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Article

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BEST PRACTICES

A BIOECONOMIC SOLUTION FOR REPLACING CHEMICAL FERTILIZERS
BY ORGANIC PROCESSES FOR ATMOSPHERIC NITROGEN FIXATION
IN SOIL

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Abstract

Large-scale use of nitrogen-based chemical fertilizers, specific to intensive agriculture, has undesirable effects consisting of soil degradation and atmospheric loading with carbon dioxide. Good practice illustrates, however, that sustainable agriculture can be achieved by substituting as much as possible chemically synthesized nitrogen with biologically fixed nitrogen.

This research is part of a study cycle that aims to use the natural symbiotic fixation models to bring into the soil and plants as much as possible atmospheric nitrogen, non-polluting, antioxidant and generator of humus and durability. We had work with a pea culture, seated in a rotation of 4 years the type pea, wheat-rape, maize, and we watched at three parameters of the symbiotic fixation between peas with species *Rhizobium leguminosarum*.

The experiments were carried out in two locations in southern Romania (Burnas Plain - on the Plateau of Alexandria, and in the Southern Plain - Modelu, Calarasi) and the results were:

- in dynamics the nodosities begin to form at 22-24 days after sowing, their number increases accentuated until the 37 days of life, after which it decreases slightly until the 70th day. The maximum number of nodosities on roots reaches a number of 57-58 per plant. The life of a nodosity is not longer than 10 days;

- the weight of dry nodosities is on the peak of biological activity higher at Modelu (5.8 g/m²) at 38 days after sowing, and less in Alexandria (4.1 g/m²) at 37 days after sowing (Figure no. 2 in-text);

- starting from the weight of the nodosities and using their transformation coefficient in the N (nitrogen) active substance (which is 4.6), and using the functional analysis of the correlations and integral calculations, the dynamics of nitrogen accumulation in the form of a high precision polynomial function (order 14) was obtained (figure no. 3 in-text), which tells us that: the highest amount of nitrogen is obtained on day 41 after sowing in the amount of 6.52 Kg N active substance/ha. Cumulative calculation obtained through the integral function the total and average quantity obtained for the two locations in 2016 – 2017 was about 116 kg N/ha (1).

$$\int_{20}^{65} f(x) d(x) = 115.89 \text{ kg} = 116 \text{ kg N/ha}$$

(1)

Keywords: biologically fixed nitrogen, natural patterns, nodosities, chemical synthesis nitrogen, bioeconomic model

JEL Classification: Q16; Q57

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Introduction

According to the University of Munster researchers (Wilhelms-Universität in Münster, 2004) and Wikipedia (2017a), in soils containing heteropolycondensate humus, such as the majority of the soils that we met in our research, the highest amount of humus and nitrogen is found in the first 10 centimetres of the soil (ground assessment under the meadow). On this depth is about approx. 1500 tons of humus, of which 1/17 meaning 0.0589×1500 tons = 88.38 tons of nitrogen, usually in organic form. This amount of nitrogen is the nutrient reserve of the soil. If a wheat crop for a 10 tones production consumes 0.25 tones N/ha, it would theoretically mean that a very good soil in 88 years: $0.25 \text{ tones N/ha} = 325$ years would become a desert and would not use it for nothing.

Because humus contains large amounts of carbon (60%) in the fullness of its ecological health, it becomes a large reservoir of carbon dioxide, water, but also an excellent habitat of microorganisms. By creating the heteropolycondensate humus but also nutritive, the earth's atmosphere impoverishes in CO₂ with a series of positive consequences that result from it sustains Raggam (2004). It is clear that landing as much nitrogen as possible from the air in organic form is a mission that research should not miss if sustainability of agricultural ecosystems is desired and a balancing of climate change. On the other hand, a numerically growing humanity has need an ever-increasing necessity of foods. At the base of growth of the vegetable agriculture production and animal farming, alongside carbon, hydrogen and oxygen is nitrogen. Nitrogen is bound in all the earth's organisms by proteins, nucleic acids, enzymes, metabolic exchange products, and so on. Life is not possible without nitrogen sustains Schröter and Lautenschläger (1986). In mineral form, nitrogen alone occupies 78.084% by volume, 75.5% by weight this determination appeared in Wikipedia (2017b). In biomolecules he is in proteins, lipids, porphyrin nucleotides.

According to Zamnesia (2015), if compared with nitrogen fertilizers of synthesis by the Haber-Bosch process, atmospheric nitrogen:

- is environmentally friendly and very suitable for nutrition;
- does not pollute the soil;
- improves soil quality, especially the microbiological one;
- is not lethal;
- is produced and absorbed according to the nutritional rhythm of the crops;
- is cheaper.

The synthesis nitrogen, in turn:

- is dangerous to soil (degrades humus);
- can be washed;
- it's evaporate;
- pollutes the soil and the environment in general;
- leaves a series of residues in the soil;
- the production is polluting;
- Nitrogen fertilizers are expensive enough.

The evolution towards the agricultural bioeconomy can be achieved by the substitution, in agricultural practice, of as much as possible quantities of nitrogen synthesized chemical with biologically fixed nitrogen, especially by symbiosis between leguminous species and fixative microorganisms (bacteria, actinomycetes, etc.).

The data published by Riou (2016) shows that a quantity of about 160 million tonnes N/year is symbiotically fixed biological on the earth, such as it is produced by synthesis (150 million tonnes N/year) or 4800 kg/second, or 60% of total fertilizers.

Two researchers, Marquard (2000) and Quispel (1982), demonstrate that the various leguminous species can symbiotically fix a large amount of nitrogen, which on average varies between 80 kg/ha for lentils, 100 kg/ha for groundnuts and soybeans, 150 kg/ha for peas, and lupine, 200 kg/ha for grain and 250 kg/ha for clover and medicago sativa (lucerne). The maximum biological fixed nitrogen levels (demonstrated) reach up to 670 kg/ha for clover and 500 kg/ha for peas.

Berca et al., (2018) sustained that in the Southern Romania according to their recent research had shown that peas fix between 115 and 160 kg N/ha. In Romania, over the past 5 years, a protein products market has been formed, among which peas (*Pisum Sativum*) occupies a growing area each year. It is therefore possible to modify the nutrition system of crops from crop rotation by increasing the biologically fixed portion of nitrogen nitrogen to the detriment of plant synthesis, with all the positive effects that result from this for the bio economic development of the agricultural system.

1. Research methodology

The purpose of this research was to create an informational model on how nitrogen accumulation by biological fixation works in the Belmondo pea variety, in the two locations in southern Romania (Alexandria and Modelu-Calarasi), in two years consecutive (2016, 2017).

The objectives proposed by experimentation and the calculation methodology were:

A. Research and construction of relational function between peas and its symbiotic (*Rhizobium leguminosarum*) using the following parameters:

- Average number of plant nodosities: occurrence, development, maximum, degradation. Determining the active period of nodosities.
- the average weight of dry nodosities expressed in g/m^2 , in dynamics, as a possible indicator of fixed nitrogen;
- the calculation of the amount of nitrogen fixed daily and the total quantity accumulated during the active period of the crop vegetation;

B. Ecological, economic and sustainability effects of crops - comparing between crop rotation systems (precursor plants).

The research was carried out in two locations in the south of the country: the Burnas Platform (Alexandria) and the Southern Plain around Calarasi (Modelu), along the Danube.

In each location, was cultivated Belmondo variety of afile type (90-100 cm high port). The precursor plant was the same in both locations, namely: maize, hybrids harvested until the end of September, allowing good soil work for the sowing of peas in the spring.

The soil, in both locations, was chernoziomic with 3.5% humus and 24% argil in Alexandria, 3.2% humus and 26% argil at Modelu (Calarasi). In the second case, the texture of the soil was much easier.

Climate: In Alexandria and Modelu (Calarasi), we have a temperate, semi-arid continental climate with an average of 10.8°C for Alexandria and 11°C for Modelu (Calarasi). Precipitations regime: 500 mm average for Alexandria and 490 mm at Modelu (Calarasi). We can say that in Modelu (Calarasi) the aridity is slightly more accentuated than in Alexandria.

The average rainfall distribution shows that in both locations, during the crop growing period, 70-72% of rainfall occurs. In the two years of our research (2016, 2017), for the both locations we have had slightly over 300 mm enough to provide a production of about 3.500 kg grains/ha.

The research was carried out by surveys in dynamics, from two fields sown early in the spring (beginning of March), for each location. The land was worked after the usual technology in the area. Since the autumn have been applied 70 kg of P2O5. Nitrogen fertilizers have not been applied. 110 grains/m² were sown but rose between 80-85 plants/m². Of these, 78-80 plants made nodosities. There were no additional symbiotes applied, the soils being well supplied with *Rhizobium leguminosarum*. Monitoring of plant growth and their relationship with *Rhizobium leguminosarum* was performed weekly.

Rhizobium leguminosarum infection has been found to occur about 20 days after sowing. Then the plants have 2-3 leaves.

It was determined in dynamics:

- number of nodosities for 40 plants (4x10). We worked with the average for 10 plants (40/4 = 10);
- the dry weight of the nodosities/m², was carried out by drying in the oven at 105 ° C ± 5 ° C for 3 hours, in special capsules;
- the balancing method, Loges et al. (2001), used by Berca et al. (2018). The coefficient calculated by the above authors, sustain that one gram of dry nodosity fixes 4.6 grams of nitrogen active substance: ratio 1: 4.6.

The data were statistically processed by analysing correlations, functions and full computation.

2. Results

It was carefully monitored both the moment of initiation of the formation of nodosities and their multiplication, the increase to a maximum level and then decreasing their number.

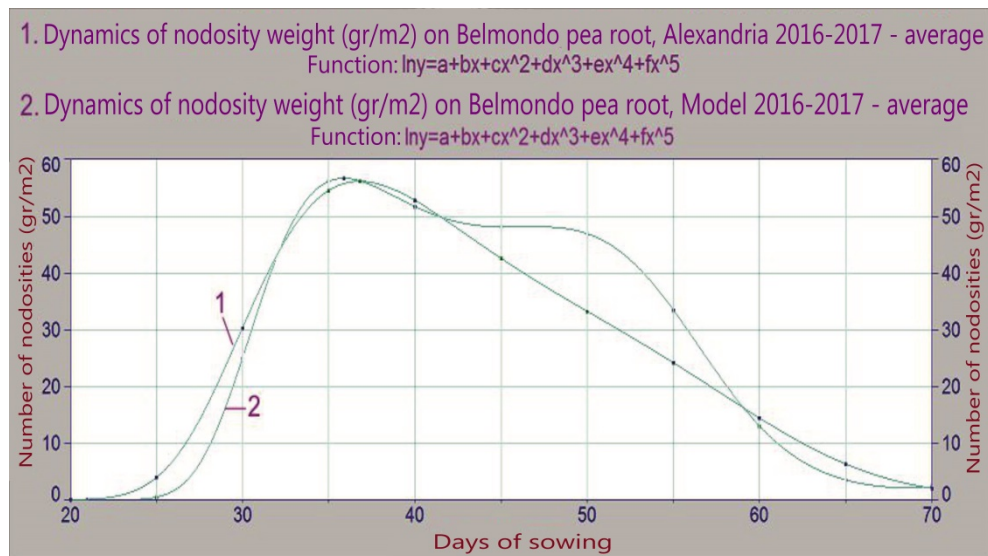
On the average for the two years, the dynamic evolution of the number of nodosities "y" according to the time elapsed since the sowing of the crop, for the two localities, is shown

in figure no. 1 ($y = f(x)$). The model is described by two logarithmic functions, one for each locality. There are basically the same functions, but the coefficients a, b, c, d, e and f are different. Functions are statistically assured, at least at a significant level, thus demonstrating their reproducibility. Our observations show that the initiation of the nodosities formation process occurs in the 2-3 leaf stage i.e. at 22-25 days after sowing, on average for the two years.

The dynamics provided by function 1 (curve 1) indicate that initiation is earlier with 2-3 days at Modelu (Calarasi) compared to Alexandria.

The increase in the number of nodosities is slightly slower in the case of Modelu (Călărași), reaching the maximum at about 37 days with a number of about 57 nodosities per plant. The decrease is sinusoidal and slow until 65-70 days after sowing. Over the period of 43 to 55 days of sunrise due to favourable climatic conditions, the formation and growth of nodosities remains high, even if other determinations have shown the fact that the activities of nodosities has to stop earlier with 2-3 days.

Curve number 2 - Alexandria, indicates a later initiation, a more pronounced increase in the number of nodosities and an achievement of the maximum one day earlier than at Modelu (Călărași). The decrease is uniform for the same period as Modelu (Călărași).



$$(1) \ln y = -99.69 + 10.32x - 0.40x^2 + 0.0077x^3 + 7.30x^4e^{-0.5} + 2.68 e^{-0.7}$$

$$(2) \ln y = -298.68 + 32.92x - 1.41x^2 + 0.0297x^3 - 0.000308x^4 + 1.25 e^{-0.5}$$

This system of equations is statistically assured by correlation reports r^2 , between 0.94 - 1.00

Curve 1 $r^2 = 1.00$
Curve 2 $r^2 = 0.94$

The statistical insurance is very significant

Figure no. 1: Expression of numerical dynamics of nodosities on the Belmondo pea's variety roots at Modelu (1) and Alexandria (2). Average for 2016-2017

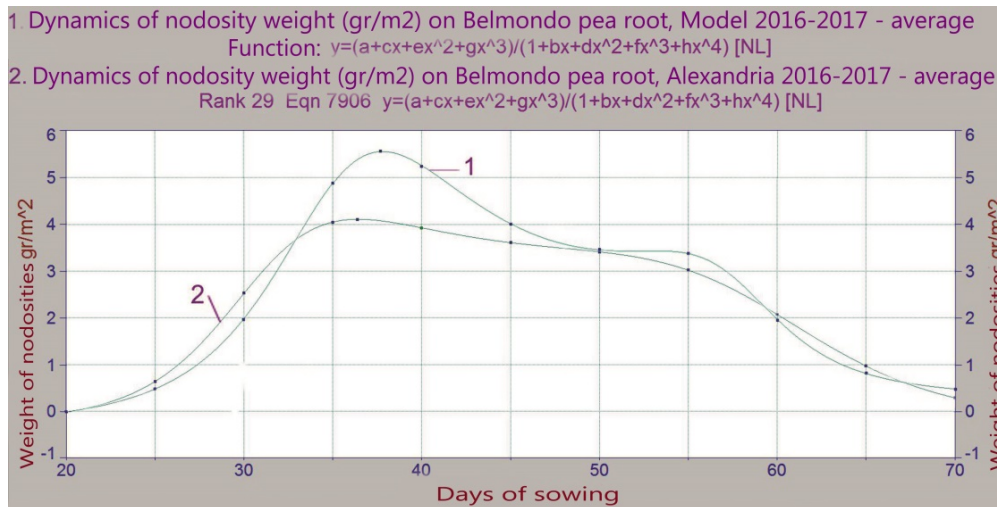
Figure no. 2 shows the dynamics of the dry nodosities weight, expressed in g/m² from the Belmondo pea roots.

Curve 1 represents Modelu (Călărași), the function being a polynomial of type NL (2), namely:

$$y = \frac{a + cx + ex^2 + gx^3}{1 + bx + dx^2 + hx^4} \quad (NL) \tag{2}$$

the numeric values of the functions are below the graph in Figure no. 2.

Curve 2 represents Alexandria and has the same form, the difference being given by the parameters (a, b, c, d, e, f, g). In both situations, dry nodosities can be weighed on the 25th day after appearance. Maximum weights are obtained at 38 days (5.8 g/m²) (order I) at Modelu (Calarasi), and at 37 days (4.1g/m²) for Alexandria. We note that both, the number of nodosities and their dry weight had the maximum at 37-38 days after sowing, the decreases being slow, allowing the plant to accumulate nitrogen up to 65-70 days after sowing. Between the number of nodosities and their dry weight we found the existence of a significant correlation which has to be calculated. We have also found that an equal number of nodosities develop differently in weight during the maximum assimilation period. Observations have shown that in Modelu (Calarasi), larger and more active nodosities have been formed than in Alexandria. The amount of "red" (leghemoglobin) was greater and more intense at Modelu (Calarasi). Probably because of a lighter and more accessible texture of soil.



$$(1) y = \frac{-0.2160 + 0.018x - 0.00040x^2 + 2.816x^3}{1 - 0.0889x + 0.029x^2 - 4.232x^3 + 2.275x^4} \quad (NL)$$

$$(2) y = \frac{-0.205 + 0.005x + 3.1095x^2 - 1.0617e^{-0.6}}{1 - 0.0925x + 0.032x^2 - 4.7794e^{-0.5} + 2.65062e^{-0.7}} \quad (NL)$$

The system of equations is complex, but is provided by a very significant correlation ratio for both equations

$$r^2 = 0.9999$$

Figure no. 2: Dynamics of nodosities weight (g/m²) on the roots of the Belmondo pea's variety at Modelu (1) and Alexandria (2), on average for 2016-2017

The amount of nitrogen accumulated by pea's plants, Belmondo variety

For the calculation of accumulation of fixed nitrogen, was used the transformation index published by Berca et al. (2018), namely:

$$N \text{ kg/ha} = \text{Nodozități kg/ha} \times 4.6$$

We mention from the outset, that this transformation only refers to the nitrogen obtained from the symbiosis with the plant and does not refer to forms of nitrogen coming into the soil through free fixation, associative fixation or degradation of organic matter. At the same time, data was accumulated from both locations and a complex function was calculated, by a specific program as shown in Figure no. 3.

We notice that the accumulation of fixed nitrogen begins after the initiation period, that is, the 25th day after sowing, after the nodosities have started to rise and leghemoglobin appears. Increased growth of accumulation is recorded from the appearance of the sixth leaf. At 8-10 leaves, growth is intense, and so does accumulation reaching a maximum (derived from order I of the function) at 42 on the day of appearance, in the amount of 6.52 kg/ha/day. After the 42 days, daily nitrogen biosynthesis decreases in the same rhythms (or almost the same) as in the case of their growth. The total fixation period is around days 32-35. The total amount of accumulated nitrogen was calculated by the integral of the function below, during the active period of the symbiosis process (fixation) (1).

$$\int_{20}^{65} f(x) d(x) = 115.89 \text{ kg} = 116 \text{ kg N/ha} \tag{1}$$

On the average for 2 years, for the two locations in southern part of Romania, for the Belmondo pea variety, was fixed an average amount of 116kg N/ha, exclusively from the fixing between the symbiote and the plant. If we take into account soil balance, the amount of nitrogen transited through soil and crops is between 160-190 kg N/ha.

The highest amount of nitrogen is fixed in the period of 30-50 days from sowing, that is in fact the period of high consumption for plants, related to flowering and the formation of pods (3).

$$\int_{30}^{50} f(x) d(x) = 96.55 \text{ kg} = 83\% \tag{3}$$

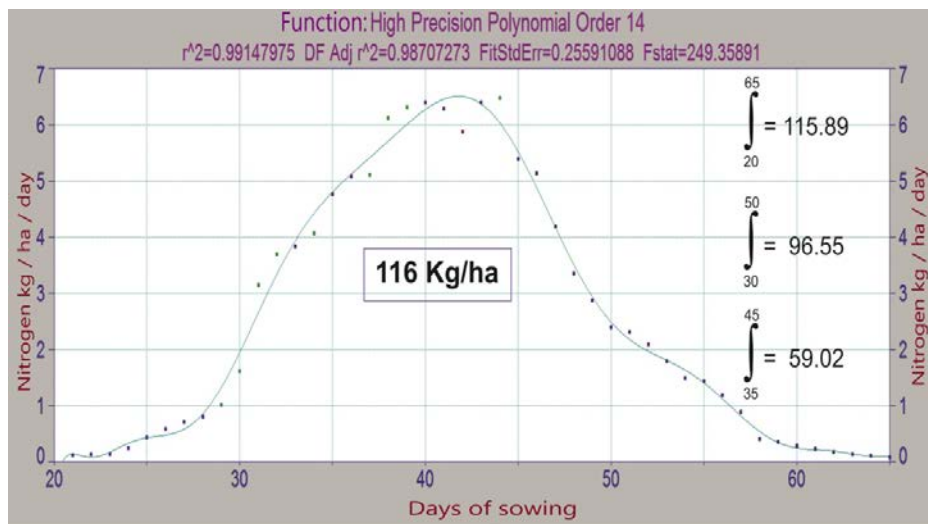


Figure no. 3: Dynamics of biologically fixed nitrogen accumulation kg s.a/ha/day, average Alexandria + Modelu 2016 – 2017

If we take into account the random factor, the biological correlation of the fixation (Figure no. 4), we can see that there may be years when the initiation and then fixation begin at 28 days after sowing, and it finishes at 55 days, i.e about 25 fixation days. But there are years when the fixation can last up to 45 days or more.

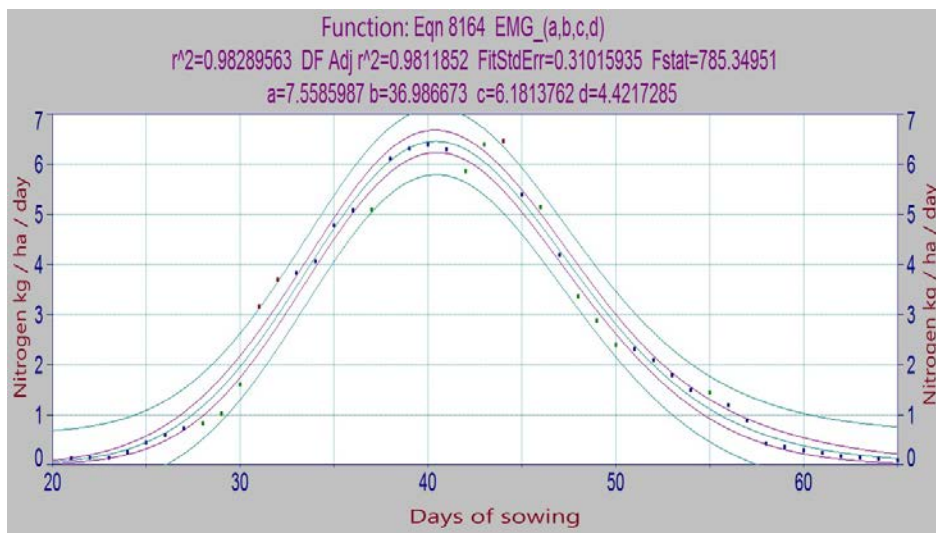


Figure no. 4: Dynamics of biologically fixed nitrogen accumulation kg/ha/day, mean Alexandria + Modelu, 2016-2017, calculated with confidence intervals for P = 5% and P = 1%

3. Discussions

The symbiotic nitrogen fixation has been studying for many years, but lately has become very useful for agricultural practice, especially when it comes about nutrition with nitrogen of crops from organic farming systems.

The FAO (2017) pays special attention to the formation of proteins from pea, in this way as a condition for obtaining a healthier and more sustainable diet. We believe that this desideratum can be a starting point for future analyses and research with bio-economic implications. The increase in the quantity of peas in systems and farming rotation of culture is supported by the specific funding of EU programs.

But very important is the technical and the ecological aspect. According to Praxisnah Zuchtung-Produktion Verwertung (2007), pea consumes a total of 51 kg N per one tonne of grain and straw. At 3.5 tonne, the consumption of nitrogen for the pea would be $3.5 \times 51 \text{ kg} = 178 \text{ kg}$. From this, 30 kg of the nitrogen returns to the soil through straw, remaining: $178 - 30 = 148 \text{ kg}$ of necessary nitrogen. If we divide $116/148 = 78\%$ coverage of nitrogen requirement from biological fixation. Pea does not need chemical nitrogen fertilizers, because the rest of about 12% takes from other forms of nitrogen, meaning other models of fixation.

From an economic point of view, we can point out that a saving of $116 \text{ kg} \times 1.3 = 151$ euro/ha is made, which makes peas a very economic and ecological culture, if we use the bioeconomic tools that this culture makes us available.

Biologically fixed nitrogen is absorbed by the whole plant, and the nodosities, microorganisms and roots become an excellent material for humus recovery. Another bio-economic implication with major potential for sustainable development for long-term for agriculture. The amount of humus that forms is equivalent to 160 kg C, this thing is highly appreciated by the EU's Cross Compliance Program.

We recall that, on a national scale, biological fixation becomes a natural model of nitrogen supply, a bio economic model, because the amount of fixed nitrogen can be increased using the symbiosis improvement at 700 kg/ha or nearly 400 euro/ha in Romania. The approximately 600,000 hectares of leguminous plants could bring the bioeconomic agriculture approximate 180,000 tons of clean Nitrogen, without cost and pollution. This would amount to 234 million euro and could, now, cover over 30% of the nitrogen requirement, without production costs and without the high cost of depollution.

Bioeconomic agriculture also involves the use of symbiotic, free or associative bacteria, that if today can bring up to 30% of the needs of nitrogen for plant nutrition, in the further the new researches can increase linear until reach the final equilibrium of nitrogen requirements of agricultural ecosystems.

Conclusions

Belmondo peas, in the southern part of Romania, fix atmospheric nitrogen using natural symbionts for a period of about 35 days.

The initiation of the fixation takes place approximately 22-25 days after sowing and stops at 60-65 days after sowing.

The number of nodosities on the roots of the plants began to form with the appearance of the third leaf. And they reach the maximum at 37-38 days. The maximum number of nodosities is about 54-58 per plant, on average.

The dynamics of the weight of the nodosities, correlates with their number, and reaches the maximum in the same period, with a size of 4 - 5.8 g/m².

The average amount of nitrogen fixed in this two years and for the two locations, for Belmondo pea variety is 116 kg N/ha and represents the 78% of the needs of crops consumption.

Nodosities are formed randomly, and they correlate closely with the soil conditions (humidity, temperature), and especially with the nitrogen content of the soil.

The savings achieved by symbiotic fixation of nitrogen, amount to more than 150 euro/ha, plus the improvement of the ecological index through the formation of humus (160 kg C), soil structure and microbiology.

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