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Original Scientific Article

IMPACT OF VEROTOXIC *E. COLI* O157 IN ANIMALS ON THE HEALTH OF SLOVENIAN HUMAN POPULATION

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Summary: Infection with verotoxic *Escherichia coli* (VTEC) is an important veterinary public health issue. Ruminants are considered the natural reservoir of VTEC and therefore meat consumption is one of the potential transmission routes. Much effort has been dedicated to monitoring programmes so as to measure the prevalence of VTEC in ruminants in Slovenia. The aim of this study was to statistically analyse data from the national monitoring system by calculating the prevalence of VTEC O157, and to test for association between the season, type of sample, age of cattle, and region with the outcome (chi square, logistic regression). Results were compared to data on human infection with VTEC. Some conclusions for further improvement of surveillance systems were drawn. Animal data were collected between 2006 and 2009 for the purposes of monitoring the prevalence of VTEC O157 in cattle (n=818), sheep (n=320), and meat in cutting plants (n=582), where specific attention was paid to sampling different carcasses/batches. Data regarding human infection with VTEC, collected from 2006 to 2008, included information on age group, gender, season of E. coli notification, and VTEC serotype. Prevalence of VTEC 0157 in cattle was 3.1% (95% CI 1.9% to 4.2%), in sheep 0.9 % (95% CI 0.2% to 2.7%), and in meat 0.2 % (95% CI 0% - 1%). Age of cattle was not significantly associated with positive tests; however, there was a statistically significant seasonal effect across all sample types (p=0.035), as positive test results were found predominantly in autumn. Out of 93 notifications of E. coli in humans, only 15 were confirmed as VTEC (16.1%). Calculated incidence rate of human cases in Slovenia was low (0.2, 0.2 and 0.4 cases per 100 000 population in 2006, 2007 and 2008, respectively), and the most frequently affected group were children. Seasonal patterns for tested animals and human cases were rather similar. In animals, however, VTEC O157 only was tested, whereas the most frequent serotypes isolated from humans were O26, O157, O103 and O111. Evidence shows that this pathogen is harboured by cattle; however, the prevalence in meat is rather low. The precise transmission route in human cases of VTEC in Slovenia remains unclear; and foodborne infection is only one of the options. It may well be that new emerging routes are shifting towards environmental transmission.

Key words: zoonosis; verotoxic *E. coli* (VTEC); ruminants; veterinary public health; Slovenia

Introduction

Verotoxic *E. coli* (VTEC) is a bacterium that produces verotoxins (*Vt*), which damage human intestinal cells, cause diarrhoea and potential life-threatening complications (1). There are two major groups of *Vt,* namely, *Vt1* and *Vt2* (1). Whilst *Vt1* is a homogeneous family of toxins, equivalent to the Shiga toxin of *Shigella dysenteriae, the Vt2* group is deemed to have a much more detrimental potential (1, 2). In addition to production of *Vt,* the bacterium needs to have mechanisms for adhering to and damaging intestinal cells. This is achieved through the AE (attaching/effacing) lesion, encoding different genes (eae genes and genes for Vt), and bacteria possessing these two virulent factors are associated with severe clinical manifestation (2). Both these virulence factors are of crucial importance in laboratory conformation methods.

The surface of *E. coli* is covered by O (somatic) and H (flagellar) antigen profiles, which constitute

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the basis for serotyping. There are more than 200 *E. coli* serotypes (2), and VTEC O157:H7 is in the worst repute of them all. Infection with historically notorious VTEC O157 serotype is strongly associated with severe complication, such as haemorrhagic uraemic syndrome (HUS)(3). HUS is caused by circulating verotoxins, damaging kidney cells, and leading to thrombosis and resulting in acute renal failure (3). Therefore, most national surveillance systems focus only on detecting this particular serotype and making it a most frequently reported one. Non-O157 VTEC serotypes, most commonly associated with severe clinical symptoms, include serotypes O26, O103, O111, O113 (4) and O145 (2).

Though a tremendous amount of knowledge on VTEC has been generated over the past 30 years, infection in humans is still a reality. Therefore, infection with VTEC is considered an emerging zoonosis. In this paper, the specific term of VTEC O157 was examined in animals, whilst in relation to humans the term of VTEC was used for all the types of verotoxin producing *E. coli*.

VTEC epidemiology in cattle and humans

VTEC is a bacterium that is part of normal gastrointestinal flora of ruminants, and thus, these animals may act as a potential reservoir of infection (1, 2, 3, 5, 7, 8, 9, 10). Animals may carry VTEC without any symptoms. There are differences in prevalence and shedding of VTEC. Studies of cattle herds in Scotland showed that the so-called super-shedding animals were responsible for the majority of transmissions (11). Other species considered as possible carriers include sheep and, to a minor extent, goats (10).

It has been reported (12) that an annual prevalence of VTEC in cattle faeces was 4.7 %. However, researchers failed to report age distribution of sampled animals, and sampling was limited to animals of up to 30 months of age. Other researchers (13) have reported a prevalence of 1.2 % in cattle, failing to provide information on how the sampling was conducted. The peak prevalence in cattle during warmer seasons has been described by several researchers studying VTEC (12, 14, 15, 16, 17). It has been reported that more animals had been found positive for VTEC in the warmer seasons (15, 16).

Studies suggest that prevalence in meat is lower than prevalence in animal faeces (18,19). The prevalence in sheep has been reported to be lower than in cattle (12,14); however, sheep may still be an important source of infection, particularly through consumption of raw milk or cheese (4).

In 1983, two human cases in Oregon and Michigan were reported, which were characterized by severe cramps and abdominal pain, where initial watery diarrhoea was followed by gross bloody diarrhoea (20). Diagram 1 outlines the pathogenesis. Diagnosis of VTEC O157:H7 should be considered in any person reporting bloody diarrhoea or HUS (21). Additionally, the incidence is greatest within the most vulnerable groups, as children and the elderly. VTEC infection is known to impair the function of kidneys; this is particularly excruciating for children, who can suffer lifelong consequences.

The immune status is a major factor in the susceptibility to VTEC infection. Children and the elderly are most severely affected, as their immunity is either in development or in decline (2). In addition to these age-related physiological factors, curtailed immunity could be due to the following facts: use of antibiotics in animal production, consumption of processed foods, urbanisation with absence of contact with animals, over-cleaning and use of disinfectants in on a daily basis. An increasing body of evidence suggests the role of acquired immunity as an important factor in preventing VTEC infection in early childhood (2). Moreover, repeated exposure of persons residing in rural environment to these organisms is linked with sub-clinical infections, acting as immunisation against the disease (1). For instance, a rural population, which had been exposed to VTEC, had elevated anti-verotoxin antibodies, as compared to a comparable urban population (2). This suggests that exposure to this bacterium, providing that a person's immune system is capable of effective defence, may have a protecting role.

Additionally, the importance of VTEC as an emerging pathogen should be viewed through costs encumbering the healthcare system. A recently published study (22) suggests that in the USA the estimated cost of foodborne illness-related issues amounts to USD 152 billion per annum in healthcare, at workplaces, in addition to other economic costs. Expenditures related to *E. coli* O157:H7 infection alone (requiring hospitalisation) were estimated at USD 14 838 per case (22). The total cost related to this pathogen was estimated at USD 993 million (with 95 % CI 296-1689).

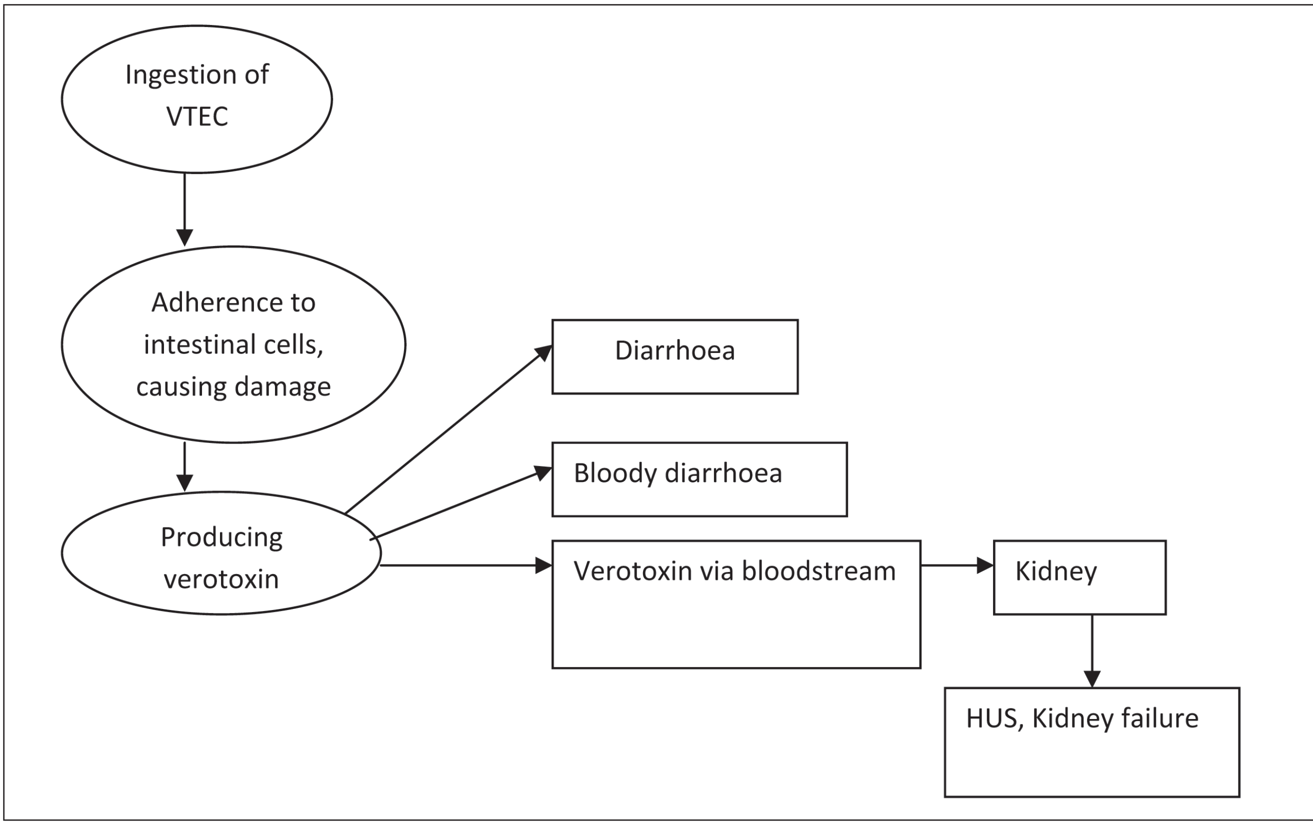

Diagram 1: VTEC infection pathogenesis

Pathways of VTEC infection transmission from animals to humans

Food of animal origin is often found to be the source of transmission to humans (2, 5, 6, 10). A particular threat is posed by undercooked minced meat; grinding of this meat helps to deliver the pathogen into the interior of the meat, where it is more likely to survive cooking (19, 5, 9). The quantity of bacteria required to cause the disease is very low, that is less than 100 cells (10). For a long time, undercooked meat had been considered as the principle source of infection. However, nowadays, knowledge suggests a wider diversity of food types potentially responsible for the spread of infection. These include apple cider and vegetables, such as lettuce, alfalfa sprouts and spinach (5). For example, an outbreak of *E. coli* O145 in USA was reported, where 26 people from multiple states were infected whilst the determinant was contaminated shredded lettuce (23). Cross contamination of vegetables usually happens by water and soil that is contaminated with animal faeces (24). This can be particularly problematic with vegetables, consumed in a raw state. As first, a foodstuff gets contaminated with VTEC (primary source, cross-contamination, or from contaminated workers, processing the produce), and in addition, it is inadequately prepared, stored or cooked, consequently allowing survival of VTEC (4). Pasteurised milk is less commonly reported as primary source of infection; however, since it is used as a raw material for milk products, cross contamination is more likely to occur, especially if the production premises are on the farm (25).

In a 10-year study of outbreaks in the USA (26) researchers report that food contaminated with VTEC was responsible for 61% of the cases. The produce and ground beef were the key sources of infection in 41% of the cases. Scottish researchers (27) found that in 54% of outbreaks caused by VTEC the transmission route was from the environment, while in 40 % it was foodborne. Other Scottish reporters observed that incidence of infection was 1.7- times higher in rural areas, and this factor was over three for children (28). This is supported by the report (29) of outbreaks in Scotland in 2004, where half of the cases were children, and contact with cattle and/or their excreta was considered the key source of infection.

According to *Trends and sources of zoonoses and zoonotic agents in EU in 2008* (30) top five VTEC serogroups include O157, O26, O103, O145 and O91. Within the EU, children accounted for 64.6% of the 144 HUS cases in 2008, and these were mainly associated with serogroup O157 (30). Infection with VTEC in Slovenia in 2005 and 2006 resulted in two deaths; one elderly man and one child (31). Later there was a fatal case of HUS following infection with O145 VTEC (32). Investigators report that meat and water samples tested positive for *E. coli*, however, not for serogroup O145 (32). They hypothesize that most likely the source of infection could be minced meat, but there were no leftovers to be tested for VTEC.

VTEC surveillance in Slovenia is passive and based on GPs' reporting of *enterohaemorrhagic E. coli* (EHEC). Notification rates are basically the number of patients presented with clinical signs for EHEC infection divided over 100 000 population, but not yet confirmed as VTEC. There is a central laboratory, where final diagnostics is carried out. Only samples from severely affected cases are submitted to central laboratory for the subsequent diagnostic testing for the presence of *Vt*, and thus, only these samples stand for confirmed cases. Incidence rate is the number of cases of VTEC divided over 100 000 population.

VTEC can never be completely eliminated from animal herds, and therefore, enormous efforts have been put into improving the knowledge of effective strategies of decreasing the transmission to humans. The aim of this study was to explore the prevalence and look for associations by statistically analysing the data available from the monitoring programme of VTEC O157 in ruminants in Slovenia.

Material and methods

Data on animals were provided by the Veterinary Administration of the Republic of Slovenia (VARS) and were part of a nationwide study aiming to investigate the baseline prevalence of VTEC O157 in animals and meat intended for human consumption. The surveillance data comprised 1720 records taken from cattle faeces (n=818), sheep faeces (n=320) and cuts of cattle meat (n=582). Sampling took place from January 2006 to December 2009 at slaughterhouses (faecal samples) and at cutting plants (meat cuts) across Slovenia (n=86). These establishments are approved and under official control of VARS. Most of the tested animals (faecal samples) originated from cattle reared within the Republic of Slovenia. Meat cuts sampled in this study were essentially from above mentioned slaughterhouses with a small number of meat cuts from other Member States. The number of samples collected from each slaughterhouse / cutting plant was proportional to its throughput. To prevent clustering of samples from the same herds, samplers were instructed that no more than one sample should be obtained from any one farm of origin on the same day. Moreover, complete records were kept on the origin of sampled animals, and additional attention was given to sampling different batches of meat. Samplers were official veterinarians, who were trained prior to sampling. Sampling of animal faeces included the cutting of a part of the caecum, including the adjacent intestinal wall, and took place immediately after the evisceration. Cuts of meat were sampled from different batches on the premises of cutting plants.

Laboratory diagnostics was carried out by the National Veterinary Institute (NVI), using the ISO 16654:2001 method for detection of VTEC O157. The date of sampling was used for the purposes of analysing the effect of season. Data were subjected to statistical analysis using the statistical software SPSS, Version 18. 0 (SPSS Inc., Chicago, Illinois). Microsoft Office Excel 2003 (Microsoft, United States) was used for calculations of proportions and rates. The variables of interest used from the dataset are the result of testing for VTEC O157, date (season) when sample was taken, age of the animal, type of the sample (cattle faeces/sheep faeces/cattle meat), and location of slaughterhouse by region in Slovenia.

First, different types of samples positive for VTEC O157 were compared with relation to animal age, season and region of sampling. χ^2 test was used to examine association between positive samples and season, and positive samples and region. Logistic regression was used to examine overall association of season with the outcome of testing positive for VTEC. Additionally, logistic regression was used in cattle subset for examination of association with season and age, respectively. Since at least 10 events per variable were required to proceed to logistic regression and vast majority of positive results were found in cattle samples (n=25), the cattle dataset was used to seek for association between age and VTEC. In winter, there were no positive samples taken from cattle. Where this is not a problem to calculate the odds ratio, it does create problems for logistic regression. Therefore, this season needed to be merged with one of the neighbouring seasons. Hence, winter was merged with spring, both being the colder parts of the year. This was then used as a reference group in logistic regression. The age trend in cattle was examined using categorisation which would allow equally sized groups. Despite the fact that such categorisation offers little practical information, categorisation was extended to ten equally sized age bands.

Ethical approval was granted by the Ethical Committee (Centre for Population Health Sciences, University of Edinburgh). Data of human infections were anonymous and partially obtained from already published sources, and steps to protect the privacy of breeders were taken as well.

Data on human infection with VTEC were obtained from reports on national surveillance of communicable diseases (31, 33) and through personal communication with experts in this field (IPH). Data on human cases were provided for the timeframe 2006-8, whilst notification rates were obtained from published sources and dated since 1997 (31, 33). Notification/incidence rates per 100 000 population were calculated by dividing the number of notifications/cases in each year with midyear census data (34). Specific information on different serotypes of VTEC isolated from patients, age, gender and season distribution of cases were provided by public health professionals from the *Institute of Public Health of the Republic of Slovenia (IPH)*.

Results

Data obtained in slaughterhouses/ meat cutting plants

a) Prevalence of VTEC O157

Data from national monitoring system revealed that there was a statistically significant difference in prevalence of VTEC O157 between types of sample (χ^2 =18.40, d.f.=2, p=0.001). The Table 1 shows that the prevalence was the highest in cattle and lowest in meat. The odds of VTEC O157 outcome were significantly higher in cattle as compared to meat (p=0.004, OR 2.48 to 135.56), but not for sheep relative to meat (p=0.141, OR 0.57 to 53.08).

Table 1: Results on VTEC O157 from different types of samples

Type of sample	N tested	n positive for VTEC O157	Prevalence (%)	95 % CI
cattle	818	25	3.1	1.9% - 4.2%
sheep	320	3	0.9	0.2% - 2.7%
meat	582	1	0.2	0% - 1%
TOTAL	**1720**	**29**	**1.7**	**1.2% - 2.4%**

b) VTEC O157 by season

Table 2 shows that the highest prevalence of VTEC O157 is in autumn (Sep-Nov). There was a statistically significant association between a positive VTEC O157 result and the season (p=0.035), with winter (Dec-Feb) as the reference group. The odds of VTEC O157 were significantly higher in autumn as compared to winter (OR 1.63 to 94.97), but not for spring and summer as compared to winter (Table 2).

Table 2: Association of VTEC 0157 with season of sample (n=1720)

Variable	N samples	n positive VTEC O157	p-value	OR (95 % CI)
Season			0.035	
Dec-Feb	381	1		Reference
Mar-May	490	6	0.152	4.71 (0.57 to 39.30)
Jun-Aug	407	8	0.056	7.62 (0.95 to 61.21)
Sep-Nov	442	14	0.015	12.43 (1.63 to 94.97)
Constant				0.003

c) VTEC O157 by age and region

In sheep, positive animals were aged 3 months, 9 months and one missing age value. The animal, whose meat tested positive, was 24 months of age. The remaining positives were found in the cattle subset, with two missing values.

Sampling took place within 10 VARS Regional Offices (each covering a respective regional unit). Region of sampling was not significantly associated with the outcome (χ^2 =12.67, d.f.=9, p=0.178).

d) VTEC in cattle by season and age

In autumn there was a significant increase in odds of cattle testing positive for VTEC O157 as compared to winter (Table 3). Percentage of cattle testing positive varies nearly 4-fold across age bands, but there is no discernible age trend (Figure1).

Table 3: Association between VTEC and season of sampling in cattle (n=818)

Variable	N	p-value	OR (95 % CI)
Season		0.021	
Dec-May	408		Reference
Jun-Aug	202	0.119	2.41 (0.80 to 7.25)
Sep-Nov	208	0.005	4.10 (1.52 to 11.09)
Constant			0.015

f) Virulence factors

Vast majority of VTEC O157 strains isolated from animals/meat (n=29) contained key virulence markers, namely, the *eae gene* (93%) for bacterial adherence and *Vt2* (86%), confirmed as being potentially pathogenic to humans (Mičunovič, personal communications).

Data on human infection

a) Notified cases

Between 1997 and 2008 there were 680 notifications of EHEC (30), however, these do not stand for laboratory-confirmed cases of VTEC.

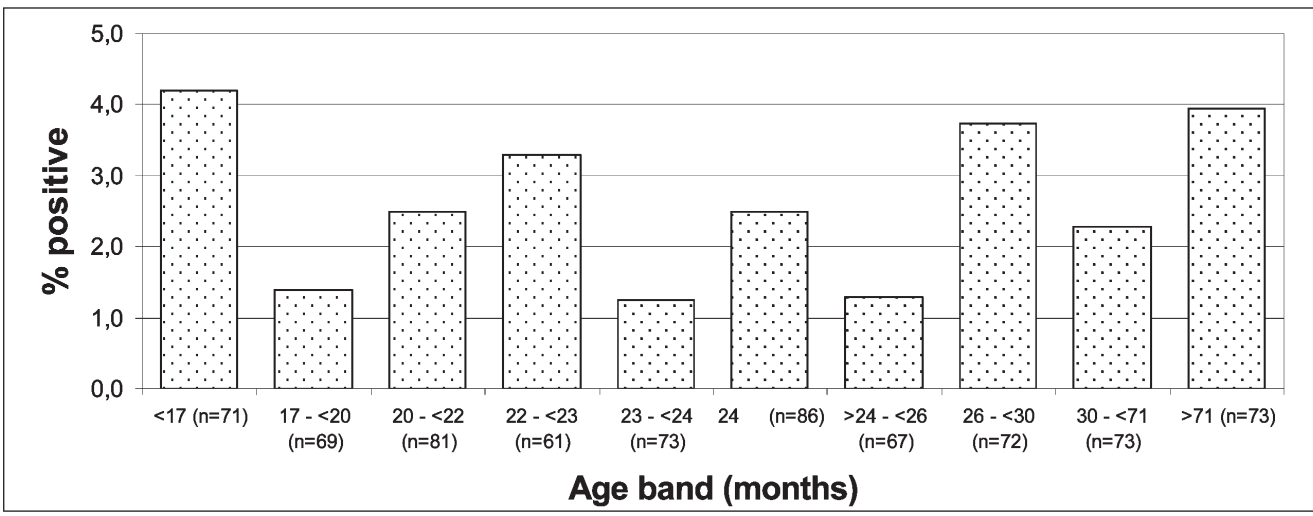

Figure 1: Percentage of cattle testing positive for VTEC O157, (n=726) with n=23 positive and n=2 missing values

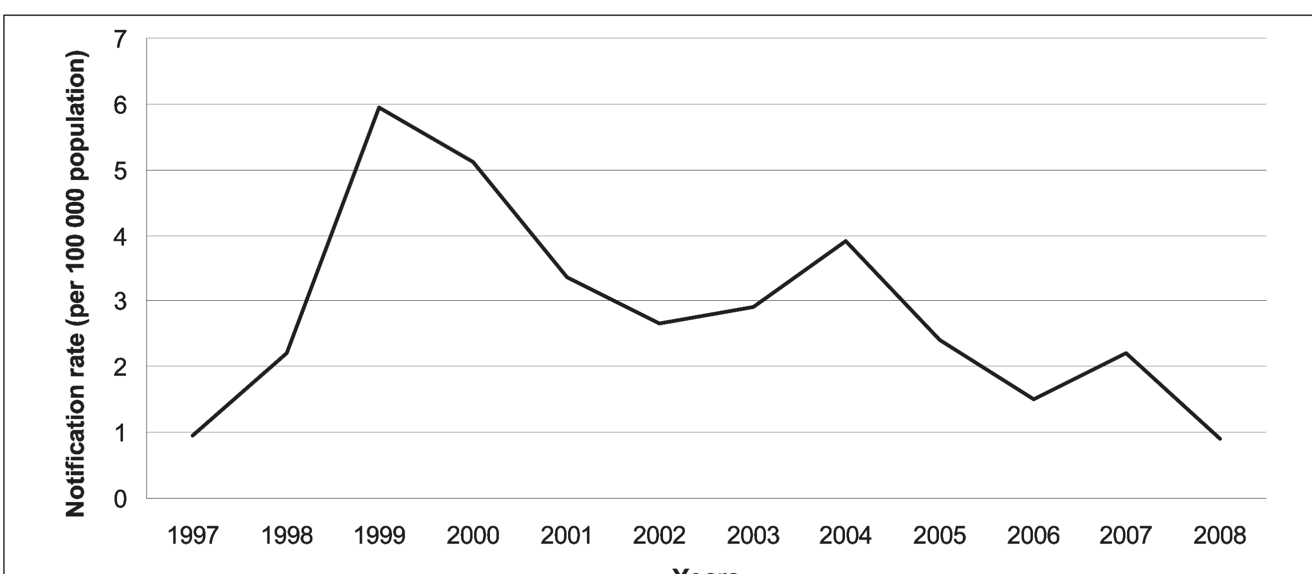

Figure 2: Notification rates of EHEC in Slovenia from 1997 to 2008 (n=680)

b) *Confirmed cases*

Data on patients infected with VTEC are provided from 2006 to 2008, with 93 notifications of EHEC, 15 cases of which were confirmed as VTEC (33, Trkov, personal communications). Majority of cases were children of up to 7 years of age, who comprise 67% of all cases. Most commonly reported gender was male (53%). More than half of the cases occurred in autumn (Sep-Nov) (53%), followed by summer (27%). Incidence rates of VTEC cases per 100 000 population are given in Table 4. Each year, approximately 10% of samples submitted for further confirmation to the laboratory carrying out molecular testing were confirmed positive for VTEC. These isolates have the ability of producing *Vt* or containing relevant genes. Serogroups isolated from patients were O26 (40%), O157 (27%), O103 (20%), O111 (6%), and not defined (6%).

Table 4: Incidence rate for VTEC cases in Slovenia, 2006-8. Data provided with the courtesy of Dr. Trkov from IPH

Incidence rate of VTEC cases per 100 000 population		
2006	2007	2008
0.2	0.2	0.4

Discussion

These findings suggest that the prevalence of VTEC O157 in sampled ruminants in Slovenia is low. Prevalence in cattle is consistent with similar research conducted elsewhere (12, 14) and with reports from the EU Member States, where prevalence in cattle ranges from 0 % to 7.2 % (29). Sheep showed lower prevalence, of 0.9% (95% CI 0.2% to 2.7%), corresponding the results of two larger studies (12,14). Meat sampled from cattle showed extremely low prevalence, of 0.2% (95% CI 0% to 1%), similar to samples from processing plants and retail (18, 19, 28). Similarly, within the EU Member States, prevalence in meat seems lower as compared to faeces, and ranges between 0% and 1.2 % (30).

The presence of VTEC O157 in cattle excreta can be influenced by the age of the animals; calves of up to four months of age seem to be the key source of infection (9). Others (14) argue that more advanced age is protective against testing positive for VTEC O157. Statistical analysis presented above did not reveal any significant association between cattle age and positive outcome.

Climate plays an important role in infection (2). Summer season association with higher prevalence in cattle has been reported (12, 14, 15, 16, 17). The seasonal effect was statistically significant, with increased odds of testing positive in autumn. This effect of season can further be explained by the fact that autumn temperatures in Slovenia (with the exception of September 2007) during the timeframe of this study were above the average (35). On the other hand, winters were among the coldest, which constitutes a possible explanation of a low number of positive results in this season.

Analyses of surveillance data of human infection with VTEC showed a relatively low incidence rate, ranging from 0.2 to 0.4 cases per 100 000 population, depending on the year. Children are most affected from all the groups. Similarly to animals, infection follows a similar seasonal pattern, i.e. an increased rate in autumn. The serogroups isolated from humans (in order of effect) were O26, O157, O103 and O111. Incidence rate is neither a rate nor a proportion; here, it was used to describe the number of new events of diarrhoea in a defined population within a specified period of time. This was a cross-sectional study, where the prevalence in animals was used as a measure of VTEC O157 occurrence. Thus, it is a point at issue, whether the rate was erroneously compared with proportion. Nonetheless, the incidence of enteric disease is rather frequently close to prevalence, as the disease is of short duration.

The data as presented above indicate that, as compared to meat, cattle are more likely to be a source of VTEC O157 infection. Thus, the emerging routes of transmission should be taken into consideration, in particular the contact with animals during farm visits, environment-related exposure (application of manure to the soil), and contaminated vegetables. It has been suggested that exposure to rural environment can have a protecting role (2). Comparably, the incidence rate in Slovenia is rather low, which is possibly due to the fact that a majority of Slovenian population lives in rural areas (36) and is thus more frequently exposed. Further research combining the veterinary and public health expertises will be most advantageous.

Further improvements in monitoring/surveillance systems

Reliable data from surveillance system are necessary for critical assessment. Limitations of this study include lack of available data, which resulted in unequal time span of animal data and human cases. Data from communicable disease surveillance systems provide information with limited analytical meaning. Animals are only tested for O157 VTEC, which accounts for one of the crucial gaps in zoonoses monitoring. There could be considerable underestimation of the true prevalence of different VTEC serogroups in animals than if the important pathogen were examined from a broader perspective. For further research on sources of infection/ transmission of VTEC in Slovenia, animal testing should necessarily be extended to non-O157 VTEC serogroups.

The above findings demonstrate that 67 % of serogroups isolated from humans belonged to non-O157 VTEC. Report of fatal case of HUS following infection with O145 VTEC in Slovenia (32) points towards extending the testing of animals as potential source of infection to this serogroup. The extended testing of animals to non-O157 VTEC serogroup can potentially lead to a better estimate of current state of VTEC in animals and consequently in the foodchain. In the final phase of drafting this Master's thesis it was brought to the authors' attention that the zoonoses monitoring programme that started in the beginning of 2010 already incorporated the testing for the above recommended serogroups. These baseline data on extended serogroups will be exceedingly valuable for the next evaluation, where the impact of animal isolates will be assessed in the light of those found in humans.

Slovenia needs an upgraded surveillance system for identifying the VTEC cases. This may be achieved through comprehensive testing of all patients suspected of infection with VTEC. Here, the availability of laboratory facilities and resources plays a pivotal role. It may well be that persons living near laboratories (in cities) are more likely to be tested and therefore included in the surveillance. It has been specified by the EU that reporting communicable diseases at the EU level should comprise the confirmed cases only, which includes any person meeting clinical and laboratory criteria (37). Another weakness in the Slovenian surveillance system is the non-exhaustive detection of VTEC infection in humans, where milder cases go unreported. In many other countries, HUS has been shown a reliable indicator of VTEC infection in the human population. For the time being, HUS that accompanies the severely affected cases is not a compulsorily notifiable disease within the Slovenian public health system. By establishing a routine HUS surveillance system, the critically affected cases would be identified as well. Capturing these cases could potentially lead to comprehensive and prompt research of underlying sources of infection. Taking into consideration all of the above, the surveillance of communicable diseases in Slovenia needs to be upgraded in order to provide a most comprehensive survey of the current state, thus facilitating a most efficient treatment of gastrointestinal diseases.

Conclusion

Despite a low prevalence it has been found that cattle and sheep in Slovenia do carry VTEC O157. As regards the Slovenian climate, data suggest that the warmer seasons stimulate the presence of VTEC O157 in animals and thus, as a possible consequence, increasing the number of human cases. Hence, specific attention should be paid to human cases during the warmer months of the year, establishing the possible interlinks with animals by thorough investigation.

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References

1. Nataro JP, Kaper JB. Diarrheagenic *Escherichia coli*. Clin Microbiol Rev 1998; 11(1):142-201.

2. Beutin L. Emerging enterohaemorrhagic *Escherichia coli*, causes and effects of the rise of a human pathogen. J Vet Med B 2006; 53(7): 299-305.

3. Tarr PI, Gordon CA, Chandler WL. Shiga-toxin-producing *Escherichia coli* and haemolytic uraemic syndrome. Lancet 2005: 365(9464): 1073-86.

4. Mainil JG, Daube G. Verotoxigenic *Escherichia coli* from animals, humans and foods: who's who?' J Appl Microbiol 2005; 98(6):1332-44.

5. Karmali MA, Gannon V, Sargeant JM. Verocytotoxin-producing *Escherichia coli* (VTEC). Vet Microbiol 2010; 140: 360-70.

6. Griffin PM, Tauxe RV. The epidemiology of infections caused by *Escherichia coli* O157:H7, other enterohemorrhagic *E. coli*, and the associ-

ated haemolytic uremic syndrome. Epidemiol Rev 1991;13: 60-98.

7. Pearce MC, Topping ME, McKendrick IJ, et al. Temporal and spatial patterns of bovine *Escherichia coli* O157 prevalence and comparison of temporal changes in the patterns of phage types associated with bovine shedding and human *E. coli* O157 cases in Scotland between 1998-2000 and 2002-2004. BMC Microbiol 2009; 9: 276.

8. Hussein HS. Prevalence and pathogenicity of Shiga toxin-producing *Escherichia coli* in beef cattle and their products. J Anim Sci 2007; 85(Suppl.13): E63-72.

9. Topping ME, McKendrick IJ, Pearce MC, et al. Risk factors for the presence of high-level shedders of *Escherichia coli* O157 on Scottish farms', J Clin Microbiol 2007: 45(5):1594-603.

10. Caprioli A, Morabito S, Brug re H, Oswald E. Enterohaemorrhagic *Escherichia coli*: emerging issues on virulence and modes of transmission. Vet Res 2005; 36(3): 289-311.

11. Matthews L, McKendrick IJ, Ternent H, et al. Super-shedding cattle and the transmission dynamics of Escherichia coli O157. Epidemiol Infect 2006; 134(1):131-42.

12. Paiba GA, Gibbens JC, Pascoe SJ, et al. Faecal carriage of verocytotoxin-producing *Escherichia coli* 0157 in cattle and sheep at slaughter in Great Britain. Vet Rec 2002; 150: 593-8.

13. Aspán A, Eriksson E. Verotoxigenic *Escherichia coli* O157 : H7 from Swedish cattle; isolates from prevalence studies versus strains linked to human infections: a retrospective study. BMC Vet Res 2010; 6(7): e2-11.

14. Milnes AS, Stewart I, Clifton-Hadley FA, et al. Intestinal carriage of verocytotoxigenic *Escherichia coli* O157, Salmonella, thermophylic Campylobacter and *Yersinia enterocolitica*, in cattle, sheep and pigs at slaughter in Great Britain during 2003. Epidemiol Infect 2008:136(6):739-51.

15. Albihn BA, Eriksson E, Wallen C, Aspán A. Verotoxinogenic *Escherichia coli* (VTEC) O157:H7: a nationwide Swedish survey of bovine faeces. Acta Vet Scand 2003; 44(1): 43-52.

16. Heuvelink AE, van den Biggelaar FL, de Boer E, Herbes RG, et al. Isolation and characterization of verocytotoxin-producing *Escherichia coli* O157 strains from Dutch cattle and sheep. J Clin Microbiol 1998; 36(4): 878-82.

17. Rhoades JR, Duffy G, Koutsoumanis K. Prevalence and concentration of verocytotoxigenic *Escherichia coli*, *Salmonella enterica* and *Listeria monocytogenes* in the beef production chain: a review. Food Microbiol 2009; 26(4): 357-76.

18. Coia JE, Johnston Y, Steers NJ, Hanson MF. A survey of *Escherichia coli* O157 in raw meats, raw cow's milk and raw cheeses in south-east Scotland. Int J Food Microbiol 2001; 66: 63-9.

19. Conedera G, Dalvit P, Martini M, et al. Verocytotoxin-producing *Escherichia coli* O157 in minced beef and dairy products in Italy. Int J Food Microbiol 2004; 96: 67-73.

20. Riley LW, Remis RS, Helgerson SD, McGee HB, et al. Hemorrhagic colitis associated with a rare *Escherichia coli* serotype. N Engl J Med 1983; 308(12): 681-5.

21. Mead PS, Griffin PM. *Escherichia coli* O157:H7. Lancet 1998; 352(9135):1207-12.

22. Scharff RL. Health related costs from foodborne illness in United States. Produce Safety Project at Georgetown University, 2010.

http://www.producesafetyproject.org/admin/assets/files/Health-Related-Foodborne-Illness-Costs-Report.pdf-1.pdf (10.7.2010).

23. Centers for disease control and prevention CDC. Investigation update: multistate outbreak of human *E. coli* O145 infections linked to shredded romaine lettuce from a single processing facility. Atlanta: Department of health and human service. (Updated May 21, 2010) http://www.cdc.gov/ecoli/2010/ecoli_o145/index.html (10.7.2010).

24. Forsythe SJ. The microbiology of safe food. Blackwell Science, 2002.

25. Schrijver KD, Buvens G, Possé B, Branden DV, Oosterlynck O. Outbreak of verocytotoxin-producing *E. coli* O145 and O26 infections associated with the consumption of ice cream produced at a farm, Belgium, 2007. Euro Surveill 2008;13: 61-4.

26. Rangel JM, Sparling PH, Crowe C, Griffin PM, Swerdlow DL. Epidemiology of *Escherichia coli* O157:H7 outbreaks, United States,1982-2002', Emerg Infect Dis 2005:11(4): 603-9.

27. Strachan NJ, Dunn GM, Locking ME, Reid TM, Ogden ID. *Escherichia coli* O157: Burger bug or environmental pathogen? Int J Food Microbiol 2006; 112: 129-37.

28. Solecki O, Macrae M, Ogden I, Strachan N. Can the high levels of human verocytotoxigenic *Escherichia coli* O157 infection in rural areas of NE Scotland be explained by consumption of contaminated meat? J App Microbiol 2007; 103: 2616-21.

29. Locking M, Allison L, Rae L, Pollock K, Hanson M. VTEC in Scotland 2004: enhanced surveil-

lance and reference laboratory data. HPS Weekly Rep 2006; 39: 290-5.

30. European Food Safety Authority. Trends and Sources of Zoonoses and Zoonotic Agents in the European Union in 2008', EFSA 2010, Parma, Italy.

31. Institute of Public Health of the Republic of Slovenia. Annual communicable disease reports, Year 2006. Ljubljana: Institute of Public Health of the Republic of Slovenia, 2006. http://ivz.arhiv.over.net/index.php?akcija=novica&n=798 (10.4.2010).

32. Kraigher A, Seme K, Lah AK, Fisher I. Fatal case of HUS after VTEC *E. coli* O145 infection in Slovenia highlights importance of testing this rare strain. Eurosurv 2005; 10:37 (2). http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=2792 (10.5.2010)

33. Institute of Public Health of the Republic of Slovenia. Annual communicable disease reports, Year 2008. Ljubljana: Institute of Public Health of the Republic of Slovenia, 2008. http://ivz.arhiv.over.net/index.php?akcija=novica&n=798 (10.4.2010).

34. Statistical Office of the Republic of Slovenia. Yearbooks 1996-2008. 2010. http://www.stat.si/publikacije/pub_letopis_prva.asp (25.7.2010)

35. Environmental Agency of the Republic of Slovenia. Klimatske značilnosti leta 2006, 2007, 2008 / Climatological Characteristics of 2006, 2007, 2008. Meteorološki letopis 2010 / Meteorological Yearbook of 2010.

http://www.arso.gov.si/vreme/podnebje/meteorolo%C5%A1ki%20letopis/meteoroloski_letopisi.htm (11.7.2010).

36. Statistical Office of the Republic of Slovenia. Upravno podeželje / Administrative Rural Territory. Ljubljana: SURS, 2010. http://www.stat.si/tema_splosno_upravno_podezelje_predstavitev.asp (25.7.2010).

37. European Commission 2002/253/EC: Commission Decision of 19 March 2002 laying down case definitions for reporting communicable diseases to the Community network under Decision No 2119/98/EC of the European Parliament and of the Council, EC 2002, Brussels.

VPLIV VEROTOKSIČNE *E. COLI* PRI ŽIVALIH NA ZDRAVJE SLOVENSKEGA PREBIVALSTVA

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Povzetek: Okužba z verotoksično *Escherichio coli* (VTEC) je pomemben problem v javnem veterinarskem zdravstvu. Prežvekovalci so naravni rezervoar okužbe, kjer je okužba z uživanjem mesa ena od morebitnih poti prenosa. Veliko truda in sredstev je bilo vloženih v programe spremljanja prevalence VTEC pri prežvekovalcih v Sloveniji. Namen te študije je bil podati statistično analizo podatkov, zbranih v okviru nacionalnega sistema monitoringa. V ta namen je bila izračunana prevalenca VTEC O157 in testirane so bile statistične povezave med spremenljivkami (chi kvadrat, logistična regresija), prav tako je bila navedena primerjava z okužbo pri ljudeh. Ugotovitve te analize so pokazale področja, kjer bi se sistem spremljanja lahko izboljšal. Zbrani podatki pri živalih so del nacionalnega sistema spremljanja VTEC O157, kjer so bili v obdobju 2006-09 odvzeti vzorci iztrebkov goveda (n=818) in iztrebkov ovac (n=320) v klavnicah ter mesa goveda (n=582), odvzetega v razsekovalnicah po Sloveniji.

Podatki o okužbi z VTEC pri ljudeh so navedeni za obdobje 2006-08 in vključujejo podatke o starostni skupini in spolu obolelih, sezoni ugotovitve okužbe ter serotipu VTEC. Prevalenca VTEC O157 pri govedu je bila 3,1 % (95 % CI 1,9 % do 4,2 %), ovcah 0,9 % (95 % CI 0,2 % do 2,7 %) in v mesu goveda 0,2 % (95 % CI 0 % do 1 %). Starost goveda ni bila statistično značilno povezana z rezultatom, pozitivnim na prisotnost VTEC O157. Nasprotno pa je bil letni čas statistično značilno povezan z vrsto vzorca (p=0,035), saj je bila večina pozitivnih rezultatov zaznana v jeseni. Od skupno 93 ugotovitev okužbe z E. coli pri ljudeh je bilo le v 15 primerih potrjeno, da gre za VTEC (16,1 %). Izračunana stopnja incidence pri slovenskem prebivalstvu je bila nizka (0,2, 0,2 in 0,4 primerov na 100 000 prebivalcev v letu 2006, 2007 in 2008). Najpogosteje zbolijo otroci. Jesen je tisti letni čas, ko je več rezultatov pozitivnih na prisotnost VTEC, tako pri ljudeh kot tudi pri živalih. Medtem ko so bile živali testirane zgolj na prisotnost serološke skupine O157, so najpogostejše serološke skupine VTEC pri ljudeh O26, O157, O103 in O111. Dokazano je, da govedo nosi povzročitelja v prebavnem traktu, vendar je prevalenca VTEC v mesu nizka. Pot prenosa za okužbo ljudi v Sloveniji ostaja nerazjasnjena, saj je okužba prek živil le ena izmed možnosti prenosa. Nove porajajoče se poti okužbe z VTEC lahko kažejo na prenos povzročitelja iz okolja.

Ključne besede: zoonoza; verotoksična *E. coli* (VTEC); prežvekovalci; javno veterinarsko zdravstvo; Slovenija