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Environmental and anthropogenic factors affecting coastal bathing water quality: preliminary study for Primorje-Gorski Kotar County (Croatia)

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ABSTRACT

Coastal bathing water, as a common good, is an economic resource of public health interest. Predictive models of coastal bathing water contamination are needed for timely prevention of pollution, warning of bathers, and activation of municipal services and utilities in case of contingencies, as well as institutional mechanism designs for common good management purposes. The goal of this research is to identify the variables that would improve predictive models of coastal bathing water bacterial contamination. The microbiological quality of coastal bathing water is affected by many variables. This research is an analysis of the following determinants: precipitation amount, seawater temperature and salinity, as well as few indicators of anthropogenic pressure on the environment such as registered population, registered tourist overnight stays and the amount of generated municipal waste, all possibly directly or indirectly affecting the bathing water quality in 17 coastal municipalities in the Primorje-Gorski Kotar County. The analysis showed that rainfall, as an instrumental confounder variable, influences salinity and seawater temperature by increasing groundwater discharge and bringing contamination i.e. increasing enterococci and *Escherichia coli* concentrations in coastal bathing water. Population as the conjectured independent variable, representing the anthropogenic cause of pollution, was once again falsified as a statistically significant determinant. For further research, longer-term sampling (preferably year-round) at micro-locations of comparable hydrogeological characteristics is recommended.

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1 Introduction

Bathing waters contaminated with faecal waste are potential sources of skin and respiratory diseases, as well as gastrointestinal, eye, ear, and nose infections [1,2]. However, coastal bathing water quality must be imposed not only as a public health issue but also as an economic issue. Namely, coastal bathing water quality is recognized to be one of the key factors influencing the choice of a vacation destination [3-6], and this is extremely important for Croatia as a tourist-oriented country.

The criterion for bathing water quality is defined as a measurable relationship between exposure to pollution and health consequences. The criteria define maximum levels of faecal pollution indicators associated with an unacceptable risk to human health. The definition of health risk acceptability, in addition to medical factors, also includes social, economic and political factors and may therefore vary greatly in various parts of the world. Water quality criteria are defined by the Bathing Water Directive 2006/07/EC, a key document for the management of bathing water quality in European union, and presently under revision [7,8].

In Croatia, systematic monitoring of bathing coastal bathing water quality began in 1989. Numerous elements of this program have been altered over several decades of its implementation: regulations, microbiological criteria and test methods, limit values of individual categories of water quality, as well as statistical methods of data processing.

Coastal bathing sites are rated as “excellent”, “good”, or “sufficient” according to limit values of microbiologi-

Table 1 Standards for the assessment of coastal bathing water quality (after each sampling campaign), according to national regulation

Indicator	Coastal bathing water quality			Test method
	Excellent	Good	Sufficient	
intestinal enterococci (CFU*/100 mL)	<60	61-100	101-200	HRN EN ISO 7899-1 or HRN EN ISO 7899-2
Escherichia coli (CFU*/100 mL)	<100	101-200	201-300	HRN EN ISO 9308-1 or HRN EN ISO 9308-3

*CFU – colony-forming unit

Source: [15-18]

cal indicators, intestinal enterococci and *Escherichia coli* (*E. coli*), in seawater samples (Table 1). In case when the number of indicator bacteria per 100 mL of sample exceeds the upper limit for category "sufficient", coastal bathing water quality is rated "poor" and it is not recommended for use.

Numerous factors influence coastal bathing water quality at a given location and temporal and spatial variations are pronounced [9-12]. Environmental factors such as precipitation, inflows of coastal and submarine springs, cloudiness, wind, seawater temperature, salinity, waves, sea currents and tides affect changes in concentration of indicator bacteria.

Contamination with municipal wastewater has the greatest impact on the deterioration of coastal bathing water quality [13,14]. Building new and upgrading existing drainage systems and urban wastewater treatment systems has resulted in a significant reduction in the number of bathing sites not meeting the minimum quality criteria. Despite significant improvement in coastal bathing water quality in the last two decades, pollution still proves to be an intermittent or persistent occurrence in certain locations. Causes of pollution should be sought in the lack of connection of certain entities to the public sewerage system, illegal connections, leaking septic tanks, leaks in the internal sewerage network of certain facilities by the sea, inadequate drainage of rainwater, specific human activities near bathing sites (ports, shipyards, and marinas), etc. Additional anthropogenic variables that affect coastal bathing water quality are tourist number and behaviour at beaches.

Finding a model for the prediction of microbiological quality is of great importance, as it would allow for a timely response from the authorities/services and would alert the bathers before the bathing waters are contaminated. We performed a preliminary study to lay the groundwork for the creation of a predictive model for coastal bathing water quality. We analysed the 2019-bathing season for coastal municipalities in the Primorje-Gorski Kotar County (PGC). In the summer months, a large number of visitors placed enormous pressure on municipal utility companies that deal with the water supply, drainage systems, sewage systems and the disposal of municipal waste [19]. Therefore, in addition to environmental indicators, the study also included anthropogenic indicators such as the number of tourists (i.e. the number of recorded overnight stays) and the amount of created municipal solid waste. The generated waste serves as a proxy variable for data about wastewater, which we were not able to obtain.

2 Data and methods

Analysis included field observations and laboratory tests. Sampling points were at locations where one can expect the largest number of bathers or where, according to the bathing water profile, the greatest risk of pollution exists. Sampling points also included sea mouths, coastal springs and submarine springs. Distance between two neighbouring sampling points depends on the type of the beach: one sampling point every 100 metres on sandy or pebble beaches, and one sampling point every 200 metres on other beaches [15].

Seawater samples were collected in sterile bottles of at least 250 mL and tested within 6 hours. The water temperature was measured *in situ* and laboratory sample tests included the determination of seawater salinity and microbiological indicators: intestinal enterococci and *E. coli*. Water at each sampling point was analysed ten times, on average every 14 days during the period between mid-May and end of September 2019. Sampling was performed at 270 locations in 17 PGC municipalities and a total of 2700 samples were analysed.

Water temperature was measured according to standard methods [20] using a thermometer with graduation intervals of 0.1 °C. Seawater salinity was determined by a conductivity electrode (Seven Multi – Mettler Toledo, Switzerland) according to salinity – conductometry method [21]. Daily precipitation data are official Croatian Meteorological and Hydrological Service data. We used them to calculate the cumulative precipitation for the time span of seven days prior to sampling.

Both microbiological parameters, *E. coli* and intestinal enterococci were analysed using the membrane filtration technique. *E. coli* cultivation was performed according to the temperature-modified ISO 9308-1 method [22,23] using Chromogenic Coliform agar (Biolife Italiana S.r.l., Milan, Italy) incubated for 4 h at 36 ± 2 °C followed by 20 h incubation at 44 ± 0.5 °C. Intestinal enterococci cultivation was performed on Slanetz-Bartley Agar (Biolife Italiana S.r.l., Milan, Italy) for 48 h at 36 ± 2 °C and on Bile-Aesculine Agar (Biolife Italiana S.r.l., Milan, Italy) for 2 h at 44 ± 0.5 °C, in case of growth of suspected colonies, as a confirmation test [17].

Bacterial counts and salinity statistical analysis (Kruskal-Wallis test and Dunn test for post-hoc analysis) was carried out in R, a freely available statistical computing and graphics software. The results were interpreted at the statistical significance level of 0.05.

For the period that coincides with the collection of seawater samples, we also collected the number of registered local inhabitants per municipality, the number of registered tourist overnight stays, and the amount of generated communal waste in kg on a monthly basis. Due to large intertwining of environmental and anthropogenic effects, we resorted to the panel ordinary least squares (OLS) analysis. In order to make the data comparable, we calculated the monthly mean values for salinity, seawater temperature, and counts of intestinal enterococci and *E. coli* as well as the average precipitation one week before sampling. For each municipality, averages were based on available data for all affiliated locations. We are aware of some parameters, such as salinity and bacteria values, being site specific and their specificity can be lost by averaging. Then again, our analysis is a panel, and our goal was to test inferential hypotheses at a more universal level without taking into account site or sampling point specificities.

In the end, our cross-section consists of 17 PGC municipalities, and the time dimension consists of 5 samples collected during the bathing season for a total of 85 observations for a balanced panel. Panel data analysis is a combination of cross-section analysis with elements of time series analysis. It is a group of statistical methods of analysis for longitudinal data that enables both falsification of hypotheses as well as coefficient calculation. Panel Granger *"causality"* testing enables falsification of *"post hoc ergo propter hoc"* statistical associations with arbitrary lag length adjustments for fine tuning according to groundwater speed. For both, the panel analysis and the panel Granger causality testing we used the statistical package E-Views.

3 Results and discussion

For all municipalities, the largest number of overnight stays was realized in July and August (Figure 1). Kostrena had the smallest number of tourist overnight stays, while the largest number overnight stays was recorded in the municipalities of Crikvenica and Mali Lošinj.

Figure 2 shows the amount of generated municipal waste in PGC during the May-September period 2019. Municipalities with small number of inhabitants relative to number of visiting tourists have the largest differences for generated waste between the season and the pre/post season months (May and September). Thus, on the island of Krk, Mošćenička Draga, Rab, Novi Vinodolski, Crikvenica and island of Mali Lošinj, the amount of generated waste in the peak months of the season increases 2 to as much as 14 times compared to the pre/post season. The smallest variation for generated waste per month was found for Rijeka, Kostrena, Lovran and Kraljevica (2-4%).

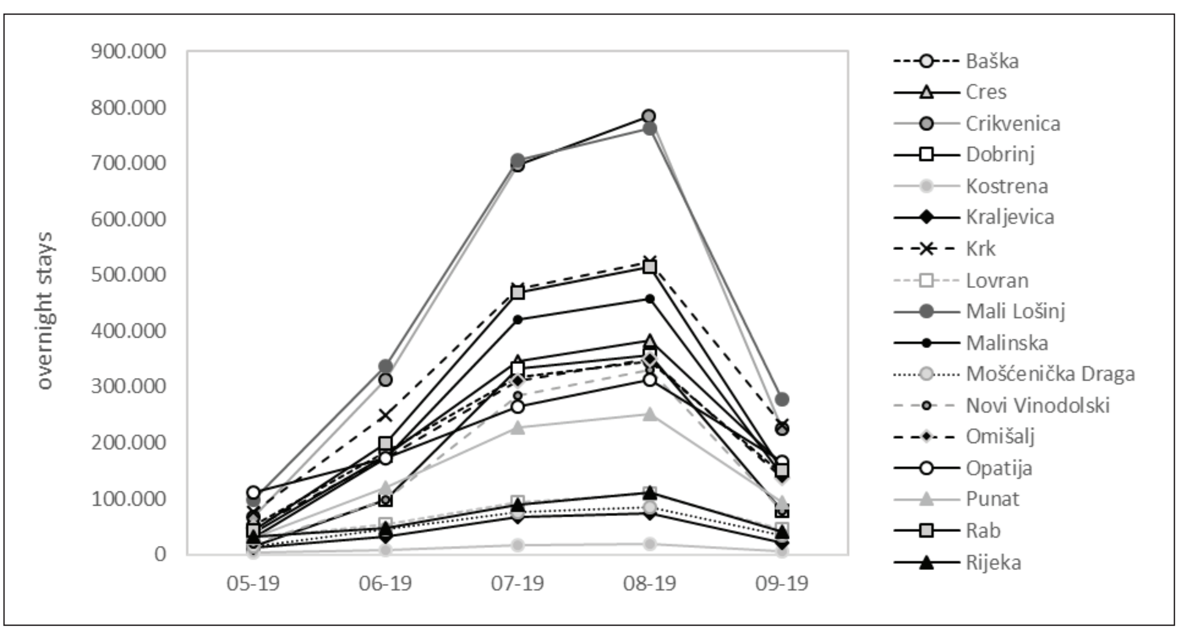

Figure 1 Tourist overnight stays in Primorje-Gorski Kotar County, May-September 2019.

Source: Authors

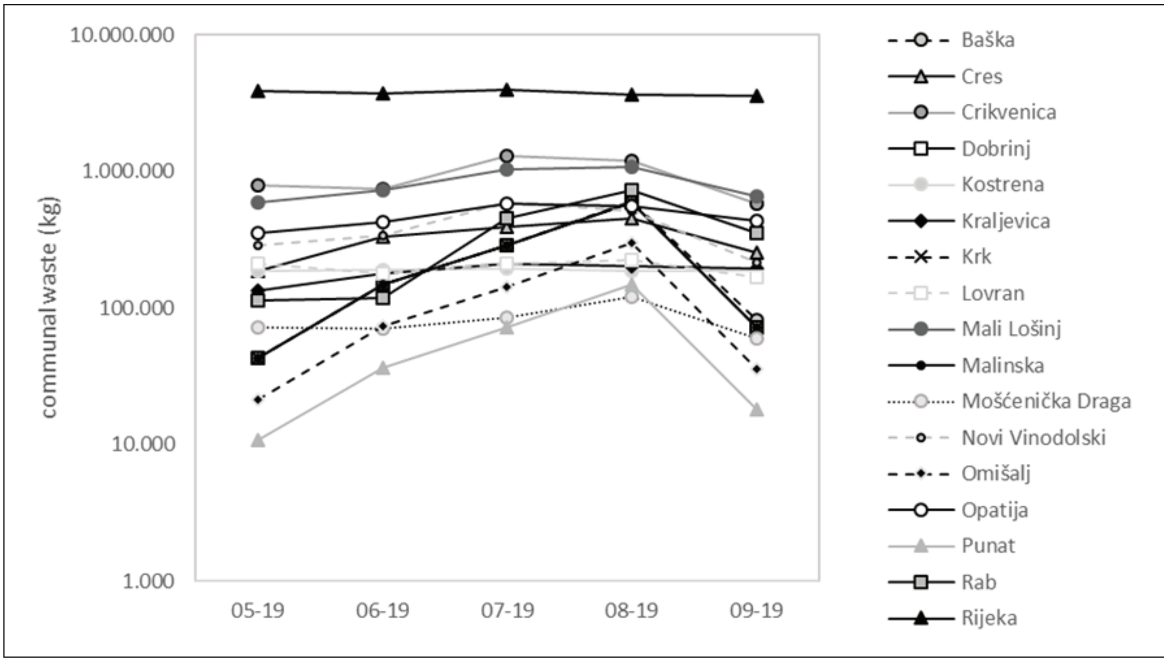

Figure 2 Generated communal waste in Primorje-Gorski Kotar County, May-September 2019.

Source: Authors

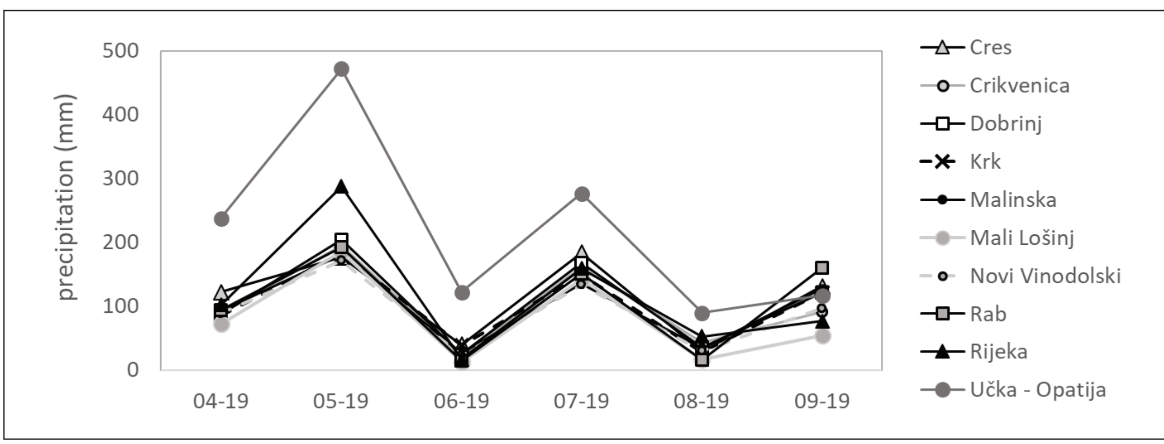

Figure 3 Monthly precipitation for the coastal area and islands Primorje-Gorski Kotar County, April-September 2019.

Source: Authors

Figure 3 shows monthly precipitation in PGC from April to September 2019. April 2019 is also included in the analysis since hydrological conditions that precede seawater sampling may also affect costal bathing water quality [24, 25]. The rainiest month in the analysed period was May, followed by July 2019. Specifically, for May 2019, recordings show the largest amount of precipitation during the second half of the month. The highest amount of precipitation for the region was recorded in Učka, i.e. in the hinterland of Opatija.

PGC can roughly be divided into three areas: Liburnia (including Mošćenička Draga, Lovran and Opatija), the Rijeka-Novi Vinodolski area and the Kvarner islands. On average, coastal bathing water has been shown to be significantly less polluted by *E. coli* on the islands (median 1.0 CFU, $p<0.0001$) compared to the areas of Rijeka-Novi Vinodolski (median 2.0 CFU) and Liburnia (median 2.5 CFU). Between the observed areas, there was also a statistically significant difference in intestinal enterococci concentration ($p<0.00001$), where the median for islands was 2 CFU, for the area Rijeka-Novi Vinodolski 3 CFU, and for Liburnia 6.5 CFU.

Figures 4 and 5 graphically represent intestinal enterococci and *E. coli* average values for PGC municipalities during bathing season 2019 (May-September). For the municipalities of Baška, Dobrinj, Kostrena, Krk, Mošćenička

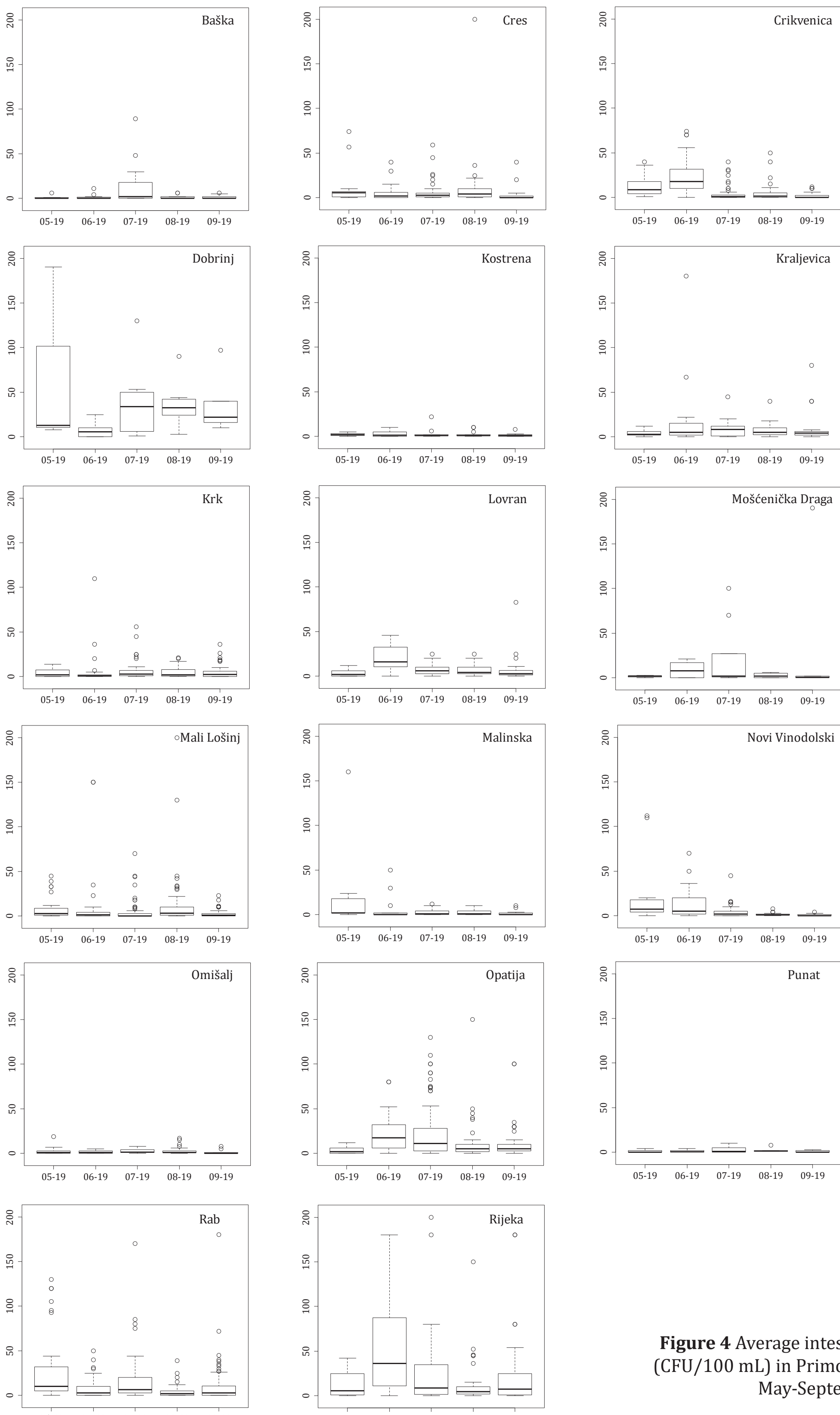

Figure 4 Average intestinal enteroccoci values (CFU/100 mL) in Primorje-Gorski Kotar County, May-September 2019.

Source: Authors

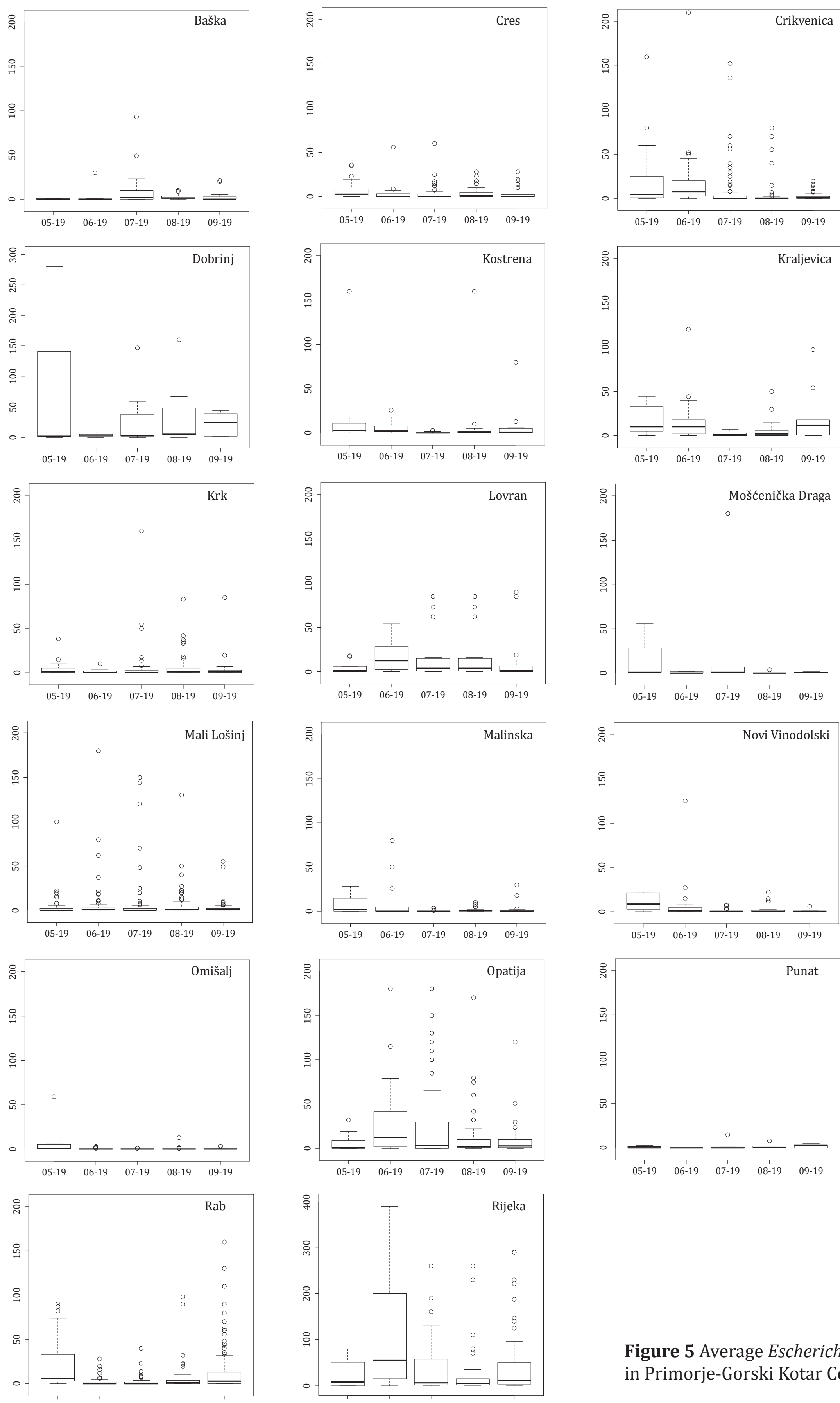

Figure 5 Average *Escherichia coli* values (CFU/100 mL) in Primorje-Gorski Kotar County, May-September 2019.

Source: Authors

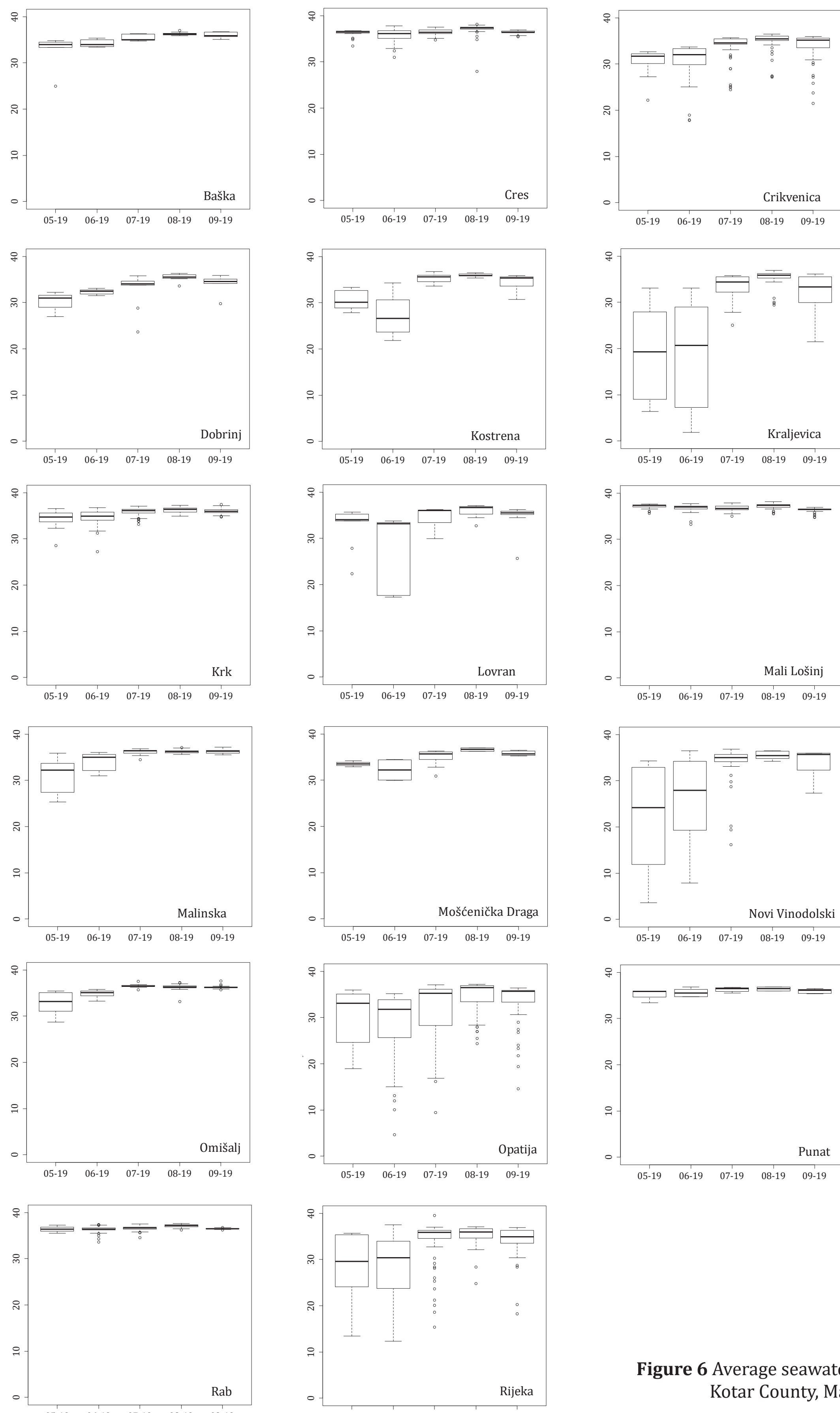

Figure 6 Average seawater salinity in Primorje-Gorski Kotar County, May-September 2019.

Source: Authors

Draga, Malinska and Punat there were no significant differences in bacteriological contamination between the months included in the analysis (p>0.05). The significant differences in the average values of both types of bacteria (p<0.05) were determined for Cres, Crikvenica, Novi Vinodolski, Omišalj, Opatija, Rab and Rijeka. Municipalities of Lovran and Mali Lošinj had significant differences in monthly contamination for intestinal enterococci, and Kraljevica for *E. coli*.

Determining the difference in the average value of bacteria between individual months was further performed by post-hoc analysis (Dunn test). On the islands, higher average levels of bacteria were recorded in May as compared to the values in June, July and August. On Rab, September was also more polluted than June, July and August (*E. coli*). Between Novi Vinodoloski and Kraljevica, both May and June were more polluted than July, August or September. In the area of Rijeka and Liburnia (Opatija and Lovran), the most polluted month of the analysed period was June.

This situation could be explained by differences in the hydrogeology of these areas [26-28]. For islands, we do not expect a large area of groundwater recharge or long retention of precipitation in the underground. Therefore, the precipitation that fell in May on the islands could have caused a rapid reaction of groundwater and their increased discharge, which brought microbial contamination material into the sea.

On the other hand, the municipalities on the mainland have large hinterlands that are recharge areas of many coastal and submarine springs. As those areas are much larger than recharge areas on islands, it takes a longer time for groundwater levels to rise high enough to carry pollution into the sea. Larger amount of water also implies a longer time of discharge, so the pollution of coastal bathing water with bacteria is present for a longer time, and therefore in this area the most polluted month was June (specifically the first half of the month).

Our assumption is that significant rain events raise groundwater levels to the point of reaching the levels of faulty septic tanks and damaged and dilapidated sewer pipes. Heavy rainwater results in increased aquifer discharges i.e. increased groundwater flows that eventually flow into the sea. Therefore, we used salinity as an indicator of fresh water presence in coastal bathing water (Fig. 6).

There is a significant difference in salinity between the observed areas. The highest average salinity is measured for the islands (average 36.16), and the lowest in the Rijeka-Novi Vinodolski area (average 32.17; p<0.0023).

The variability of salinity for the islands (coeff.var. 3.7%) was low in relation to Liburnia (coeff.var. 17.2%) and Rijeka-Novi Vinodolski area (coeff.var. 18.3%).

The minimal salinity for coastal bathing water for Kvarner islands (23.7) was recorded in June. In most cases, the average salinity of islands' coastal bathing water in May and June was lower compared to other months of the bathing season. That is consistent with the fact that May was the rainiest month of the season and the month with the highest bacteria concentrations. On island Rab, a significantly lower salinity was recorded in September compared to August (p<0.00001).

For the area of Rijeka-Novi Vinodolski, May and June are the months with a large range, and significantly lower average values of salinity compared to other months of the bathing season. This is especially pronounced for Kraljevica, Novi Vinodolski and Rijeka (Fig. 6). The minimum value was recorded in Kraljevica in June (1.9). For the salinity of the coastal bathing water in Liburnia, only June differs significantly from July, August and September. The minimum value of 4.7 was also recorded in June. For both, Liburnia and Rijeka-Vinodol, the lowest salinity coincides with the highest bacteriological pollution.

We also investigated potential multicollinearity, and the major source of interdependence is the relation between the number of inhabitants (local population) and the quantity of municipal solid waste (R=0.96). This posed a problem for the simultaneous use of these two variables during our modelling, so we decided that the number of inhabitants is statistically more significant and instrumental. Additionally, we also tested for signs, i.e. if the direction of causality is commensurate with theory. The usual method

Table 2 Correlation matrix

	Enterococci	*E. coli*	Population	Rainfall	Salinity	Sea temp.	Tourists
Enterococci	1.000						
E. coli	**0.842***	1.000					
Population	**0.347***	**0.502***	1.000				
Rainfall	0.139	0.209	-0.001	1.000			
Salinity	**-0.378***	**-0.441***	-0.139	**-0.353***	1.000		
Sea temp.	-0.054	-0.160	-0.029	-0.420	**0.465***	1.000	
Tourists	-0.186	-0.232	-0.135	-0.416	0.401	0.606	1.000
Waste	0.316	0.460	**0.963***	-0.086	-0.085	0.083	0.083

Note: Statistically significant (p<0.05) correlations are shown in bold with an asterisk (*).

Source: Authors

Table 3 Panel Ordinary Least Squares of *E. coli* and enterococci as dependent variables

Dependent	Independent	Coefficient	Std. Error	t-Statistic	Prob.	R^2
E. coli	Salinity	-2.511	0.561	-4.476	0.0000	0.19
	Constant	99.545	19.05454	5.224	0.0000	
E. coli	Rainfall	0.223	0.046947	4.752	0.0000	0.27
	Waste	$1.20 \cdot 10^{-5}$	$2.09 \cdot 10^{-6}$	5.758	0.0000	
E. coli	Population	0.0003	$6.17 \cdot 10^{-5}$	5.172	0.0000	0.39
	Salinity	-2.156	0.495	-4.356	0.0000	
	Constant	83.656	16.928	4.942	0.0000	
Enterococci	Salinity	-1.401	0.377	-3.718	0.0004	0.14
	Constant	58.539	12.799	4.574	0.0000	
Enterococci	Rainfall	0.173	0.034	5.034	0.0000	0.08
	Waste	$6.55 \cdot 10^{-6}$	$1.53 \cdot 10^{-6}$	4.286	0.0000	
Enterococci	Population	0.0001	$4.52 \cdot 10^{-5}$	3.071	0.0029	0.23
	Salinity	-1.247	0.362605	-3.439	0.0009	
	Constant	51.628	12.401	4.163	0.0001	
Salinity	Rainfall	-0.0489	0.014	-3.436	0.0009	0.12
	Constant	35.527	0.635	55.944	0.0000	
Salinity	Sea temperature	0.478	0.088	5.418	0.0000	0.60
	Constant	23.553	1.905	12.362	0.0000	
Salinity	Sea temperature	0.425	0.117	3.648	0.0005	0.25
	Rainfall	-0.0265	0.015	-1.810	0.0740	
	Constant	25.625	2.778	9.223	0.0000	
Sea temperature	Rainfall	-0.053	0.012	-4.215	0.0001	0.18
	Constant	23.274	0.558	41.730	0.0000	
Waste	Population	0.944	0.016	58.779	0.0000	0.97
	Tourist stays	0.932	0.059	15.785	0.0000	

Source: Authors

is to show a correlation matrix of all variables (Table 2). As expected, in most cases, where contamination with intestinal enterococci is found, there is also contamination with *E. coli*. Thus, we analyzed the intestinal enterococci and the *E. coli* as dependent variables.

Municipal solid waste is entirely of anthropogenic origin, and this is mostly created by the local population (R=0.96). For a detailed study of municipal solid waste generation on the Croatian coast, see [19]. Municipal solid waste is thus a good proxy for anthropogenic environmental pressure on the coastline. The signs in front of the correlation coefficients between bacteria and salinity are negative as expected, as well as between bacteria and seawater temperature.

Rainfall is positively correlated with intestinal enterococci and *E. coli* bacteria. Rainfall is thus our primary antecedent variable, serving as a medium for pollutants. At the same time, rainfall is also a confounder variable, influencing both the independent variables (salinity and temperature) as well as both dependent variables: intestinal enterococci and *E. coli* concentrations. Thus, rainfall is the antecedent confounding variable in our analysis.

We proceed with the panel OLS analysis of faecal bacteria as dependent variables and a selection of independent variables that were highly statistically significant. Here we emphasize once again that this is only a preliminary study, with only 85 samples in the data set. Data being statistically significant at $p<0.001$ level for all statistical associations shown in Table 3 is quite an achievement for such a small data set. When we look at the different influences of factors on bacteriological contamination of the coastal bathing water, we see that *E. coli* and intestinal enterococci are mostly positively affected by the domicile population and negatively by salinity.

According to the results shown in Table 3, we constructed a flow chart (Fig. 7) showing the conjectured relationships between rainfall as the antecedent confounding variable negatively influencing seawater temperature and salinity with a secondary positive inference going from seawater temperature to salinity. Without a mediator variable such as water isotopic composition [24, 25], no statistical evidence of direct causal-mechanical link between rainfall and bacterial contamination could be found in the panel OLS.

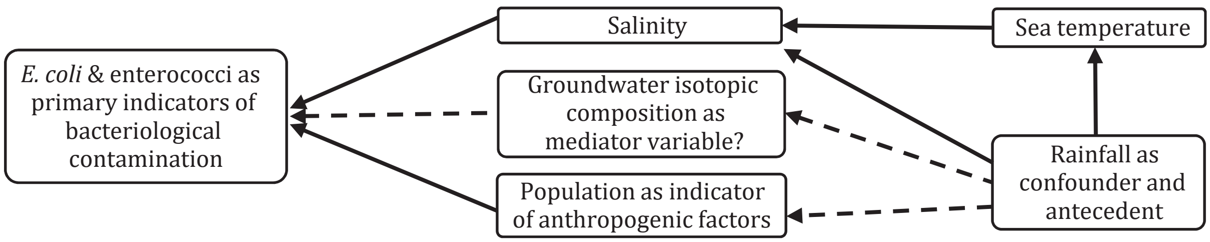

Figure 7 Flow chart with indicators and factors leading to bacterial contamination. Solid lines show the determinants. Dashed lines show the indicators (proxy variables).

Source: Authors

Table 4 Pairwise Granger "causality" tests

Null Hypothesis:	F-Statistic	Prob.
SALINITY does not Granger cause ENTEROCOCCI	9.48367	0.0030
LOCAL POPULATION does not Granger cause ENTEROCOCCI	12.8711	0.0006
WASTE does not Granger cause ENTEROCOCCI	10.1516	0.0022
RAINFALL does not Granger cause ESCHERICHIA COLI	4.16047	0.0454
SALINITY does not Granger cause ESCHERICHIA COLI	4.82168	0.0317
LOCAL POPULATION does not Granger cause ESCHERICHIA COLI	45.6699	$5 \cdot 10^{-9}$
WASTE does not Granger cause ESCHERICHIA COLI	34.9989	$1 \cdot 10^{-7}$
SEA TEMPERATURE does not Granger cause SALINITY	6.09215	0.0162
SEA TEMPERATURE does not Granger cause TOURISTS	4.76900	0.0326
LOCAL POPULATION does not Granger cause WASTE	16.4317	0.0001
TOURISTS does not Granger cause WASTE	8.16329	0.0057
RAINFALL does not Granger cause SALINITY	2.98009	0.0890
RAINFALL does not Granger cause ENTEROCOCCI	3.05751	0.0851

Source: Authors

Analysing the causal relationships, the best results were obtained with a time lag of just one, thus 85 observations – 17 cross-sections = 68 final observations in the Panel Granger "causality" test (Table 4). The highest F-Statistic is shown for the successful rejection of the null hypothesis that local population (as in registered local population) does not Granger "cause" the *E. coli* bacteria in the seawater sample (F-Statistic=45.67, and $p<1\cdot10\text{-}8$). Commensurate results were gained for the enterococci bacteria (F-Statistic=12.87, and $p<0.001$).

The panel pairwise Granger causality test with a time lag set to two weeks, could not reject a null hypothesis of a rainfall Granger causing the *E. coli* faecal bacteria contamination at $p<0.05$. Local population is collinear with the disposed waste and since population better represents the anthropogenic pollution, it was kept in the model instead of the disposed waste.

Pairwise Granger "causality" test falsifies the causal conjecture by testing the intertemporal statistical association of variables controlling for autoregression. The null hypothesis is of no causation, thus all inferences with $p<0.05$ are rejected. It follows from Table 4 that we may not reject the claims that salinity, local population, waste and rainfall, have an impact on faecal contamination of coastal bathing water. However, the Granger causality between rainfall and salinity, and rainfall and enterococci bacterial contamination are only significant at $p<0.1$ level. The former is probably due to a lack of freshwater sources near sampling points on the islands. The latter is probably due to a smaller sensitivity of the enterococci vs the *E. coli* indicator [29]. The time lag of 2 weeks barely captures the enterococci at the $p<0.1$ level. This leads to the conclusion that the analysis is context- and site specific, and it is difficult to come to universal conclusions about causal relationships.

4 Conclusions

In this preliminary study, we analysed the impact of environmental and anthropogenic factors on the level of bacteriological pollution on popular bathing sites in

Primorje-Gorski Kotar County. The analysis included 17 coastal municipalities during the 2019 bathing season (May-September). Results show that the domicile population is the principal instrumental variable which has a positive impact on the bacteriological contamination, i.e. increases it, and that seawater salinity is the principal determinant which decreases the contamination.

Salinity does not affect bacteriological contamination directly but is an indicator of fresh water inputs containing bacterial contamination after rainfall events. The average and maximum salinity is higher on the islands, whereas its variability is much lower. This can be accounted for by the fact that sampling points on islands have fewer freshwater sources in the vicinity. These findings indicate that the primary sources of decreasing salinity are freshwater sources bringing the rainfall and bacteriological contamination with it. Nevertheless, statistical modelling captured a statistically significant positive association between temperature and salinity that cannot be disregarded.

Greater influence of other variables on coastal bathing water quality probably went unnoticed because the study covered a large area of versatile hydrogeology. In addition, there are large differences in the number of officially registered inhabitants across municipalities: from 1,500 (Mošćenička Draga) up to 128,600 (Rijeka). For certain municipalities/locations, there is persistent microbiological pollution (probably due to poor infrastructure), which could offset other impacts.

In this paper, we attempted to give an overview of the variables describing and explaining the process of anthropogenic sea contamination at bathing locations in the Primorje–Gorski Kotar County. Building on this knowledge, we conclude that, in the future, prediction for bacteriological contamination should be modelled for micro-locations and include dummy variables representing the presence and quality of the local sewage infrastructure. For better understanding of how environmental and anthropogenic factors impact coastal bathing water quality, year-round sampling should be carried out to cover periods of drought/heavy rainfall, high and low seawater temperatures, large number of tourists, etc. For such a study, we suggest sampling locations to be in areas where elevated bacteria levels were present throughout the bathing season.

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