

# Road salts effects on soil chemical and microbial properties at grassland and forest site in protected natural areas

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## ABSTRACT

Road salting is used as a dominant way to keep road safety in winter, even in the protected natural areas. In our study, possible effects of winter road salting on soil microorganisms in close road vicinity were investigated. Soil chemical and microbial properties were monitored at a forest site in the Krkonoše Mountains national park and at a grassland site in the Kokořínsko protected landscape area (both located in the Czech Republic) in two sampling campaigns (autumn and spring). Effects of road salting on soil chemical properties ( $\text{Na}^+$  and  $\text{Cl}^-$  levels, pH, base saturation etc.) were clearly apparent at both sites. The most affected plots were 1 and 5 m from the road (increased pH, base saturation, and  $\text{Na}^+$  accumulation). At these plots, changes of microbial parameters were observed in both autumn and spring sampling, which suggested influence of salts. Increased value of metabolic quotient ( $q\text{CO}_2$ ) indicated stress and potential ammonification was inhibited even 5 m from the road at the forest site. Hence, possible influence on soil biological quality should be considered when assessing the ecological risks of this kind of road treatment, especially in natural protected areas.

**Keywords:** soil microbial biomass; soil respiration; ammonification; road salting; soil chemistry

Snow and ice removal from roads is definitely necessary to improve safety of traffic in winter periods. Sodium chloride ( $\text{NaCl}$ ) is the most common deicing agent due to low price, easy availability, storage and handling. Despite all benefits for traffic safety, there are also many adverse effects, especially related to increased levels of chloride and sodium ions in the surrounding environment.  $\text{NaCl}$  from road runoff is responsible for increased salinity or osmolality of surface and ground waters (Thunqvist 2004) and negative effects on freshwater organisms are frequently reported (Benbow and Merritt 2004, Environment Canada 2004, Ramakrishna and Viraraghavatan 2005).

In soil environment, salt transport and effects depend on a variety of factors and local conditions, such as roadside slope, soil type and texture, and vegetation cover (Ramakrishna and Viraraghavatan 2005). Generally, the most affected area is up to 5–10 m from the roadside (Lundmark and Olofsson 2007, Zehetner et al. 2009). Effects of  $\text{NaCl}$  ad-

ditions on soil chemistry are serious: increasing amounts of  $\text{Na}^+$  and  $\text{Cl}^-$  affect soil pH, soil structure, permeability, hydraulic conductivity, air conditions, and osmotic potential. This leads to a loss of soil stability, fertility and to osmotic stress for vegetation and soil macro- and microorganisms. Several studies reported negative effects of road salts on vegetation (Butler and Addison 2000, Czerniawska-Kusza et al. 2004).

Only few studies have dealt with the impact of deicing salts on intrinsic soil microorganisms although they are crucial in the maintenance of quality and fertility of soils. Therefore, the aim of our study was to investigate effects of road salting on soil microorganisms at two sites with different vegetation cover and to relate the microbial biomass changes to the soil chemistry conditions. Two sampling campaigns (spring and autumn) can help to decide if the effects are only temporary (spring sampling after snowmelt) or persistent (autumn sampling after the most salt is washed out).

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## MATERIAL AND METHODS

The sampling sites were situated in the Czech Republic, in the Kokořínsko protected landscape area (KOK site) and in the Krkonoše Mountains national park (KRK site), nearby the roads with very intensive traffic. Both roads are intensively treated by salting during winters (about 6.6 and 13 t/km each winter at the KOK site and KRK site, respectively). Sampling plots were placed at the transect lines perpendicular to the roads. At both sites, sampling plots closest to the road (1 m) were at the roadside slope. They were typical Anthrosols, covered by grass and they were sampled from 0–15 cm depth. At the KOK site, soils (Gleyic Fluvisol) were sampled at distances of 5, 10, and 15 m from horizontal meadow (mowed reed) below the road. Soil was sampled from two layers: 0–5 cm and 5–15 cm. At the KRK site, soils (Haplic Cambisol) were sampled at distances 5 and 20 m at the slope below the road and also 20 m at the slope above the road (as control). All plots at the KRK site were covered by spruce-beech forest and soil was sampled from fermentation organic top horizons (Of; 1–5 cm thick) and from humic organic top horizons (Oh; 2–7 cm thick). Sampling was performed in autumn 2009 and spring 2010. Soils were sampled as 3–5 sub-samples at each plot which were then mixed. Soil samples were manipulated and stored in accordance with the ISO 10381-6. For the microbiological analyses, fresh soils were sieved (2 mm) and stored at 4°C in darkness with air exchange. For chemical analyses, soils were air-dried at laboratory temperature and sieved (2 mm).

Several chemical properties of soil samples were measured in accredited commercial laboratory: organic carbon content –  $C_{org}$  (ISO 14235), total nitrogen –  $N_{tot}$  (ISO 11261), soil pH active –  $pH(H_2O)$  and exchangeable –  $pH_{KCl}$  (ISO 10390), concentrations of chlorides ( $Cl^-$ ) in soil water extract 1:5 (ISO 9297), concentration of cations  $Ca^{2+}$ ,  $K^+$ ,  $Mg^{2+}$ ,  $Na^+$ , cation exchange capacity, and base saturation – BS in three types of soil extracts: water 1:5 (active concentration), in Mehlich III extracts (potential concentration), and 0.1 mol/L  $BaCl_2$  extracts according to Gillman (exchangeable concentration) (ISO 13536, Zbíral et al. 2004).

Three basic microbial parameters of soils were measured: microbial biomass content ( $C_{bio}$ ) by the chloroform fumigation extraction method according to the ISO 14240-2, basal (BR) and substrate-induced (SIR) respiration by gas chromatography according to the ISO 16072 and ISO 14240-1. In spring 2010 also potential ammoni-

fication (PAMO) was measured according to Alef and Kleiner (1987). All microbial measurements were carried out in triplicates. From the measured parameters, eco-physiological coefficients were calculated – microbial coefficient  $C_{bio}/C_{org}$  and metabolic quotient  $qCO_2$  – as  $BR/C_{bio}$  (Anderson and Domsch 1989, 1993) and also  $qCO_2$ -SIR as  $SIR/C_{bio}$  (Dilly 2006).

For each layer, statistical analyses (Statistica for Windows 10, StatSoft, Inc., Tulsa, USA) of effect of distance from the road and differences between plots were performed by the analysis of variance ANOVA followed by the Tukey's test. Trends were tested by linear regression with least squares method. Layers were compared at each plot by the Student *t*-test.

## RESULTS AND DISCUSSION

Effects of road salting on soil chemistry were clearly apparent at both sites (Table 1). The most apparent effects were detected at 1 m, rarely at 5 m distance from the road, which confirmed the literature findings (Lundmark and Olofsson 2007, Zehetner et al. 2009) that effects of road salting on soil chemistry are mostly limited to the distance up to 10 m.

At both sites, pH values were apparently increased at 1 m plot. At the KOK site, pH was continuously decreasing with increasing distance from the road. At the KRK site, pH dropped to naturally acidic values at 5 m with no further change. At 1 m plot,  $Na^+$  concentrations were several times higher than at more distant plots, at both sites in all types of extracts. At the KRK site, strong increase of  $Na^+$  was also clearly apparent at 5 m from the road when compared with 20 m plot or control. From increased  $Na^+$  levels, increased BS might be expected, because this relationship was reported several times for salinized soils (Czerniawska-Kusza et al. 2004, Ramakrishna and Viraraghavatan 2005). This was also confirmed by our results: increased BS (both in  $BaCl_2$  and Mehlich III) at 1 m plot was observed when compared to more distant plots. However, this was much more evident at the KRK site, because forest soil was naturally less base-saturated. There was continuous BS decrease with increasing distance from the road at the KOK site. Increase of  $Na^+$  and pH were stronger in spring sampling which indicates actual influence after winter road maintenance. On the other hand, BS changes were similar in autumn and spring which indicated that the changes in soil sorption

Table 1. Soil chemical properties at Kokořínsko protected landscape (KOK) and Krkonoše natural park (KRK) sites in autumn 2009 and spring 2010

Site	Sam- pling time	Distance from the road (m)	Layer	C <sub>org</sub>	N <sub>tot</sub>	Water 1:5				Gilman (0.1 mol/L BaCl <sub>2</sub> )				Mehlich III									
						pH <sub>H<sub>2</sub>O</sub>	pH <sub>KCl</sub>	Cl <sup>-</sup>	Na <sup>+</sup>	Ca <sup>2+</sup>	Na <sup>+</sup>	K <sup>+</sup>	Ca <sup>2+</sup>	Na <sup>+</sup>	K <sup>+</sup>	Ca <sup>2+</sup>	Mg <sup>2+</sup>	BS	CEC	BS	CEC		
				(%)				(mg/kg)				(meq/kg)				(%)							
KOK	autumn	1		5.3	0.33	7.4	6.6	18	153	95	184	230	3580	199	204	98	490	466	4383	192	277	97	
		5		5.1	0.50	7.2	6.8	100	121	226	148	78	5260	220	293	97	164	124	5052	184	293	95	
		10	0-5	5.4	0.59	6.6	6.4	153	137	149	196	47	5160	225	292	95	367	77	4427	415	311	88	
	spring	15		7.5	0.76	6.5	6.1	185	114	251	152	94	6060	349	341	98	122	151	5875	271	365	89	
		5		3.6	0.34	7.2	6.9	84	115	175	78	27	4520	183	248	97	88	46	4540	147	253	96	
		10	5-15	4.0	0.36	7.1	6.7	91	98	94	72	23	3480	158	230	82	223	30	3424	144	210	92	
	KRK	autumn	15		3.1	0.38	6.5	6.0	86	48	101	120	35	3980	199	245	88	268	36	3728	187	249	86
			1		4.0	0.31	7.7	7.1	19	318	98	347	293	4920	88	303	87	616	600	4319	181	283	96
			5		2.8	0.56	7.3	6.9	195	113	472	53	109	8180	183	399	107	95	237	6133	184	352	94
		spring	10	0-5	5.4	0.60	6.9	6.5	51	91	208	43	43	6840	208	365	99	40	112	4668	195	290	88
			15		9.6	0.88	6.6	6.3	263	88	448	155	109	8040	279	412	104	104	231	6259	165	381	88
			5		2.4	0.45	7.5	7.0	48	96	287	65	12	6760	158	356	99	92	79	5517	147	311	94
		autumn	10	5-15	3.8	0.40	6.7	6.4	39	82	131	36	< 4	4880	140	310	82	29	51	3423	135	212	87
			15		5.5	0.51	6.4	6.0	52	62	149	130	< 4	5240	151	245	112	< 1	52	3737	153	234	85
			1		4.3	0.22	7.4	6.3	16	154	23	428	31	1716	336	129	89	477	42	1310	304	119	94
KRK		autumn	5		36.2	1.46	3.7	2.8	77	119	85	146	98	1314	284	207	46	242	207	909	241	308	26
			20	Of	40.4	1.79	3.6	2.9	47	18	52	10	74	1066	184	152	46	133	253	747	155	354	18
			C		36.5	1.68	4.5	3.5	56	22	60	116	129	276	93	230	13	235	249	287	92	292	13
		spring	5		19.8	0.95	3.8	2.9	73	136	222	112	51	370	113	174	19	373	100	327	103	164	27
			20	Oh	22.3	1.12	3.6	2.7	16	17	67	4	55	326	90	188	13	80	118	319	81	233	12
			C		28.7	1.46	4.0	3.5	24	16	41	92	62	366	79	199	15	139	118	257	76	186	15
	autumn	1		4.1	0.29	7.8	6.6	NA	309	33	904	< 4	954	151	106	68	1230	72	1032	176	131	92	
		5		41.7	1.71	4.4	3.8	103	230	162	352	168	2820	351	290	62	341	336	2036	359	379	41	
		20	Of	42.6	0.91	4.0	3.6	74	46	143	10	203	1476	201	258	37	< 1	436	1192	199	426	20	
	spring	C		42.9	1.73	5.9	5.2	78	51	166	< 2	230	678	118	244	20	< 1	436	647	129	337	16	
		5		27.3	0.60	3.9	3.0	84	250	103	304	66	644	201	299	20	334	176	575	166	283	22	
		20	Oh	23.0	0.55	3.5	2.7	30	30	116	< 2	59	384	90	288	10	< 1	164	467	78	263	13	
	C		33.6	1.78	4.1	3.3	90	115	109	113	152	310	103	296	11	< 1	316	445	119	318	13		

properties are rather persistent. In general,  $\text{Na}^+$  levels as well as BS were lower or same in deeper soil layer at the most plots.

Chlorides showed strongly increased levels at plots further from the road which confirms they are more mobile in the soil than  $\text{Na}^+$ . Increased levels were apparent at 5 m plot at the KRK site when compared to the control plot and 20 m plot. Levels of chlorides at the KOK site increased generally with the increasing distance from the road with exception of 10 m plot in spring. Chloride content was generally lower in deeper soil layers.

Chloride and  $\text{Na}^+$  levels at both sites and most of the plots might be alarming; it was reported that plant damage may occur at 16 mg/kg  $\text{Na}^+$  and 30 mg/kg  $\text{Cl}^-$  (Environment Canada 2004).

$\text{C}_{\text{org}}$  and  $\text{N}_{\text{tot}}$  were apparently increasing with increasing distance from the road at the KOK site. This is probably not attributed to the road salting but this behavior should be considered when interpreting soil microbial parameters.

Soil microbial parameters were under focus in our study to observe if winter road salting can negatively affect soil microbial communities. It was found, that generally microbial parameters showed differences between sampling plots and some interesting trends related to the distance from the road (Figures 1 and 2).

At both sites, microbial biomass ( $\text{C}_{\text{bio}}$ ) was always significantly higher at more distant plots than at 1 m plot. While this observation was found only for top layer at the KOK site, it was valid for both layers at the KRK site. Significant positive correla-

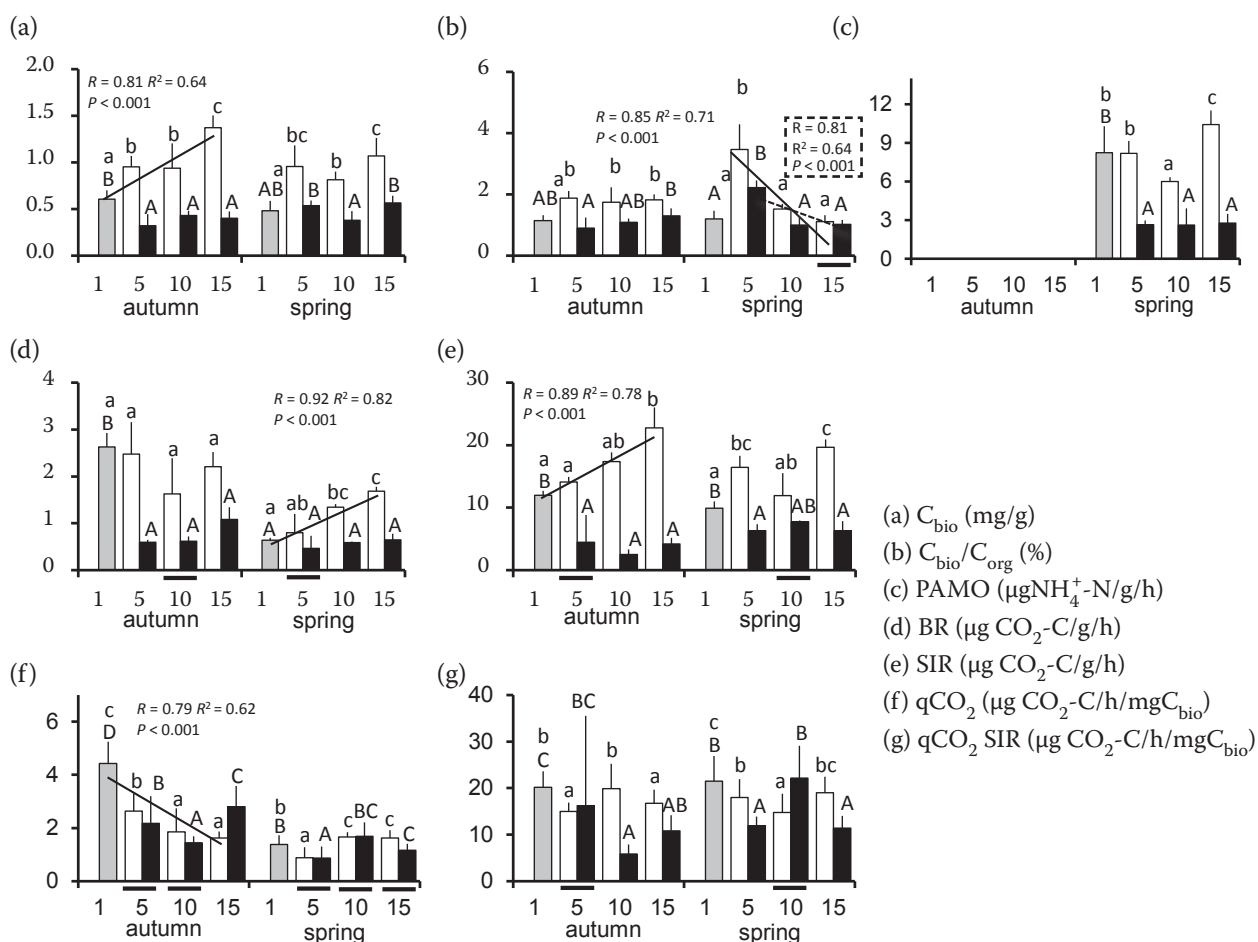


Figure 1. Soil microbial properties at Kokořínsko protected landscape (KOK) site in autumn 2009 and spring 2010. Numbers below the columns show the distance from the road in meters. Grey column shows the 1 m plot (0–15 cm layer). White and black columns show layers of 0–5 cm and 5–15 cm, respectively. Bars show standard deviations. Same letters above the columns indicate an insignificant ( $P > 0.05$ ) difference between plots based on the Tukey's test. Small or capital letters are used for the top or deeper layer, respectively. Thick line below couple of white and black columns indicates an insignificant ( $P > 0.05$ ) difference between the layers based on the Student  $t$ -test. Skew lines and text boxes show linear regression of parameter versus distance, where significant (solid or dashed line and box border are used for the top or deeper layer, respectively)

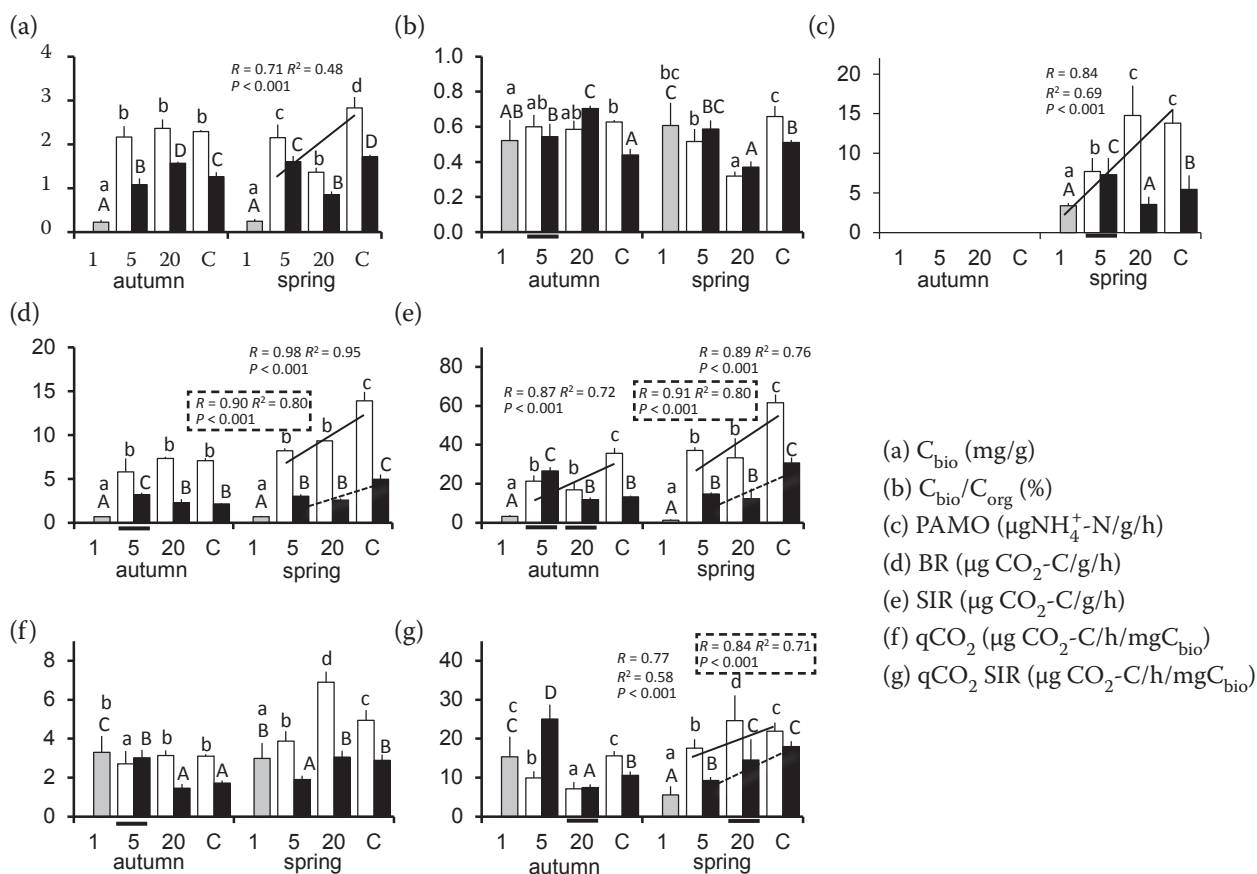


Figure 2. Soil microbial properties at Krkonoše natural park (Krk) site in autumn 2009 and spring 2010. Numbers below the columns show the distance from the road in meters and C is the control plot. Grey column shows the 1 m plot (0–15 cm layer). White and black columns show Of and Oh horizons, respectively. Bars show standard deviations. Same letters above the columns indicate an insignificant ( $P > 0.05$ ) difference between plots based on the Tukey's test. Small or capital letter are used for the top or deeper layer, respectively. Thick line below couple of white and black columns indicates an insignificant ( $P > 0.05$ ) difference between the layers based on the Student  $t$ -test. Skew lines and text boxes show linear regression of parameter versus distance, where significant (solid or dashed line and box border are used for the top or deeper layer, respectively)

tion with the distance was observed at the KOK site in autumn ( $R = 0.81$ ) and at the Krk site in spring ( $R = 0.71$ ). These trends must be interpreted carefully, because soil microbial biomass is strongly influenced by organic carbon in soil ( $C_{org}$ ), which may mask the effects of salting. To eliminate the impact of  $C_{org}$  on the observed trends, microbial quotient  $C_{bio}/C_{org}$  was calculated. Significantly lower values of  $C_{bio}/C_{org}$  at 1 m plot than at further plots were found at the KOK site in autumn and spring. At the Krk site, the microbial quotient was not systematically changing with the distance. A significant decrease of  $C_{bio}/C_{org}$  from 5 m plot to 15 m plot at the KOK site in spring was found in both layers ( $R = 0.85$  and  $R = 0.81$ ), but this was probably caused by  $C_{org}$  trend at this site (Table 1). At the Krk site in spring, strongly decreased value was observed at 20 m plot from unknown reasons. Decreased values of the microbial quotient indicate

a stress situation (Anderson and Domsch 1989), which might be the case at 1 m plot at the KOK site in our study.

With only exception of basal respiration at the KOK site in autumn, both basal respiration (BR) and substrate induced respiration (SIR) showed a significant increase of the values with the increasing distance from the road or, at least, significantly higher values at further plots than at 1 m plot (Figures 1 and 2). These trends were not apparent for deeper layers with exception of the Krk site in spring (significant positive correlation with increasing distance from the road;  $R = 0.91$ ). The observed trends for BR and SIR could originate from trends of  $C_{bio}$ . To avoid this influence, biomass-specific respiration was calculated – metabolic quotient  $qCO_2$  and  $qCO_2$ -SIR. They showed much less clear relationships with the distance from the road in most cases (Figures 1 and 2), which confirms that



$C_{\text{bio}}$  was partially responsible for trends of BR and SIR. This was not the case for SIR at the KRK site in spring, where trends of SIR were confirmed. Interesting observation was found for  $q\text{CO}_2$  at the KOK site in autumn: an increase of the values with decreasing distance to the road ( $R = 0.79$ ). This might indicate stress from road salting, because an increase of the  $q\text{CO}_2$  values was reported in literature as indication of higher maintenance energy of soil microorganisms induced by stress conditions (Anderson and Domsch 1993).

Potential ammonification (PAMO) as a marker of the important process of N-transformation in soil showed a significant increase with increasing distance ( $R = 0.84$ ) at the KRK site (Figure 2). Significantly decreased PAMO was observed at 5 m plot when compared with the control or 20 m plot. This might be an inhibition due to increased  $\text{Na}^+$  level and consequent changes in soil chemistry. However, this trend was not found for PAMO in deeper layers.

A negative impact of soil salinization on soil microbial characteristic (biomass,  $q\text{CO}_2$ ) was observed also in other studies. Yuan et al. (2007) reported that higher salinity in arid soil in China resulted in a smaller, more stressed, microbial community, which was less metabolically efficient. Soil nitrification was significantly reduced at a concentration of about 100 mg/kg sodium and 150 mg/kg chlorides in study of Butler and Addison (2000). Černohlávková et al. (2008) reported a significantly reduced microbial biomass and respiration activity at the roadside with increased levels of  $\text{Na}^+$  (up to 100 mg/kg) and pH (up to 8).

Effects of road salting on soil microbes were also confirmed by our results. In several cases, microbial biomass or/and its activities increased significantly with increasing distance from the road and in one case there was a stress indication by eco-physiological coefficient  $q\text{CO}_2$ . Some changes might indicate long-term effects (decrease of microbial biomass and organic carbon in soil) and other changes short-term effects (changes in PAMO or stress indication by  $q\text{CO}_2$ ). Although other case studies are necessary, it is now apparent that soil microbial communities must be considered when ecological effects of road salting are assessed.

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