Prevalence of Escherichia coli *O157:H7 isolated from fecal samples of diarrheic camels in Tunisia*

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Veterinaria Italiana 2022, **58** (3), 315-323. doi: 10.12834/VetIt.2555.16997.2
Accepted: 07.03.2022 | Available on line: 31.12.2022

Keywords
E. coli O157:H7,
Camels,
Shiga-like toxin genes,
Enterohemorrhagic *Escherichia coli*,
Enterohaemolysin (ehxA),
Antimicrobial resistance.

Summary
Shiga-toxin-producing *E. coli* (STEC) is a foodborne pathogen associated with outbreaks worldwide that can be identified in the feces and in the meat of food-producing animals. Our study aimed to evaluate the incidence of *E. coli* O157:H7 in the feces of diarrheic camels (*Camelus dromedarius*) in Tunisia. From January 2018 to April 2019, 120 unduplicated fecal samples were obtained from diarrheic camels located in southern Tunisia. Non-sorbitol-fermenting colonies were confirmed as *E. coli* O157 via latex agglutination test and were screened for the presence of *rfbEO157*, *fliCH7*, *stx1*, *stx2*, *eaeA*, and *ehxA* genes by PCR. All isolates were examined for their susceptibility to 21 antibiotics. Of the 70 *E. coli* isolates that were recovered from 120 diarrheic camels, 4 (5.7%) were identified as STEC O157:H7. All isolates harbored *ehxA* and *eae* genes. Shiga toxin genes *stx2* and *stx1* were present in 50% and 25% of isolates, respectively. All *E. coli* O157:H7 isolates were sensitive to amoxicillin/clavulanic acid, cefotaxime, cefepime, aztreonam, colistin, and sulfamethoxazole-trimethoprim. All isolates belonged to the phylogroup E. This is the first report of *E. coli* O157:H7 isolates from diarrheic camels in Tunisia with a prevalence of 4 isolates (3.3%) amongst 120 fecal samples. This study supports the necessity for a platform purposed for regular screening and surveillance programs in food-producing animals and meat products, to perform early and rapid identification of food-borne pathogens.

Introduction

Escherichia coli is a Gram-negative, rod-shaped bacterium, that normally colonizes the intestine of human and most animals and is considered an opportunistic pathogen (Tayh *et al.* 2016). Some strains of *E. coli* are capable to cause severe diseases in the human gut. They are recognized as enterohemorrhagic *E. coli* (EHEC) belonging to intestinal pathogenic (diarrheagenic) strains that cause gastroenteritis. These strains cause illness in humans by producing an effective toxin known as Shiga toxin (Meng *et al.* 2012).

Shiga-toxin-producing *E. coli* (STEC) still known as VTEC (verotoxin-producing *Escherichia coli*) brings together several serotypes like EHEC pathogenic to humans, as food-borne bacteria linked with outbreaks worldwide. They can be associated with severe bloody diarrhea, hemolytic uremic syndrome (HUS), and/or hospitalization (Al-Ajmi *et al.* 2019, Falup-Pecurariu *et al.* 2019). These bacteria represent a significant public health concern and have the efficiency to produce Shiga toxin type 1 (*stx1*) and Shiga toxin type 2 (*stx2*), which are very potent toxins and are the main virulence determinants of this pathogen. Importantly, *stx2*-producing strains cause more severe infections than *stx1*-producing strains (Ogura *et al.* 2015). In fact, purified *stx2* is 1,000 times more toxic to human renal endothelial cells than *stx1*. The other main virulence factors produced by these serotypes are enterohaemolysin (*ehxA*) and intimin (*eae*) (Sperandio and Nguyen 2012). STEC O157 is the most clinically important serogroup but some serogroups non-O157 are also clinically significant foodborne pathogens, including STEC O26, O45, O103, O111, O121, and O145 (Al-Ajmi *et al.* 2019, Hegde *et al.* 2012).

According to the Centers for Disease Control and Prevention (CDC), the dissemination of the STEC to humans might take place through contaminated food such as beef meat, fruits and vegetables, contaminated water, or via contact with contaminated animals or persons (CDC 2019). It is found in the intestines of healthy cattle, goats, and sheep which are considered natural reservoirs and feed may be contaminated with livestock manure (Ferens and Hovde 2011). According to El-Gallas and colleagues (El-Gallas *et al.* 2006), in Tunisia, 3.4% of isolates from human stool samples were *E. coli* O157:H7.

However, the incidence reports of *E. coli* O157:H7 in camels (*Camelus dromedaries*) are rare. Studies from the United Arab Emirates (UAE) (Moore *et al.* 2002, Al-Ajmi *et al.* 2019), Iran (Rahimi 2012), Kenya (Baschera *et al.* 2019) and Iraq (Mohammed Hamzah *et al.* 2013) failed to isolate these bacteria among camel fecal samples. Furthermore, the same failure to detect this pathogen was reported in African countries (Egypt, Somalia, Djibouti, Kenya, and Sudan) (El-Sayed *et al.* 2008). However, in some countries, the prevalence of STEC O157:H7 isolates among healthy camel feces, has been reported, as in UAE was 4.3 (6/140) (Al-Ajmi *et al.* 2020), Saudi Arabia (2.4% and 11.5%) (Al Humam 2016, Bosilevac *et al.* 2015) and Iran (2%) (Sami and Adeli 2013), as well as from diarrheic camel feces in Egypt (17.9%) (El-Hewairy *et al.* 2009).

No data are available on the prevalence of *E. coli* O157:H7 among camels in Tunisia. We selected diarrheic camels to determine whether *E. coli* O157 is an agent of diarrhea as this has not been studied in Tunisia and can potentially pose a risk to people in contact with diarrheic animals. Therefore, this study aimed to determine the prevalence, virulence factors, and antimicrobial resistance profiles of STEC O157:H7 among fecal samples of diarrheic camels. To the best of our knowledge, this is the first report of STEC O157:H7 in camels in Tunisia.

Materials and methods

Samples collection

One hundred and twenty fecal samples were collected from diarrheic camels of southern Tunisian regions (Douz, Tozeur, Gabes and Ben Guerden) from January 2018 to April 2019. Only one specimen per animal was included. These samples were transported appropriately to the Microbiology Laboratory at the National School of Veterinary Medicine of Sidi Thabet for bacterial isolation and further investigations.

Bacterial isolation and identification

Fecal samples were enriched in buffered peptone water overnight at 37 °C, and then 10 µl were plated on MacConkey agar for 18-24 hrs at 37 °C. The *E. coli* isolates were identified by conventional biochemical tests. *E. coli* isolates were stored at - 20 °C in brain heart infusion broth supplemented with 20% glycerol.

Identification of *E. coli* O157:H7

One isolated strain was streaked on a plate of Sorbitol MacConkey with Cefixime Tellurite (SMAC-CT) agar. The bacterial plate was incubated overnight at 37 °C. After incubation, all colonies which are not able to ferment sorbitol (colorless/white colonies) were selected as probably *E. coli* O157. All *E. coli* colonies of non-sorbitol-fermenting on SMAC-CT medium were tested for the presence of the O157 antigen by agglutination test (DrySpot™ *E. coli* O157 Latex Agglutination Test, Thermo Fischer Scientific).

DNA extraction of bacteria genome

The extraction of the *E. coli* genome was carried out by boiling method. Bacterial colonies were suspended in 1 ml of sterile distilled water. After centrifugation at 13,000 rpm for 5 min, the supernatant was removed, replaced by 100 ml of sterile distilled water, heated at 95 °C for 10 minutes, and then kept at - 20 °C to be used for amplification by polymerase chain reaction (PCR). The PCR was carried out in this study including positive (*E. coli* O157:H7 isolated from cattle in our laboratory) and negative (DNA-free water) controls.

Detection of O157 and *fliCh7* genes by PCR

PCR amplification was used to detect the *rfbE* and *fliCh7* genes encoding for O157 and H7 antigens of *E. coli* O157:H7 strains, respectively, using oligonucleotide primers listed in Table I. DNA amplification reactions were carried out using a DNA thermal cycler (2720 thermal cycler, Applied Biosystem by Life Technologies, Singapore) with the following program: one cycle of denaturation at 94 °C for 5 min; 35 cycles of denaturation at 94 °C for 45 sec, annealing for 45 sec at 52 °C and 60 °C for O157 for detecting *rfbE* and extension at 72 °C for 45 sec; and a final extension at 72 °C for 10 min for detecting *fliCh7*. The PCR amplification products were separated by gel electrophoresis with 1.5% agarose and visualized under ultraviolet (UV) light using ethidium bromide staining.

Detection of virulence factors

The presence of *stx1*, *stx2*, and *ehxA* virulence genes with *uidA* gene was screened in *E. coli* O157:H7 isolates by the multiplex PCR method and *eae* by the simplex method using primers listed in Table I. The PCR thermocycler apparatus was used to perform PCR reactions. The reaction consists of 25 cycles of DNA denaturation for 1 min at 95 °C, primer annealing for 1 min at 56 °C, and first extension for 1 min at 72 °C. The last step is the final extension for 5 min at 72 °C. The separation of PCR bands was performed by electrophoresis using 2% agarose gel, the PCR products were watched under UV light by ethidium bromide stain.

Antimicrobial sensitivity test

The antimicrobial sensitivity was studied by the disk-diffusion method on Mueller-Hinton agar plates according to the guidelines and clinical breakpoints of the Antibiogram Committee of the French Society (CASFM-Vétérinaire 2018) using twenty-one antibiotics discs belonging to eight classes comprising µg/disk (Bio Rad, France): twelve β-lactams [(amoxicillin (25), amoxicillin/clavulanic acid (20/10), ticarcillin/clavulanic acid (75/10), cefotaxime (30), ceftazidime (30), cefepime (30), cefoxitin (30), aztreonam (30) ertapenem (10), piperacillin (30), cefalotine (30), cefuroxime (30)], and nine non-β-lactams antibiotics [chloramphenicol (30), gentamicin (15), nalidixic acid (30), enrofloxacin

Table I. *Primers for PCR amplification of* E. coli *O157:H7.*

PCR reaction	Gene	Primer sequence (5'-3')	Size of PCR product (bp)	Annealing temperature (°C)	Reference
		Phylogenetic genes			
Quadruplex	*chuA*	chuA.1b: ATGGTACCGGACGAACCAAC chuA.2: TGCCGCCAGTACCAAAGACA	288	60	Clermont *et al.* 2013
	yjaA	yjaA.1b: CAAACGTGAAGTGTCAGGAG yjaA.2b: AATGCGTTCCTCAACCTGTG	211	60	Clermont *et al.* 2013
	TspE4C2	TspE4C2.1b: CACTATTCGTAAGGTCATCC TspE4C2.2b: AGTTTATCGCTGCGGGTCGC	152	60	Clermont *et al.* 2013
	arpA	AceK.f: AACGCTATTCGCCAGCTTGC AceK.r: TCTCCCCATACCGTACGCTA	400	60	Clermont *et al.* 2013
Group E	*arpA*	ArpAgpE.f: GATTCCATCTTGTCAAAATATGCC ArpAgpE.r: GAAAAGAAAAAGAATTCCCAAGAG	301	57	Clermont *et al.* 2013
Group C	*trpA*	trpAgpC.1: AGTTTTATGCCCAGTGCGAG trpAgpC.2: TCTGCGCCGGTCACGCCC	219	59	Clermont *et al.* 2013
Internal control	*trpA*	trpBA.f: CGGCGATAAAGACATCTTCAC trpBA.r: GCAACGCGGCCTGGCGGAAG	489	57	Clermont *et al.* 2013
		Virulence factors			
Shiga toxin type 1	*stx1*	F: CAGTTAATGTGGTGGCGAAGG R: CACCAGACAATGTAACCGCTG	348 bp	56	Sjöling *et al.* 2015
Shiga toxin type 2	*stx2*	F: ATCCTATTCCCGGGAGTTTACG R: GCGTCATCGTATACACAGGAGC	584 bp	56	Sjöling *et al.* 2015
Enterohaemolysin	*ehxA*	F: GCATCATCAAGCGTACGTTCC R: AATGAGCCAAGCTGGTTAAGCT	534 bp	56	Grispoldi *et al.* 2017
Enteropathogenical attachment and effacement	*eae*	F: TGCGGCACAACAGGCGGCGA R: CGGTCGCCGCACCAGGATTC	629 pb	58	Ranjbar *et al.* 2017
		Others			
Part of O-antigen 157	*O157*	F: CGGACATCCATGTGATATGG R: TTGCCTATGTACAGCTAATCC	259 bp	52	Paton and Paton 1998
Encoding H7 flagellar antigens	*fliCH7*	F: GCGCTGTCGAGTTCTATCGAGC R: CAACGGTGACTTTATCGCCATTCC	625 bp	60	Al-Ajmi *et al.* 2020
Beta-glucuronidase	*uidA*	F: ATCACCGTGGTGACGCATGTCGC R: CACCACGATGCCATGTTCATCTGC	486 bp	56	Heininger *et al.* 1999

(5), tetracycline (30), sulfamethoxazole/trimethoprim (1.25/23.75), streptomycin (10), florfenicol (30) and colispot].

Detection of phylogenetic groups

The phylogenetic groups (A, B1, B2 and D) were identified in O157 isolates by quadruplex PCR using *chuA*, *yjaA* genes and the *TspE4-C2* fragment. The phylogroups C and E were detected by PCR using *arpA* and *trpA* genes. The primers and the method were previously described by Clermont and colleagues (Clermont *et al.* 2013).

Data analysis

The data of the camels; age, gender and origin were analyzed with the frequency of *E. coli* O157 by the Statistical Package for the Social Sciences (SPSS) version 26 software (IBM Corporation, Somers, NY). The comparison of data was performed using Pearson's Chi-square with a $P < 0.05$ value of statistical significance.

Results

A total of 70 non-duplicate isolates isolated from 120 diarrheic camel samples from four major cities in southern Tunisia were identified by conventional biochemical tests as *E. coli*. Testing of *E. coli* isolates on SMAC-CT and by agglutination test, revealed four isolates (5.71%) considered as presumptive *E. coli* O157:H7 isolates.

Thus, 4 *E. coli* O157 were identified out of 120 camels (3.33%). The frequency of isolation in this study was different between males and females with statistical significance (P = 0.045) (Table II). The difference between the rate of isolation according to the age of the animals was not statistically significant (P = 0.657) (Table II). Furthermore, there was no statistically significant difference between the isolation frequency of O157 according to animal's origin (P = 0.231) (Table II).

The confirmation of *E. coli* O157 by specific genes showed that *uidA*, O157 and *fliCh7* were present in all isolates. The virulence genes *eae* and *ehxA* were present in the four isolates; *stx1* and *stx2* were present in one and two isolates, respectively (Table III). All isolates belong to the phylogroup E.

All isolates were susceptible to cefoxitin, enrofloxacin, florfenicol, colistin, and sulfamethoxazole-trimethoprim. Furthermore, 75% of the isolates were susceptible to piperacillin, cephalotin, cefotaxime, nalidixic acid and tetracycline. However, 50% of the isolates were

Table II. *Prevalence of* E. coli *O157:H7 in Tunisian camels according to gender, age groups and region.*

Factors	No. (%) samples	No. E. coli O157	Rate
Gender			
Male	61 (50.8)	4	6.56%
Female	59 (49.2)	0	0
Total	120 (100)	4	3.33%
P-value = 0.045			
Age (years)			
< 1	70 (58.3)	2	1.67%
1-5	28 (23.3)	2	1.67%
6-10	6 (5.0)	0	0
≥ 10	16 (13.4)	0	0
Total	120 (100)	4	3.34%
P = 0.657			
Origin (city)			
Gabes	51 (42.5)	4	7.84%
Douz	34 (28.3)	0	0
Tozeur	19 (15.8)	0	0
Ben Guerden	16 (13.4)	0	0
Total	120 (100)	4	3.34%
P = 0.231			

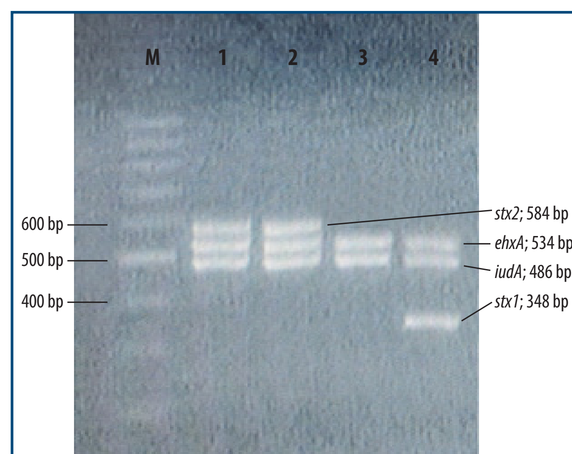

Figure 1. *PCR for detection of virulence genes.*
Lane M = Ladder 100 bp; lane 1, 2, 3, 4 = Positive samples; Agarose gel concentration: 2%.

Table III. *Characteristics of the 4* E. coli *O157:H7 isolated in Tunisian camels.*

Bacterial code	Specific genes			Virulence genes			
	O157	*fliCH7*	*uidA*	*stx-1*	*stx-2*	*eae*	*ehxA*
D5	+	+	+	-	+	+	+
D10	+	+	+	-	+	+	+
D25	+	+	+	-	-	+	+
D42	+	+	+	+	-	+	+

Table IV. *Antimicrobial susceptibility of* E. coli *O157:H7 Tunisian camel isolates.*

Antimicrobial	Susceptible	Intermediate	Resistant
Ampicllin	25%	75%	0
Piperacillin	75%	25%	0
Cephalotin	75%	25%	0
Cefuroxime	50%	0	50%
Cefoxitin	100%	0	0
Cefotaxime	75%	25%	0
Ceftazidime	25%	50%	25%
Cefepime	50%	50%	0
Aztreonam	25%	75%	0
Ertapenem	50%	50%	0
Amoxicillin/ clavulanic acid	25%	75%	0
Ticarcillin/ clavulanic acid	50%	0	50%
Gentamicin	50%	50%	0
Streptomycin	25%	25%	50%
Nalidixic acid	75%	25%	0
Enrofloxacin	100%	0	0
Chloramphenicol	50%	50%	0
Florfenicol	100%	0	0
Colistin	100%	0	0
Sulfamethoxazole/ trimethoprim	100%	0%	0%
Tetracycline	75%	0	25%

resistant to cefuroxime, ticarcillin-clavulanic acid and streptomycin (Table IV).

Discussion

Escherichia coli O157:H7 infections in humans originally came from food of animal or vegetable origin via food contamination with feces containing this pathogen. This particularly concerns ruminants such as cattle, sheep and goats which are considered the major natural reservoirs for this serovar and mainly implicated in human infections (Jo *et al.* 2004).

Due to the consumption of camel meat in Tunisia, specifically in the southern part, and given the absence of studies on this subject, it was an established decision to study the prevalence of *E. coli* O157:H7 in camels. The choice of diarrheic dromedaries is related to the lack of data on this pathology in Tunisia and on the zoonotic risk of serotype O157:H7, in order to raise awareness of this risk among individuals responsible for the maintenance and care of dromedaries (breeders, veterinarians, butchers, slaughterhouse employees etc). To our knowledge, in Tunisia, only one study has focused on *E. coli* isolated from diarrhoeic camels but has not targeted STEC (Bessalah *et al.* 2016). Interestingly, in Iran, Salehi and colleagues (Salehi *et al.* 2011) detected the serotype O157:H7 in two dromedaries with haemorrhagic diarrhea.

The occurrence of *E. coli* O157 was investigated among 70 *E. coli* isolated from feces of 120 diarrheic animals in the main southern Tunisian cities. This is the first report of *E. coli* O157 isolation from camels in Tunisia and also the first report in animals in Tunisia.

The incidence of *E. coli* O157:H7 in camel's fecal samples was 5.71%, with four isolates amongst 70 strains. The occurrence studies of this bacterium in camels are limited; a study conducted in the UAE was unsuccessful in detecting *E. coli* O157:H7 in fresh feces of racing camels (Moore *et al.* 2002). The same failure to identify *E. coli* O157 was among 400 camel fecal samples obtained from African countries (Egypt, Somalia, Djibouti, Kenya, and Sudan) (El-Sayed *et al.* 2008) and also from 40 camel samples in Iran (Rahimi 2012). More recently, in a study in Kenya, none of 163 fecal samples of grazing dromedaries contained STEC O157:H7 (Baschera *et al.* 2019).

An Iraqi study was performed on a zoo animals in Baghdad; they detected 24 *E. coli* O157:H7 out of 174 fecal samples from bear, deer, pony, horses, zebra, ostrich, hyena, llama, goat, and jaguar. However, no *E. coli* O157:H7 was identified among camels (Mohammed Hamzah *et al.* 2013).

Our methods used for the detection and isolation of O157:H7 are considered appropriate but non-selective, so the results might be an underestimate of the actual prevalence of O157:H7 among our samples. The ISO 16654:2001 method is the standard for detection of *E. coli* O157 but it requires separation and concentration using immunomagnetic beads coated (Tozzoli *et al.* 2019).

Our findings were compatible with the results of a study in the UAE (Al-Ajmi *et al.* 2020) in which the prevalence of *E. coli* O157 was 4.3%. They used an isolation method like the ISO 16654 method. The demonstration of *E. coli* O157 in camels was also reported in Saudi Arabia among 200 camel fecal samples in six isolates (11.5%) from 52 *E. coli* isolates (Al Humam 2016). In Iran, they reported 2% of 150 healthy camel feces were *E. coli* O157 (Sami and Adeli 2013). Another report from Iran demonstrated that one (1.3%) strain was *E. coli* O157 out of 75 camel meat samples using the protocol of selection of sorbitol negative colonies and PCR amplification of the O-antigen encoding region of the O157 gene and flagellar H7 gene (*fliC*) (Hajian *et al.* 2011). A high prevalence (17.9%) was recorded among 60 Egyptian camel calves; half of them suffered from diarrhea (El-Hewairy *et al.* 2009). In Iraq, Al-Gburi (Al-Gburi 2016) detected *E. coli* O157:H7 with 19% in healthy camels by the same protocol as our study.

Many reports confirmed that the prevalence of *E. coli* O157 isolated from camel feces was lower than that of those reported in cattle (Bosilevac *et al.* 2015, Sami and Adeli 2013) suggesting that camels cannot be considered the major natural reservoir for this pathogen in comparison with cattle.

Considering the low number of *E. coli* O157:H7 in Tunisia and few positive reports on camels in other studies, it can be concluded that the camel is not a significant source of STEC O157:H7 infection; practically, the studied camels share environments or pastures or pens with cattle, goats, and sheep. These ruminants can therefore be the source of STEC contamination of dromedaries. Surveillance and screening programs on camel products remain necessary for the prevention of outbreaks, particularly for food-borne infections, especially if those camels share spaces with ruminants.

The frequency of *E. coli* O157 was greater in males than in females with statistical significance, while there was no significant difference according to age or region. Our results correlate with another study in Saudi Arabia which reported that the prevalence of *E. coli* O157:H7 was significantly greater in young males than among females (Al-Ajmi *et al.* 2020). On the contrary, Baschera and colleagues (Baschera *et al.* 2019) reported from a Kenyan study a prevalence of 14.5% (17/117) of female camels and 6.7% (3/45) of male camels. Another study has found no statistically significant difference according to gender and age among camel samples (Al-Ajmi *et al.* 2020, Al-Gburi 2016).

In the present study, no statistically significant relationship was found between *E. coli* O157:H7 prevalence and the camel's origin. All *E. coli* O157:H7 were isolated from animals sampled in Gabes and these results highlight the high risk of the spread of this pathogen among animals from that region. Moreover, this finding strongly encourages us to carry out investigations on a larger scale of camels from Gabes and to try to find possible particularities of the camel farms of this region, and further investigations are necessary to study *E. coli* O157:H7 portage in cattle, sheep and goats from Gabes.

Serotype O157:H7 might carry *stx1* and *stx2* genes encoding Shiga toxins. Strains that possess *stx2* are associated with human illness more frequently than strains carrying *stx1* (Bosilevac *et al.* 2015). Both factors play important roles in the progress of hemolytic uremic syndrome. Epidemiological evidence of STEC O157 has revealed that strains carrying *stx2* are associated with more severe human infections than strains carrying *stx1* (Manning *et al.* 2008). Furthermore, STEC possesses hemolysin encoded by plasmid-carried enterohaemolysin gene (*ehxA*) which is associated with diarrheal disease and HUS (Fu *et al.* 2018). This factor is an important epidemiological marker for the rapid detection of STEC strains (Schwidder *et al.* 2019). Another important virulence factor is intimin protein (*eae*) which plays an important role in the attaching of host intestinal mucosa within the colonization and causes severe infections (Sperandio and Nguyen 2012).

In the present study, the STEC O157:H7 was confirmed by the identification of specific genes *rfb-O157* and *fliCH-7* using PCR as well as assessment of Shiga toxins genes (*stx1*, *stx2*) and enterohaemolysin (*ehxA*). Our finding revealed that *eae* and *ehxA* genes were present in all isolates; *stx2* and *stx1* were present in two and one isolates, respectively.

In a recent study, out of the 12 *E. coli* O157 isolates from fecal samples of healthy camels, *stx2* was found in all isolates, *eae* in 11 (91.7%), *hlyA* in 11 samples (75%) and *stx1* was absent in all isolates (Al-Ajmi *et al.* 2020). In a study conducted in Saudi Arabia, they found one isolate carrying both Shiga toxins genes (*stx1* and *stx2*), and like to our results, all 6 isolates of *E. coli* O157:H7 possessed the *eae* gene (Bosilevac *et al.* 2015). Rahimi and colleagues (Rahimi *et al.* 2012) confirmed 2.0% of 50 camel samples as *E. coli* O157:H7 but they failed to identify any virulence gene (*stx1*, *stx2*, *hlyA* and *eae*).

Antimicrobials have saved millions of human lives and their use has made a significant contribution to the improvement of the health of animals and humans. Antimicrobial resistance development is known as a global health threat. Antimicrobial agents are used in animals intended for human consumption to make animals healthier, reducing diseases and mortality (Oliver *et al.* 2011). The increased quantity of antibiotic residues in food-producing animals participates in the increase of antimicrobial resistance problems. Misuse and overuse of antimicrobials constitute a serious health enigma in many countries, such as using them for growth progression and infection prevention; furthermore, in several regions, the antibiotic quantities used in animals are four times larger than the quantities used in humans (Hudson *et al.* 2017).

The results of this study revealed that all isolates were susceptible to most tested antibiotics except cefuroxime, ticarcillin/clavulanic acid and streptomycin with resistance detected in 50% of the isolates and some isolates were intermediate to antibiotics which might progress to resistance. The results of an Iraqi study showed resistance of *E. coli* O157:H7 isolates to all the antibiotics tested in the study (doxycycline, cephalexin, erythromycin, clarithromycin, ceftriaxone, ampicillin, cloxacillin, rifampin and carbenicillin) except to trimethoprim (Al-Gburi 2016). A report in Iran revealed the resistance of *E. coli* O157:H7 to most tested antibiotics (ampicillin, erythromycin, gentamycin, nalidixic acid,

doxycycline, streptomycin, kanamycin, tetracycline, chloramphenicol and amoxicillin) and susceptible to cefuroxime (Tanzifi *et al.* 2015). In a UAE study, *E. coli* O157 isolated from the fecal samples of camels were 100% susceptible to cefotaxime, chloramphenicol, ciprofloxacin, norfloxacin, and polymyxin B (Al-Ajmi *et al.* 2020). The results of our study showed that the *E. coli* O157 isolates belonged to the phylogroup E, which is in agreement with other studies (Coura *et al.* 2015, Tenaillon *et al.* 2010).

Antibiotic resistance in these isolates can be used as an indicator of antibiotic use in the animals or their environment. These resistances can be useful for studying the isolates, but O157:H7 infections in humans are not treated with antibiotics as this can induce expression of Shiga toxins and worsen the disease.

Conclusions

This is the first report of *E. coli* O157:H7 isolated from diarrheic camels in Tunisia. The presence of this pathogen in our study and some positive reports in the region suggest that camels might be considered as the natural reservoir and play an important role in the infection of humans with this pathogen. Our results supply some baseline data concerning the occurrence of *E. coli* O157:H7 in camels and their antimicrobial resistance that could be used in future studies. This work leads to highly recommended surveillance and screening programs on camel products to prevent outbreaks. Furthermore, it supports the necessity for a platform purposed to regular screening and surveillance programs in food-producing animals and meat products, to allow early and rapid identification of food-borne pathogens. More studies should be performed to characterize the prevalence of STEC O157 and other non-O157 pathogens in animals and meat food products with a large number of isolates.

Funding

This work was supported by the research project PEER 7-349 funded by the USAID "Monitoring of antimicrobial resistance of bacteria for a better health of animals in Tunisia".

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