EFFECT OF N FERTILIZER FORMS AND SOIL MOISTURE LEVELS ON THE N GASEOUS LOSSES

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Abstract. In a pot experiment using brown forest soil with clay eluviation (40 kg soil/pot) from Keszthely the gaseous losses of nitrogen (total N, N_2, N_2O NO NO_2) caused by denitrification were tested. Gas-collecting traps were placed at a depth of 20 cm in the soil. At the same fertilizer N input (6250mgN/pot, 150mg N/kg soil), the effects of two fertilizer forms (KNO_3, NH_4Cl) and two soil moisture levels (field water holding capacity, WHC= 65% and 100%) were evaluated to the gaseous losses of nitrogen with and without sowing of maize hybrids (two Stira and two Mv 355/pot) as test plants. The composition of the N-containing gases was determined by gas chromatography. From the soil-atmosphere the gas samples were taken 8 times, the sampling times were on the 2, 16, 30, 54, 68, 82, 110, 138-th days of the experiment. Statistical evaluation of the results was performed by analysis of variance. The N gaseous loss in the planted pots was the 12% of the applied fertilizer dose, while in unplanted pots it was nearly the double of it (22%) Consequently, there was a 50% decrease in the denitrification gaseous losses of fertilizer N due to plant N uptake. The N-gaseous loss was significantly higher in cases of KNO_3 applications and at the WHC=100% soil moisture than in NH_4Cl treatments and at the WHC= 65%, probably due to more intensive denitrification. In the unplanted pots the amount of N_2 and N_2O increased the most (by 95% and by 70%) compared with planted pots. It can be concluded that the improperly applied fertilizer causes significant GHG emissions in the absence of plants.

Keywords: *N-gaseous losses, nitrous-oxide, GHG emission, fertilizer treatment, soil moisture*

Introduction

The relative contributions of the three main greenhouse gases (GHG): CO_2, CH_4 and N_2O to overall global radiative forcing are 63.5%, 18.1% and 6.2%, respectively. Soils are one of the major sources of GHG contribution both to anthropogenic emissions due to land use change and agricultural management (up to 22.5% of all anthropogenic sources) (Blagodatsky and Smith, 2012). Yienger and Levy (1995) estimate for annual above-canopy emissions is 5.5Tg N (NO_x) with a range of 3.3-7.7 Tg N. Globally the strongest emitters are agriculture, grasslands, and tropical rain forests, accounting for 41%, 35% and 16% of the annual budget, respectively."Pulsing" (the emissions burst following the wetting of a dry soil) contributes 1,3 Tg N annually. In temperate regions, agriculture dominates emission, and in tropical regions, grassland dominates. By the year 2025, the increasing use of nitrogen fertilizer may raise total annual emissions to 6.9 Tg N with agriculture accounting for more than 50% of the global source. The agriculture is a significant contributor to the increasing N_2O concentration of air (Delmas et al., 1997; Hellebrand et al.,2003; Ruser et al., 2006). The agriculture is estimated to contribute more than two-thirds of total anthropogenic N_2O emissions (Pattey et al., 2007). The agricultural soils emit 3.3 Mt N_2O per year (Mosier et al.,1998). As the global warming potential of N_2O is 298 times as strong as that of CO_2, N_2O emissions from soils thereby exert an important impact on the environment. Soil microbial processes, nitrification and denitrification contribute significantly to the agricultural N_2O emission (Stange and Döhling, 2005). N-gaseous emissions from agricultural soils, speciation of predicted gas N flux into N_2O and N_2 depend on the N-fertilizer dose and type, the soil type and pH, the soil moisture, the oxygen supply and the C/N ratio of the soil, application practice, the vegetation and the temperature (Ma et al., 2007; Chu et al., 2007, Gu et al, 2009). Field response to fertilizaton is variable; some plots have enhanced emissions for prolonged periods, whereas others have sharper initial increases that decay over time. In general, however a positive linear correlation seems to be between fertilizer use and emission, and over the course of a growing season, total NO_x emissions change between 1-10% of added nitrogen fertilizer (Williams et al., 1992; Shepard et al., 1991, Cardenas et al., 1993). The exact fertilizer form which is more stimulatory depends on the site, although evidence suggests that a mixed form, such as NH_4NO_3, generates strongest emission (Sanhueza, 1992). In a pot experiment the total N gaseous losses were higher at KNO_3 treatment than NH_4Cl treatment in brown forest soil with clay eluviation (Debreczeni et al. 1998). The N_2O emissions from NH_4^+-N fertilizer were higher than from urea and NO_3^--N fertilizers in coarse- textured soils (Gu et al, 2009). Kramer et al.(1999) studied greenhouse gas emissions of Dutch agricultural crop production by using a life cycle approach. It was determined that the emissions of N_2O are mainly caused by the production and application of synthetic nitrogen fertilizer. The use of other nitrogen sources manure, compost or limiting the use of synthetic nitrogen could positively influence the emissions of N_2O. In contrast, Jones et al. (2005) found that during growing season the cumulative total N_2O flux from manure treatments was 25 times larger than that from mineral fertilizers. Large inputs of manure N have a potential to contribute a very high N_2O emissions, particularly in wet soil conditions. The ratio of NO_x gases depends on the soil moisture. In an experiment testing the tropical rain forest ecosystem during dry periods the NO-N: N_2O-N ratio was as high as 60:1, whereas for wetter periods it decreased to <7:1. (Butterbach et al, 2004). In Hungary, where the proportion of agricultural area to the total area of the country is quite high (63%) it is essential to investigate agricultural contribution to greenhouse gas emission.

Material and methods

Brown forest soil with clay eluviation from Keszthely (pH_{KCl} =7.7, C=1.1 %, NO_3-N = 17.5 mg kg^{-1}, NH_4-N = 12 mg kg^{-1}, humus = 1.9 %) was filled in large plastic pots (40 kg soil/pot). N fertilizers (KNO_3, NH_4Cl) were applied at a high rate of approx. 150 mg N kg^{-1} soil (6 250 mg N/pot) to trace the precise transformation processes of N, P fertilizer in all treated soils, and K fertilizer in NH_4Cl treatments was also used (N:P:K= 1:1:3.35). All experiments were carried out at two soil moisture levels (field water holding capacity, WHC= 65 % and 100 %) with and without sowing of maize as test plants (two Stira and two Mv355 plants/pot) (Table1). For statistical calculations, results of maize hybrids were taken as replicates. The plants were grown until full ripening. Maize (*Zea mays L.*) was sown in 48 pots, and 32 pots were left without plant.

***Table 1**. Setting up of the experiment*

		With maize						**Without maize**					
Maize hybrid	**Treatment**	**Control**		**KNO_3+P**		**NH_4Cl+PK**		**Control**		**KNO_3+P**		**NH_4Cl+PK**	
Stira	**WHC %**	**100**	**65**	**100**	**65**	**100**	**65**	**100**	**65**	**100**	**65**	**100**	**65**
MV 355													

All treatments were replicated four times. Gas-collecting traps of 1800 cm^3 capacity with silicon pipe outlets were placed at a depth of 20 cm in the soil. The traps collected the N gaseous losses (NO, NO_2, N_2O, N_2) released during the denitrification process. Gas samples were taken 8 times by syringe (5 cm^3) in 3 replicates from the soil atmosphere. For soil atmosphere analysis the composition of the N-containing gases (N_2, NO, NO_2, N_2O) was determined by a Carlo Erba 2350 type gas chromatograph. From these data the total gaseous N losses were calculated (mg N /pot or mg N/1800 cm^3) for each sampling point. At the end of the experiments the cumulative gaseous N losses and N_2O-N losses were also calculated by the difference method (N content difference between treated and control gas samples) and by the calculations of the relative % gaseous N losses. Results were analyzed by the means of analysis of variance (MANOVA). The statistical analyses were performed with a Microsoft Excel Macro (Aydinalp et al., 2008; Vágó et al., 2008)

Results and discussion

The total N and total N_2O gaseous losses were found to depend significantly ($LSD_{5\%}$=16,7 and 5,1) on the presence or absence of the plants (*Table 3*). In control samples in the planted pots the total N gaseous losses were 1,4 times higher (at 65% and 100% WHC), the N_2O gaseous losses were 1,6 times (at 65%WHC) and 1,9 times (at 100 %WHC) higher than in the unplanted pots (*Table 2*). Similar results were obtained by Heltai et al. (2013) in a pot experiment in control samples. In contrast to the control, in the unplanted pots treated with fertilizer the total N and N_2O gaseous losses were higher (1,4-1,7 times) than in the planted pots. Calculated by difference method the total N gaseous losses were higher in the unplanted pots by 78-84% at KNO_3 treatment and by 71-73% at NH_4Cl treatment while the total N_2O gaseous losses were higher by 67-72% and by 62-63% compared with planted pots.

The total N and total N_2O gaseous losses were significantly higher in the N-fertilizer treatments than in control samples (*Table 3*: $LSD_{5\%}$=20,5 and 6,3). The difference of total N gaseous losses was 7 times higher in KNO_3 and NH_4Cl treatment in the planted pots while in the unplanted pots it was 16-17 times higher than in the controls (*Table 2*). The difference of N_2O gaseous losses was 5-6 times higher in the planted pots and 14-15 times higher in the unplanted pots in fertilized pots than in the controls. The total N and total N_2O gaseous losses were more intensive in case of KNO_3 treatment than in NH_4Cl treatment (*Table 3:* $LSD_{5\%}$=20,5 and 6,3).

***Table 2.** Total N and total N_2O gaseous losses (Nmg/$1800cm^3$)(+ with plant, - without plant)*

Treatment	**Maize**	**WHC %**	**$\sum N$**	**$\sum N_2O$**
Control	+	**100**	**131,5**	**20**
		65	**119,5**	**16,5**
KNO_3	+	**100**	**921**	**107**
		65	**868,5**	**96**
NH_4Cl	+	**100**	**907,5**	**102**
		65	**868,5**	**93,5**
Control	-	**100**	**91,5**	**10,5**
		65	**85,5**	**10**
KNO_3	-	**100**	**1494**	**156**
		65	**1461,5**	**146,5**
NH_4Cl	-	**100**	**1438**	**144,5**
		65	**1369**	**135**

***Table 3.** Analysis of variance of total N and total N_2O gaseous losses according to plant presence, soil moisture and treatment (N and N_2O mg/$1800cm^3$)*

Plant presence	**$\sum N$**	**$\sum N_2O$**
Maize +	636	73
Maize -	990	100
$LSD_{5\%}$	**16,7**	**5,1**

Soil moisture (WHC%)	**$\sum N$**	**$\sum N_2O$**
100%	831	90
65%	795	83
$LSD_{5\%}$	**16,7**	**5,1**

Treatment	**$\sum N$**	**$\sum N_2O$**
Control	107	14,3
KNO_3	1186	126
NH_4Cl	1146	119
$LSD_{5\%}$	**20,5**	**6,3**

The difference was significant in the unplanted pots only (*Table 4:* $LSD_{5\%}$=29 and 8,8). Similar results were obtained in other pot experiments using same soil samples from Keszthely by Debreczeni et al.(1995, 1998) and Nótás et al. (2003).

***Table 4.** Analysis of variance of total N and total N_2O gaseous losses according to plant presence x treatment, plant presence x soil moisture and treatment x soil moisture (N and N_2O mg/$1800cm^3$)*

Plant presence X Treatment		**$\sum N$**	**$\sum N_2O$**
Maize +	Control	126	18,3
	KNO_3	895	102
	NH_4Cl	888	98
Maize -	Control	89	10,3
	KNO_3	1478	151
	NH_4Cl	1404	140
$LSD_{5\%}$		**29**	**8,8**

Plant presence X Soil moisture		**$\sum N$**	**$\sum N_2O$**
Maize +	100%	653	76,3
	65%	619	68,7
Maize -	100%	1008	103,7
	65%	972	97,2
$LSD_{5\%}$		**23,6**	**7,2**

Treatment X Soil moisture		**$\sum N$**	**$\sum N_2O$**
Control	100%	111,5	15
	65%	102,5	13
KNO_3	100%	1207	132
	65%	1165	121
NH_4Cl	100%	1173	123
	65%	1119	114
$LSD_{5\%}$		**29**	**8,8**

The total N and total N_2O gaseous losses were higher at 100%WHC than 65%WHC (*Table 3*: $LSD_{5\%}$=16,7 and 5,1). This difference of total N loss was equally significant in the planted and unplanted pots (*Table 4*: $LSD_{5\%}$= 23,6), but the total N_2O gaseous loss was significantly higher only in the planted pots (*Table 4*: $LSD_{5\%}$= 7,2). The effect of the soil moisture was significant in the fertilized samples only, in the control samples it was not (*Table 4:* $LSD_{5\%}$=29 and 8,8). The differences were dependant on the presence or absence of the plants in control samples 7-10% and 5-20%, these are not significant; while in KNO_3 treatment 2-6% and 7-12%, in NH_4Cl treatment 5% and 7-9%, these are significant.

Figure 1 and *2* (on the basis of *Table 5*) show the rate of total N gaseous losses increase in the planted pots (*Fig. 1*) and in the unplanted pots (*Fig. 2*). Results of maize hybrids were averaged.

Table 5. *Total N gaseous losses in the sampling time, ∑Nmg/1800cm3 (- unplanted pots)*

Treatment	**Maize hybrid**	**WHC %**	**Sampling time (day)**							
			2.	**16.**	**30.**	**54.**	**68.**	**82.**	**110.**	**138.**
Control	**Stira**	**100**	47	70	96	126	127	130	123	122
		65	37	62	7	108	113	122	112	115
	Mv 355	**100**	48	71	91	126	124	133	121	116
		65	31	65	80	101	105	117	100	109
KNO_3	**Stira**	**100**	150	344	480	616	719	931	918	876
		65	110	305	448	587	619	848	836	844
	Mv 355	**100**	124	365	504	615	743	911	903	863
		65	116	337	474	589	692	889	858	874
NH_4Cl	**Stira**	**100**	121	337	475	589	604	918	894	855
		65	107	297	426	546	587	846	827	844
	Mv 355	**100**	119	333	492	595	701	893	897	870
		65	102	303	475	579	624	850	839	891
Control	**-**	**100**	40	47	58	76	84	94	86	89
		65	38	40	52	78	75	89	73	82
KNO_3	**-**	**100**	108	345	651	911	971	1156	1494	1493
		65	101	304	614	829	895	1125	1343	1473
NH_4Cl	**-**	**100**	108	342	658	891	956	1114	1415	1461
		65	93	300	614	824	852	1096	1352	1386

It can be established that the sampling time significantly influences the production rates of total N gaseous losses. The change of total N gaseous losses growth can be characterized by two peaks, in the planted pots *(Fig.1)* on the 16th and 82nd day of the experiment and in the unplanted pots *(Fig.2)* later, on the 30th and 110th day. This differerence was probably caused by the increase of microbiological activity due to the effect of root secretion in the planted pots. Similar results were obtained in other pot experiments using the same soil type from Keszthely Debreczeni et al.(1995). The minimum values were received on the 68th day in the planted and unplanted pots equally. The N gaseous emission fluctuations may be caused by periodic changes of soil bacteria's microbiological activity, life cycle and temperature.

In our experiment with maize the total N gaseous losses changed between 749-789 mgN/1800 cm^3 in the planted pots and between 1304-1408 mgN/1800 cm^3 in the unplanted pots (*Fig. 3*). The total N gaseous losses in the planted pots were 12-13% of the applied fertilizer N while in the pots without maize it was about twice as much, 21-22%.

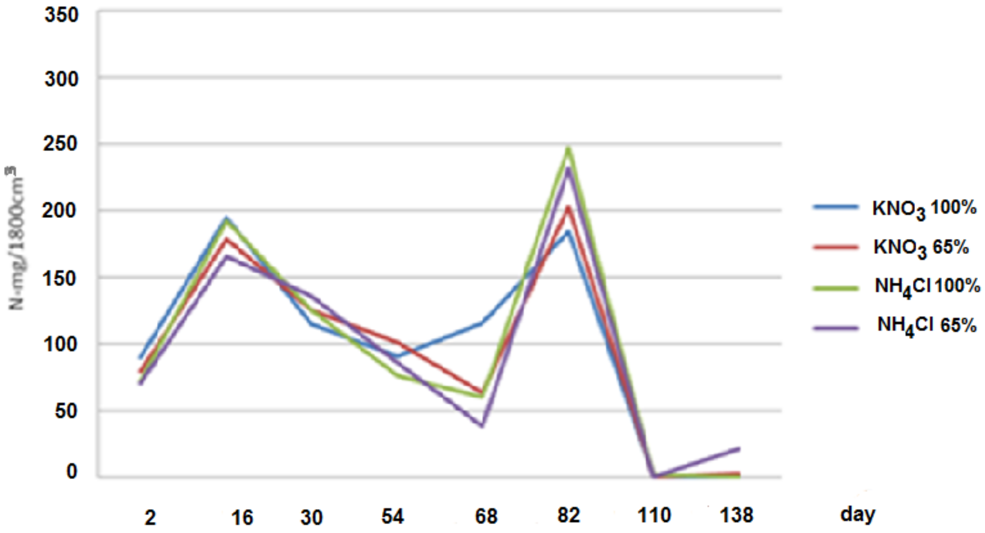

Figure 1. *The change of total N gaseous losses increase in the planted pots in the function of sampling time*

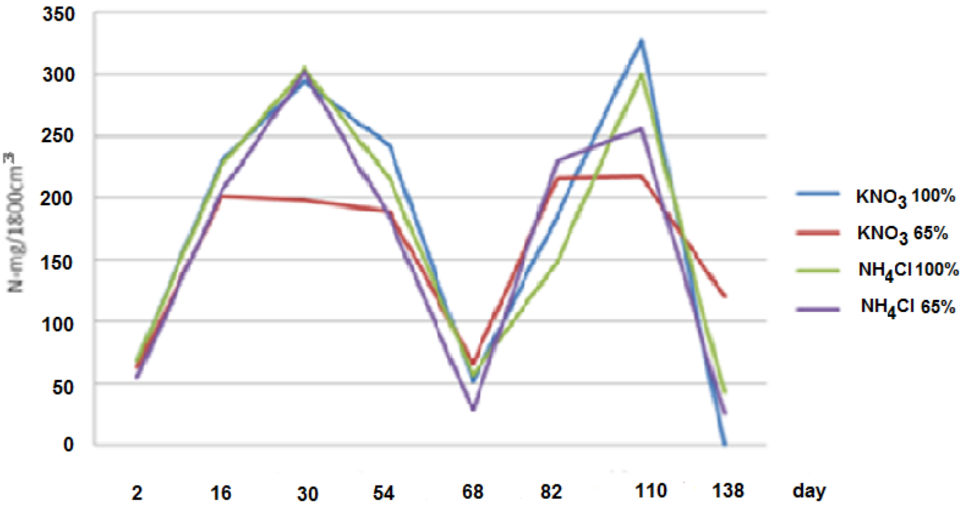

Figure 2*. The change of total N gaseous losses increase in the unplanted pots*

The change of the relative total N gaseous losses (*Fig.4*) was found to be highly influenced by the presence of maize, the fertilizer treatment and soil moisture. It was higher at KNO_3 treatment and 100% WHC soil moisture (except 30th day), than NH_4Cl treatment and at 65% WHC. Between the 2nd and 68th day the 100%WHC increased more intensively the total N gaseous losses in unplanted pots than with maize compared to 65%WHC while KNO_3 treatment enhanced the gaseous N losses stronger in planted pots than without maize compared to NH_4Cl. The forms of the curves are similar among the 2nd and 68th sampling times. The effect of KNO_3 and 100%WHC is the strongest at the start of the experiment (2nd and 16th day) and on the 68th day compare to NH_4Cl and 65%WHC.

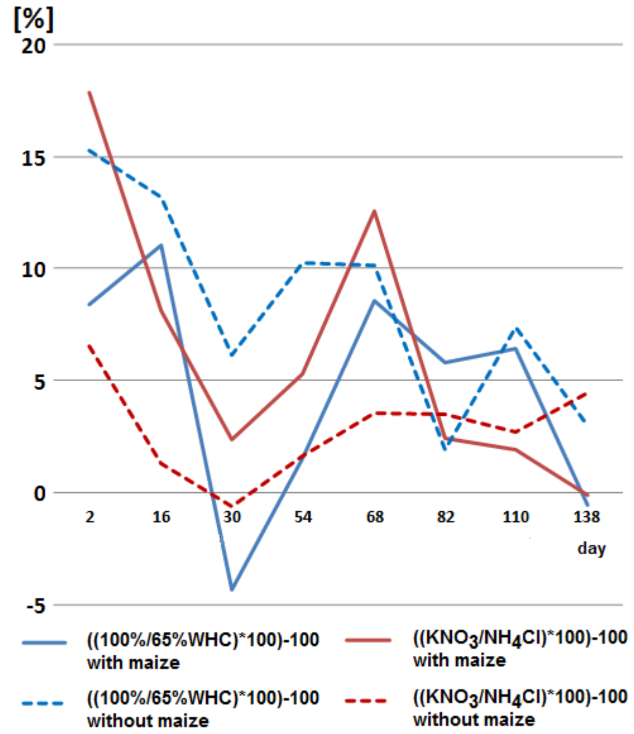

Figure 3. *The total N gaseous losses (+ planted pots, -unplanted pots)(LSD5%= 41mgN/1800 cm3)*

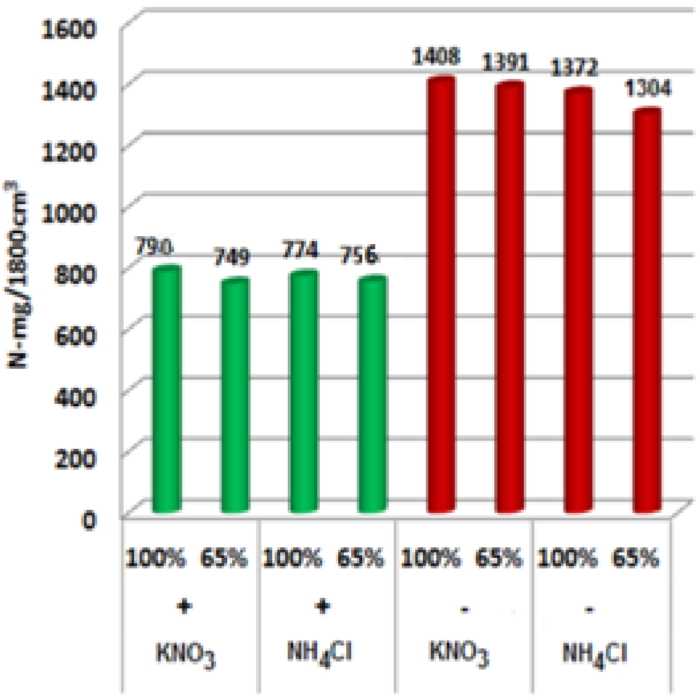

***Figure 4**. Distribution of the relative total N gaseous losses in the planted and unplanted pots*

The ratio of NO_x gaseous losses was different in the planted and unplanted pots (*Fig.5*). In the unplanted pots the ratio of N_2 gas was higher (78%) while the ratios of N_2O, NO_2 and NO were lower than in planted pots. The N_2 and N_2O gaseous losses were higher by 95% and by 70% in the unplanted pots than in the planted pots (N_2 and N_2O: $LSD_{5\%}$= 17 and 5,1 N and N_2O mg/1800cm^3) (*Fig.6*).

***Figure 5**. The % distribution of N gaseous losses in planted (left) and unplanted pots (right)*

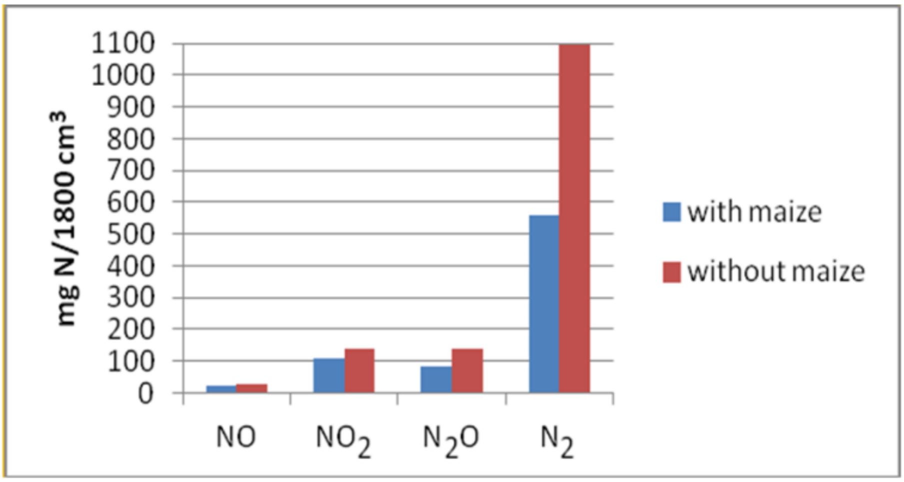

***Figure 6.** The distribution of N gaseous losses in the planted and unplanted pots*

After the 16th day of the experiment the N_2O gaseous losses were much higher in the unplanted pots than in the planted pots and reached the maximum value (140 mgN/1800 cm^3) on the 82nd and 110th day of the experiment depending on treatment (*Fig.7*) and soil moisture (*Fig.8*) while in the planted pots the maximum value (about 80 mgN/1800 cm^3) was measured on the 82nd day. The N_2O gaseous losses were significantly higher at KNO_3 treatment than NH_4Cl treatment in the unplanted pots between the 54th -110th day while in the planted pots between the 2nd – 30th day and on the 82nd day (*Fig.7*). It was found that at WHC=100% soil moisture the N_2O gaseous losses were significantly higher than at WHC= 65% in all pots in most of the sampling time (*Fig.8*).There are two exceptions: in the unplanted pots on the 82nd day when same values were measured at both soil moisture and in the planted pots on the 54th day when the N_2O gaseous loss was significantly higher at WHC= 65% than at WHC=100% soil moisture.

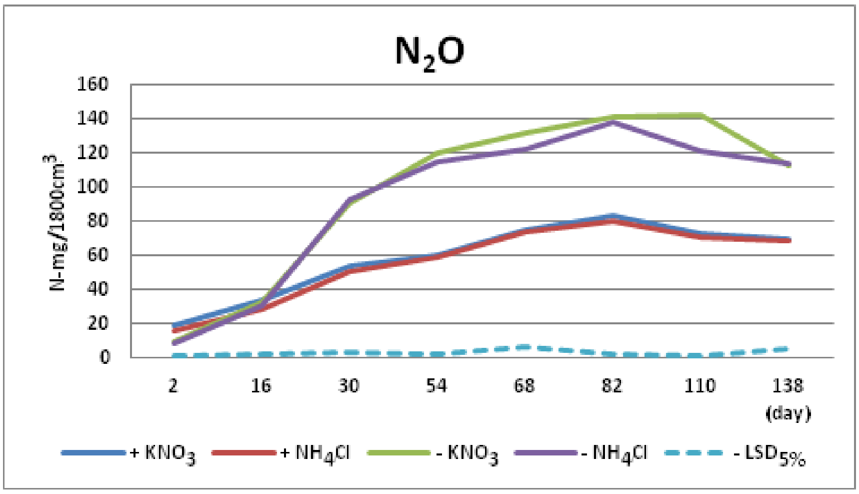

Figure 7. *Change of N2O gaseous losses in the sampling time depend on N fertilizer forms (+in planted pots, - in unplanted pots)*

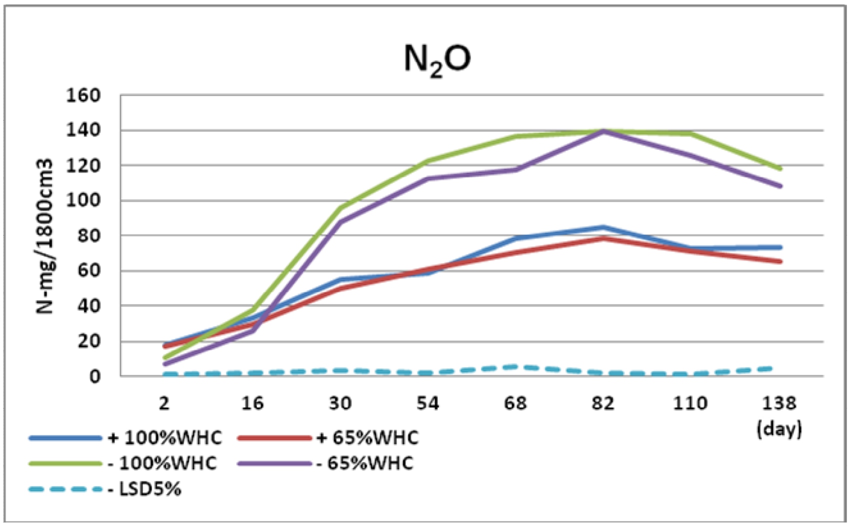

Figure 8. *Change of N2O gaseous losses in the sampling time depend on soil moisture (+in planted pots, - in unplanted pots)*

Conclusions

The total N and N_2O gaseous losses were significantly higher in the unplanted pots than in the planted pots in treated samples. In our experiment with maize the total N gaseous loss was 12-13% of the applied fertilizer N while in the pots without maize it was about twice as much 21-22%. Plant N uptake reduced the N gaseous losses by about 50%.

In contrast to the treated samples in control samples in the planted pots the total N and N_2O gaseous losses were higher than in the unplanted pots due to soil drying-rewetting intensifying microbiological activity, which is more intensive in the rhizosphere around roots (Kuzyakov et al.2000).

The total N and N_2O gaseous losses were significantly higher in the N-fertilizer treatments than in control samples due to the increased activity of bacteria, accelerated mineralization and release of nutrients by the large fertilizer N dose (Kuzyakov et al.2000).

The total N and N_2O gaseous losses were more intensive in case of KNO_3 treatment than in NH_4Cl treatment in most cases as an assumed consequence of the intensive denitrification.

The total N and N_2O gaseous losses were significantly higher at 100%WHC than 65%WHC due to more intensive denitrification processes.

In the unplanted pots the ratio of N_2 gas was higher (78%) than in the planted pots (72%). The N_2 and N_2O gaseous losses were higher by 95% and by 70% in the unplanted pots than in the planted pots due to the absence of plants and more intensive denitrification processes.

It can be concluded that the improperly applied fertilizer causes significant GHG emissions in the absence of plants.

The gaseous N losses were found to be influenced significantly by the presence or absence of plants, the N fertilizer form and the soil moisture.

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