STUDY ON THE EFFICIENCY AND MOBILITY OF DIFFERENT FORMS OF NITROGEN FROM FOLIAR FERTILIZERS BY USING OF ^{15}N ISOTOPE

Daniela MIHALACHE[1], Carmen Eugenia SÎRBU[1], Traian Mihai CIOROIANU[1], Adriana Elena GRIGORE[1], Nicoleta MĂRIN[1], Ana Maria STĂNESCU[1], Bogdan RUJOI[1], Mariana IANCU[1]

e-mail: daniela.mihalache@icpa.ro

Abstract

The objective of the study was to establish (by using as tracer the ^{15}N isotope) the contribution of foliar complex fertilizer containing natural organic compounds for improving the efficiency of different forms of nitrogen fertilizers applied to the soil. We used the isotope in order to track the efficiency use of different forms of nitrogen fertilizer.

Efficient use of nitrogen has a positive effect on the quality of the crop which was accompanied by a corresponding decrease of the pollutant impact of chemical fertilization on the environment.

There were used fertilizers with NPK matrix containing meso and micro-nutrients, and we incorporated organic substances, such as protein hydrolysates with biostimulatory role due to the presence of free aminoacids, and we obtained maize green mass yiled increases, which are stastistically significant.

The nitrogen present in various forms, such as ammonia N-NH4, nitric N-NO3, amide N-NH2 was labelled with ^{15}N isotope. We observed that there were increases when nitrates and ammonia forms were used, when was compared with the unfertilized control variant. In fact, this study demonstrates the efficiency of protein hydrolysates.

Key words: foliar fertilizer, protein hydrolysates, ^{15}N isotope.

Studies concerning the rational use of fertilizers and nutritional analysis processes can be addressed both in the case of conventional root and unconventional (foliar) application by using of stable and/or radioactive isotopes.

Notable results were achieved in investigating the sources of nutrients, the movement speed within the plant and the interaction between them, as factors of particular importance in determining the efficiency of nutrients inputs within the cropping system *(*Blair G. and Tiel R., 2000; Tanemura K. *et al*, 2005). As for absorption and transport of aminoacids in the plant organs (ears of barley), studies were carried out with aminoacids labelled with 15N in order to determine the relationship between aminoacid transport in the ear and their proteinogenic nature (Axente D *et al*, 2010).

Using of nuclear techniques allow knowing accurately the quantities of nutrients absorbed differently by plants from the soil and fertilizers following their evolution in plants, as well as changes occurring in the complex soil-plant-fertilizer system (Reddy G. B., Reddy K.R, 1980; Vasilas B.L., Legg O.J., Wolf D.C., 1980; Stevens W.B., Hoeft R.G., Mulvaney R.L., 2005).

Using of fertilizer labelled with 15N to assess the nitrogen absorption in different parts of the plant has been described by Tatiele A.B. Fenilli et al., within an experiment conducted in a coffee culture which was fertilized with ammonium sulphate labelled with 15N isotope.

Rawluk C.D.L, Grant C. A. and Racz G.J. also demonstrates the effects of urea on protein content in wheat grain by using an urea solution labelled with 15N. Using isotopic techniques and 15N isotope in urea they determined the relative efficiency of foliar application of urea fertilizer solution and was compared to soil application.

Also, in studies where 15N isotope was used, the penetration and translocation mechanisms in different plant organs of the three existing forms of nitrogen in fertilizers (e.g. amide, nitrate, ammonia) are analyzed (Soare M. *et al*, 2009).

In generall, the products with bioregulatory role are organic substances that, applied in low concentrations, participates in the physiological processes of plants growth and development with beneficial effects on crops, both quantitative and qualitative, while reducing losses due to transport and products preservation, at the same time with reducing the impact of pollutant chemical

[1]National Research and Development Institute for Soil Science, Agro-chemistry and Environment (ICPA) Bucharest

fertilization on the environment (Cioroianu T.M. and Sîrbu C., 2011).

MATERIAL AND METHOD

To establish the contribution of complex foliar fertilizers enriched with natural organic substances, by using the 15N isotope, in order to improve the efficiency use of different forms of nitrogen in fertilizer applied to the soil, there were obtained three fertilizers of NPK type with meso, micro nutrients and natural organic substances. There were used as chelating and bio-stimulator compounds, the components resulting from the neutral hydrolysis of protein substances containing proteins, peptides and amino acids of animal or vegetal nature.

Figure 1 shows the technological process for obtaining the experimental NPK fertilizers with meso, micro elements and natural organic substances, and with extraradicular application.

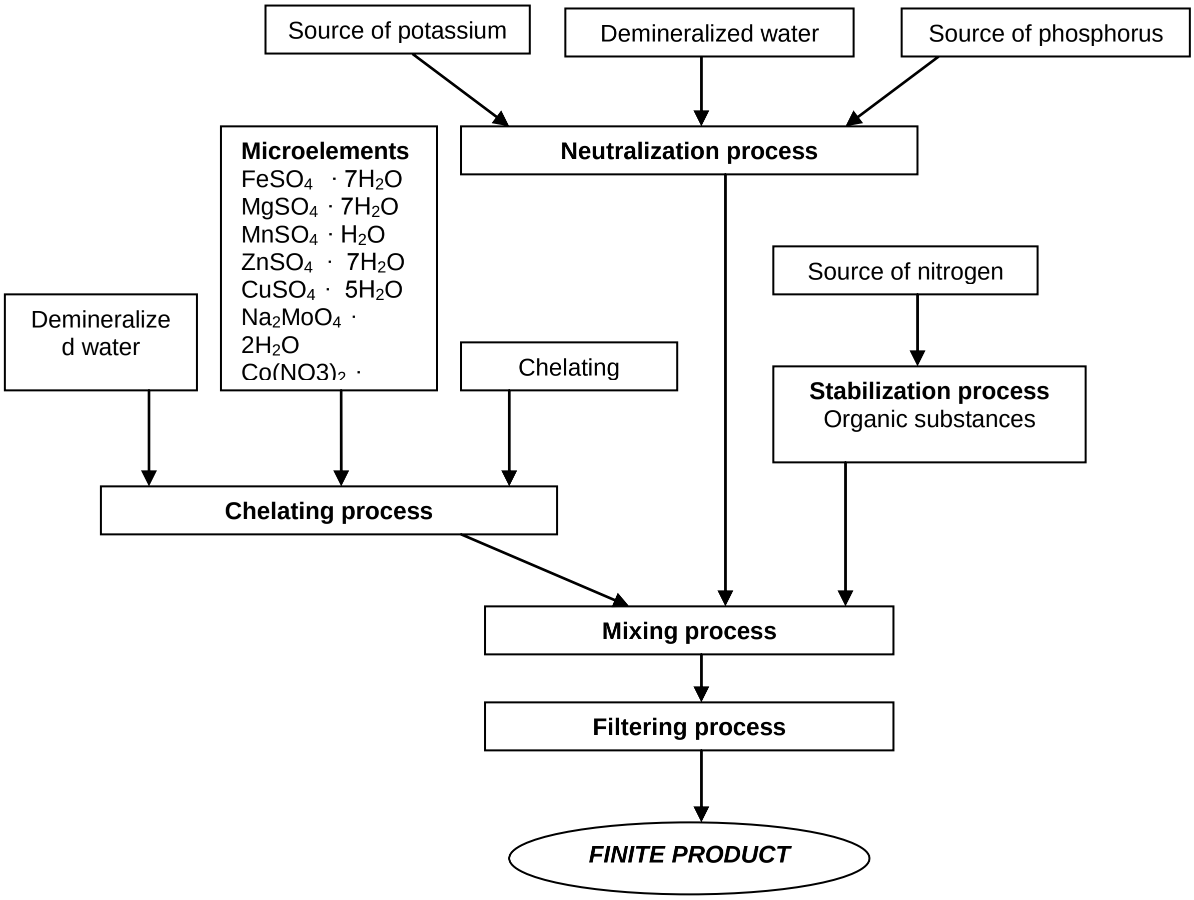


Figure 1 **Scheme for obtaining of liquid fertilizers of NPK type, with/without organic matter**

The experimental liquid fertilizers were obtained in the laboratory phase, and were characterized from physico-chemical point of view and then were tested from agrochemical point of view.

To establish the agrochemical efficiency of obtained extraradicular fertilizers there were conducted tests in vegetation pots, at the same time with experiments on translocation of different forms of nitrogen from the soil in plant by using as marker the 15N nitrogen isotope. Agrochemical experiments were performed on a *Albic Aluvisol* with following characteristics: pH 6,63; Humus 9,84 % and organic carbon 5,71 %.

Experimental conditions used:

Vegetation pots with 10 liters volume, filled with 10 kg of soil Maize crop – CORTES hybrid.

Experiments were performed in 3 replicates. V1 is a version of control variant without basic fertilization. Variants between V2 - V13 are variants with basic fertilization, and were fertilized with NPK 15:15:15 complex fertilizer, physically equivalent to 300 kg fertilizer/ha.

The nutrients dose applied to the basic fertilization is: $N_{45}P_{45}K_{45.}$

The amount of nitrogen applied as urea or ammonium nitrate in the pot was: 300 mg/pot.

The amount of ^{15}N isotope applied as urea or ammonium nitrate in the pot was: 30 mg/pot.

In these experiments there were used the following fertilizers labelled with 15N:

- urea with 10% amidic nitrogen ^{15}N labelled (-NH_2);
- ammonium nitrate with 10% nitric nitrogen ^{15}N labelled (-NO_3);
- ammonium nitrate with 10% ammonia nitrogen ^{15}N labelled (-NH_4).

There were applied the following experimental foliar fertilizers:

- F1 with hydrolyzed animal protein;
- F2 with hydrolyzed vegetal protein;
- F3 with the NPK, meso and micro-nutrients matrix used as control variant.

Samples of green mass from maize (harvest I and II) were analyzed in the LAFC - ICPA Bucharest, by measuring the contents of N, P, K, Ca, Mg, Zn, Cu, Fe and Mn, and the total nitrogen content and the ^{15}N isotope in the USA, at Cornell University - Isotope Laboratory (COIL) by using a mass spectrometer of Thermo Delta V type (IRMS) which has an interface for elemental analysis NC 2500.

In *Table 1* the experimental variants are presented.

Table 1

Explaining the variants and the fertilization applied

No. of variant	Fertilization	Fertilization description
V1	Basic NF	Without basic fertilization
V2 F1	Basic F	Basic fertilization with NPK 15.15.15 ($N_{45}P_{45}K_{45}$), foliar non-fertilization
V3 F1	FB + F1	Basic fertilization with NPK 15.15.15 ($N_{45}P_{45}K_{45}$) and Urea labelled with ^{15}N 10% (NH_2) - 300mg/pot; foliar fertilizer applied Variant F1 (HA- animal hydrolyzed)
V4 F1	FB + F1	Basic fertilization with NPK 15.15.15 ($N_{45}P_{45}K_{45}$) and Nitrate labelled with ^{15}N 10% (NO_3) - 300mg/pot; foliar fertilizer applied Variant F1 (HA- animal hydrolyzed)
V5 F1	FB + F1	Basic fertilization with NPK 15.15.15 ($N_{45}P_{45}K_{45}$) and Nitrate labelled with ^{15}N 10% (NH_4) - 300mg/pot; foliar fertilizer applied Variant F1 (HA- animal hydrolyzed)
V6 F2	Basic F	Basic fertilization with NPK 15.15.15 ($N_{45}P_{45}K_{45}$), foliar non-fertilization
V7 F2	FB + F2	Basic fertilization with NPK 15.15.15 ($N_{45}P_{45}K_{45}$) and Urea labelled with ^{15}N 10% (NH_2) - 300mg/pot; foliar fertilizer applied Variant F2 (HV- vegetal hydrolyzed)
V8 F2	FB + F2	Basic fertilization with NPK 15.15.15 ($N_{45}P_{45}K_{45}$) and Nitrate labelled with ^{15}N 10% (NO_3) - 300mg/pot; foliar fertilizer applied Variant F2 (HV- vegetal hydrolyzed)
V9 F2	FB + F2	Basic fertilization with NPK 15.15.15 ($N_{45}P_{45}K_{45}$) and Nitrate labelled with ^{15}N 10% (NH_4) - 300mg/pot; foliar fertilizer applied Variant F2 (HV- vegetal hydrolyzed)
V10 F3	Basic F	Basic fertilization with NPK 15.15.15 ($N_{45}P_{45}K_{45}$), foliar non-fertilization
V11 F3	FB + F3	Basic fertilization with NPK 15.15.15 ($N_{45}P_{45}K_{45}$) and Urea labelled with ^{15}N 10% (NH_2) - 300mg/pot; foliar fertilizer applied Variant F3 (control – classic fertilizer with the same NPK, meso and micro-nutrients matrix)
V12 F3	FB + F3	Basic fertilization with NPK 15.15.15 ($N_{45}P_{45}K_{45}$) si Nitrate labelled with ^{15}N 10% (NO_3) - 300mg/pot; foliar fertilizer applied Variant F3 (control – classic fertilizer with the same NPK, meso and micro-nutrients matrix)
V13 F3	FB + F3	Basic fertilization with NPK 15.15.15 ($N_{45}P_{45}K_{45}$) si Nitrate labelled with ^{15}N 10% (NH_4) - 300mg/pot; foliar fertilizer applied Variant F3 (control – classic fertilizer with the same NPK, meso and micro-nutrients matrix)

RESULTS AND DISCUSSIONS

Table 2 presents the estimated and determined physico-chemical characteristics for the fertilizers obtained in laboratory and agrochemical tested, namely fertilizers of NPK

type to which protein organic substances were added.

Table 2

Characteristics of the applied fertilizers in agro-chemical experiments

Compositio n, g/l	Variant F1		Variant F2		Variant F3	
	Estimated	Determined	Estimated	Determined	Estimated	Determined
Nitrogen, N	65	67.22	65	67.46	65	68.12
Phosphorus	65	66.16	65	65.82	65	66.22
Potassium,	58	59.32	58	59.18	58	58.96
Boron, B	0.2	0.22	0.2	0.21	0.2	0.22
Cobalt, Co	0.005	0.005	0.005	0.005	0.005	0.005
Copper, Cu	0.2	0.23	0.2	0.22	0.2	0.22
Iron, Fe	0.35	0.37	0.35	0.36	0.35	0.37
Magnesium,	0.3	0.31	0.3	0.32	0.3	0.31
Manganese,	0.25	0.26	0.25	0.27	0.25	0.25
Molybdenu	0.005	0.005	0.005	0.005	0.005	0.005
Sulfur, SO_3	1.5	1.78	1.5	1.86	1.5	1.96
Zinc, Zn	0.12	0.13	0.12	0.12	0.12	0.13
Protein substances	52	52	56	56	-	-
Free	0.55	0.55	6.1	6.1	-	-
Nonionic	1	1	1	1	1	1

The results interpretation pursued the following aspects:

- green mass production of maize obtained at first and second harvest;
- content of N, P, K, Ca, Mg, Zn, Cu, Fe and Mn;
- the isotopic ratio 15N:14N in the collected samples;
- the parameter δ15N, which represents the accumulation of isotope δ15N in samples;
- labelled nitrogen export, translocation from soil to maize samples, according to applied nitrogen fertilization and foliar fertilization respectively.

Isotope ratio: is the ratio between the number of heavy isotopes of an element and the number of light isotopes within a sample (e.g. 15N:14N, usually noted with the letter R).

The parameter δ15N, which represents the accumulation of isotope within a sample is given by the following formula:

$$\delta 15N = 1000\ (R_{sample} - R_{standard}) / R_{standard}$$

From each experimental variant were collected two samples of green mass which were weighed and then the N, P, K, Ca, Mg, Zn, Cu, Fe and Mn contents, isotopic ratio (R) 15N: 14N and δ15N parameter were determined according to basic nitrogen fertilization application and the extraradicular fertilizer used.

Some of the experimental results obtained are presented below.

The *Figures 2 – 4* present the green mass yield obtained for the first and second harve st and average yield of the harvests according to the nitrogen source applied by incorporation into the soil and the used foliar fertilizer.

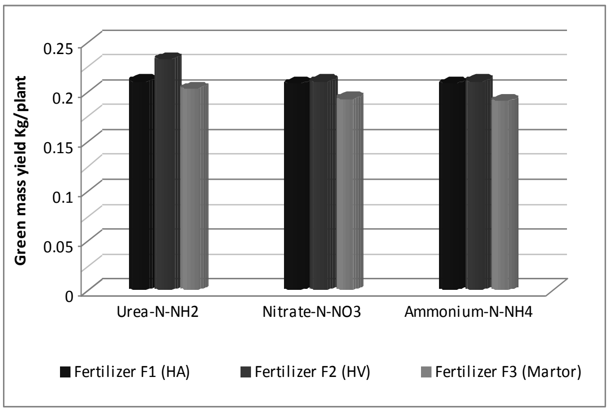


Figure 2 **Variation of the green mass yield depending on the source of nitrogen fertilizer incorporated into the soil and the applied extraradicular fertilizer (first harvest)**

Although there were differences in green mass yield both between the applied foliar fertilizers and the nitrogen source used as fertilizer, they were not statistically significant.

Evolution of the yields increases when compared to the control without basic fertilization (NFB) and for the variants with basic fertilization and basic and foliar fertilization are shown in *Figure 5*.

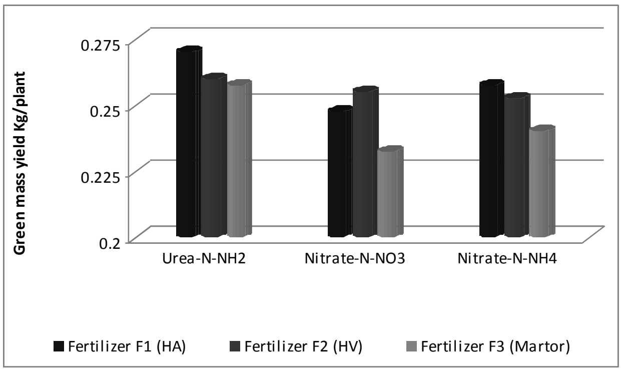


Figure 3 **Variation of the green mass yield depending on the source of nitrogen fertilizer incorporated into the soil and the applied extraradicular fertilizer (second harvest)**

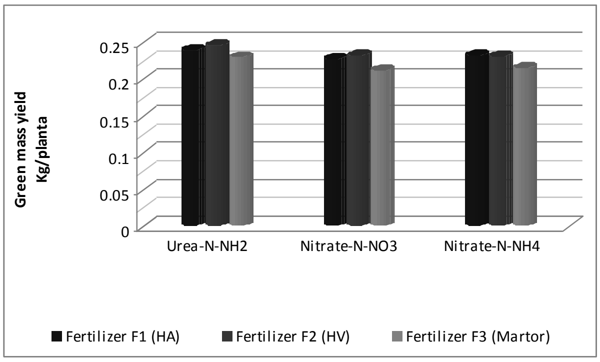
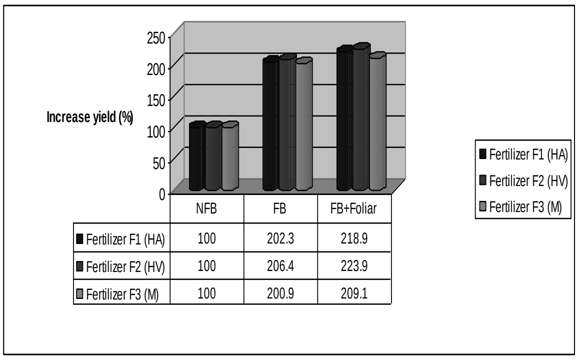


Figure 4 **Variation of the green mass yield depending on the source of nitrogen fertilizer incorporated into the soil and the applied extraradicular fertilizer (the average yield of the harvests)**



	NFB	FB	FB+Foliar
Fertilizer F1 (HA)	100	202.3	218.9
Fertilizer F2 (HV)	100	206.4	223.9
Fertilizer F3 (M)	100	200.9	209.1

Figure 5 **Evolution of the green mass yields (obtained average values) according to the applied fertilization as compared to the control variant (foliar unfertilized)**

Increases obtained when compared to control with basic fertilization (FB) are within the range frequently obtained in case of foliar fertilizers application to maize crop. It may be noted however the effect of incorporation into the matrix of NPK type with meso and micro-nutrients of the organic substances, protein hydrolysates, substances that have a biostimulatory role in the presence of free aminoacids, resulted in increases of 18.2% for fertilizer with animal protein hydrolyzate (HA) and of 23.9% for fertilizer which contain vegetal protein hydrolyzate (HV), increases that are statistically significant.

The *Figures 6 – 8* present the yield increases due to basic fertilization and foliar application of the three fertilizers, increases due to basic fertilization application of nitrogen as ammonium nitrate and urea respectively, as well as those due to foliar fertilization and basic fertilization.

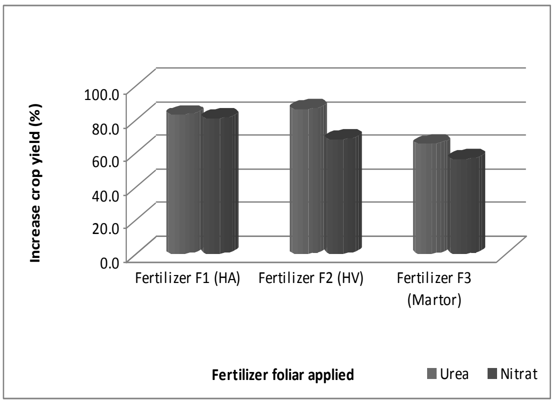


Figure 6 **Yield increases as a function of the nitrogen type applied (nitrate or urea) for each foliar fertilizer applied (first harvest)**

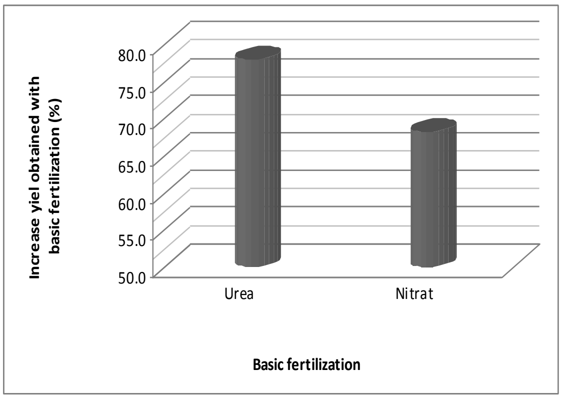


Figure 7 **Yield increases as a function of the nitrogen type applied additionally as compared to the control with basic fertilization in dose of $N_{45}P_{45}K_{45}$**

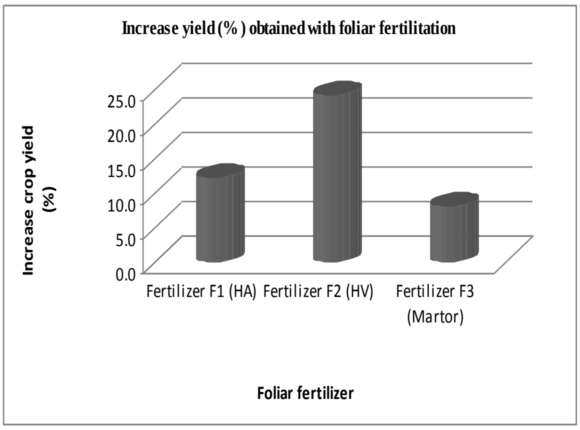


Figure 8 **Yield increases as a function of application of foliar fertilization (basic fertilization with urea)**

In this study it was found also that the nitrogen fertilization with amide type (urea) provided increases by approx. 10% higher than in case of fertilization with nitrogen as nitrate and ammonia type (ammonium nitrate). Foliar fertilization combined with a basic fertilization with urea had a contribution in increasing yields ranging from 5.5% for control + foliar fertilizer F3 and to 21% for fertilizer with vegetal hydrolyzed F2 (HV) due to the presence of free aminoacids in the NPK matrix type.

CONCLUSIONS

1. Three extraradicular fertilizers were obtained and there were physically and chemically characterized. These fertilizers are extraradicular applied and stands out by a complex composition through a combination of NPK or NK-type matrix with micro-nutrients (Fe, Cu, Zn, Mn and Mg) and natural substances which have a role of chelating agent but bio-stimulant.
2. Technological schemes have been designed and mass balances have been produced for fertilizers obtained from the laboratory phase.
3. Incorporation into the NPK matrix with meso and micro-nutrients of the organic substances, protein hydrolysates, substances that have a biostimulatory role due to the presence of free aminoacids, led to yield increases by 18.2% for green mass in case of the fertilizer which contain animal protein hydrolyzed (HA) and by 23.9% in case of the fertilizer with vegetal protein hydrolyzed (HV), increases that are statistically significant.
4. Foliar fertilization combined with a basic fertilization with urea, had a contribution in increasing yields ranging from 5.5% in case of control foliar fertilizer F3 and 21% in case of the fertilizer which contain vegetal hydrolyzed F2 (HV) due to the presence of free aminoacids in the NPK matrix.

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