

UDC: 631.452; 631.95

METHODOLOGICAL ASPECTS OF DETERMINING THE TREND OF ORGANIC MATTER MINERALIZATION ↔ SYNTHESIS PROCESSES IN CROPLANDS

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Received January 11, 2019 / Received February 15, 2019 / Accepted March 22, 2019

Aim. To determine the peculiarities of N_2O and CO_2 soil emissions under different systems of crop fertilization.

Methods. Field experiment, gas chromatography. **Results.** The data obtained during the permanent field experiment on leached chernozem with crop rotation including potatoes, spring barley, peas and winter wheat have shown that both absolute (N_2O) and specific (relative to carbon losses in the form of CO_2) losses of nitrogen depend on crop fertilization system. The introduction of raw organic material to the soil in the form of cattle manure or lupine (green manure) has led to the increased CO_2 emission levels. However, specific nitrogen losses in the form of N_2O (g $N-N_2O/kg$ C- CO_2) have remained at the control level. The application of mineral fertilizers (under the absence of raw organic material) have triggered N_2O emissions and more intense production of CO_2 (up to 67 % in the variant with the highest dose of mineral fertilizers ($N_{120}P_{120}K_{120}$ kg/ha of active ingredient), which can be caused by the mineralization of humus compounds. In organo-mineral fertilization system the specific losses of nitrous oxide have not exceeded the control and fallow. Based on the obtained results the authors propose the method of determining the “mineralization-synthesis” indices of organic matter in agricultural soils for estimation of the orientation of biological processes under different crop growing conditions. **Conclusions.** Systemic application of mineral fertilizers without introduction of raw organic material leads to the misbalance of mineralization and synthesis processes in soils. Under such conditions the mineralization of organic matter is prevailing. The use of organic and organo-mineral fertilizers has balanced these processes in the soil. The proposed methodological approach for determination of mineralization-synthesis indices is based on the emission ratio of g $N-N_2O/kg$ C- CO_2 compared to the reference values and can provide an objective view of the tendency of mineralization (negative ratio values) and synthesis (positive ratio values) processes in the soils. It provides grounds to the decision-making principles of agricultural crops fertilization or introduction of certain agronomic techniques.

Keywords: N_2O soil emissions, CO_2 soil emissions, crop fertilization, mineralization, synthesis, organic matter, humus.

DOI:

INTRODUCTION

The mineralization of organic matter in soil is known to occur with the release of carbon dioxide and ammonium [1]. In case of optimal ratio of C : N in soil, the processes, following mineralization and preventing the loss of nitrogen and carbon, are the processes of synthesis of raw organic matter, the first among them being the immobilization of nitrogen by newly-formed

cells of microorganisms. Under conditions of deficient raw organic matter in soil, the released ammonium gets involved in nitrification and further on – in denitrification. The processes of mineralization ↔ synthesis of organic matter are closely interconnected and, depending on their direction, regulated by the C : N ratio; there is either domination of mineralization over synthesis, resulting in nitrogen loss, decrease in the content of organic matter in the soil, including humus, or, vice versa, prevalence of synthesis processes, ensuring the increase in soil fertility [1]. These processes are more

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balanced in natural phytocenoses, on contrary to the croplands. It allows to use the direction of the processes of mineralization ↔ synthesis of organic matter is a key indicator in the evaluation of measures, targeted at the restoration of soil fertility.

The scientific approaches to the objective nature of microbiological processes of mineralization ↔ synthesis of organic matter are based on different solutions. For instance, there is widely applied determination of mineralization-immobilization coefficients, using the ratio of the number of microorganisms, mainly consuming mineral compounds of nitrogen, to the number of ammonifiers [2]. V.D. Mukha [3] improved this method, having suggested the consideration of both the ratio of the number of microorganisms of the mentioned groups and their total number.

However, the mentioned methodological approaches do not take into consideration the fact that the number of microorganisms cannot always reflect their functional activity objectively. Thus, it is relevant to determine the functional activity of key processes in soil that characterize the direction of mineralization ↔ synthesis reactions of organic matter.

MATERIALS AND METHODS

The research was conducted in 2018 in a permanent (started in 2009) 1 ha experimental field of the Institute of Agricultural Microbiology and Agroindustrial Manufacture, NAAS, on leached chernozem under short crop rotations (potato, 0.25 ha – spring barley 0.25 ha – pea 0.25 ha – winter wheat 0.25 ha). The agrochemical characteristics of soil determined by the standard methods described by Fomin G.S. and Fomin A.G. [4] were as follows: pH_{salt} – 5.3; humus content – 3.03 %; easily hydrolysable nitrogen – 95 mg/kg of soil; mobile phosphorus compounds (P₂O₅) – 150 mg/kg of soil (according to Kirsanov); content of exchangeable potassium (K₂O) (according to Kirsanov) – 108 mg/kg of soil.

During our investigations, the crops were grown under the following fertilization systems: 1) no fertilizers; 2) organic No. 1, (see below); 3) organic No. 2 (see below); 4) mineral low; 5) mineral medium; 6) mineral intense; 7) organic-mineral (see below). The control (“reference” plot) in the studies was permanent fallow land (since 2009) with the area of 0.75 ha, located next to the permanent experimental field.

Cattle manure was introduced once during the crop rotation cycle (for potatoes) within organic system No.1, with a dosage of 40 t/ha. Organic system No.

2 envisaged the application of 13 t/ha of the biomass of an intermediate lupine green manure crop grown on the same field prior to potatoes.

Mineral fertilization systems were characterized with the application of compound nitrogen, phosphorus and potassium fertilizer (N16P16K16 per 100 kg of product) for potatoes, with a dosage of 40, 80, and 120 kg/ha of the active ingredients; the same mineral fertilizers were used for barley, pea and wheat in the doses of 30, 60, and 90 kg/ha.

The organo-mineral system envisaged the single annual application of cattle manure (for potatoes, 40 t/ha), and mineral fertilizers (40 kg/ha of nitrogen, P₂O₅ and K₂O each). The after effect of manure in the combination with the direct effect of mineral fertilizers, applied in the medium dose (60 kg/ha) for barley and wheat, and in the low dose (30 kg/ha) for peas, was investigated for the crops, following in the crop rotation.

The area of the single experimental plot was 6.4 sq.m. (7.2 × 12.0 m), the experiment was conducted in randomized four repeats. In total there were 112 plots (4 crops × 7 variants × 4 repeats).

The closed chamber method was used to estimate the emission of nitrous oxide and carbon dioxide in the “soil-plant” system according to references 5, 6 and 7 (pages 267–268) after our modification [8, pages 286–290]. Our adaptation involved the use of plastic 10-liter buckets, the bottom of which was drilled to install a rubber plug to isolate gases, and 50 ml syringe that was used to take air samples.

The chambers were installed in soil at the depth of 5 cm. A water plug was made around the chamber for better isolation of the system and prevention of the loss of gases, accumulated in the chambers. A weighing cup with water was put inside the chamber with the addition of 20 g of calcium acetylide prior to the exposition (the reactions of calcium acetylide with water result in the formation of acetylene, inhibiting the nitrous oxide reductase enzyme and terminating the process of dissimilation of NO₃⁻ and NO₂⁻ at the stage of nitrous oxide reduction [7]). The exposition lasted three hours. The selected gas samples were placed into previously vacuumed vials with rubber plugs, then taken to the laboratory and analyzed using gas chromatography.

The amount of N₂O in the samples was detected using a Tsvet-500 M gas chromatograph (Dzerzhynsk, Russia) with an electron capture detector and 3-meter-long steel sorption columns filled with sorbent Paropak Q

60–80 mesh (Waters Corporation, USA). The column temperature was 40 °C, the evaporator temperature – 120 °C, that of the detector – 330 °C. Carrier gas (argon with methane 95/5) usage was 35 cc/min.

The investigation on the content of CO₂ in gas samples was conducted using the same gas chromatograph Tsvet-500 M as described above with a thermal conductivity detector (bridge current of 130 mA) and a 30 m steel sorption columns filled with sorbent Paropak Q 60–80 mesh. The temperature of columns was 25 °C and that of the detector – 40°C. The carrier gas (helium) usage was 20 cc/min.

The emission of N₂O was estimated according to the formula:

$$\frac{E \times V_1}{V_2 \times S \times t}$$

where: E – amount of nitrous oxide in the sample under analysis, nmol N₂O; V₁ – volume of chamber, cc; V₂ – volume of the sample, introduced to the chromatograph, cc; S – area of chamber cross section, sq.m.; t – exposition time.

The same formula was used to determine the emission of CO₂, but E was estimated as the amount of carbon dioxide in the sample under analysis, nmol.

The losses of nitrogen and carbon caused by the emission of the gases studied were estimated for hectare per day, taking into consideration the molecular mass of N₂O and CO₂ and the exposure time of the chambers in the field.

The planning and conducting of field experiments was conducted according to B. Dospekhov [8, pages 61–72] The statistical processing of the experimental data was conducted using ANOVA disperse analysis method.

RESEARCH RESULTS

The determination of the intensity of N₂O and CO₂ production from soil under potatoes demonstrated rather considerable gas emission in the variants with organic and organo-mineral fertilization (Table 1). This is obvious evidence of the presence of raw organic matter and its active mineralization. There is quite a logical increase in the emission of nitrous oxide from soil under the introduction of mineral fertilizers into soil. However, the increase in the intensity of CO₂ production (under conditions of the deficient raw organic matter) may demonstrate active mineralization of humus compounds.

While analyzing the obtained data, we decided to detect how many grams of nitrogen in the form of N₂O were used per one kilogram of carbon in the form of CO₂, *i.e.* to estimate the emission ratio of the investigated gases. The obtained results were rather surprising, demonstrating that the indices of gas emission ratio in the variants with organic and organo-mineral fertilization were almost at the control level. In other words, though in case of the intake of organic matter in soil the absolute values of gas emission exceeded the data of the control variant considerably, the specific loss of nitrogen did not exceed the control indices. At the same time, the investigated ratio exceeded control values by 35–37 % in the variants, where only mineral fertilizers were used. The lowest indices of specific loss of N-N₂O were noted in the fallow.

The increased emission of nitrous oxide compared to the emission indices of carbon dioxide demonstrated the prevalence of mineralization processes in soil. The decrease in specific emission losses of N₂O demonstrated the fact that a considerable amount of nitrogen was immobilized in soil and thus the processes of mineralization ↔ synthesis of organic matter in soil tend to balance.

Taking into consideration the fact that natural phytocenoses (virgin soils, fallows) are characterized by balanced processes, the more objective approach is to compare gas emission to relevant indices of “reference” soils instead of control variants of experiments.

Table 1. The emission of gases and the emission ratio (g N-N₂O/kg C-CO₂) in potato agrocenoses soils depending on fertilization system

Experiment variants	Emission of N-N ₂ O, g/ha in 24 h	Emission of C-CO ₂ , kg/ha in 24 h	Emission ratio of g N-N ₂ O/kg C-CO ₂
No fertilizers, control	99.29	60.53	1.64
Manure, 40 t/ha	226.04	138.47	1.63
N ₄₀ P ₄₀ K ₄₀	185.28	84.02	2.21
N ₈₀ P ₈₀ K ₈₀	201.06	89.29	2.25
N ₁₂₀ P ₁₂₀ K ₁₂₀	226.87	100.64	2.25
Manure, 40 t/ha + + N ₄₀ P ₄₀ K ₄₀	266.30	166.47	1.60
Green manure (lupine)	161.22	87.02	1.85
Fallow	36.2	22.7	1.59
HIP ₀₅	21.0	11.4	

In our case, this is soil of the fallow, located next to the experiment. The notion of soil reference was substantiated in the monitoring investigations on the condition of soils [9] and is actively used in the practice of ecological studies [10].

Based on the emission ratio $\text{g N-N}_2\text{O/kg C-CO}_2$, we suggest estimating the mineralization-synthesis indices as the ratio of the module of the difference between the emission ratio $\text{g N-N}_2\text{O/kg C-CO}_2$ of the “reference” plot soil (fallow, virgin soil) and the soil of croplands, to the emission ratio index $\text{g N-N}_2\text{O/kg C-CO}_2$ of the “reference” plot:

$$I_{m-s} = (E_{r.p.} - E_{agro})/E_{r.p.}$$

where I_{m-s} – mineralization-synthesis index; $E_{r.p.}$ – emission ratio of $\text{g N-N}_2\text{O/kg C-CO}_2$ of the “reference” plot; E_{agro} – emission ratio of $\text{g N-N}_2\text{O/kg C-CO}_2$ in croplands.

Under intense mineralization processes in soil, the indices of I_{m-s} will have negative values. The approxima-

tion of index values to “0” will demonstrate the state of balance for the investigated processes. On condition that synthesis processes in soil will prevail over the mineralization processes, the obtained results will be positive.

The estimation of I_{m-s} in potato agrocenoses (Table 2) demonstrated that the absence of fertilizers (in the control variant) as well as the application of organic fertilizers ensures insignificant prevalence of mineralization processes over the synthesis ones.

The use of only mineral fertilizers leads to considerable prevalence of the processes of organic matter mineralization over synthesis ones. At the same time, the combination of an inconsiderable dose of mineral fertilizers and manure (organo-mineral fertilization) ensures the optimization of the processes of mineralization \leftrightarrow synthesis of organic matter (index I_{m-s} approximates “0”).

The emission ratio of $\text{g N-N}_2\text{O/kg C-CO}_2$ in plots of spring barley, the following crop of the crop rotation, demonstrates the increase in indices in the variants of direct impact of mineral fertilizers (Table 3).

The indices are getting higher along with the increase in agrochemical burden. In case of the aftereffect of organic fertilizers and organo-mineral fertilization, the emission ratio of nitrogen and carbon approximated the indices of fallow which demonstrated positive changes in the ratio of mineralization \leftrightarrow synthesis of organic matter. The estimation of mineralization-synthesis indices confirmed this conclusion. For instance, the positive index demonstrated an insignificant prevalence of synthesis processes in soil under organo-mineral fertilization, and in case of aftereffect of organic fertilizers the mineralization-synthesis index approximated zero values.

Table 2. The indices of mineralization-synthesis in potato agrocenoses depending on crop fertilization

Experiment variants	I_{m-s}
No fertilizers, control	-0.03
Manure, 40 t/ha	-0.03
$N_{40}P_{40}K_{40}$	-0.39
$N_{80}P_{80}K_{80}$	-0.42
$N_{120}P_{120}K_{120}$	-0.42
Manure, 40 t/ha + $N_{40}P_{40}K_{40}$	-0.01
Green manure (lupine)	-0.16
Fallow	0

Table 3. The emission ratio of $\text{N-N}_2\text{O/C-CO}_2$ and the mineralization-synthesis indices in agrocenoses of spring barley depending on fertilization system

Experiment variants	Emission of $\text{N-N}_2\text{O}$, g/ha in 24 h	Emission of C-CO_2 , kg/ha in 24 h	Emission ratio of $\text{g N-N}_2\text{O/kg C-CO}_2$	I_{m-s}
No fertilizers, control	83.60	33.10	2.53	-0.18
First year, manure aftereffect	174.85	77.13	2.27	-0.06
$N_{30}P_{30}K_{30}$	125.51	45.25	2.77	-0.29
$N_{60}P_{60}K_{60}$	162.87	50.87	3.20	-0.49
$N_{90}P_{90}K_{90}$	206.43	56.94	3.63	-0.69
First year, aftereffect of manure + $N_{60}P_{60}K_{60}$	185.99	89.28	2.08	+0.03
First year, aftereffect of lupine	104.25	47.68	2.19	-0.02
Fallow	34.6	16.1	2.15	0
LSD ₀₅	15.4	7.8		

Table 4. The emission ratio of g N-N₂O/kg C-CO₂ and the mineralization-synthesis indices in agroecosystems of peas depending on fertilization system

Experiment variants	Emission of N-N ₂ O, g/ha in 24 h	Emission of C-CO ₂ , kg/ha in 24 h	Emission ratio of g N-N ₂ O/kg C-CO ₂	I _{m-s}
No fertilizers, control	40.7	21.46	1.90	-0.10
Second year, manure aftereffect	74.7	50.61	1.48	+0.14
N ₃₀ P ₃₀ K ₃₀	79.5	34.21	2.32	-0.35
N ₆₀ P ₆₀ K ₆₀	143.7	44.54	3.23	-0.88
N ₉₀ P ₉₀ K ₉₀	219.4	53.45	4.11	-1.39
Second year, aftereffect of manure + N ₃₀ P ₃₀ K ₃₀	82.2	51.42	1.60	+0.07
Second year, aftereffect of lupine	33.6	31.79	1.06	+0.38
Fallow	36.2	16.5	1.72	0
LSD ₀₅	16.5	7.9		

Table 5. The emission ratio of g N-N₂O/kg C-CO₂ and the mineralization-synthesis indices in agroecosystems of winter wheat depending on fertilizers

Experiment variants	Emission of N-N ₂ O, g/ha in 24 h	Emission of C-CO ₂ , kg/ha in 24 h	Emission ratio of g N-N ₂ O/kg C-CO ₂	I _{m-s}
No fertilizers, control	37.6	18.02	2.09	-0.22
Third year, manure aftereffect	86.9	51.02	1.70	+0.01
N ₃₀ P ₃₀ K ₃₀	73.9	26.72	2.77	-0.61
N ₆₀ P ₆₀ K ₆₀	91.4	32.59	2.81	-0.63
N ₉₀ P ₉₀ K ₉₀	166.0	35.83	4.63	-1.69
Third year, aftereffect of manure + N ₆₀ P ₆₀ K ₆₀	99.7	51.42	1.94	-0.13
Third year, aftereffect of lupine	33.0	22.47	1.47	+0.15
Fallow	28.4	16.50	1.72	0
LSD ₀₅	13.8	8.0		

Similar peculiarities were noted in agroecosystems of peas (Table 4). The studies demonstrated rather a prolonged impact of manure and lupine on the optimization of biological processes in soil. At the same time, in case of only mineral fertilizers (without any additional provision of raw organic matter for soil), mineralization processes dominate in agroecosystems and get activated with the increase in the norm of solid fertilizers. This fact is confirmed by the estimation of mineralization-synthesis indices for organic matter (Table 4). For instance, in the second year the aftereffect of manure and lupine promoted obtaining the positive indices of mineralization-synthesis which demonstrated the prevalence of synthesis processes in soil under these conditions along with the restoration of its fertility.

Similar peculiarities of the impact of fertilizers on the mineralization-synthesis processes in soil were

also observed while growing winter wheat, when the aftereffect of organic fertilizers on the third year was studied along with the direct impact of mineral fertilizers (Table 5). According to the obtained mineralization-synthesis indices, the synthesis processes in cropland soils were observed even on the third year of the aftereffect of organic fertilizers (values with “+” in Table 5).

At the same time, the application of only mineral fertilizers led to the prevalence of mineralization processes in soil. Their intensity increased along with the increase in the doses of mineral fertilizers.

CONCLUSIONS

Systemic application of mineral fertilizers in the technologies of growing agricultural crops without providing the soil with organic matter leads to misbal-

ance in the mineralization ↔ synthesis processes. The mineralization of organic matter dominates under these conditions. Organic and especially organo-mineral fertilization ensures the balance of the mentioned processes in soil.

The suggested methodological indices regarding the determination of mineralization-synthesis indices, based on the emission ratio of g N-N₂O to kg C-CO₂ and the comparison of indices against the value of “reference” soil, may ensure objective vision of the intensity of mineralization and synthesis processes for the process of taking decisions on the fertilization of agricultural crops or application of specific agronomic techniques. The application of gas chromatography, remarkable for high accuracy and analysis velocity, while determining the emission of nitrous oxide and CO₂ promotes fast tracking of changes in the direction of soil-biological processes.

This article does not relate to any studies using humans and animals as investigation subjects.

Conflict of interests. *The authors deny any conflict of interests.*

Financing. *This study did not receive any specific grant from the financing institutions in state, commercial or non-commercial sectors.*

Методичні аспекти визначення спрямованості процесів мінералізації ↔ синтезу органічної речовини в ґрунтах агроценозів

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Мета. Визначити особливості перебігу емісії N₂O і CO₂ з ґрунту за різних систем удобрення сільськогосподарських культур. **Методи.** Польового дослідження, газохроматографічні. **Результати.** У польовому стаціонарному досліді на чорноземі вилуженому при вирощуванні в сівозміні картоплі, ячменю ярого, гороху та пшениці озимої встановлено, що як абсолютні втрати азоту (N₂O), так і питомі (відносно втрат вуглецю у формі CO₂) залежать від систем удобрення. За надходження до ґрунту свіжої органічної речовини (гній, біомаса проміжної сидеральної культури – люпину вузьколистого) зростає емісія CO₂. Проте питомі втрати азоту в формі N₂O (г N-N₂O/кг C-CO₂) при цьому залишаються на рівні контролю. Мінеральні системи удобрення (за дефіциту в ґрунті свіжої органічної ре-

човини) призводять до зростання емісії N₂O. За цих умов спостерігається також інтенсивне продукування CO₂ (до 67 % у варіанті з найвищою дозою мінеральних добрив N₁₂₀P₁₂₀K₁₂₀), що можна пояснити мінералізацією гумусових сполук. За органо-мінерального удобрення питомі втрати закису азоту не перевищують показників контролю та перелогу. На основі отриманих результатів пропонується методика визначення індексів «мінералізації-синтезу» органічної речовини в ґрунтах агроценозів для оцінки спрямованості біологічних процесів за різних агроприйомів. **Висновки.** Системне застосування мінеральних добрив без забезпечення ґрунту свіжою органічною речовиною призводить до розбалансування процесів мінералізації ↔ синтезу. За цих умов домінує мінералізація органічної речовини. Органічне і, особливо, органо-мінеральне удобрення забезпечує врівноваження в ґрунті зазначених процесів. Запропоновані методичні рішення щодо встановлення індексів мінералізації-синтезу, які базуються на емісійному співвідношенні г N-N₂O/кг C-CO₂ і порівнянні показників зі значеннями «еталонного» ґрунту, можуть забезпечити об'єктивне бачення інтенсивності мінералізаційних та синтетичних процесів для прийняття рішень щодо принципів удобрення сільськогосподарських культур або застосування певних агрономічних прийомів.

Ключові слова: емісія N₂O і CO₂ з ґрунту, системи удобрення сільськогосподарських культур, процеси мінералізації та синтезу в ґрунті, органічна речовина, гумус.

Методические аспекты определения направленности процессов минерализации ↔ синтеза органического вещества в почвах агроценозов

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Цель. Определить особенности эмиссии N₂O и CO₂ из почвы под влиянием различных систем удобрения сельскохозяйственных культур. **Методы.** Полевой опыт, газохроматографические. **Результаты.** В полевом стационарном опыте на черноземе, выщелоченном при выращивании в севообороте картофеля, ячменя ярого, гороха и пшеницы озимой, показано, что как абсолютные потери азота (N₂O), так и удельные (относительно потерь углерода в форме CO₂) зависят от систем удобрения. При поступлении в почву свежего органического вещества (навоз, биомасса промежуточной сидеральной культуры – люпина узколистого) возрастает эмиссия CO₂. Однако удельные потери азота в форме N₂O (г N-N₂O/кг C-CO₂) при

этом остаются на уровне контроля. Минеральные системы удобрения (при дефиците в почве свежего органического вещества) приводят к увеличению эмиссии N_2O . В этих условиях происходит также интенсивное продуцирование CO_2 (до 67 % в варианте с наибольшей дозой минеральных удобрений $N_{120}P_{120}K_{120}$), что можно объяснить минерализацией гумусовых соединений. При органо-минеральном удобрении удельные потери закиси азота не превышают показателей контроля и залежи. На основании полученных результатов предлагается методика определения индексов «минерализации ↔ синтеза» органического вещества в почвах агроценозов для оценки направленности биологических процессов при осуществлении различных агроприемов. **Выводы.** Системное применение минеральных удобрений без обеспечения почвы свежим органическим веществом приводит к разбалансированию процессов минерализации ↔ синтеза. В этих условиях доминирует минерализация органических соединений. Органическое и, особенно, органо-минеральное удобрение обеспечивает сбалансированное состояние в почве исследуемых процессов. Предложенные методические решения по определению индексов минерализации-синтеза, базирующиеся на эмиссионном соотношении $g\ N-N_2O/kg\ C-CO_2$ и сравнении показателей со значениями «эталонной» почвы, могут обеспечить объективное видение интенсивности минерализационных и синтетических процессов для принятия решений относительно принципов удобрения сельскохозяйственных культур или применения определенных агрономических приемов.

Ключевые слова: эмиссия N_2O и CO_2 из почвы, системы удобрения сельскохозяйственных культур, процессы минерализации и синтеза в почве, органическое вещество, гумус.

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