukanur S, Meundi M, Bajaj A, Kotigadde S (2015) Co-relation between virulence factors and antibiotic resistance of *E. coli*, with special reference to uropathogenic *E. coli*. IOSR-JDMS 14(3):15–21

Lamuka OP, Njeruh FM, Gitao GC, Abey KA (2017) Camel health management and pastoralists' knowledge and information on zoonoses and food safety risks in Isiolo County, Kenya. Pastoralism 7:20

Nato SM, Matofari JW, Bebe BO, Huelsebusch C (2018) Effect of predisposing factors on microbial loads in camel milk along the pastoral dairy value chain in Kenya. Pastoralism 8:16

Naveen R, Mathai P (2005) Some virulence characteristics of uropathogenic *E. coli* in different patient groups. Indian J Med Res 122(2):143–147

Neto JN, Silva LD, Martins AC, Tiraboschi RB, Domingos AL, Suaid HJ, Tucci S, Cologna AJ (2003) Prevalence and susceptibility of hospital acquired urinary tract information. Acta Bras Cir 18(Suppl 5):36–38

Nogueria KS, Daur AV, Reason IT, Gales AC, Costa LM (2011) Cefepime versus extended spectrum β-lactamase-producing Enterobacteriaceae. Braz J Infect Dis 15(2):167–169

Noor IM, Guliye AY, Tariq M, Bebe BO (2013) Assessment of camel and camel milk marketing practices in an emerging peri-urban production system in Isiolo County, Kenya. Pastoralism 3:28

Patel T, Levitin A (2014) *Escherichia coli* adaptive resistance to clinical antibiotics. JSM Microbiol 2(1):1007

Paterson DL, Bonomo RA (2005) Extended-spectrum β-lactamases: a clinical Update. Clin Microbiol Rev 18(4):657–686

Shaikh S, Fatima J, Shakil S, Mohd S, Rizvi D, Kamal MA (2015) Antibiotic resistance and extended spectrum beta-lactamases: types, epidemiology and treatment. Saudi J Biol Sci 22:90–101

Short EC, Kurtz HZ (1971) Properties of haemolytic activity of *Escherichia coli*. Infect Immun 3(5):678–687

Tamma PD, Cosgrove SE, Maragakis LL (2012) Combination therapy for treatment of infections for gram-negative bacteria. Clin Microbiol Rev 25(3):450–470

Vincent CP, Boerlin P, Daignault D, Dozois CM, Dutil L, Galanakis C, Reid-Smith RJ, Tellier PP, Teliss PA, Ziebell TK, Manges AR (2010) Food reservoir for *Escherichia coli* causing urinary tract infections. Emerg Infect Dis 16(1):88–95

Wanjohi MC, Gitao G, Bebora L (2013) Subclinical mastitis affecting hygienic quality of marketed milk from North-Eastern Province, Kenya. Microbiol Res Int 1(1):6–15

Wiedemann B, Heisg A, Heisg P (2014) Uncomplicated urinary tract infections and antibiotic resistance—epidemiological and mechanistic aspects. J Antibiot 3:341–352