

Monitoring of nitrates in drinking water from agricultural and residential areas of Podravina and Prigorje (Croatia)

Jasna Nemčić-Jurec · Miljenko Konjačić ·
Anamarija Jazbec

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Abstract Nitrates are the most common chemical pollutant of groundwater in agricultural and suburban areas. Croatia must comply with the Nitrate Directive (91/676/EEC) whose aim is to reduce water pollution by nitrates originating from agriculture and to prevent further pollution. Podravina and Prigorje are the areas with a relatively high degree of agricultural activity. Therefore, the aim of this study was, by monitoring nitrates, to determine the distribution of nitrates in two different areas, Podravina and Prigorje (Croatia), to determine sources of contamination as well as annual and seasonal trends. The nitrate concentrations were measured in 30 wells ($N=382$ samples) in Prigorje and in 19 wells ($N=174$ samples) in Podravina from 2002 to 2007. In Podravina, the nitrate content was

24.9 mg/l and 6 % of the samples were above the maximum available value (MAV), and in Prigorje the content was 53.9 mg/l and 38 % of the samples above MAV. The wells were classified as correct, occasionally incorrect and incorrect. In the group of occasionally incorrect and incorrect wells, the point sources were within 10 m of the well. There is no statistically significant difference over the years or seasons within the year, but the interaction between locations and years was significant. Nitrate concentrations' trend was not significant during the monitoring. These results are a prerequisite for the adjustment of Croatian standards to those of the EU and will contribute to the implementation of the Nitrate Directive and the Directives on Environmental Protection in Croatia and the EU.

J. Nemčić-Jurec (✉)
Institute of Public Health of Koprivnica-Krizevci County,
Trg Tomislava Bardeka 10/10,
48 000 Koprivnica, Croatia
e-mail: nemicic-jurec@inet.hr

M. Konjačić
Department of Animal Science and Technology,
Faculty of Agriculture, University of Zagreb,
Svetošimunska 25,
10 000 Zagreb, Croatia
e-mail: mkonjacic@agr.hr

A. Jazbec
Department for Forest Inventory and Management,
Faculty of Forestry, University of Zagreb,
Svetošimunska 25,
10 000 Zagreb, Croatia
e-mail: ajazbec@sumfak.hr

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Introduction

Nitrates are the most frequent chemical pollutant of groundwater in agricultural and suburban areas. The effects of agriculture and soil utilisation on deterioration of groundwater have been reported by numerous scientists (Krapac et al. 2002; Widory et al. 2003; Cetindag 2005; Nemčić-Jurec and Vadla 2010; Thorburn et al. 2003).

Agricultural activities affect the nitrate concentration in groundwater in two ways: directly and indirectly. Direct

increase is a result of constant perennial nitrate transfer from agrochemicals into aquifer. Indirectly, agrochemicals cause physical, chemical and biological changes in the soil, the increase of soil acidity, resulting in increased leaching from natural soil sources, predominately from humus. It is very important to influence the indirect factors in order to decrease infiltration and leaching (Elhatip et al. 2003; Hooker et al. 2008).

Specific habitat factors such as climate, hydrogeology and soil can cover up general effects of soil utilisation. It is considered to be important to determine the relationship between the nitrate leaching from soil under different soil management in agriculture and nitrate concentration in groundwater (McLay et al. 2001). As nitrate compounds are mobile and present in agricultural, industrial and household wastes, they are considered to be the indicators of the pollution of groundwater. High average nitrate concentration of 57.3 mg/l as a result of human activity was reported by Caballero et al. (2003). The anthropogenic effects on groundwater were also found by Obeidat et al. (2007). By measuring nitrate concentration in water on 88 different locations, they found that 9 % of the locations had concentration above the maximum available value (MAV). Also, between 2001 and 2006, the increase of nitrate concentration (from 10 to 330 mg/l) was reported in 16 wells ($N=248$ samples). More than 92 % of the samples contained more than 20 mg/l nitrate and this was assumed to be the result of human activities. In the areas of developing urbanisation, a decrease of nitrate concentration in aquifer was found in agricultural and suburban areas (Xu et al. 2007; Showers et al. 2008; Drake and Bauder 2005).

In Croatia, there are few studies investigating the effects of agricultural and suburban areas on groundwater quality through longer period of time and in different hydrogeological areas (Nemčić et al. 2007). The aim of the present study was to detect and compare the nitrate concentrations in two different areas of the northwest Croatia (the Podravina and Prigorje regions) and to examine and compare the seasonal and annual patterns of nitrate contamination in both regions. According to the dynamics of the monitored timeline, the aim was also to predict the trends of nitrate concentration in the future and to estimate the possible pollution of groundwater with nitrates.

Materials and methods

The research area

Hydrogeological characteristics

Podravina, the area of the town of Đurđevac and its nearby settlements, is situated in the aquatic region of Middle Podravina. The area of an old alluvial terrace consists of the river Drava and its influents' deposits, and terrace sediments. From the hydro geological point of view, the most important is quaternary gravel–sand aquifer, which is 30 m, and in certain parts of the aquifer area even 70 m thick. The top part—roof of the aquifer—consists of the following: dust, sand and clay with significant occurrence of live sands and predominantly swamp loess. The thickness of this layer and its texture strongly affect the permeability and possibility of leaching of nitrates and other components from surface layers, i.e. agricultural soil to aquifer. There is no unique hydrogeological regime existing in the aquatic region. The effect of the river Drava on the direction of the groundwater flow is obvious in the zone width of 2 to 3 km along the river Drava. In this zone, groundwater imitates the Drava regime during the year. The supply of the aquifer layer is done through infiltration of precipitations through the low permeable cover, as well as through leaching of the Drava riverbed in the upstream area. The quality of natural water changes from location to location. According to some data, it is assumed that water quality in the first aquifer layer is endangered by the regular application of agrochemicals all across the area.

Prigorje, the area of the town of Krizevci, also includes the area of the Kalnik highlands with the dominant landscape body of Kalnik. Morphologically most exposed parts are built of the Palaeogene and Neogene rocks, while the northern part consists of the Mesozoic sediments. The Kalnik highland massive consists of well-permeable dolomite breccias and permeable to low permeable sediments. On the surface, the sediments are collectors of precipitations, and in deeper layers, there is a reservoir of groundwater, the dolomite lime aquifer. The other part of the settlements which belong to the Prigorje region stretches on Pleistocene terrace. Under 7 to 9 m of covering clay, there is a 9-m-thick aquifer layer that consists of gravel and sand of different granulation. Under this layer, there is a thin layer of coal, clay and loess clay. Although the roof

layers, soil and Pleistocene loam under fluvial–glacial effects are low permeable materials protecting the aquifer, it can be assumed that water in this aquifer layer is also endangered due to the intensive application of agrochemicals.

Climate, water balance and soil utilisation

Moderate continental climate prevails in the research area and the average annual amount of precipitations is about 820 mm. In the annual cycle of precipitations, there are two maximums. The largest amount of precipitations occurs during the summer (July), which is a characteristic of the continental climate, and during the autumn (September), which is the effect of maritime influence of the Mediterranean Sea and the Atlantic Ocean. The primary maximum is in July reaching between 80 to 100 mm, and the secondary maximum appears in September with 90 to 110 mm at the measuring stations. Monthly and annual precipitations (in millimetre) in the regions of research Prigorje and Podravina are shown in Table 1. The average annual temperature is around 11 °C showing characteristics of a moderately warm climate (data obtained from the Metrological and Hydrological Service). As meteorological elements, precipitations are the most important factor that can potentially affect nitrate leaching into groundwater.

By calculating the water balance in the soil according to the Thornthwaite method, potential and actual evapotranspiration as well as water deficit and water surplus were determined for perennial average in the meteorological stations Krizevci and Đurđevac. Water deficit was

found by analysing data in the research area in certain months of the warmer part of the year, i.e. potential evapotranspiration was higher than precipitations. Water deficit found in Krizevci during July and August amounted to 88.7 mm, and similar was for the Đurđevac meteorological station where water deficit was 63.8 mm. Water surplus was found during the cold months, in no vegetation period. Water surplus in Krizevci amounted to 189.3 mm and in Đurđevac to 190.5 mm.

The Koprivnica-Krizevci County covers the surface of 177.464 ha, out of which 104.011 ha or 58.61 % are agricultural areas. The rest are forests (32.03 %) and unfertile soil, and settlements and aquatic areas (9.32 %). Agricultural areas include arable areas (cultivated fields and gardens, orchards, vineyards and meadows) and pastures. Arable land makes 98.9 % of the total agricultural area (102.858 ha). The rest of the agricultural area, i.e. pastures, make 1.1 % (2.15 ha). The data were obtained from the Croatian Bureau of Statistics, Annual statistics for 2009. The research was conducted in eastern (Podravina) and western (Prigorje) part of the county.

Sampling and water analysis

The research was conducted in the area of about 65,000 inhabitants and includes 49 wells from shallow aquifers (Fig. 1). In accordance with the aim, the wells were divided into two spatial groups. Thirty wells were situated in the area of Prigorje (*N*=382 samples) and 19 wells were situated in the area of Podravina (*N*=174 samples). The distribution of nitrate concentration in drinking water was researched in wells situated in suburban or urban

Table 1 Month and annual precipitations (in millimetre) in the region of Prigorje and Podravina

	Months												Sum
	1	2	3	4	5	6	7	8	9	10	11	12	
Prigorje													
Average	36.6	39.2	49.1	60.6	58.1	68.6	83.1	76.5	94.2	72.2	68.2	61.2	767
Min	7.2	1.3	2.6	8.0	12.0	45.0	23.0	1.0	24.2	2.6	32.4	20.8	564
Max	70.7	78.6	111.6	119.0	106.2	90.3	172.2	165.9	205.4	172.7	117.3	101.8	1057
Podravina													
Average	40.7	45.6	53.4	65.1	63.9	81.9	85.7	81.1	101.3	63.9	73.9	62.0	818
Min	5.8	2.9	6.8	7.0	11.8	32.6	19.3	5.8	21.7	2.5	39.1	24.9	608
Max	100.4	135.5	110.1	133.4	103.6	150.0	185.5	149.3	195.2	147.8	113.1	114.7	1071

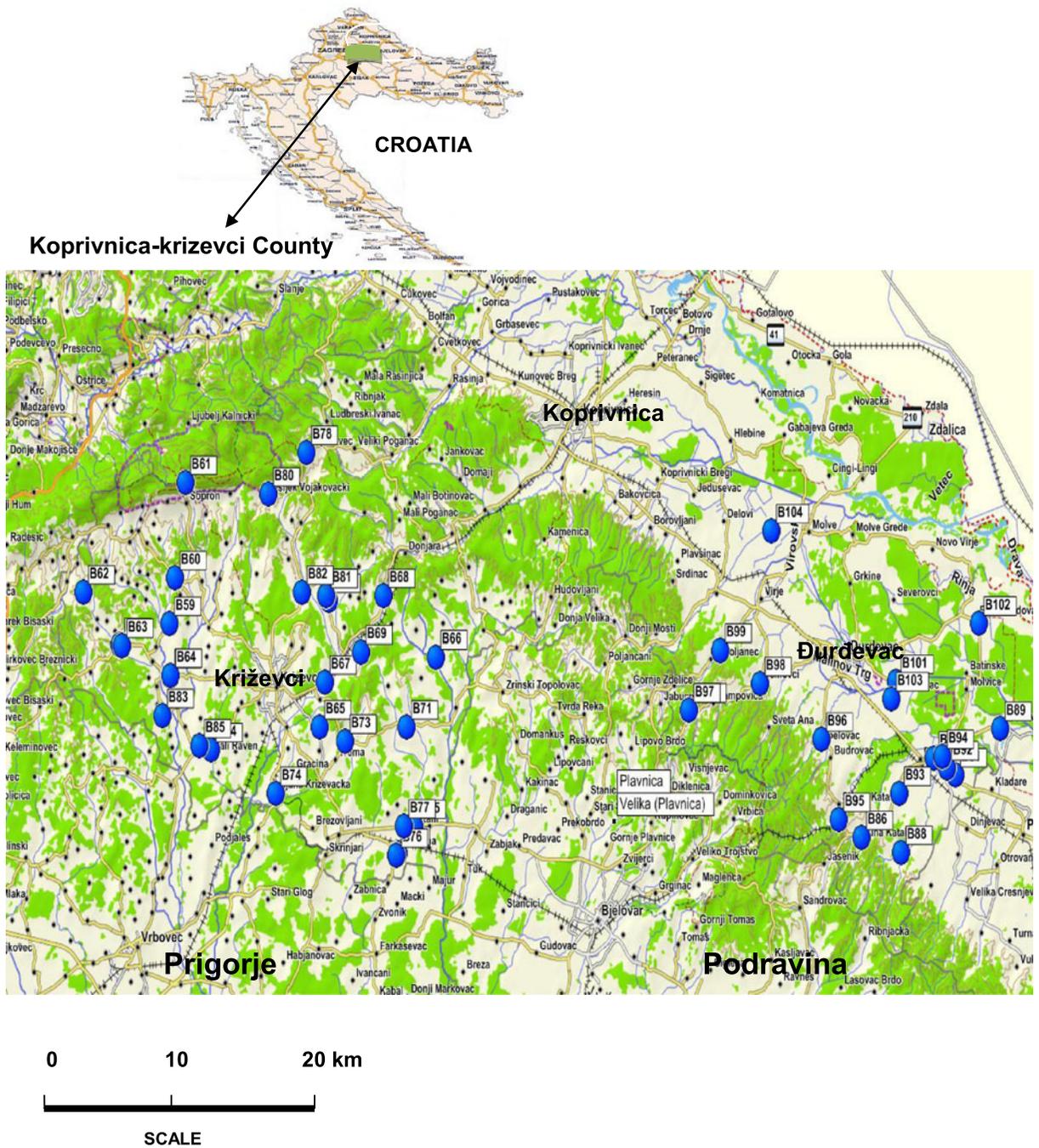


Fig. 1 Locations of the wells included in monitoring

areas near to the point sources of contamination—septic tanks and sewer systems—or in wells that were near to the point sources of agricultural origin—farms, stables, organic manure depots.

Sampling was done according to HRN ISO 5667:2000. Water sampling from wells was done after releasing of water for 2 to 3 min on the outlet of the water supply system, in the clean polyethylene vessel of 1 l volume. The

samples were stored in the portable fridge, in the dark at 1 to 5 °C for transportation. After delivery to the laboratory, the samples were stored at the same temperature until analysis (within 24 h). The nitrate content in drinking water was determined using the ion chromatography method (HRN ISO 10304–1:1998). Prior to injection into the analyser, ion chromatograph ICS 3000, Dionex (USA), samples were filtered through a membrane filter (of pore 0.45 µm) to remove any particulate matter. After establishing the calibration function, samples were injected into the ion chromatograph and the peaks were measured in accordance with the manufacturer's instructions. The chromatograph system consists of the following components: auto sampler device, sample injection system (incorporating sample loop of 50 µl), gradient pump, liquid chromatography device, conductivity detector and eluent generator. AS 15 anion separator column and appropriate precolumn were used for separation. ASRS ULTRA II-4 mm performed chemical suppression. Thirty-eight millimolars NaOH was used as eluent and flow was 0.5 ml/min. The whole system was supervised by Dionex Chromeleon Softver. Water testing was conducted during the period from 2002 to 2007 in all seasons in order to determine time trends.

Data analysis

Descriptive statistics was done for all the data and it was compared to the MAV Regulation (Regulation of Health Safety of the Drinking Water 2008), i.e. recommended level value (RLV) of Nitrate Directive (European Union 1991). Nitrate concentration was set into three groups—low (<25 mg/l), moderate (25–50 mg/l) and high (>50 mg/l).

The relationship between the average nitrate concentration in wells and distances from point pollution sources of the wells was determined by the Spearman rank correlation (Sparks 2000). By distributing the samples according to water quality from shallow aquifers and the distance from point sources (χ^2 test), the wells were classified as correct—nitrate content regularly in category low or moderate (below MAV), occasionally incorrect—nitrate content present in all categories, and incorrect—nitrate content regularly in category high.

The nitrate concentration measurements through seasons included testing of samples taken in spring (March, April and May), summer (June, July and August), autumn (September, October and November) and winter (December, January and February). In order to determine

leaching of water from soil into groundwater during seasons, water balance was calculated according to the Thornthwaite method (Kos et al. 1993). To compare the levels of nitrate by wells at two locations and for the analysed period, we used the repeated measure analysis of variance (ANOVA) (Davis 2002).

For trend analysis of nitrate during the research period (the winter of 2002 to winter 2007), we used the Mann–Kendall test. In order to analyse and present wells in respect to their annual average concentrations of nitrate, we have made principal component analysis (PCA) based on the annual averages for the period from 2002 to 2007 (Sparks 2000).

Descriptive statistics, correlation, the χ^2 test, the repeated measure ANOVA and part of the Mann–Kendal test were done using the SAS 9.2. PCA was done using the STATISTICA 8.0 (2011) and graphical presentations were done using both statistical packages.

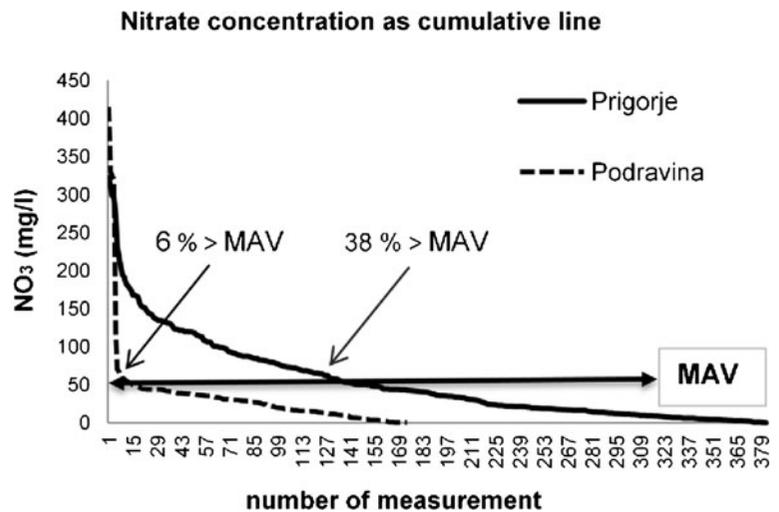
Results

Distribution of nitrate concentration

In the region of Podravina, the town of Đurđevac and its nearby settlements, the average nitrate content was 24.9 mg/l and in 6 % of the samples the nitrate content was above MAV of the regulation. Average nitrate content in the region of Prigorje, the town of Krizevci and its nearby settlements, was 53.9 mg/l. The proportion of the samples above MAV in this area was 38 %. The distribution of nitrate concentration as a cumulative line in both areas is shown in Fig. 2.

According to the MAV of the regulation and RLV of the Directive, a categorisation of wells into three groups was made in order to determine the distribution of the relative nitrate content in the research area (Table 2). From 30 wells analysed altogether, the average nitrate concentration in 40 % of the wells was below RLV of the Directive, low, in 17 % of the wells it was between RLV and MAV of the regulation, moderate. In 43 % of the wells, it was above MAV, high. In the Podravina region, the results showed lower nitrate concentrations in the wells. From 19 wells analysed altogether, 47 % of the wells had an average nitrate concentration below RLV—low, 42 % had moderate nitrate concentrations, and only 11 % had high nitrate concentration. It is assumed that an

Fig. 2 Nitrate concentration as cumulative line in Podravina and Prigorje from 2002 to 2007



anthropogenic effect was present in the areas of moderate and high concentration.

Agriculture and suburban area—sources of water contamination with nitrates

The Spearman rank correlation between the distance from point source (in metre) and the average nitrate concentration of wells for years (2002, 2003, 2004, 2005, 2006 and 2007) are as follows, respectively: -0.788 , -0.867 , -0.886 , -0.852 , -0.867 and -0.863 for Prigorje and -0.745 , -0.532 , -0.579 , -0.530 , -0.712 and -0.784 for Podravina, all statistically significant ($p < 0.05$). From Table 3, it can be seen that the average nitrate concentration in wells that were within 10 m from the point sources ($N=9$) was 94.3 mg/l and the highest average nitrate concentration of all the measurements in the individual wells was 152.2 mg/l. The average nitrate concentration in the wells within 10 to 20 m of the sources of contamination was 45.2 mg/l ($N=4$) with the highest average of the wells of 58.7 mg/l. Distances further than 20 m show average nitrate concentration of individual well as 57.3 mg/l. The proportion of wells with average nitrate concentration above MAV (>50 mg/l) was 78 % when wells were within 10 m of contamination sources, which additionally confirms the case of an incorrect well. The proportion of wells with average nitrate concentration above MAV in distance from 10 to 20 m from contamination source was lower—50 %, while the lowest proportion was of wells with average nitrate concentration above MAV and further than 20 m from contamination source—16 %.

Table 4 presents distribution of samples according to quality, i.e. health safety of water from wells and distance from point sources of contamination from the wells. In the group of correct wells, the highest number of wells with safe samples ($N=275$) was on a distance of more than 20 m between the point sources and the wells. On a distance between 10 to 20 m ($N=15$), the number of safe samples decreases, and the smallest number of safe samples is from the wells within 10 m of point contamination sources ($N=3$). In the group of occasionally incorrect and incorrect wells, the highest number of samples was within 10 m of point contamination sources, then follow those situated from 10 to 20 m away, and the smallest number of samples was in a distance further than 20 m from contamination sources. Causal relationship of point sources of agricultural origin and septic tanks with nitrate content from shallow aquifers was determined according to the results of analysis. Namely, point sources within 10 m of the wells affected nitrate concentration in the wells.

Seasonal and annual changes of nitrate concentrations

The results of the repeated ANOVA measuring show the difference between locations, but there is a level of significance of 8 % ($p=0.0777$). The reason for this is the high variability of nitrate concentration especially in Prigorje as seen in Fig. 3. There is a statistically significant variability in nitrate concentration of wells within each location ($F=58.43$, $p < 0.0001$, Table 5). The results also show that there is no statistically significant difference over the years or seasons within

Table 2 Distribution of the nitrate content compared with MAL and RLV (MAV—50 mg/l; RLV—25 mg/l)

Well	NO ₃ (mg/l)			
	Average	Median	Skewness	Kurtosis
Prigorje				
PR12	1.7	0.9	1.11	0.88
PR25	3.9	3.3	0.89	0.77
PR3	9.2	8.2	1.31	1.38
PR18	9.5	7.0	0.76	-0.77
PR23	11.4	8.5	2.00	5.00
PR7	12.2	12.1	0.40	-0.79
PR28	12.4	13.6	-0.43	-1.36
PR2	12.7	11.0	0.75	-0.82
PR6	17.8	16.8	0.73	0.29
PR14	20.4	21.0	-0.96	0.91
PR20	21.9	20.1	1.46	2.10
PR11	22.7	21.3	0.81	-0.04
PR5	28.4	21.2	0.82	-0.45
PR15	81.4	90.1	-0.52	0.28
PR8	32.8	32.7	-0.49	-0.85
PR16	38.9	42.0	-1.37	2.00
PR27	41.0	40.6	0.08	-1.85
PR17	57.4	52.8	0.73	-0.12
PR10	77.7	65.5	1.37	0.27
PR29	78.5	77.6	0.4	-0.65
PR21	58.7	58.1	0.82	1.31
PR30	49.6	50.0	0.01	-0.31
PR24	89.1	86.3	0.34	-0.5
PR26	92.6	87.5	0.30	-0.88
PR9	103.4	104.6	-0.12	-1.30
PR1	106.6	106.0	0.28	1.38
PR22	121.5	119.9	0.80	1.19
PR19	124.2	129.4	-0.58	0.70
PR4	152.5	166.5	-0.43	-0.95
PR13	245.7	220.1	0.87	0.47
Podravina				
PO19	0.6	0.2	1.85	3.39
PO10	1.1	0.5	1.34	1.13
PO15	22.6	4.4	1.22	-0.84
PO12	5.6	5.5	1.47	1.49
PO18	12.5	13.6	-1.05	0.02
PO5	12.9	12.7	0.52	0.19
PO3	14.7	15.2	-2.57	8.41
PO17	16.0	15.4	-0.11	0.03
PO8	22.5	21.7	0.75	-1.77
PO2	28.7	29.5	0.79	0.36

Table 2 (continued)

Well	NO ₃ (mg/l)			
	Average	Median	Skewness	Kurtosis
PO13	30.4	30.2	-1.0	1.09-
PO11	32.8	33.0	-0.01	-1.47
PO7	33.0	35.4	-1.42	2.42
PO6	35.4	37.8	-0.92	-1.0
PO16	37.2	38.2	-1.74	3.5
PO4	38.0	40.0	-1.32	1.66
PO14	50.2	50.0	0.55	0.12
PO1	54.6	47.8	2.98	9.4
PO9	60.7	60.5	0.31	1.51

the year. However, the interaction between locations and years was statistically significant ($F=7.25$; $p<0.0001$) which indicates the fact that the concentration of nitrate over the years do not behave the same in Prigorje and Podravina.

The Mann–Kendall trend test shows that there is no statistically significant trend in test data (nitrate concentrations) at both locations ($Z_{Prigorje}=0.48$; $Z_{Podravina}=-1.14$; $n=23$) as can be seen from Fig. 4.

In order to compare wells from both locations with respect to their average annual concentration (2002–2007), we used the PCA (Fig. 5). From Fig. 5, it can be seen that there is only one significant factor which explains 92.58 % of the total variability which is to be expected because neither trend is statistically significant. From the Fig. 5, we can also see how individual wells stand out and are grouped based on the average annual nitrate concentrations.

Discussion

Low average nitrate concentration was found in Podravina (24.9 mg/l) which is similar to the results reported by other authors (Hallberg 1989; Kazemi 2004; Jalali 2005; Aelion and Conte 2004). However, higher average nitrate concentration was found in Prigorje (53.9 mg/l) in water from shallow aquifers, which is similar to the results reported for some other research areas (Kattan 2001). The results presented in this study show differences of two geographically different regions within the Koprivnica-Krizevci County, which are probably affected by different environmental

Table 3 Relationship between the average nitrate concentration in wells and distances of point source pollution from wells

Distance from point source (m)	Well number	Nitrate concentration average (mg/l)	Maximal nitrate concentration average (mg/l)	Proportion of wells (%) >MAV
< 10 m	9	94.3	152.5	78
10 to 20 m	4	45.2	58.7	50
> 20 m	36	29.8	57.3	16

and anthropogenic conditions. The anthropogenic effect is bigger in the Prigorje region which is manifested in higher proportion of wells with high nitrate concentration (40 %) above MAV. The proportion of wells in the Podravina region with nitrate concentration above MAV was 11 %. Very similar results showing 9 % of the wells with nitrate concentration above MAV were reported by Obeidat et al. (2007). It is considered that these results were caused by human activity.

Since plant and livestock production are well developed in the research region of Podravina, and especially Prigorje, and the point sources of contamination in suburban areas are very often near to the wells, the presented results were expected. But the results presented in this study are lower than the ones reported by some other authors (Woo-Jung et al. 2007). Significant difference ($p < 0.05$) of nitrate concentration in drinking water from wells was found depending on the distance from point sources. When the wells were within 10 or 20 m of the point source, higher nitrate content (above MAV) was found, while further distance from the point source, more than 20 m, showed no effect on water quality. Similar results obtained by Gardner et al. (2005) reported that vicinity and number of septic tanks, dense population and livestock production contribute to increased

nitrate concentration in the water. Assumptions on point sources of contamination in similar areas with similar characteristics were reported in other studies as well (Nemčić et al. 2007; Drake and Bauder 2005; Widory et al. 2003).

ANOVA shows the difference between locations but not statistically significant at 5 % level than at 8 % and the reason for this is the high variability of nitrate concentration especially in Prigorje. The results also show that there is no statistically significant difference over the years or seasons within the year. However, the interaction between locations and years was statistically significant which indicates the fact that the concentrations of nitrate over the years do not behave the same in Prigorje and Podravina. Some scientists explained the reasons for such results by the fact that excessive nitrogen application leads to high nitrate concentration in soil in dissolved form; nitrogen is not absorbed in soil colloid complex but penetrates soil, enters groundwater and is rinsed with rain (Šimunić et al. 2002). Maxwell et al. (2010) pointed out that the higher concentrations of nitrate occur in the rainy seasons and lower in the dry seasons. According to our data, the precipitations are higher from spring to autumn which according to Maxwell would increase the concentration of nitrate

Table 4 Distribution of samples according to water quality in wells and distance from source points of contamination

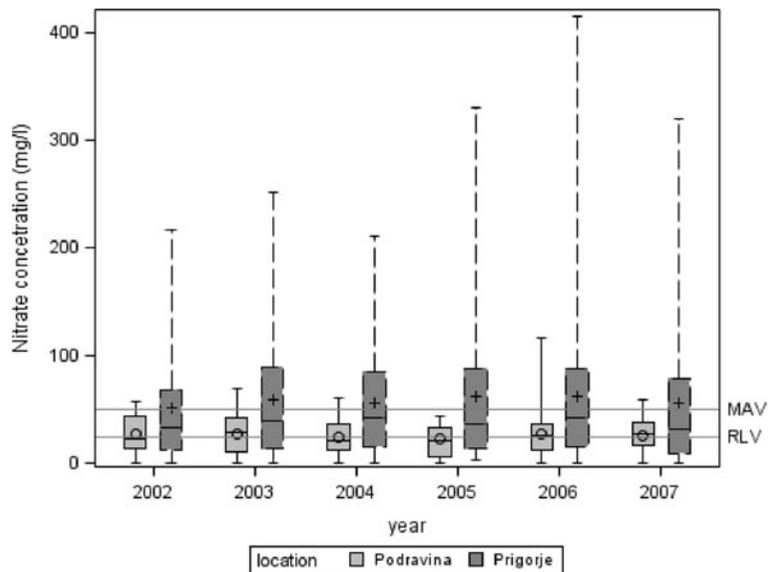
Distance from source points (m)	Type of wells			
	Correct ^a	Occasionally correct ^b	Incorrect ^c	Total
<10 m	3 (4 %)	17 (22 %)	57 (74 %)	77 (14 %)
10-20 m	15 (31 %)	34 (69 %)	0 (0 %)	49 (9 %)
>20 m	275 (64 %)	123 (29 %)	32 (7 %)	430 (77 %)
Total	293 (53 %)	174 (31 %)	89 (16 %)	556

^a Nitrate content regularly below 50 mg/l

^b Nitrate content occasionally below 50 mg/l and occasionally above 50 mg/l

^c Nitrate content regularly above 50 mg/l

Fig. 3 Box plot of annual nitrate concentration (in milligram per litre) by location indicating RLV (recommended level value) and MAV (maximum allowed value). (Boxes mean, lines median, boxes Q1–Q3, whiskers min–max)



in these seasons. However, according to the available water balance data, the highest water surplus is recorded in autumn and winter, and deficiency in spring and summer, which explains the variability of concentrations over the seasons. It is assumed that certain rainy cases contribute to variability regardless of the season (Hooker et al. 2008). Hooker et al. also reported that climate and vegetation, depending on season and rainfall, affect nitrate leaching from soil and its concentration in water. The same authors reported that the lowest nitrate concentration is expected in spring and summer as vegetation is the highest, and it is suggested that in regions with mild climates, the growth of vegetation should be prolonged in order to cause potential decrease of nitrate concentration in water. These explanations can be partially applied in

this study. The lowest nitrate concentrations were obtained during the period of vegetation, meaning that plants consume higher proportion of nitrogen, but only in certain years. Atxotegi et al. (2003) reported that besides vegetation, precipitations and temperatures have also an important effect. Higher amounts of precipitations cause denitrification, and higher temperatures increase nitrate fixation in the soil that can lead to lower nitrate concentrations in spring and summer. Such an explanation can be partially applied to this study.

Literature offers other explanations too. Seong-Chun et al. (2005) found in their research area especially high nitrate concentrations in spring and summer as a result of increased nitrogen input by fertilisation and increased leaching into groundwater,

Table 5 Results of repeated measure analysis of variance for nitrate concentration

Source of variation	DF	Type III SS	Mean square	F value	Pr>F
Between subject					
Location	1	84,053.47663	84,053.47663	3.25	0.0777
Well (location)	47	1,214,759.352	25,845.944	58.43	<0.0001
		Type I SS	Mean square	F value	Pr>F
Within subject					
Year	5	2,998.1058	599.6212	0.22	0.9553
Season (year)	17	22,795.3363	1,340.9021	0.49	0.9596
Location × year	6	120,200.2086	20,033.3681	7.25	<0.0001

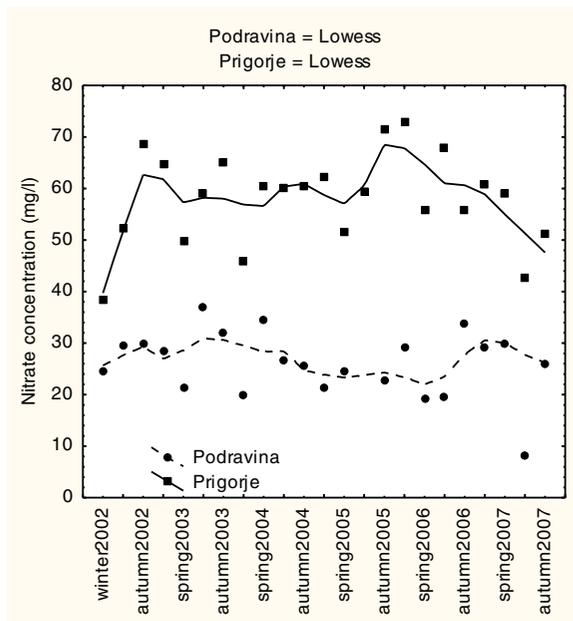


Fig. 4 Scatterplot of average nitrate concentration (in milligram per litre) per season and year for both locations with Lowess smoothing line

while in other seasons, lower concentrations were explained by denitrification. Banks et al. (1998) reported lower nitrate leaching during the winter due to the lower temperatures and frozen soil. Lower levels of nitrate in the winter are also found in certain years during the course of the study. It can also be the result

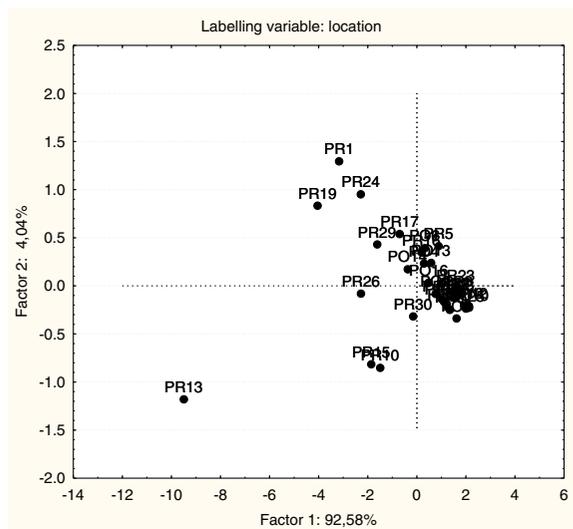


Fig. 5 Projections of the wells from Podravina (*PO*) and Prigorje (*PR*) in respect to the first two factors (PCA), and this in respect to the average annual nitrate concentrations

of low temperatures. Large variability of nitrate concentration over the years or seasons within the year can be explained by the combination of the previously mentioned factors, especially in Prigorje.

In this study, the Mann–Kendall trend test shows that there is no statistically significant trend in nitrate concentrations at both locations while some researchers showed different results. Nitrate increase in the 5-year research period was reported by Obeidat et al. (2007). Perennial monitoring of nitrate concentration in agricultural area was undertaken by Gardner and Vogel (2005). They reported higher nitrate concentrations in the research area than in any other areas. Increase of nitrate concentration in aquifer was reported by Xu et al. (2007) and it was explained as a result of increased nitrate content during the monitoring period. Drake and Bauder (2005) also reported increase of nitrate content through decades. Increase was especially noticeable on easy permeable soils and locations with high population density and numerous septic tanks. However, they highlight that nitrate concentration did not significantly increase in the whole monitored area, which is similar to the results reported in this study. In the Podravina region, the nitrate content did not change significantly through time and was considered to be constant while the Prigorje region showed increased variability which causes a slight increase of concentration, but not statistically significant. Woo-Jung et al. (2007) found over the period of 3 years an increase of nitrate concentration and proportion of samples above MAV by monitoring the areas with different agricultural activities. The lowest nitrate concentration increase was reported in natural areas, then in areas with crops, and the highest increase was found in the areas with crops and farms, which was assumed to be the cause of nitrate concentration increase in Prigorje region as well.

In conclusion, high average nitrate content was found in the Prigorje region (53.9 mg/l), while in the Podravina region, the average nitrate content was lower than 24.9 mg/l. Categorisation of wells showed that in the Prigorje region 40 % of the wells showed average concentration above MAV, i.e. category high, while in Podravina 11 % of the wells belong to the category high. The distance of point sources of contamination within 10 m contributes greatly to deterioration of water quality in respect to nitrate concentration from shallow aquifer. There is no statistically significant difference over the years or seasons within the year. Nitrate concentrations' trend was not significant during the monitoring. By

comparing water quality in respect to nitrate concentration in two different areas of the County, worse water quality was found in the area of Prigorje region than in the Podravina region. If no preventive measures are taken in Prigorje, there is a further danger of water contamination from point sources.

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