

VIRAL CONTAMINATION IN MUSSEL PRODUCTION CHAIN ON THE SLOVENIAN COASTLINE

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Summary: Noroviruses (NoVs) and Hepatitis A virus (HAV) are significant agents of foodborne human viral illness worldwide, both as agents of greatest concern from the consumption of shellfish. In the framework of official national monitoring shellfish samples have been collected since 2013 to determine the spread of NoVs and HAV. Contamination of shellfish samples with NoVs varied from 25% in year 2014 to 40% in year 2015. HAV was not found in any of the analyzed samples, which correlates with the low number of human infections in Slovenia. Alongside official surveillance, semi-structured interviews were carried out with local shellfish farmers regarding this subject. The qualitative analysis highlighted that Slovenian shellfish farmers are aware of food safety hazards, especially associated with hazards to primary production, but only in the context of *Escherichia coli* as an indicator microorganism, and not in the concept of viral food safety. Despite detected foodborne viruses in shellfish on the Slovenian market, local farmers are not aware of or do not recognize foodborne viruses as distinctive food safety hazards. It can be concluded that local farmers possess knowledge and information of critical points in the mussels food supply chain connected to food safety in general. However, in particular, they are not familiar with viruses that represent barrier and consequently critical point to food safety management practices. Training and education on all important aspects of viral food safety according to the current recommendations is strongly recommended for all actors within the shellfish supply chain.

Key words: mussel farmer; Slovenia; official control; food safety; foodborne viruses; semi-structured personal interviews

Introduction

Mariculture is a traditional activity in the Primorska region. Mediterranean mussels (*Mytilus galloprovincialis*) are the main harvested species and, of a smaller quantity, also warty venus (*Venus verrucosa*). Mussel farming takes place in a standard manner in lines of floating buoys linked together, with longline nets hung from them. In Slovenia, within 46.6 km of coastline there are three official harvesting areas of Mediterranean

mussels: Seča, Strunjan, and Debeli rtič, with 56 registered shellfish farmers on a total area of 46 ha. In 2013, 231 persons were involved in aquaculture activities in Slovenia, and only 39 persons were involved in marine fish and shellfish farming. The aquaculture sector in Slovenia is mainly characterized by small self-employed family farms, most of which have one employee, and some are assisted by unpaid family members. Total shellfish production in 2013 was 311 tonnes, and current production covers mainly the needs of the domestic market. The major cultured shellfish species, Mediterranean mussel, accounts

for 83 percent of total mariculture production in Slovenia (1, 2). Next to that there were about 153 tons of imports of mussels in Slovenia but only 23 tons of export, what is a negligible quantity in alimentation compared to consumption of fresh fruit and vegetables, which are also recognized as vulnerable to viral contamination. Yet, the level of shellfish consumption by country is very different. In some countries, the mussel consumption per capita is over 3 kg per year, while it is not even part of the local diet in others (3). Per capita consumption of fresh fruit in Slovenia was in 2013 75 kg and 73 kg of fresh vegetables. However, there were no contaminated samples with NoVs or HAV of fresh produce found within Slovenian national monitoring (27). Shellfish specialties are commonly part of the culinary and gastronomic specialties only along the Slovene coast and are generally prepared and served in restaurants.

The filter-feeding nature of bivalves and the traditional way of consuming them (often raw or slightly cooked) make shellfish one of the most common vehicles of viral foodborne illness. Shellfish are filter-feeding animals, which ingest and accumulate human pathogens (5). Lees (6) reported that shellfish grown in sewage-polluted waters tend to bio-accumulate environmentally stable enteric viruses. Processing interventions such as depuration do not eliminate viral particles (7), and food consumption practices increase the health risk related to shellfish consumption. The increasing amount of data on virus detection in shellfish (8, 9, 10) and shellfish-borne viral outbreaks (11) indicates the necessity of a constant surveillance system in European countries, including Slovenia (12). The management of the harvesting areas continues to rely uniquely on bacterial standards, such as *Escherichia coli*, despite the proven fact of being unreliable tools to indicate the viral presence in harvesting areas or to control the efficiency of the process, such as depuration (13, 14). From a virological point of view, shellfish safety continues to be a sanitation challenge and to protect consumers the EU strives towards establishing legislation on this matter.

With better diagnostic technology and investigative epidemiology, it is now accepted that enteric viruses are major contributors to foodborne disease as well. Enteric viruses are transmitted through contaminated food, but also in combination with person-to-person contact or through environmental contamination. They

have been increasingly recognized a significant cause of foodborne disease, despite the measures already in place, mainly targeted at reducing bacterial contamination, because of the increasing consumption of ready-to-eat foods, raw and/or minimally processed shellfish, fruits, and vegetables. This is because products are often imported from areas lacking strict hygienic measures, they are often eaten uncooked, and they often come into contact with potentially contaminated animal manure, water, ice, human hands and surfaces from the “farm-to-table” continuum (10). Most foodborne viruses are more resistant (15) than bacteria to commonly used control measures, (e.g. refrigeration, freezing, pH, drying, UV radiation, heat, pressure, disinfection, etc.). There are currently no effective, realistic and validated risk management options to eliminate viral contamination prior to consumption without changing the normally desired characteristics of the food. Because of concerns about virus persistence during food processing, effective control strategies need to focus on the prevention of contamination. From the limited available information, foodborne viruses have a low infectious dose and are dispersed in stool or emesis in high numbers. Only a few viral/infectious particles are needed to cause an infection that may lead to illness (10, 15, 17).

Shellfish aquaculture is a marine-based industry that is affected by other land users such as tourism, recreation, forestry, agriculture, and urban development. In many cases, the public is unaware of the detrimental impact their activities have on the aquaculture sector and, consequently, also on shellfish food safety. Food safety embraces the absence or acceptable and safe levels of contaminants, adulterants, naturally occurring toxins or any other substances that can make food dangerous to human health. Microbial food safety is considered a significant public health issue but historically has focused mostly on the control of bacterial contamination; however, enteric viruses have been increasingly recognized as an important cause of foodborne disease, and control measures are being developed (16, 17). The food supply chain from stable to table includes activities such as production, processing, distribution, retail, packaging and labeling of foodstuffs, which are governed by a mass of laws, regulations, codes of practice and guidance. Nowadays, the distance that food travels from producer to consumer has increased as a result of globalization in the food

trade. Moving these food products safely and efficiently from farm to fork requires a highly coordinated series of links in a long chain of trading partners. Food miles, as a term that refers to the distance food is transported from where it is grown or raised to where it is purchased by a consumer, is part of the broader issue of sustainability that deals with a large range of environmental, social and economic issues. Therefore, keeping safety and quality along the food supply chain has become a significant challenge, whereas good traceability systems, defined as the ability to trace and follow a food, feed, food-producing animal or substance intended to be, or expected to be incorporated into a food or feed, through all stages of production, processing and distribution (18), help to minimize the production and distribution of unsafe or poor quality products.

Epidemiology of foodborne viruses

Although shellfish consumption can contribute to a healthy diet they are often associated with outbreaks of foodborne disease. Viral foodborne outbreaks associated with shellfish consumption have occurred in many countries (11) despite existing strategies to prevent contamination. They are often attributed to water contamination by sewage and/or during processing and serving. According to epidemiological evidence, NoVs as the predominant agents of nonbacterial gastroenteritis in humans along with Hepatitis A virus (HAV), both as agents of greatest concern from the consumption of shellfish, are important agents of foodborne human viral illness worldwide (17, 19, 20, 21, 22, 23).

Of the approximately 600 million cases of illness caused by foodborne hazards in 2010 worldwide, infectious agents that cause diarrheal diseases accounted for the vast majority (550 million), in particular noroviruses (120 million cases) and Hepatitis A virus 14 million cases (24).

A total of 5251 foodborne outbreaks were reported in 2014 in the EU (20) within the framework of member states' national monitoring. In 2014, food-borne viruses were, for the first time, identified as the most commonly detected causative agent in the reported food-borne outbreaks. 1070 food-borne outbreaks caused by viruses were reported in 2014, implicated 11740 cases, 2486 hospitalizations and 2 deaths. In

strong-evidence outbreaks caused by viruses, 'crustaceans, shellfish, mollusks and products thereof' was the most commonly implicated food vehicle (44.7% of outbreaks), followed by 'buffet meals' (15.8% of outbreaks), 'mixed food' (13.2%) and 'fruit' and 'berries and juices' (both 5.3%). The place of exposure most frequently reported was 'restaurant, café, pub, bar, hotel', followed by the household. Norovirus was the most commonly reported virus implicated in the strong-evidence outbreaks and accounted for 97.6% of cases.

National statistics on foodborne viral disease are not easily available and, where present, likely to reflect significant under-reporting (17), because there is a lack of systematic surveillance for foodborne viral disease (25). Considering the scientific opinion from the EFSA, RASFF notifications and results of official controls, since 2013 the Slovenian National Zoonoses Monitoring Programme has included food sampling for the presence of NoVs and HAV in live shellfish at the retail level and distribution of local and foreign origin, which are recognized as potentially zoonotic viruses (26). In Slovenia, the Zoonoses Monitoring Programme (27) has been conducted at the national level since 1985. It is designed for the systematic collection, monitoring, analysis and communication of data on the emergence of zoonosis, zoonotic agents, and related antimicrobial resistance and comprises the recently emerging zoonotic agents, including foodborne viruses. The ultimate purpose is to capture high-quality information about infections in humans as well as in animals and the contamination of foods, providing important information that is integrated across sectors. It should provide a fundamental basis for making public health decisions with actions for reducing the risks to public health, document the impact of an intervention, track progress towards specified goals, and elucidate the epidemiology of health problems.

The results obtained within Slovenian official national monitoring have shown that live shellfish, purchased at retail stores in Slovenia during independent sampling times throughout the year were contaminated with NoVs, but HAV was not found in any of the samples analyzed (Table 1), which correlates with the low number of human infections in Slovenia (Table 2). In studies, contamination of mussels' samples varies from 16.9% (9), to 34.4% NoVs in Italy (28), to 35.0% of contaminated mussels in France (29). Henigman et al. (8) reported

Table 1: Presence of NoVs and HAV in live shellfish samples within Slovenian official surveillance

Year	Virus	
	NoVs	HAV
2013	5/17 (29%)	0/15
2014*	3/12 (25%)	0/12
2015*	3/10 (40%)	0/10

*Preliminary results

Table 2: Reported infections caused by NoVs and HAV in humans in Slovenia from 2007–2014 (27)

N° of cases	Year							
	2007	2008	2009	2010	2011	2012	2013	2014*
NoVs	1094	1043	1393	2012	2231	1611	2146	1316
HAV	15	17	12	9	12	11	23	10

*Preliminary results

that mussels collected in Slovenian coastal waters were contaminated with NoVs, the highest at Debeli Rtič (25.9%), 21.2% in Strunjan, and only 8.1% in the Seča harvesting area. The difference in positive results is interlinked to the location of harvesting areas due to sea current, dense shipping, and the influx of streams and rivers.

Control and prevention of foodborne viruses

Virus contamination as a consequence of human handling can occur at any stage of food production, processing, and even preparation. At present, we are faced with insufficient knowledge and awareness of food safety issues among food handlers and accompanied by consumers being insufficiently informed about food safety principles in the home.

Today, we manage food safety through good practices at different levels within a food supply chain that can be described as a network of food-related businesses involved in the creation and consumption of food products that move from farm to table and are linked by information, material, and capital flows. Good practices are described in several different codes of practice designed by producers' organizations, importers and retailer's consortia and government bodies at different levels of production, processing and consumption within the food supply chain. All current active

practices are segregated along the food supply chain and are not connected to a comprehensive system, resulting in the existence of exposure to potential of food hazards, especially emerging hazards, such as viruses (12, 30).

The development in different areas within the food production chain and in particular in technological and technical means is moving very quickly. We encounter innovations in materials, and supporting measures almost daily. Consequently, the gap between knowledge and skills is widening. We are willing to accept the paradigm that drifting is the most dangerous challenge in analytical instruments. However, it is also extremely influential in technological practices. It occurs side by side with "industrial blindness", which develops as a personal characteristic of employees who do not see particular items although they are commonly present in routine operations.

Consumers play an important role in the transmission of hazards, including viruses. Implemented viral food safety guidelines (12) are not purposely designed for informing consumers, although studies in recent years have highlighted gaps in food safety knowledge and some critical safety violations regarding food handling at home (31, 32, 33, 34, 35, 36). Consumer behaviour and attitudes toward food safety have shown that the levels of understanding, motivation and trust need to be further cultivated, and their training and informing due to changes in lifestyle and food

consumption patterns encouraged (30, 37, 38, 39).

EFSA reported (20) that viral foodborne outbreaks most frequently occurred in ‘restaurant, café, pub, bar, hotel’, followed by the household. However, outbreaks of foodborne illness occurring in private homes are less likely to be reported than those in commercial and public premises, and it is believed that infections attributed to private homes are three times more frequent than those attributed to canteens (40).

Food handlers also play an important role in the transmission of enteric viruses in the shellfish supply chain (16, 20, 41, 42), especially because shellfish specialities are generally prepared and served in restaurants.

During production, harvest and packaging preparation, food can become contaminated with viruses by food handlers or after contact with virus-contaminated water and surfaces. A major contributor to the spread of disease in food production is poor hygiene practices or being in contact with faecal material or vomit (15). Food handlers are unaware of controls specific to enteric viruses (16). That is why training on all important aspects of NoVs and HAV according to the recently developed Codex Alimentarius guidelines to control viruses in food is strongly recommended. The primary purpose of the codex guidelines for the control of viruses in food is to give guidance on how to prevent or minimize the presence of human enteric viruses in food, especially NoVs and HAV, and to emphasize that management strategies regarding foodborne viruses and associated illnesses should be different from those for bacterial pathogens.

In 2011 Poklar Vatovec with co-workers (42) carried out the research to evaluate the offer of shellfish specialities in Slovene Istria restaurants and to assess food safety knowledge and behaviour of food handlers in preparing shellfish dishes. Results indicated poor food safety knowledge regardless the education of food handlers. The origin of shellfish is important in ensuring food safety; and restaurants should be convinced of good raw meat to exclude foodborne poisoning. Therefore, shellfish should be bought only at registered plants, since these are under official supervision. However they observed that shellfish were not always bought at registered plants, but supplied from the so called illegal »black market«. Next to that it was also observed that employed personnel were hardly acquainted with HACCP

principles which represent major food safety hazard. Cooking (at least 90 °C for at least 90 seconds) is a critical point for ensuring food safety. The survey showed that the mid temperature was measured by only 26.8% of the interviewees with formal education and 7.3% with informal education. The remaining did not perform this procedure or it was not known whether it was performed. The results of the survey demonstrated that only 4.9% of the interviewees, regardless their education, are familiar with the correct temperature for heat treatment of shellfish. Research also pointed out that food handlers employed in Slovene Istria restaurants have insufficient knowledge on storing temperatures, storing time and the adequate methods of storing shellfish.

Pilot study: Semi-structured interview with Slovenian mussel farmers

Pilot study illustration

In order to determine eventual connection between comprehension of viral food safety and the shellfish growing practices, the four semi-structured interviews were carried out with Slovenian mussel farmers. The interview guide covered the following topics:

- Factors responsible for food safety within the shellfish food supply chain,
- Conditions related to food safety hazards with an emphasis on foodborne viruses due to virus-commodity combination, which has been identified as one of the greatest public health concerns.

In this pilot study, an empirical grounding was important because an exploration of local farmers’ viral food safety perceptions and their good hygiene practices, together with results from the National Zoonoses Monitoring Programme, outlined and gave insight into the current situation in Slovenia. The semi-structured interviews were chosen due to the sensitivity and complexity of the subject discussed.

The semi-structured interview started with questions concerning food safety in general. The first open thematic question was: “Tell me as much as you can about the importance of the mussels production process, and about the factors that could affect its food safety.” Discussions continued with the questions: “Could you please

explain what food safety means from your point of view and when mussels are considered safe for a consumer?” and: “Have you ever heard about viruses that are transmitted by shellfish?” Follow-up questions were posed to complement and facilitate the dialogue. The discussions were concluded with the question: “Do you use working documents that have resulted from food safety legislation, like the HACCP plan, good practices that includes viruses?” The semi-structured interview ended with a question on whether the interviewee had something to add. Efforts were made to create trust, since issues of guilt and failure may easily arise. The results of national monitoring are supported by the responses recorded by the interviewers, which were clearly marked due to the assurance of anonymity. The letter “I” (I1) signifies “interview”, while the number represents a running number of interviews.

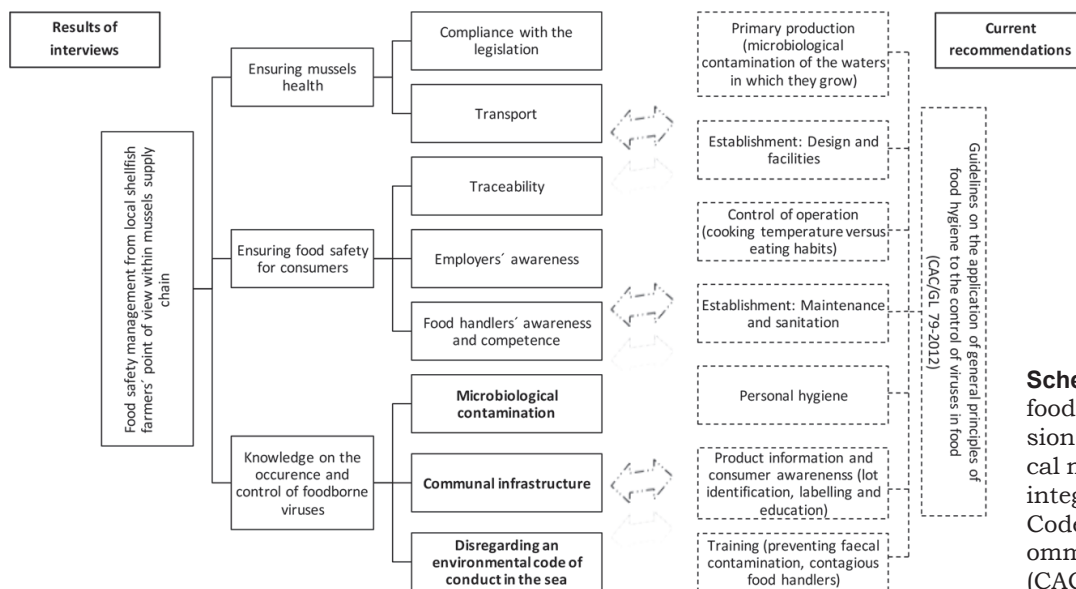
The semi-structured interviews with the local mussel farmers were qualitatively analyzed, using the grounded theory method. This theory produces theoretical models of individuals’ perspectives of a given phenomenon and the strategies they use to resolve or cope with the problem in a distinct and bounded context (43, 44, 45). Interviews were recorded using a Dictaphone and later transcribed. Transcripts of the interviews were analyzed by coding the statements of the respondents using identified notions. These notions were gathered into topic categories (44, 45, 46). The validity is justified by triangulation. Three researchers

with different disciplinary basic knowledge and different experiences in research were included in the analysis and interpretation process.

Findings from semi-structured interviews

Eight topics (Scheme 1) were identified following text analysis of the respondents’ answers during semi-structured interviews: compliance with the legislation, transport, traceability, employer’s awareness, food handlers’ awareness and competencies, microbiological contamination, communal infrastructure, and disregarding an environmental code of conduct in the sea. These topics were obtained after identifying the topics in the statements. The interviewee’s answers were very guarded; consequently, more comprehensive data processing cannot be achieved.

With the intention to show comprehension within interviewee reflections, these identified topics (Scheme 1) were integrated with a specified section in Guidelines for the control of viruses in food (15) as follows: Primary production/Harvesting area; Establishment: Design and facilities; Control of operation; Establishment: Maintenance and sanitation; Establishment: Personal hygiene; Product information and consumer awareness; and Training. Specific topics (microbiological contamination, communal infrastructure, and disregarding an environmental code of conduct in the sea) represent an unrecognized threat to



Scheme 1: Topics of viral food safety comprehension among Slovenian local mussel farmers and its integration within current Codex Alimentarius recommendations key points (CAC/GL 79-2012)

food safety in the mussels supply chain from viral point of view and are marked bold in the Scheme 1 due to their significance.

The identified topics are in accordance with the Codex Guidelines (15) sections, but not from the viral point of view, but in the context of bacterial contamination associated with hygiene practices. The results indicated that the respondents rarely comprehend viral food safety separately, but view it in different combinations with already obtained knowledge and skills. This can be demonstrated by the question “Are you familiar with viruses, which are transmitted by shellfish?”, which yielded no answers dealing with foodborne viruses. With other questions, dialogue was maintained, but replies always approached local farmers’ familiar topics in the field of food safety as ensuring cold chain management practices; microbiological contamination connected to biotoxins and *E. coli*, but *E. coli* in conjunction with gulls’ and cormorants’ excrement and as obligatory indicator microorganisms; and food handlers’ awareness. This aspect is most obvious in the following statement:

Citation III (I3) [...] A couple of years ago, biotoxins, now viruses [...] we depend on water, because mussels grow themselves [...] the biggest problem is the buoys [...] in summer every buoy is covered with gulls’ and cormorants’ faeces, and you are not able to see the color of the buoy [...] I was asking if this may be an *E.coli* reservoir, but they said no [...]

Local farmers link food safety to employers’ awareness and food handlers’ awareness and competencies, which often intersect and obstruct food safety system implementation. They associated food safety with compliance with legislation and regulations and transport practices, which can be illustrated by answers to the question “What is important for food safety within the mussels food supply chain?”. This aspect is the most obvious in the following statement:

Citation I (I1) [...] if anything goes wrong, it can be seen immediately due to inspection control and traceability issues, because we export all harvested mussels to wholesalers [...] Anyway, I would not sell mussels, which I would not give to my children to eat [...]

Citation II (I2) [...] I think that the problem is not only in growing conditions but also in the awareness of employees, especially in restaurants.

Respondents also pointed out the quality of

growing waters and its linkage to the communal infrastructure and sewage discharge, and failures to comply with hygiene practices on the sea was also observed. This aspect is seen in the statement:

Citation IV (I3) [...] heavy rainfall and storms may flush sewage overflow or farm run-off into the growing waters [...]

Recreational and economic activities in the sea were also pointed out as hazards:

Citation V (I4) [...] tourists are rascals and throw garbage and discharge sewage into the sea, even though they know that it is prohibited [...] next to that problems are large transport ships, which are regularly present in the area with and river estuary [...] we only exploit what nature offers to us, and the quality of growing waters is not solely under our responsibility.

This overview has clearly indicated that the development of new concepts is far from sufficient to enhance implementation in real practice. It is a fact that approximately one third of the live shellfish bought on the Slovenian market are contaminated with NoVs. The qualitative analysis alongside official surveillance highlighted that Slovenian mussel farmers are aware of food safety hazards in the mussel food supply chain connected to compliance with the legislation requirements. Despite detected foodborne viruses in samples bought on the Slovenian market, local farmers are not aware of or do not recognize foodborne viruses as distinctive food safety hazards, which represent barrier and consequently critical point to food safety management practices. Despite the fact that guidelines on viruses in food (15) are enforced are mostly unknown to professionals. There is a need to disseminate current guidelines as good viral food safety practice via food safety authorities and professional associations, chambers and societies, because we have demonstrated that even professionals in the field are generally unaware of its recommendations or even existence. Generally, despite a quite long tradition of aquaculture in Slovenia, there is no leading research institution dealing with fisheries and aquaculture. The research programmes are dispersed to different government and public institutions. Non-government institutions and farmers are only exceptionally included in research activities. Advanced level training in aquaculture is not well developed; consequently, shellfish farmers are thus more or less self-educated in accordance with the requirements of existing legislation.

Conclusions

The filter-feeding nature of shellfish and their tendency to concentrate any environmental or man-made contaminant present in their growing waters requires attention to these food safety issues and compliance with applicable requirements. As viruses do not grow in food, do not cause deterioration of the product, and the organoleptic properties of the food are not affected, it is questionable if control measures aiming at microbial growth inhibition are effective to reduce viral contamination. There is a need to assess whether the control measures in place for bacterial hazards require adjustments to be effective against viruses. For the time being, HACCP studies need to address prerequisite programs, such as good hygiene, agricultural and aquacultural practices, especially the origin and quality of water used in food supply chains, and adequate hand hygiene as the most effective prevention measure. Compliance with prerequisite programs, such as codex guidelines, is essential to reduce the risk of contamination. It is also beneficial to have the harmonized integration of monitoring and control to be able to routinely monitor that compliance measures are being undertaken effectively.

Food safety education is most effective when messages are targeted at changing the behaviors most likely to result in foodborne illness, such as personal hygiene, adequate cooking, avoiding cross-contamination, keeping food at safe temperatures, and avoiding foods from unsafe sources. Food safety education is most likely to be effective if the messages are targeted toward specific audiences. The results emphasize the need for tailored educational programs to improve awareness with respect to viruses and to implement innovations into good practices. Not just connected to hygiene, but even more to integrate it into comprehensive good aquaculture practice.

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References

1. Food and Agriculture Organization of the United Nations (FAO). National aquaculture sector overview: Slovenia. http://www.fao.org/fishery/countrysector/naso_slovenia/en (31. Mar. 2016)
2. Statistical Office Republic of Slovenia (SURSTAT). Fishery and aquaculture. Ljubljana : SURSTAT, 2016. <http://www.stat.si/StatWeb/en/field-overview?idp=94&headerbar=0#> (31. Mar. 2016)
3. Monfort MC. The European market for mussels. GLOBEFISH Research Programme: Vol. 115. Rome: Food and Agriculture Organization of the United Nations (FAO), 2014: 65 pp.
4. Kmetijski inštitut Slovenije. Poročilo o stanju kmetijstva, živilstva, gozdarstva in ribištva v letu 2013. Pregled po kmetijskih trgih. Ljubljana : KIS, 2016. http://www.kis.si/Porocila_o_stanju_v_kmetijstvu_OEK. (15. Mar. 2016)
5. Rippey SR. Infectious diseases associated with molluscan shellfish consumption. Clin Microbiol Rev 1994; 7(4): 419–25.
6. Lees D. Viruses and bivalve shellfish. Int J Food Microbiol 2000; 59(1/2): 81–116.
7. Loisy F, Atmar RL., Guillon P, et al. Real-time RT-PCR for norovirus screening in shellfish. J Virol methods 2005; 123(1): 1–7.
8. Henigman U, Biasizzo M, Vadnjal S, et al. Molecular characterisation of noroviruses detected in mussels (*Mytilus galloprovincialis*) from harvesting areas in Slovenia. New Microbiol 2015; 38: 225–33.
9. Diez-Valcarce M, Kokkinos P, Söderberg K, et al. Occurrence of human enteric viruses in commercial mussels at retail level in three European countries. Food Environ Virol 2012; 4(2): 73–80.
10. EFSA. Scientific Opinion on an update on the present knowledge on the occurrence and control of foodborne viruses. EFSA J 2011; 9(7): e2190 (96 pp.) <http://www.efsa.europa.eu/en/efsajournal/pub/2190>
11. Bellou M, Kokkinos P, Vantarakis A. Shellfish-borne viral outbreaks: a systematic review. Food Environ Virol 2013; 5(1): 13–23.
12. Ambrožič M, Božič T, Jevšnik M, et al. Compliance of proposed Codex Alimentarius Guidelines for virus management with principles of good practice. Acta Aliment Hung 2011; 40 (3): 364–75.
13. Richards GP, McLeod C, Le Guyader FS. Processing strategies to inactivate viruses in shell-

fish. *Food Environ Virol* 2010; 2: 183–93.

14. Chalmers JWT, McMillan JH. An outbreak of viral gastroenteritis associated with adequately prepared oysters. *Epidemiol Infect* 1995; 115: 163–7.

15. CAC/GL 79-2012. Guidelines on the application of general principles of food hygiene to the control of viruses in food. Codex Alimentarius. International Food Standards. Rome : FAO ; WHO, 2012: 13 pp.

16. Verhoef L, Gutierrez GJ, Koopmans M, et al. Reported behavior, knowledge and awareness toward the potential for norovirus transmission by food handlers in Dutch catering companies and institutional settings in relation to the prevalence of norovirus. *Food Control* 2013; 34(2): 420–7.

17. Koopmans M. Foodborne viruses from a global perspective. In: *Improving food safety through a one health approach: workshop summary*. Washington : National Academies Press, 2012: 225–57.

18. Regulation (EC) No. 178/2002 of the European Parliament and of the Council of 28 January 2002 laying down the general principles and requirements of food law, establishing the European Food Safety Authority and laying down procedures in matters of food safety. *Off J Eur Commun* 2002; L 31: 1–24.

19. Centers for Disease Control and Prevention (CDC). Surveillance for foodborne disease outbreaks, United States, 2013, Annual Report. Atlanta, Georgia: US Department of Health and Human Services, 2015: 20 pp.

20. EFSA. The European Union summary report on trends and sources of zoonoses, zoonotic agents and foodborne outbreaks in 2014. *EFSA J* 2015; 13(12): e4329 (191 pp) <https://www.efsa.europa.eu/en/efsajournal/pub/4329>

21. Kirk M, Ford L, Glass K, et al. Foodborne illness, Australia, circa 2000 and circa 2010. *Emerg Infect Dis* 2014; 20(11): 1857–64.

22. Scallan E, Hoekstra RM, Angulo FJ, et al. Foodborne illness acquired in the United States: major pathogens. *Emerg Infect Dis* 2011; 17(1):7–15.

23. Mead PS, Slutsker L, Dietz V, et al. Food-related illness and death in the United States. *Emerg Infect Dis* 1999; 5(5): 607–25.

24. Havelaar AH, Kirk MD, Torgerson PR, et al. World Health Organization global estimates and regional comparisons of the burden of foodborne disease in 2010. *PLoS Med* 2015; 12(12): e1001923

(23 pp.). <http://journals.plos.org/plosmedicine/article?id=10.1371/journal.pmed.1001923>

25. Newell DG, Koopmans M, Verhoef L, et al. Foodborne diseases: the challenges of 20 years ago still persist while new ones continue to emerge. *Int J Food Microbiol* 2010; 139(1): S3–15.

26. Wilhelm B, Waddell L, Greig J, et al. A scoping review of the evidence for public health risks of three emerging potentially zoonotic viruses: hepatitis E virus, norovirus, and rotavirus. *Prev Vet Med* 2015; 119(1/2):61–79.

27. Program monitoringa zoonoz in povzročiteljev zoonoz 2015. Ljubljana : Ministrstvo za kmetijstvo, gozdarstvo in prehrano Republike Slovenije ; Ministrstvo za zdravje Republike Slovenije http://www.uhvvr.gov.si/si/delovna_podrocja/zivila/zoonoze (25. Mar. 2016)

28. Suffredini E, Lanni L, Arcangeli G, et al. Qualitative and quantitative assessment of viral contamination in bivalve molluscs harvested in Italy. *Int J Food Microbiol* 2014; 184: 21–6.

29. Le Guyader F, Haugarreau L, Miossec L, et al. (2000). Three-year study to assess human enteric viruses in shellfish. *Appl Environ Microbiol* 2000; 66(8): 3241–8.

30. Raspor P, Ambrožič M, Jevšnik M. Food chain safety management systems: the impact of good practices. In: Yanniotis S, ed. *Advances in food process engineering research and applications*. New York : Springer, 2013: 607–25. (Food engineering series)

31. Wills WJ, Meah A, Dickinson AM, et al. I don't think I ever had food poisoning'. A practice-based approach to understanding foodborne disease that originates in the home. *Appetite* 2015; 85: 118–25.

32. Taché J, Carpentier B. Hygiene in the home kitchen: changes in behaviour and impact of key microbiological hazard control measures *Food Control* 2014; 35(1): 392–400.

33. Byrd-Bredbenner C, Schaffner DW, Abbot JM. How food safe is your home kitchen? A self-directed home kitchen audit. *J Nutr Educ Behav* 2010; 42: 286–9.

34. Jevšnik M, Hlebec V, Raspor P. Consumers' awareness of food safety from shopping to eating. *Food control* 2008; 19(8): 737–45.

35. 41. Jevšnik M, Hlebec V, Raspor P. Food safety knowledge and practices among food handlers in Slovenia. *Food Control* 2008; 19(12): 1107–18.

36. Jevšnik M, Hlebec V, Raspor P. Meta-anal-

ysis as a tool for barriers identification during HACCP implementation to improve food safety. *Acta Aliment Hung* 2006; 35(3): 319–53.

37. Ergönül B. Consumer awareness and perception to food safety: a consumer analysis. *Food Control* 2013; 32: 461–71.

38. Raspor P. Total food chain safety: how good practices can contribute. *Trends Food Sci Technol* 2008; 19(8): 405–12.

39. Raspor P, Jevšnik M. Good nutritional practice from producer to consumer. *Crit Rev Food Sci Nutr* 2008; 48(3): 276–92.

40. Scott E. Food safety and foodborne disease in 21st century homes. *Can J Infect Dis Med Microbiol* 2003; 14: 277–80.

41. Boxman ILA, Verhoef L, Hägele G, et al. Environmental testing for norovirus in various institutional settings using catering companies as sentinels for norovirus prevalence among the gen-

eral population. *Food Control* 2015; 47: 98–102.

42. Poklar Vatovec T, Jakus T, Bizjak M. Seashell specialties and food handling in Slovene Istria restaurants. *Acta Agric Slov* 2014; 104(2): 81–90.

43. Stave C, Törner M. Exploring the organisational preconditions for occupational accidents in food industry: a qualitative approach. *Safety Sci* 2007; 45: 355–71.

44. Strauss LA. *Qualitative analysis for social scientists*. Cambridge : University Press, 1987: 319 pp.

45. Glaser B, Strauss A. *The discovery of grounded theory: strategies for qualitative research*. Chicago : Aldine, 1967.

46. Hollway W, Jefferson T. *Doing qualitative research differently: free association, narrative and the interview method*. London, Thousand Oaks, New Delhi : Sage, 2003: 166 pp.

OKUŽBA Z VIRUSI V PROIZVODNO-OSKRBOVALNI VERIGI ŠKOLJK NA SLOVENSKI OBALI

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Povzetek: Med živili, ki so pogosto povezana z norovirusnimi izbruhi in izbruhi hepatitisa A svetovnih razsežnosti, so tudi školjke. V okviru uradnega nadzora v Sloveniji od leta 2013 vzorčimo školjke, pri katerih se ugotavlja prisotnost norovirusov in virusov hepatitisa A. Prisotnost norovirusne RNK je bila ugotovljena v 25 % testiranih vzorcih v letu 2014 do 40 % v letu 2015. Prisotnosti virusov hepatitisa A ni bilo možno potrditi v nobenem od analiziranih vzorcev, kar povežujemo z nizkim številom okužb pri ljudeh v Sloveniji. Poleg ugotavljanja prisotnosti norovirusov in virusov hepatitisa A v školjkah smo opravili tudi polstrukturirane intervjuje s slovenskimi školjkarji. Kvalitativna analiza je razkrila, da se slovenski školjkarji zavedajo možnih tveganj na področju gojenja školjk, ampak samo v povezavi s prisotnostjo bakterije *Escherichia coli* kot indikatorskega mikroorganizma in biotoksinov. Kljub ugotovljeni prisotnosti norovirusne RNK v školjkah, prisotnih na slovenskem tržišču, lokalni školjkarji ne prepoznajo virusov kot dejavnikov tveganja, pomembnih za zagotavljanje varnosti živil. Ugotovitve kažejo, da se lokalni školjkarji zavedajo možnih mikrobioloških tveganj na področju gojenja školjk. Vendar pa kljub temu ne prepoznajo virusov kot možnih dejavnikov tveganja, kar izpostavi pomembnost kontinuiranega, rednega usposabljanja in izobraževanja pri obvladovanju virusnih okužb v proizvodni in oskrbovalni verigi školjk.

Ključne besede: školjkar; Slovenija; uradni nadzor; varnost živil; virusi v hrani; polstrukturirani pogovor