INFLUENCE OF SOIL TILLAGE SYSTEMS AND INOCULATION ON SOYBEAN NODULATION AND YIELD

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Abstract

A field study was conducted over two agricultural years, 2015 - 2016, and was aiming to establish the influence of some soil tillage systems and inoculation on soybean nodulations and grain yield. The experimental design of the research, was organized using a split plot method and the following factors were analyzed: Factor A -soil tillage system: Conventional tillage (CT): Ploughing at 20 cm (control, A_{20}); Minimum Tillage (MT): Chisel plow at 20 cm (C_{20}), Chisel plow at 40 cm (C_{40}), Disking at 10 cm (D_{10}), Disking / Ploughing at 20 cm (D/A_{20}) and Disking/ Chisel at 40 cm (D/C_{40}); Factor B -soybean varieties from different maturity groups: Carla, 000, PR92B63, 0; Factor C -soybean seeds inoculation with Nitragin Bac Soya: non - inoculated; inoculated.

Influenced by nitrogen - fixing bacteria and soil tillage systems, the soybean nodulation recorded an increase of 37.9%. After two years of research, the average grain yield for soybean varieties sowed in variants treated with Bradyrhizobium japonicum, ranged from 2177 kg/ha (D) to 2044.6 kg/ha (D/C_{40}). For variants, where soybean seeds weren't inoculated with nitrogen - fixing bacteria, the grain yield ranged in average from 2044.6 kg/ha (D) to 2422.7 kg/ha (D/C_{40}). An increase of soybean grain yield with 5.4 % was brought by seed inoculation with nitrogen-fixing bacteria.

Key words: soybean, minimum tillage, conventional tillage, nodulation, yield,

INTRODUCTION

As а basic component of sustainable agriculture, the unconventional soil tillage systems (Marin et al., 2011), reduce the soil erosion (Guş and Rusu, 2011; Marin, 2011; Li et al., 2017), has a favorable influence on soil water conservation (Hatfield and Stewart, 1994; Marin et al. 2007; Busari et al., 2015), involves a lower energy consumption (Rusu et al. 2009; Rusu, 2014) and costs (Höflich et al., 1999; Ignea et al., 2012; Li et al, 2017), and on a long-term supports crops' productivity (Soane et al., 2012; Pittelkow et al., 2015; cited by Li et al. 2017).

Researches carried out in Romania, regarding the influence of unconventional soil tillages systems on soybean grain yield, reported increases between 1.6 % and 7% (Guş and Rusu, 2011; Guş et al., 2013; Căpățână and Ciocan, 2016) compared to the conventional system.

All the physical, chemical and biological properties of the soil are influenced by the

tillage systems (Jităreanu and Ailincai, 1999; Feiza and Cesevicius, 2006; Cara et al., 2008; Cociu and Alionte, 2011; Dogan et al., 2011; Sheibani and Ahangar, 2013). Compared to the conventional system, minimum tillage system favors the microbial activity (Höflich et al., 1999), of the arable layer. As an important element of a sustainable agricultural system, the symbiotic nitrogen fixation (SNF) by leguminous plants has a major contribution in influencing cycle of the nitrogen in nature (Ferguson, 2013) and is also an important source of renewable energy (Rotaru, 2009). On long-term. SNF improves, soil fertility (Bohlool et al., 1992; Cass et al., 1994; Tago et al., 2011; Matsumiya et al., 2013; Ferguson, 2013), can successfully replace the synthetic nitrogen fertilizers (Kovačević et al., 2011) and has a positive influence on crops' grain yields (Căpătână et al., 2017).

Soybean *Glycine max* L. (Merrill) is an important commercial crop at a global level (Subramanian and Smith, 2013), primarily cultivated due to its high content of protein and

oil (Hymowitz et al., 1972) and also due to the plants' capacity to fix atmospheric nitrogen (SNF), through roots' nodules resulted from the symbiosis with *Bradyrhizobium japonicum* nitrogen - fixing bacteria (Chetan et al., 2014).

MATERIALS AND METHODS

This study, conducted during 2015 - 2016, presents results, regarding the influence of conventional tillage (CT) and minimum tillage (MT) on number of root-nodules and grain yield, of soybean crop. The experiment was established on a chromic luvisol with a clay-loam texture, a moderately acid reaction (pH 5.2 - 5.4) and a low humus content that ranged between 2.1% and 2.2%, located at Moara Domneasca Didactic Farm, Ilfov County (Mihalache et al., 2010) based on a split splot set-up with the following factors tested:

Factor A - Tillage system with six graduations: *Conventional system (CS)*:

a1 - Ploughing at 20 cm (A20 - Control, C);

Minimum tillage (MT):

 a_2 - Chisel plow at 20 cm (C₂₀); a_3 - Chisel plow at 40 cm (C₄₀); a_4 - Disking at 10 cm (D₁₀); a_5 - Disking at 10 cm / Ploughing at 20 cm (D/A₂₀); a_6 - Disking at 10 cm / Chisel at 40 cm (D/C₄₀).

Factor B - soybean varieties:

b₁ - Carla, 000 (ISTIS 2015);

b₂ - PR92B63, 0;

Factor C - seeds inoculation with Nitragin Bac Soya before sowing:

c₁ - non-inoculated seeds;

c2 - inoculated seeds.

For D/A₂₀, and D/C₄₀, the soil tillage were applied alternative, as follows: D for the previous crop and A₂₀ and C₄₀ for the soybean crop. During the two years of research, the soybean varieties were sown on April 16th 2015 and on April 9th 2016, using an SPC sowing machine at 50 cm between rows. The fertilization was assured by a complex fertilizer N₄₅P₆₀K₄₅ (kg/ha a.s), applied at seedbed preparation. Prior to soybean sowing, seeds were treated with Nitragin Bac Soya (pure bacterial culture of *Bradyrhizobium japonicum*) at a dose of 300 g/ha. Weed control was performed, preemergent with *S-metolachlor* 960 g/l (Dual Gold, 1.5 l/ha). After plants' emergence, the control of dicotyledonous weeds was assured by using 480 g/l *bentazone* + 150 g/l *wettol* (Basagran, 2 l/ha) and for the monocotyledonous weeds *quizalofop-P-teflil* 40 g/l (Pantera, 1.2 l/ha) was used.

Harvesting date of varieties was different, influenced by the maturity group and by the climatic conditions of the area: a1 - Carla 000 (ISTIS 2015), September 07th 2015 (144 days after sowing), September 10th 2016 (154 days after sowing); a₂ - PR92B63 0, October 19th 2015 (186 days after sowing), October 21th 2016 (195 days from sowing). The number of nodules per plant for sovbean varieties was determined once at every two weeks, from May 31 to June 30 (2015 - 2016). At June 30th, determination of nodules number per plant was conducted when soybean varieties were in different stages of development, Carla - R3 (beginning of pods formation) and PR92B63 -R1 (beginning of blooming).

Climatic conditions of the area significantly influenced soybean plants development. The average amount of annual rainfall was 662.4 mm, higher than the normal values of the area with 106.3 mm and the average temperature was 12.2°C compared to normal of the area 10.5°C (Table 1).

Table 1. Climatic conditions, Moara Domneasca, Ilfov County. Average 2015 - 2016

| | Average 2 | 2015 - 2016 |) | |
|-----------------|-----------|-------------|---------|--------|
| | Tempera | ture (°C) | Rainfal | l (mm) |
| Month | Average | | Average | |
| wonun | 2015 - | Normal | 2015 - | Normal |
| | 2016 | | 2016 | |
| October | 11.3 | 11.0 | 67.1 | 35.8 |
| November | 6.8 | 5.3 | 79.9 | 40.6 |
| December | 2.2 | 0.4 | 43.2 | 36.7 |
| January | -2.5 | -3.0 | 48.0 | 30.0 |
| February | 1.9 | -0.9 | 28.5 | 32.1 |
| March | 6.9 | 4.4 | 66.4 | 31.6 |
| April | 13.0 | 11.2 | 33.3 | 48.1 |
| May | 17.2 | 16.5 | 52.3 | 67.7 |
| June | 21.7 | 20.2 | 85.8 | 86.3 |
| July | 24.7 | 22.1 | 4.7 | 63.1 |
| August | 23.7 | 21.1 | 68.6 | 50.5 |
| September | 18.9 | 17.5 | 84.6 | 33.6 |
| Avg. / Sum Oct. | 12.2 | 10.5 | 662 1 | 556.1 |
| - Sept. | 12.2 | 10.5 | 002.4 | 550.1 |
| Avg. / Sum Apr. | 19.9 | 18.1 | 329.3 | 349.3 |
| - Sept. | | | | |

During the vegetative period (April -September), average temperature was 19.9°C, 1.8°C higher than the normal value of the area (18.1°C). Average rainfall recorded during the vegetative period was 329.3 mm, close to normal values of the area (349.3 mm), (Table

1). The rainfall distribution during the vegetative period had influenced the development of soybean varieties. On average during the two years of research (2015 - 2016). July rainfalls' recorded a critical value of 4.7 mm, which represented 7.4% of the normal values of the area (63.1 mm), having a negative influence on varieties' yield, with them being in the reproductive period, Carla R3 - R5, and PR92B63 R1 - R3.

RESULTS AND DISCUSSIONS

Influence of soil tillage systems on soybean number of nodules

On average in two years of research (2015 - 2016) under the climatic conditions at Moara Domneasca, in variants where the soybean

seeds were inoculated before sowing, the soybean number of nodules per plant at 30^{th} of June, recorded differences from -2.3 (D) to 7 (C₄₀) nodules/plant at Carla and between -2.7 (D) and 6.8 (C₄₀) nodules/plant at PR92B63 statistically significant compared to control (A₂₀).

For the Control (A₂₀, C) variant, where the conventional system was applied, soybean varieties recorded on average 33.1 Nodules nodules/plant. number per plant increased with very significant values, statistically assured, for variants in which minimum tillage systems with C_{20} , C_{40} , and D/C₄₀, were applied. The highest number of nodules per plant was recorded by PR92B63 variety, 41.7 nodules/plant for variant C₄₀, with an increase of 19.6 % compared to A_{20} , (Table 2).

 Table 2. Soil tillages influence on the number of nodules per plant in variants treated with Nitragin Bac Soya, 30thJune.

 Average 2015 - 2016

| | CARLA | | | | | PR92 | B63 | | Average - Varities | | | | |
|-------------|------------------|------------|------------|-------|-----------------|------|-------|-------|--------------------|------|-------|-------|--|
| Variants | No. Nod./pl. | Diff | % | Signf | No. Nod./pl. | Diff | % | Signf | No. Nod./pl. | Diff | % | Signf | |
| A20 | 31.3 | С | 100.0 | - | 34.8 | С | 100.0 | - | 33.1 | С | 100.0 | - | |
| C20 | 35.8 | 4.5 | 114.3 | *** | 39.4 | 4.6 | 113.2 | *** | 37.6 | 4.5 | 113.7 | *** | |
| C40 | 38.3 | 7.0 | 122.4 | *** | 41.7 | 6.8 | 119.6 | *** | 40.0 | 6.9 | 120.9 | *** | |
| D | 29.0 | -2.3 | 92.7 | 000 | 32.2 | -2.7 | 92.3 | 000 | 30.6 | -2.5 | 92.5 | 000 | |
| D/A20 | 31.8 | 0.5 | 101.7 | - | 35.1 | 0.3 | 100.8 | - | 33.5 | 0.4 | 101.2 | - | |
| D/C40 | 35.2 | 3.9 | 112.4 | *** | 38.2 | 3.4 | 109.8 | *** | 36.7 | 3.6 | 111.0 | *** | |
| LSD 5% = 1. | $0: LSD \ 1\% =$ | 1.3: LSD (| 0.1% = 1.7 | 7 | | | | | | | | | |

*Note: C - control. ns - not significant. * positive significance. 0 negative significance; *No Nod./pl = Number of. Nodules / plant.

For the non - inoculated variant (Table 3), the number of nodules for soybean varieties recorded statistically significant increases from 5.2 % to 18.6 %. For both varieties, the number of nodules per plant recorded lower values compared to A_{20} with negative differences between -0.6 (Carla) and -1.2 (PR92B63) for

the variants where the minim tillage with D was applied. Under the soil tillage influence at June 30^{th} , the varieties sown in non - inoculated variant recorded between 22.9 and 28.2 nodules/plant, compared to Control (A₂₀) (Table 3).

Table 3. Soil tillages influence on the number of nodules per plant in variants not treated with Nitragin Bac Soya, 30^{th} June. Average 2015 - 2016

| | | | PR92 | B63 | | Average - Varities | | | | | | |
|----------|----------------|------|-------|-------|---------------|--------------------|-------|-------|---------------|------|-------|-------|
| Variants | No. Nod /pl | Diff | % | Signf | No Nod /pl | Diff | % | Signf | No Nod /pl | Diff | % | Signf |
| 1.20 | 22.0 | 0 | 100.0 | | 24.6 | 0 | 100.0 | | 22.0 | 0 | 100.0 | |
| A20 | 23.0 | C | 100.0 | - | 24.6 | C | 100.0 | - | 23.8 | C | 100.0 | - |
| C20 | 26.3 | 3.3 | 114.3 | *** | 27.6 | 3.0 | 112.2 | *** | 27.0 | 3.1 | 113.2 | *** |
| C40 | 25.4 | 2.3 | 110.1 | *** | 27.2 | 2.6 | 110.5 | *** | 26.3 | 2.5 | 110.3 | *** |
| D | 22.4 | -0.6 | 97.3 | 0 | 23.4 | -1.2 | 95.3 | 000 | 22.9 | -0.9 | 96.2 | 00 |
| D/A20 | 24.2 | 1.2 | 105.2 | *** | 25.9 | 1.3 | 105.4 | *** | 25.1 | 1.3 | 105.3 | *** |
| D/C40 | 27.2 | 4.2 | 118.3 | *** | 29.2 | 4.6 | 118.6 | *** | 28.2 | 4.4 | 118.5 | *** |

LSD 5% = 0.6; LSD 1% = 0.8; LSD 0.1% = 1.1

*Note: C – control. ns – not significant. * positive significance. 0 negative significance; *No Nod./pl = Number of. Nodules / plant.

Influence of Nitragin Bac Soya on soybean number of nodules

Influenced by Nitragin Bac Soya treatment, the number of nodules per plant recorded an very significant increase of 9.7 nodules/plant (37.9 %), very significantly positive, compared to non-inoculated, Control (C). In two years of research (2015 - 2016), the Nitragin Bac Soya inoculation brought an statistically significant increase of nodules number per plant with values between 7.7 and 13.7. For variants were the conventional tillage system (A_{20}) was applied, inoculation with Nitragin Bac Soya, generated an increase of 9.3 nodules/plant compared to the control. The average number of nodules per plant (2015 - 2016) for the variant inoculated with Nitrogen Bac Soya, ranged from 30.6 (D) to 40 (C₄₀) nodules/plant (Figure 1).



Figure 1. Influence of Nitragin Bac Soya treatment on the nodules number. Average 2015 -2016

Influence of soil tillage systems on soybean grain yield

Under the influence of soil tillage grain yield for the variant treated with Nitragin Bac Soya, in two years of research (2015 - 2016) recorded statistically significant increases ranging from 93.6 kg/ha (PR92B63, D/A₂₀) to 120.1 kg/ha (Carla, D/C₄₀), compared to A₂₀ (Table 4). According to data presented in Table 4, varieties' average grain yield compared to control recorded differences from -279.0 kg/ha (very significantly negative, D) to 115.4 kg/ha (distinctly significant positive, D/C_{40}).

Negative differences, statistically assured were compared to compared to A_{20} were recorded for both varieties in variants where the minimum tillage with D and C₂₀ were applied. Influenced by soil tillage and Nitragin Bac Soya inoculation, the highest grain yield was 2639.0 kg/ha (PR92B63, D/C₄₀) and the lowest was 2083.2 kg/ha (Carla, D).

 Table 4. Soil tillage influence on soybean grain yield (kg/ha), in variants treated with Nitragin Bac Soya.

 Average 2015 - 2016

| | CARLA | | | | | PR92B | 53 | | Average - Variety | | | | |
|---------------------------------|---------------|-------------|-------------|---------------|-------------|--------|-------------|---------------|-------------------|--------|-------|-----|--|
| Variants GY Diff kg/ha kg/ha | % | Signf | GY kg/ha | Diff kg/ha | % | Signf | GY kg/ha | Diff kg/ha | % | Signf | | | |
| A20 | 2383.6 | C | 100.0 | - | 2528.2 | C | 100.0 | - | 2455.9 | C | 100.0 | - | |
| C20 | 2305.3 | -78.3 | 96.7 | 0 | 2449.4 | -78.8 | 96.9 | 0 | 2377.4 | -78.6 | 96.8 | 0 | |
| C40 | 2492.8 | 109.2 | 104.6 | ** | 2607.5 | 79.3 | 103.1 | * | 2550.2 | 94.2 | 103.8 | ** | |
| D | 2083.2 | -300.4 | 87.4 | 000 | 2270.7 | -257.5 | 89.8 | 000 | 2177.0 | -279.0 | 88.6 | 000 | |
| D/A20 | 2421.3 | 37.6 | 101.6 | - | 2621.8 | 93.6 | 103.7 | ** | 2521.6 | 65.6 | 102.7 | - | |
| D/C40 | 2503.8 | 120.1 | 105.0 | ** | 2639.0 | 110.8 | 104.4 | ** | 2571.4 | 115.4 | 104.7 | ** | |
| LSD 5% = 6 | 8.9 kg/ha; LS | D 1% = 92.8 | 8 kg/ha; LS | SD 0.1% = | 123.1 kg/ha | | | | | | | | |

*Note: C – control. ns – not significant. * positive significance. 0 negative significance;

According to Table 5, the highest grain yield for non-inoculated variant was 2492.9 kg/ha (PR92B63, D/A₂₀), with an increase of 2.9 %

compared to control variant (A_{20}) . Compared to control lower yields were recorded for variants where the minimum tillage with D was applied

(Table 5) by both Carla and PR92B63, with significant negative differences between -297.7 kg/ha (Carla) and -315.2 kg/ha PR92B63. On average, due to the minimum

tillage system soybean varieties registered differences in yield statistically assured between -306.5 kg/ha at D and 71.7 kg/ha at D/C_{40} (Table 5).

Table 5. Soil tillages influence on soybean grain yield (kg/ha), in variants not treated with Nitragin Bac Soya. Average 2015 - 2016

| | CARLA | | | | | PR92B | 63 | | Average - Variety | | | | |
|----------|--------|--------|-------|-------|--------|--------|-------|-------|-------------------|--------|-------|-------|--|
| Variants | GY | Diff | % | Signf | GY | Diff | % | Signf | GY | Diff | % | Signf | |
| | kg/ha | kg/ha | | | kg/ha | kg/ha | | | kg/ha | kg/ha | | | |
| A20 | 2278.9 | С | 100.0 | - | 2423.2 | С | 100.0 | - | 2351.0 | С | 100.0 | - | |
| C20 | 2191.1 | -87.8 | 96.1 | 0 | 2330.3 | -92.9 | 96.2 | 00 | 2260.7 | -90.3 | 96.2 | 00 | |
| C40 | 2358.4 | 79.5 | 103.5 | * | 2467.4 | 44.2 | 101.8 | - | 2412.9 | 61.9 | 102.6 | - | |
| D | 1981.2 | -297.7 | 86.9 | 000 | 2108.0 | -315.2 | 87.0 | 000 | 2044.6 | -306.5 | 87.0 | 000 | |
| D/A20 | 2335.6 | 56.8 | 102.5 | - | 2492.9 | 69.7 | 102.9 | * | 2414.3 | 63.2 | 102.7 | - | |
| D/C40 | 2366.0 | 87.1 | 103.8 | * | 2479.5 | 56.3 | 102.3 | - | 2422.7 | 71.7 | 103.0 | * | |

Influence of Nitragin Bac Soya on soybean grain yield

During 2015 - 2016, inoculation with Nitragin Bac Soya had a favorable influence on soybean yields (Figure 2). Thus, under the influence of Nitragin Bac Soya inoculation influence, the average grain yield of varieties recorded increases between 104.9 kg/ha and 148.6 kg/ha, compared to non-inoculated, statistically assured, Figure 2. The average grain yield increase brought by inoculation was 124.5 kg/ha, higher with 5.4 % compared to the non - inoculated variant. The highest grain yield was recorded at D/C_{40} with a value of 2571.4 kg/ha, by 6.1% higher than control (non-inoculated).



Figure 2. Influence of Nitragin Bac Soya treatment on soyabean grain yield. Average 2015 - 2016

CONCLUSIONS

For the two varieties tested in South - East of Romania, during the agricultural years of 2014/2015 - 2015/2016, the highest number of nodules per plant in variants treated with Nitragin Bac Soya, was 29.2 (PR92B63, D/C40) and the lowest was 22.4 (Carla, D). On average, during the two years of research, under the inoculation with Nitragin Bac Soya (pure bacterial culture of *Bradyrhizobium japonicum*), the number of nodules per plant recorded an increase of 37.9%. The average

grain yield of the two varieties (2015-2016), under the influence of soil tillage systems along with inoculation, ranged between 2177.0 kg/ha and 2571.4 kg/ha, and for the non-inoculated variant the average yield varied between 2044.6 kg/ha and 2422.7 kg/ha.

The lowest grain yield was 1981.2 kg/ha recorded by Carla at D, non-inoculated variant, and the highest was 2639.0 kg/ha recorded by PR92B63 at D/C_{40} inoculated variant. For variants inoculated with Nitragin Bac Soya, grain yield recorded increases compared to A₂₀ (C) between 2.7 % and 4.7 % for the minimum

tillages systems (MT) C₄₀, D/C₄₀, D/ A₂₀. For variants were conventional tillage (CT) was applied the average grain yield of varieties ranged from 2351.0 kg/ha and 2455.9 kg/ha. Regardless of the Nitragin Bac Soya inoculation, the grain yield of varieties recorded for D/C₄₀ had on average higher values statistically assured compared to A₂₀ with 71.7 kg/ha (non-inoculated variant) and with 115.4 kg/ha (inoculated variant).

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