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Research paper

# The efficiency of Nitrogen utilization and root nodules' life cycle in Alfalfa after various mineral fertilization and cultivation of soil

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ABSTRACT

The efficiency of the Nitrogen utilization and root nodules' life cycle in alfalfa after various mineral fertilizing and cultivation of soil methods have been investigated. The field test has been performed in the Forage Crops Institute, Pleven, Bulgaria on leached chernozem subsoil kinds and with no irrigation. The next cures have experimented: 1) for fertilization as below: N0P0K0 (controlling); N60P100K80 (an admitted technology); N23P100K35 (nitrogen has been used 1/2 in the first year of grow and 1/2 in the third year); N23P100K35 (nitrogen has been provided pre-sowing); N35P80K50, and Amophose – 250 kg/ha, estimated at fertilization rate N27P120K0; ii) for soil cultivation as below: soil losing 10-12 cm, plow at depth 12 to 15 cm, 22 to 24 cm (an admitted technologies), 18 to 22 cm and 30 to 35 cm. It has been seen that the cultivation of soil and mineral fertilizing had an impact on nitrogen utilization efficiency and the root nodules' life cycle in Alfalfa. Nitrogen utilization efficiency has been discovered for being maximum at N23P100K35 and plow at the depth of 22 to 24 cm. The root nodules' life cycle has been the longest at N35P80K50 and plows at the depth of 18 to 22 cm. The more useful root mass for nodule number ratio has been seen at N23P100K35 and plow at the depth of 22 to 24 cm.



## Introduction

Artificial nitrogen fertilizers' utilization in farming is important for product fertility (Tilman et al., 2002, Mueller et al., 2012, Sinclair and Rufty, 2012). They are a piece of "green revolution" after that although not sufficiently, while the plant protein have to be discovered growing (Lassaletta et al., 2014a). Nevertheless, now almost the entire half of nitrogen utilized for fertilization is lost to the surroundings. For nearly all of Europe, the losses are further fifty kg N ha/yr. This deduction has been accomplished by comprehensive study on nitrogen utilization performance (used to the products as artificial fertilizers, manure, symbiotic nitrogen consolidation and atmospheric confession).

This investigation has been accomplished according to the FAO database and 124 counties have been applied (Power and Alessi, 1971; Tilman et al., 2002; Herridge et al., 2008; Bouwman et al., 2009). Outcomes demonstrated which nitrogen utilization performance has been typically high to farming techniques by high ratio of N inputs obtained from symbiotic nitrogen consolidation. Contrarily, nitrogen utilization performance has been typically more down for a high ratio of artificial fertilizers in whole fertilization system. The high nitrogen utilization performance coonected to nitrogen consolidation is probably described by a high performance in the incorporation with legumes of the self-provided nitrogen (Herridge et al., 2008).

However, considerable progress in nitrogen utilization performance has appeared in some countries, a gain of nitrogen fertilization could be resulted in a disproportionately low growth of production with more environment modifications (Lassaletta et al., 2014b). Utilizing nitrogen inputs by low performance causes environment issues like as contamination of groundwater by nitrates, ammonia release and greenhouse gases.

Thus, the source utilization efficiency is important and its advancement is the critical challenge for endurable farming. Promising, whereas at the identical time environmental and social endurable, agroecosystems are required. Appropriate plans of fertilization and soil cultivation techniques are between the elements, which are essential for nitrogen and phosphorus utilization performance progress. A growth of nitrogen consolidation possibility as a nitrogen resource could contribute for improving nitrogen utilization performance in the local and global scale (Herrero et al., 2010, Lassaletta et al., 2014a, Bonaudo et al., 2014, Soussana and Lemaire, 2014).

Phosphorus is the 2-nd considerable essential plant nutrient separated from nitrogen, whereas for legumes, it assumes prior importance that recreates an essential role in root increase (Tairo and Ndakidemi, 2013). Richardson and Berea (2009) noted that growing, yield and yield details in legumes enhanced especially by good store of phosphorus. One of the most significant forage legume kinds is Alfalfa (Medicago sativa L.). It related to order Family Fabaceae, genus Medicago. The acceptable name is Medicago sativa L. It is a practical protein resource for animals by highly nutritive amount and rich in minerals (Barnes et al., 1995; Kertikova, 2008; Keskin et al., 2009). As a legume, Alfalfa is nitrogen consolidation crop and the possibility for nitrogen consolidation by symbiosis is approximately 450 kg/ha/year (Heichel and Henjum, 1991; Starchenkov and Kot's, 1992).

The additional introduction issue of nitrogen to Alfalfa and how the nitrogen utilization performance would be is arguable in the literature (Oliveira et al., 2004; Werner and Newton, 2005). Considerable writers backing the idea for nitrogen fertilization utilization (Cihacek, 1994; Petkova, 1994; Sharma and Sharma, 1995; Butorac et al., 1988; Raun et al., 1999; Trepachev, 1999; Delgado et al., 2001; Pachev, 2001; Tufenkci et al., 2006; Vasileva et al., 2006; 2011; Vasileva and Ilieva, 2011). Raun et al. (1999) have established the nitrogen fertilization demand. They have discovered an extra nitrogen fertilizer using (once in the spring) improved dry mass fertility in the cuts, harvested later, that is in line by decreased then nitrogen consolidation capability.

In the identical time high doses of mineral nitrogen inhibit the biological consolidation procedure (Streeter, 1985a,b; Kot's et al., 1990, 1996; Streeter, 1993; Kot's, 2001; Vasileva, 2004). This study objective has been investigation the nitrogen utilization performance and root nodules life cycle after various mineral fertilization and soil cultivation methods in Alfalfa.

**Methodology**

The investigation has been performed in the empirical area of the Institute of Forage Crops, Pleven, Bulgaria (2003-2006) on filtered chernozem soil subtype by not considering irrigation. Alfalfa diversity Obnova ten has been buried. Long plot technique has been utilized and plots size has been ten m$^2$. The cures being four-times reproduced existed: i) to fertilize as below: N0P0K0(controlling); N60P100K80 (by obtained technology); N23P100K35 (nitrogen has been used 1/2 in first year of grow, 1/2 in third year, and P and K provide); N23P100K35 (nitrogen has been provided pre-sowing, K provide, P by 1/3 in the first, second and third year of grow); N35P80K50, and Amophose - 250 kg/ha, estimated in fertilization rates N27P120K0; ii) to soil cultivation as below: soil loos10-12 cm, plow in depth 12-15 cm, 22-24 cm (an admitted technology), 18 to 22 cm and 30 to 35 cm. Most significant agrochemical features of the soil (specified by Page et al., 1982), have been as below: N, 31.5/1000 g soil; P (P2O5), 5.19 mg/100 gsoil;K (K2O), 25.4 mg/100 g soil; pH (H2O), 6.95; humus, 1.77 percent.

One cut in the first practical year and 2 cuts in the next 3 years have been gathered. Nitrogen utilization performance (kg/kg) has been estimated utilizing instructions of Bowen and Zapata (1991) and Yousefi and Mohammadi (2011), equation (1) Phosphorus utilization performance (kg/kg) has been estimated utilizing instructions of Bowen and Zapata (1991), equation (2):

$$Nitrogen\ utilization\ performance = dry\ mass\ yield\ \frac{\frac{kg}{ha}}{nitrogen}\ applied\ \left(\frac{kg}{ha}\right) \quad (1)$$

$$\text{Phosphorus}\ utilization\ performance = dry\ mass\ yield\ \frac{\frac{kg}{ha}}{\text{Phosphorus}}\ applied\ \left(\frac{kg}{ha}\right) \quad (2)$$

Soils of soil profile have been chosen (20/30/40 cm). Root mass has been cleaned by tap water and calculated (Beck et al., 1993): the life cycle period of root nodules – documented on the basic of inner color (days) (Milev, 2014), roots to number of root nodules ratio has been estimated (g roots/number root nodules) - dry root mass, dried at 60oC has been separated to number of root nodules. Empirical database have been averaged for the investigation time and statistical processed utilizing SPSS software program (2012).

**Discussion and Results**

Agro meteorological situations for the investigation period have been unfavorable according to Table 1. Long drought time happened in the first year. Early spring drought also unevenly dispersed rain appeared in the 2-nd empirical year. The 3-nd year has been specified as desirable (by better and evenly dispersed rain).

Table. 1. Agro meteorological situation for the investigation period

| Months | 1-st year | | 2-rd year | | 3-rd year | | 4-th year | |
|---|---|---|---|---|---|---|---|---|
| | t | rain | t | rain | t | rain | t | rain |
| | ºc | 1/m2 | ºc | 1/m2 | ºc | 1/m2 | ºc | 1/m2 |
| 1 | -0.1 | 53.4 | -2.2 | 26.1 | 2.4 | 55 | -2.9 | 8.8 |
| 2 | -3.4 | 27.6 | 3.2 | 23.9 | -1.7 | 46.3 | 0.6 | 121 |
| 3 | 4.7 | 10.2 | 7.4 | 41.6 | 5.1 | 75.8 | 6.8 | 176 |
| 4 | 10.9 | 83.6 | 13.1 | 6.9 | 12.4 | 57.4 | 13.2 | 30.9 |
| 5 | 20.5 | 74.5 | 16.1 | 87.5 | 17.5 | 101.0 | 17.8 | 33.2 |
| 6 | 23.9 | 12.8 | 20.3 | 70.3 | 19.8 | 95.9 | 21.0 | 47.3 |
| 7 | 23.7 | 49.7 | 23.5 | 38.5 | 22.6 | 115 | 23 | 78.4 |
| 8 | 25.5 | 1.4 | 22.6 | 82.4 | 21.4 | 156 | 23.2 | 63.4 |
| 9 | 17.7 | 67.6 | 18.4 | 42.2 | 17.4 | 225 | 18.7 | 48.5 |
| 10 | 10.7 | 107 | 14 | 14.6 | 17.4 | 25.5 | 17.4 | 25.5 |
| 11 | 7.4 | 24.8 | 7.9 | 21.6 | 5.1 | 24 | 5.1 | 24 |
| 12 | 1.2 | 28.2 | 2.6 | 43.3 | 2.7 | 41.7 | 3.3 | 32.5 |

Low rain and drought observed the last year.Nitrogen utilization performance is an agronomical element expressing the ratio among yield gained and nitrogen provided. Outcomes demonstrated greater nitrogen utilization performance for lower doses of nitrogen fertilizing, which nitrogen input has been more acceptable according to Table 2.

Table. 2. Nitrogen utilization performance of Alfalfa after various fertilization speeds and soil cultivation

| Cures | Soil Losing | | | Plow | |
|---|---|---|---|---|---|
| | 12 to 15 cm | 12 to 15 cm | 22 to 24 cm | 18 to 22 cm | 30 to 35 cm |
| | Kg | | | | |
| N60P100K80 | 67.33 | 72.17 | 78.17 | 61.83 | 70.67 |
| N23P100K35 | 210 | 196.09 | 210.87 | 182.61 | 203.91 |
| N23P100K35 | 208.26 | 209.13 | 208.26 | 188.26 | 194.35 |
| N35P80K50 | 126 | 117.43 | 127.43 | 120 | 110 |
| Amophose | 161.11 | 155.56 | 167.41 | 155.93 | 156.30 |
| SE | 26.8 | 25.3 | 25.2 | 23.3 | 25.4 |
| Maximum | 210 | 209.13 | 210.87 | 188.26 | 203.91 |
| Minimum | 67.33 | 72.17 | 78.17 | 61.83 | 70.67 |

Hartwig and Soussana (2001) discovered which Alfalfa utilized the soil or fertilizer nitrogen in the prior growth of the plants, since nitrogen merger requires low levels of CO2 and energy as in comparison to nitrogen fixation procedure, so needs using nitrogen. Vasileva and Pachev (2015) propose that the immediate growth of Alfalfa plants need nitrogen for avoiding the retention of root development. Justes et al. (2001) supposed that Alfalfa plants by not considering nitrogen fertilizing had a remarkably low root dry mass in comparison with plants by nitrogen fertilizing.

There have been small discrepancies in nitrogen utilization performance in N23P100K35, however nitrogen has been used in various ways (1/2 in first year of growing, 1/2 in third year or pre-sowing). Rather larger discrepancies have been discovered for the most superficial and most resounding soil plow. For various soil cultivation depths the nitrogen utilization performance for N23P100K35 can be organized in reducing manner as below: p;ow in the depth of 22 to 24 cm, soil losing in the depth of 12 to 15 cm, plow in the depths 12 to 15, 30 to 35 and 18 to 22 cm. The yield reactions to N addition can be restricted by imbalances by other nutrients like as phosphorus (van der Velde et al., 2014).

Phosphorus is essential to aboveground and root mass grow of Alfalfa. It has a critical position in the nodule elements like number, size and movement (Armstrong, 1999). Vasileva and Pachev (2009) and Jing-Wei Fan (2015) discovered both high nodulation capacity and dry mass fertility in Alfalfa plants that have been nicely provided by phosphorus. Data in the Table 3 demonstrated that phosphorus utilization performance has been discovered for being high for low doses of phosphorus fertilizing and vice versa. So, the phosphorus utilization performance for whole soil cultivation depths has been the maximum in the fertilizing dose of N35P80K50.

Table. 3. Phosphor usutilization performance of Alfalfa after various fertilization speeds and soil cultivation

| Cures | Soil Losing | | | Plow | |
|---|---|---|---|---|---|
| | 12 to 15 cm | 12 to 15 cm | 22 to 24 cm | 18 to 22 cm | 30 to 35 cm |
| | Kg/kg | | | | |
| N60P100K80 | 40.40 | 43.30 | 46.90 | 37.10 | 42.40 |
| N23P100K35 | 48.30 | 45.10 | 48.50 | 42 | 46.90 |
| N23P100K35 | 47.90 | 48.10 | 47.90 | 43.30 | 44.70 |
| N35P80K50 | 55.13 | 51.38 | 55.75 | 52.50 | 48.13 |
| Amophose | 36.25 | 35 | 37.67 | 35.08 | 35.17 |
| SE | 3.30 | 2.75 | 2.88 | 3.03 | 2.29 |
| Maximum | 55.13 | 51.38 | 55.75 | 52.50 | 48.13 |
| Minimum | 36.25 | 35 | 37.67 | 35.08 | 35.17 |

It differed in a narrow limitations for the remainder empirical doses, that the provide by phosphorus has been 100 kg/ ha. Phosphorus utilization performance has been the high at the plow in the depth of 22 to 24 cm and N35P80K50.

Symbiotic nitrogen consolidation happen in the root nodules (Serraj et al., 1999). Pink colored and placed in the major root nodules are sign for their efficacy (Beck et al., 1993; Athar and Johnson, 1996; Athar and Shabbir, 1997; Kostov and van Cleemput, 1997). Life cycle period of root nodules is essential for the normal function of the structures and fix of more nitrogen also. Milev (2014) discovered the amount of this indicator in pea has been instantly proportionate to the vegetation rain in the year and the period has been among thirty and fifty-two days. Life cycle period of root nodules in this investigation differed among 32.2 and 41.6 days according to Table 4. Life cycle period of root nodules has been the minimum in the controlling. The most extended period of 39.1 days has been discovered at the shallow soil cultivation and maximum nitrogen provide (N60P100K80).

Table. 4. The life cycle period of root nodules of Alfalfa after various fertilization speeds and soil cultivation

| Cures | Soil Losing | | | Plow | |
|---|---|---|---|---|---|
| | 12 to 15 cm | 12 to 15 cm | 22 to 24 cm | 18 to 22 cm | 30 to 35 cm |
| | days | | | | |
| N0P0K0 | 34.1 | 32.2 | 34.2 | 38.4 | 36.0 |
| N60P100K80 | 39.1 | 37 | 35.4 | 39 | 39.2 |
| N23P100K35 | 35.4 | 38.6 | 39.5 | 38.8 | 37.3 |
| N23P100K35 | 36.3 | 33.7 | 38.6 | 40.2 | 39.8 |
| N35P80K50 | 35 | 34.6 | 35.8 | 41.6 | 38.3 |
| Amophose | 34.5 | 37.4 | 35.6 | 41.4 | 39.6 |
| SE | 0.74 | 1 | 0.84 | 0.56 | 0.6 |
| Maximum | 39.1 | 38.6 | 39.5 | 41.6 | 39.8 |
| Minimum | 34.1 | 32.2 | 34.2 | 38.4 | 36 |

More extended life cycle of root nodules has been noticed in the plants generously provided by phosphorus (for the plow at the depths of 22 to 24, 18 to 22 and 30 to 35 cm). The results of Asuming-Brempong et al. (2013) have been identical for cowpea. Keeping in mind the features contained in the analysis of root mass to nodule number ratio, low amounts of this ratio demonstrated more suitable condition of root mass by root nodules according to Table 5. The better assurance of root mass by root nodules has been discovered at N23P100K35 (nitrogen has been used 1/2 in first year of grow, 1/2 in third year) and plow at the depth of 22 to 24 cm. Nitrogen utilization performance and the more suitable root mass for nodule number proportion in Alfalfa has been discovered to be most elevated at N23P100K35 (nitrogen has been used 1/2 in first year of grow, 1/2 in third year) and plow in the depth of 22 to 24 cm. Growing speed such as biomass yield of alfalfa have to be investigated additionally detail. forage high quality after producing by speeds N35P80K50 has been discovered by Naydenova and Pachev (2009).

Table. 5. Root mass to nodule number ratio of Alfalfa after various fertilization speeds and soil cultivation

| Cures | Soil Losing | | | Plow | |
|---|---|---|---|---|---|
| | 12 to 15 cm | 12 to 15 cm | 22 to 24 cm | 18 to 22 cm | 30 to 35 cm |
| | g roots/number root nodules | | | | |
| N0P0K0 | 2.6214 (4.58/1.8) | 0.7608 (4.56/6.0) | 0.5462 (4.64/8.5) | 0.6756 (5.74/8.5) | 0.6758 (5.23/7.8) |
| N60P100K80 | 0.9690 (5.08/5.3) | 0.6387 (4.95/7.8) | 0.5344 (4.81/9.0) | 0.6703 (5.69/8.5) | 0.4759 (5.23/11.0) |
| N23P100K35 | 2.4857 (4.35/1.8) | 0.5046 (4.41/8.8) | 0.4288 (5.14/12.0) | 0.6732 (5.72/8.5) | 0.6746 (5.90/8.8) |
| N23P100K35 | 2.2089 (4.97/2.3) | 0.9150 (4.57/5.0) | 0.5092 (4.96/9.8) | 0.6850 (5.82/8.5) | 0.5498 (5.91/10.8) |
| N35P80K50 | 2.2422 (5.04/2.3) | 0.9895 (4.94/5.0) | 0.6144 (5.53/9.0) | 0.6897 (6.55/9.5) | 0.6568 (6.07/9.3) |
| Amophose | 1.6438 (5.34/3.3) | 0.6213 (4.81/7.8) | 0.7348 (5.69/7.8) | 0.6514 (5.70/8.8) | 0.7315 (6.03/8.3) |
| SE | 0.2523 | 0.0007 | 0.0004 | 0.0005 | 0.0003 |
| Maximum | 2.6214 | 0.9895 | 0.7348 | 0.6897 | 0.7315 |
| Minimum | 0.9690 | 0.5046 | 0.4288 | 0.6514 | 0.4759 |

**Conclusion**

Soil cultivation and mineral fertilizing had influence on nitrogen utilization performance and life cycle of root nodules in Alfalfa. Nitrogen utilization efficiency has been discovered to be high at N23P100K35 (nitrogen has been used 1/2 in first year of grow, 1/2 in third year) and plow at the depth of 22 to 24 cm; and the phosphorus utilization performance has been the most increased at the identical depth of soil cultivation and N35P80K50. Root nodules' life cycle has been the longest at N35P80K50 and plow at the depth of 18 to 22 cm. The better root mass to nodule number ratio has been discovered at N23P100K35 (nitrogen has been applied 1/2 in first year of grow, 1/2 in third year) and plow in the depth of 22 to 24 cm.

**Conflict of interest**

The authors declare that they have no conflict of interest.

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