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ESCUELA DE AGRONOMÍA

"EFECTO DE LA APLICACIÓN DEL BIOFERTILIZANTE TWIN N EN UN CULTIVO DE PAPA (*Solanum tuberosum* L.) VARIEDAD DESIREÉ ESTABLECIDO EN UN ANDISOL DE LA REGIÓN DE LA ARAUCANÍA"

Tesis de grado presentada a la Facultad de Recursos Naturales como parte de los requisitos para optar al título de:

INGENIERO AGRÓNOMO

MARÍA JOSÉ HUENCHULEO HUENTECURA

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“Effect of the application of Twin N biofertilizer on potato crop (*Solanum tuberosum* L.) Desireé variety established in an Andisol of the Araucanía Region”

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SUMMARY

The negative effects caused by chemical fertilizers on the environment have led in recent years, research on ecological crop management, as biofertilizers, using nitrogen-fixing microorganisms alternatives. During the 2013-2014 season, a field trial at the Experimental Station Pillanlelbún that aimed to assess the effect of biofertilizer Twin N in a potato crop, variety Desiree was established. An experimental design of blocks completely randomized with three treatments (T0 = control, T1 = 50 kg N to aporca and T2 = Twin N) and four repetitions. At harvest, plants yield per hectare, number of tubers, and weight distribution was quantified by size, plus health status and sprouting along with the effect of biofertilizer on arbuscular mycorrhizal fungi (AMF). The total yield increased significantly by 41%, added with Twin N treatment than the control, this strongly influenced by the performance of the highest caliber. Disease incidence in differences in dry rot and craters found. Regarding fungal parameters, treatment with N she had the lowest Twin colonization in roots and a low amount of spores in the rhizosphere. It is concluded that the addition of biofertilizer Twin N to a potato crop could partially replace chemical nitrogen fertilization reducing pollution problems in the soil.

Keywords: nitrogen fixing bacteria, chemical pollution, sustainable agriculture, bio-fertilizers.

INTRODUCTION

The potato (Solanum tuberosum L.) is a di cot annual herbaceous plant belonging to the family Solanaceae. The cultivation of this edible tuber, it is spread throughout the world (CIP, 2008) as the fourth important food product after corn, wheat, rice and constituting the main non-cereal food for mankind. In 2008, the World potato production exceeded 314 million tons, with a cultivated area of 18,192,405 hectares (PASO, 2010) area. According to the VII Agricultural Census in the country annually they are grown around 50,000 hectares occupying potato fourth with the highest number of associated farmers (about 59,600). The production is almost entirely for the domestic market where it occupies an important place in the diet of Chileans (PASO, 2015). From a regional perspective, in the 2013/2014 season as potato acreage is concentrated in the region of La Araucania, with around 13,050 ha, corresponding to one of the main items of family farming (AFC) and with 70% of the area cultivated by smallholders (PASO, 2015). However, in this stratum of farmers growing potatoes poses serious technical problems that affect yields and profitability. Among them, one of the major constraints is the development of diseases that cause significant losses in production and product quality, which severely affects the marketing, either in the internal and external market (SAG, 2002).

In the south, the incidence of fungi and bacteria in the rhizosphere cause recurrent disease in the tubers as rizoctoniasis (Rhizoctonia solani), dry rot (Fusarium spp.), Silver scurf (Helminthosporium solani), powdery scab (underground Spongospora), scabies Common (Streptomyces scabies) and soft rot (Erwinia carotovora) (Acuna and Santos, 2006). To combat these diseases, some basic precautions can help avoid great losses, including crop rotati on, using tolerant varieties and healthy, certified seed tubers (Castro and Contreras, 2011).

An important agricultural practice developed in the south for the potato crop and allows significantly increase the yield and quality of tubers is the use of chemical fertilizers, especially when they aim to achieve high levels of productivity, affecting the economy producers by cost overruns (Moraga, 2008). The cultivation requires high availability of nitrogen (N) for a certain period of time the shortfall can cause chlorosis of leaves, while excess produces an intense green color causing delay in crop maturity. Also, phosphorus (P) is required in the initial growth of the plant tissues, particularly the estate and its deficiency produces small purplish plants by accumulation of anthocyanins; whereas, potassium (K) is involved in the process of tuber, so its deficiency severely affects crop yield (Inostroza, 2009). However, at present there is growing concern about the indiscriminate use of chemicals in agro ecosystems, orienting agriculture towards a more rational use of available resources (Alarcon and Ferrera, 2000) and the use of fertilizers to maintain a biological balance and physicochemical soil, such as the so-called bio-fertilizers. These fertilizers could replace synthetic, significantly reducing pollution problems generated by the excessive use of chemical fertilizers (Cuenca et al., 2007).

Bio-fertilizers are microorganism preparations that applied to the soil and / or plant partially or completely replace the traditional fertilization allowing reduce pollution from agrochemicals (Armenta et al., 2010). The microorganisms used in these products can be divided into two groups: a) those with ability to synthesize substances that promote plant growth fixing atmospheric nitrogen, solubilizing iron (Fe) or inorganic P and improving tolerance to drought stress, salinity, excess toxic metals and pesticides b) other can reduce or prevent the effects caused by pathogenic microorganisms (Bashan and Holguin, 1998; Lucy et al, 2004)..

In order to improve crop yields and reduce the use of chemicals (Davis et al., 2005) sustainable agriculture systems increasingly is further reducing the use of chemical fertilizers backing itself in the increased use of bio-fertilizers. Some microorganisms in the rhizosphere, as diazotrophic bacteria (found in Twin N) and arbuscular mycorrhizal fungi, establish beneficial interactions with plant roots. The plant association - rhizosphere microorganisms is important for the development and protection from plant diseases (Jeffries et al., 2003).

Agricultural management practices can affect the composition and diversity of AMF communities (Mathimaran et al., 2007) causing a negative effect on the association with plants (Gosling et al., 2006). The fungal abundance is reduced by the application of phosphate fertilizer and crops, mainly due to mechanical disturbance caused by soil tillage and changes in host plants in crop rotation systems (Preger et al., 2007).

Moreover, the bacteria can fix atmospheric diazotrophic transforming N ammonium which stimulates plant growth (Postgate, 1998). In the market there are several products that contain bacteria Azotobacter and Azospirillum type, which benefit plants and soil, as Twin N® (INIFAP, 2009), a product which includes strains from different regions of the world and can be applied in different crops and growing conditions. We recommend reducing the dose use of mineral N and apply in times of higher demand for the crop nutrient (Mabiotec Agribiotec Pty, 2006). This group of bacteria do not penetrate the root system of plants, they are only drawn to a set of exudates acting as a source of carbon and energy, providing between 25% and 50% of the N requirements of crops (Elmerich et al ., 1992; and Craswell Peoples, 1992; Kannalyar and Lahda, 1997). These microorganisms maintain large populations for 90 to 100 days, gradually reducing them by depletion of nutrients from the root exudates, because of aging crop and antagonism with other microorganisms in the rhizosphere (Dibut, 2001).

The aim of this study was to evaluate the effect of the use of bio-fertilizer TwinN on yield, quality and health mycorrhization of tubers in a potato crop (S. tuberosum) established in a variety Desiree Andisol the Araucania Region.

MATERIALS AND METHODS

The trial was established during the agricultural season 2013/2014, at the Experimental Station Pillanlelbún, belonging to the Catholic University of Temuco, located in the town of Pillanlelbún (38 ° 39'17,24 "lat;. 72 ° 27'00 , 79 "long. West), 14 km northeast of Temuco, La Araucania. This sector corresponds to a agroclimatic Central Valley, which is characterized by a cool te mperate Mediterranean climate, with low temperatures in winter (minimum 4.1 ° C average in July) and with peak summer temperatures (average 27.1 ° C in January). The frost-free period is 90 days mainly comprising the months of December to February. Average annual rainfall is around 1220 mm and water deficit spans 3-4 months (NRC, 2003). According to information reported by the Center for Research of Natural Resources (CIREN) provides that this area is characterized by soils used in agricultural activities, with the ability to use the class IV, ie, soils suitable for upland crops and livestock.

The field trial was established in a Andisol Temuco belonging to the series, characterized by thin soils, silty clay loam and drainage varies from good to excessive (Rouanet, 2005). Planting took place on 6 November 2013 used the commercial potato variety Desiree, with a current seed of a caliber of 4.5 to 5.5 cm. This variety is characterized by a smooth skin bright red, cream-colored pulp and production of elongated oval tubers. The plant is semi-erect, with good vigor, with dark green foliage gray, with abundant pale pink flowers with medium late cycle with good tubers for fresh consumption and for processing. It has a dormancy period of four to five months and presents a plant health, high to dry rot (Fusarium spp.) Average resistance to late blight (Phytophthora infestans) and very high, the black foot (Erwinia carotovora subsp. Atroseptica). It also displays average resistance to nematodes and medium to high to PVA , PVM, PVX and PVY virus (Kalazich et al., 1997).

Soil preparation for testing consisted of two disc harrow harrowing with off-set, leaving a two-week interval between the two and then rastraje with tiller, which yielded a well fluffy and weed-free soil. The tillage was conducted in mid-October to plant in November 2013. The seed tubers before planting fludioxonil were disinfected with 2.5% w / v (CELEST) at doses of 1.0 L t -1 of seed . Sowing was done manually, with a density of 45,000 plants ha-1 at a distance of 0.75 m between row upon row of 0.30 m. The plot size was 3 m long and 2.1 m wide, each with an area of 6.3 m2. Each plot included three rows with 22 plants. Subsequently, weed control work carried performed two aporcas with foliar application fungicide chlorothalonil 72% w / v (1 L ha-1) and sprinkler irrigation (50 mm per irrigation). For fertilization of the crop sowing fertilization a base corresponding to a dose of 1200 kg was used ha-1 11:30:11 mixture equivalent to 132 kg N ha-1; 360 kg ha-1 P2O5 and 132 kg K2O ha-1 (T0). Furthermore a second dose of chemical fertilizer was applied to the aporca, 50 kg of N as urea (T1) and finally at the beginning of flowering the dose of 1 g ha-1 Twin N biofertilizer (T2) (Table applied 1).

Table 1. Nitrogen fertilizer used for growing potatoes established in Andisol of the Araucanía Region.

Tratamientos	Chemical Fertilizer (kg ha^{-1})		Bio-fertilizante
	planting	Top dress	Flowering
T0	132	-	-
T1	132	50 (urea)	-
T2	132	-	Twin N (1 gr ha^{-1})

Before application of bio-fertilizer Twin N (1 g ha-1) rehydration of the freeze-dried bacteria with 100 mL of water was performed, then 3 g of sugar were added, allowing them to stand up to his application, which was made with knapsack adding 2.52 mL dilution in 20 L of water (water 200 L ha-1). Twin N is a biofertilizer developed based on N-fixing bacteria, the genus Azospirillum lyophilized and other diazótropos in a concentration of 1 × 1011 CFU g-1 pc (endophyte) providing an inoculation period, from 30 to 110 kg N ha-1, depending on seasonal conditions and soil moisture and crop needs. That is, the plant is inoculated and colonized externally and internally, for highly efficient species for fixing N.

After seven months of development of the crop, the harvest took place on May 12, 2014 in manually removing each plot five floors of the central bank along with a sample of rhizosphere soil and roots of each plant, for analysis AMF spores and colonization, respectively.

Evaluations were performed at the Laboratory of Soils belonging to the School of Agronomy of the Catholic University of Temuco. At harvest, the parameters measured were:

a) On the floor: bulk density, pH and number of AMF spores.

b) In the roots of plants: AMF colonization.

c) In the tubers: number of tubers per plant, tuber yield and distribution by size, presence of disease and overall performance.

At 72 and 105 days after harvest quality of stored potatoes we were evaluated, by counting the number of shoots of each tuber and measuring its length using a ruler (cm), after separating each tuber sprouting.

Physico-chemical parameters

a) Humidity (%). It was obtained by weight difference between the moist and dry soil in an oven with air circulation (Arquimed ZRD-5055) at 105 ° C for 24 h.

b) Bulk density. Measured by the method of the specimen.

c) pH of the rhizosphere soil. It was measured potentiometrically in a soil suspension: water ratio of 1: 2.5 by pH meter reading (Thomas Scientific TS 625).

Fungal parameters

a) Quantification of AMF colonization in roots of potato

The roots were washed thoroughly with water to remove any soil stuck to the surface and then pieces of about 1 cm long were cut, which were placed in glass jars. These pieces were covered and clarified with a solution of KOH 2.5% w / v leaving for 3 days at room temperature. Subsequently, the KOH was removed and the roots were rinsed with water to remove excess reagent. Then, covered with a solution of HCl 1% v / v for 24 h at room temperature. After this time the acid was removed and the roots were stained with try pan blue 0.05% w / v for one day at room temperature (Phillips and Hayman, 1970). Finally, try pan blue was removed and the roots were rinsed with water, quantifying under the AMF colonization stereomicroscope according to the line intercept method (Giovanetti and Mosse, 1980).

b) Accounting for the number of AMF spores in the rhizosphere soil planted with potatoes

Spores were isolated from the ground following the methodology of Brundrett et al. (1996) which consists of passing 20 g of ground water through a sieve set of 460 microns, 250 microns, 160 microns and 53 microns. The content of the last two sieves is transferred to a funnel and water to a polyethylene centrifuge tube of 50 mL. Then, a solution of sucrose 70% w / v is added to form a double layer between water and sugar are retained area where spores (Sieverding, 1991). The tubes were centrifuged at 3000 rpm for 5 min and the supernatant was passed through the sieve of 53 microns by rinsing with distilled water. Finally, spores retained in the latter sieve plate to Doncaster where they were recorded as number of spores moved HMA 100 g ss-1 or AMF spore number ss 100 mL-1.

Plant parameters

Tubers

From each plant all tubers were harvested, counted and weighed to determine yield. Then they were classified by size according to its equatorial diameter <2.8 cm; 2.8 to 3.5 cm; 3.5 to 4.5 cm; 4.5 to 5.5 cm; 5.5 to 6.5 cm and> 6.5 cm.

Presence of diseases

The preparation and classification of potato tubers and subsequent analysis of diseases was conducted at the Laboratory of Plant Protection, Faculty of Agricultural Sciences of the Catholic University of Temuco, La Araucania.

The diseases were determined by observing each tuber with symptoms separating and classifying sick and incidence rate.

In each tuber, the percentage of skin covered with each of the following conditions are observed: black scab, common scab, silver scurf, dry rot and craters comparing with a scale designed to assess each disease (Ministry of Agriculture Fisheries and Food, 1976), whose ranges are: 0 = healthy; 1% of damaged area; 10% of the damaged area; 25% of the damaged area; 50% of the damaged area and> 75% of damaged area.

Black Sarna

The evaluation was made by observing the percentage of skin covered by the sclerotia which generates the pathogen Rhizoctonia solani.

Common scab

This disease is caused by the pathogen S. scabies, which was assessed for injuries caused in the tubers, which can be superficial or deep. The percentage of damaged area caused by the bacteria and compared to the scale was observed.

Silver scurf

The cause of the disease is H. solani, which generates the tuber surface in an area with a characteristic silvery sheen.

Dry rot

It is produced by Fusarium fungi and the most common symptoms is presented in tubers, with seed rot in the field or dry rot in storage.

Craters

The cause nematodes that infect tubers and develop gills with a warty appearance.

Following the determination of the percentage of area covered by signs of pathogens, it was separated from the tubers of incidence range to classify and count them.

In Figure 1, the field test potato tubers harvested in Andisol the Araucania Region, according to the degree of infestation they had shown.

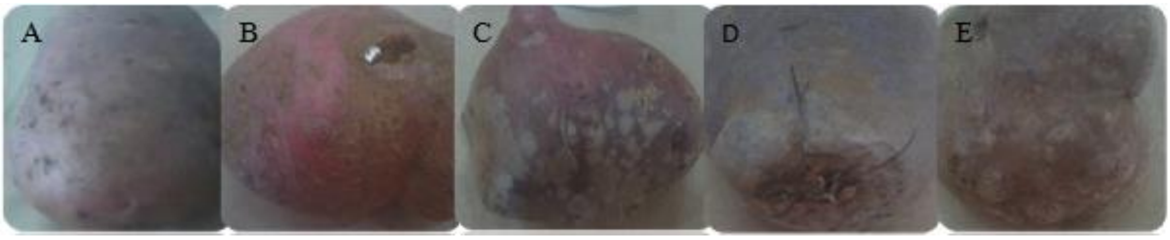


Figure 1. Incidence of some skin diseases of potato variety Desiree tubers established in Andisol of the Araucanía Region: A) black scab, B) common scab, C) silver scurf, D) dry rot, and E) craters.

Statistic analysis

The experimental design was randomized complete block with four replications. The results were subjected to a test of normality and homogeneity, and then make an analysis of variance using one-way ANOVA and multiple comparison test of Tukey ($p \leq 0.05$). The percentage data were transformed by √arcoseno x. For the statistical analysis of the results, the R-project software version 3.1.2 for Windows was used.

RESULTS AND DISCUSSION

The bulk density was low in all treatments ranging from 0.56 to 0.58 g mL-1, consistent with results reported by Sierra et al. (2002), who note that most trumaos soils have values below 0.8 g mL-1 density, which gives them a high porosity and friability optimal. This parameter is used as an indicator of the degree of compaction of soil and restrictions on root development of plants (Selles et al., 2012). The degree of soil structure can vary by driving conditions, such as machinery or farming. In relation to the above, the HMA play an important role in soil structure by producing a specific glycoprotein called recalcitrant Glomalin (Wright et al., 1996) that increases the aggregation (Wright and Upadhyaya 1998;. Wright et al, 1999 ;. Rillig et al, 2001) decreasing soil erosion (Rillig et al, 2002)..

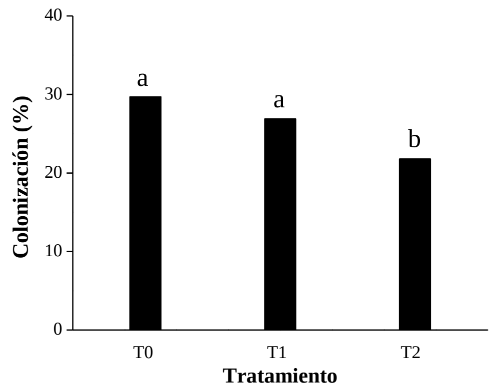
At harvest, the treatments had high acidity (pH: T0 = 5.22; T1 = 5.15; T2 = 5.25) and when soils are very acidic (<5.5) tend to have toxic levels aluminum and manganese. Moreover, the activity of soil microorganisms is inhibited in highly acidic soil and agricultural crops for the ideal pH value is approximately 6.5 (FAO, 2015). The beneficial soil microorganisms are also affected by the acidity decreases the mineralization of organic matter and nitrogen fixation by rhizobia growing associated with the roots of legumes, which results in a reduced availability of nutrient (Frioni 1999) .

Fungal parameters

Percentage of AMF colonization in roots

In Figure 2, the percentage of AMF colonization observed in the roots of potato plants harvested, finding differences ($p \leq 0.05$) between T2 (treatment with Twin N) had a significantly lower percentage than control and T1. In general, colonization of potato HMA was lower compared with other traditional crops such as cereals (Montano et al., 2001).

Regarding the other fungal parameter evaluated, the number of spores was quantified as much T1, treatment corresponding to the application of chemical fertilization, with significant difference ($p \leq 0.05$) than T2 and control.



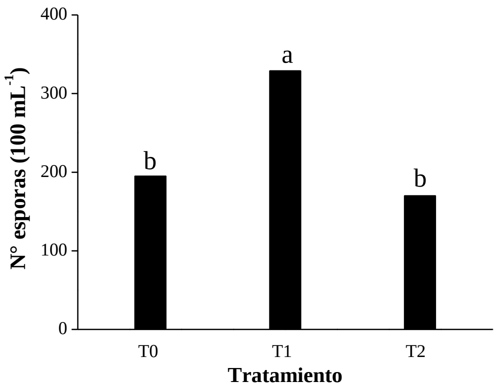


Figure 2. Parameters fungal roots and rhizosphere of potato variety Andisol established in the Region of La Araucania Desiree: A) Percentage AMF colonization and B) Number of spores HMA. Bars followed by different letters are significantly different according to Tukey test ($p \leq 0.05$).

Figure 2. Fungal parameters on roots and rhizosphere of potato variety Desiree established in Andisol of the Araucanía Region: A) Percentage AMF colonization and B) Spore number HMA. Bars followed by different letters are Significantly different according to Tukey test ($p \leq 0.05$).

Arbuscular mycorrhizal symbiosis helps the survival of the plants increase in the uptake of nutrients and water absorption, resistance to pathogens and production of plant hormones (Jeffries et al., 2003).

When in soil N availability is high, the microorganisms do not fix atmospheric nitrogen capturing the available soil and therefore the symbiotic association is established. For arbuscular mycorrhizal fungi extra radical transported through the mycelium, among other nutrients, P and N to the root system of the plants through more efficient use of chemical fertilization (Sutton, 1973; Barea and Azcón-Aguilar, 1983; Read, 1998; Selosse and Le Tacon, 1998).

Distribution of tubers per plant

Regarding the number of potato tubers per plant (Tabl e 2), no significant differences between treatments were observed, showing a homogeneous distribution with an average of two tubers per caliber; presenting T1 and T2 equal amount of tubers per plant. These results agree with those reported by Veitia et al. (2003) who accounted for a similar average of 14 tubers per plant.

Table 2. Distribution of the number of tubers per potato variety Desiree caliber established in Andisol of the Araucania region.

Treatment	Distributión de númbers de tubérs por calibre diámeter tubérs (cm)						N° tubérs/ plant
	<2,8	2,8-3,5	3,5-4,5	4,5-5,5	5,5-6,5	>6,5	
T 0	2	2	3	1	2	2	**12**
T 1	1	2	3	2	1	4	**13**
T 2	3	2	2	2	2	2	**13**

Tubers by size distribution

In general, although no significant differences between treatments were observed, the presence of urea increased the potatoes out of calibration (category discard) and between 4.5 to 5.5 cm. The witness had as many tubers in size between 5.5 to 6.5 cm. Twin N increased the number of tubers in the sizes 2.8 to 3.5 cm and 3.5 to 4.5 cm (Figure 3). However, in the more than 6.5 cm caliber, significant differences between treatment with Twin N (T2) and added treatment chemical fertilizer (T1) were found as well, with the control (T0). It is remarkable that this treatment increased by about 150% return over the control.

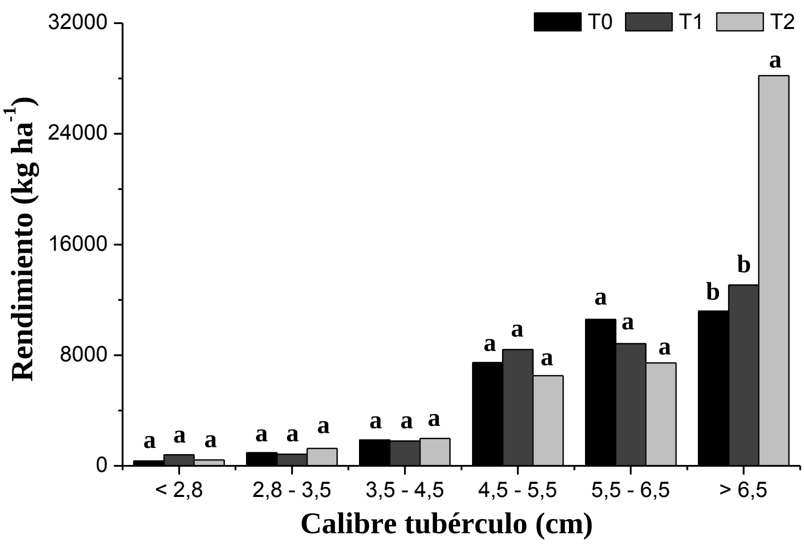


Figure 3. Yield (kg ha-1) according to distribution of tubers per caliber.

Similar results obtained Glendinning (2008), who with two Twin N applications increased to more than double the yield of tubers larger between 4.5 and 6.5 cm, while the smaller tubers were cut in half, with respect to the control. These results are contrary to those reported by Talamas (1998); quoted by Basly (2003), who evaluated the effect of applying a bio estimulante seaweed based on the performance of a potato cultivar found no statistically significant differences in yields by size of the treated plants. Moreover, Terry et al. (2005) evaluated bacteria in a tomato crop and found that the largest genus in the rhizosphere was Azospirillum bacteria also present in Twin N. These authors with artificial inoculation had a positive effect on the size and nutritional status The plant, with a yield of 11% higher than the control without inoculation.

Twin N bio-estimulante effect on the health condition of harvested tubers

Concerning the health status of harvested tubers (Table 3) shows that the treatments did not differ, with low incidence levels, ranging between 1% and 2% of lesions on the skin of the tuber.

However, in the case of black scab and mange the presence of N Twin presented incidence rates of 87% and 81%, respectively, concentrate to a minimum which does not significantly affect tuber.

Table 3. Incidence (%) of scabs in tubers harvested potato variety Desiree established in Andisol of the Araucania region.

Incidence (%)	Presence of scabs (%)								
	Black scab			common scab			silver scab		
	T0	T1	T2	T0	T1	T2	T0	T1	T2
0	52	41	13	48	41	19	52	54	41
1	44	52	76	49	46	71	37	37	57
10	4	6	9	2	10	8	7	3	2
25	-	1	-	1	-	1	5	6	-
50	-	-	2	-	2	1	-	-	-
> 75	-	-	-	-	1	-	-	-	-

Values are not statistically different ($p \leq 0{,}05$).

Concerning the health status of harvested tubers (Table 4) shows that for Fusarium applying urea (T1) increased the incidence rate in levels of 25%, with 5% of the tubers in this category. In contrast, the incidence of craters concentrated in the control (T0) levels of 10%, with 5% of patients and tubers (T2) levels of 25%, with 11% of damaged tubers .

Table 4. Incidence (%) of other diseases in potato tubers variety Desiree established in Andisol of the Araucanía Region.

Incidence (%)	Presence of diseases (%)					
	Dry rot			craters		
	T0	T1	T2	T0	T1	T2
0	99a	95a	99a	94a	90a	88a
1	-	-	-	-	-	-
10	-	-	-	5a	1 b	1 b
25	1b	5a	1b	1b	9 ab	11a
50	-	-	-	-	-	-
> 75	-	-	-	-	-	-

Different letters indicate significant differences according to Tukey test ($p \leq 0.05$).

Total Return

In Figure 4, the total expressed in kilograms per hectare is presented shows that significant differences between T2 (Twin N) for control and T1 is found. Applying TwinN, replaced the 50kg N in urea, with a 41% increase in performance. The highest yield is based on a significant growth of the tubers in the caliber of greater value. These results agree with those obtained by Araujo et al. (2010) in a potato crop using the bio-fertilizer Azotobacter sp., where performance was similar to that seen with traditional fertilization. Moreover, Abarca et al. (2012) Using Twin N in a corn crop, found that the bio-fertilizer did not affect performance, allowing dose reduction of nitrogen fertilization. The bio-fertilizer helped to reduce chemical fertilization avoiding the undesirable effects that generate excessive applications of N in the soil. It has been established that the activity of the microorganisms is of vital importance in soil fertility and sustainability of the agro ecosystem (Ferrera-Cerrato and Alarcón, 2001).

The results of this field test, resemble those reported by Marinao (2011), who evaluated three commercial bio-stimulants on the performance of potato variety Desiree, he found that foliar application of bio-estimulante Fartum was superior to the control and others bio-estimulantes. Moreover, Epuin (2004) evaluated three commercial bio-stimulants on the performance of four varieties of potatoes, he found that the application of any bio-estimulante was favorable, increasing the total yield between 7% to 18% relative to the control.

Conversely, Basly (2003) obtained in two potato cultivars with radicular lower yields and foliar application of an organic bio-stimulant derived from seaweed.

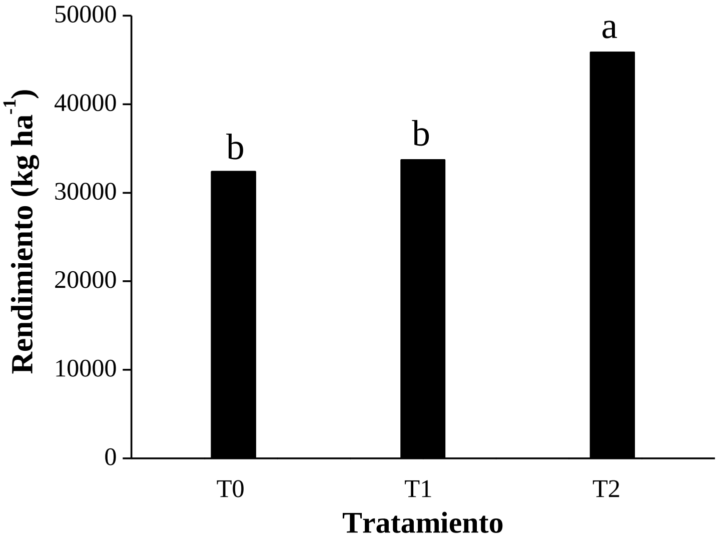


Figure 4. Total yield of potato variety Desiree established in Andisol in the Region of La Araucania. Different letters Indicate significant Differences bars ACCORDING TO Tukey's test ($p \leq 0.05$).

Quality stored tubers

In Table 5, you can see the number and length of sprouts from tubers stored at 72 and 105 after harvest.

At 72 CDD, on the lower level (<0.5 cm) in diameter, T2 reached the highest value of outbreaks with significant statistical differences, which reached 200% over the control. Instead T1, presented fewer outbreaks in the lower levels.

A DDC 105, Similarly the distribution of recorded outbreaks show that all treatments significantly increased the number of sprouts, focusing on the levels of 1 to 1.5 cm no significant differences between treatments.

In this regard, Santos and Orena (2006) report that since the beginning of the formation of tubers to harvest time and initial part of the storage period, the potatoes are in a state of "dormancy"; ie do not grow to be under the effect of natural inhibitors. This period can last from one to several months depending on the variety, growing conditions, damage and the storage temperature. The sprouting of tubers as it progresses causes very high weight loss. The outbreak itself, is a direct loss of dry matter, which negatively affects the industrial process, depending on the purpose of processing; since, tubers for potato chips are low in reducing sugars and high dry matter (Gomes and Wong, 1998).

Table 5. Number of Outbreaks in stored potato tubers variety Desiree established in Andisol of the Araucanía Region.

Shoot length (cm)	N° of shoots					
	72 DDC			105 DDC		
	T0	T1	T2	T0	T1	T2
0-1	11 ab	8 b	34 a	33 a	27 a	39 a
1-1,5	4 a	2 a	3 a	35 a	20 a	28 a
1,5-2,5	0 a	0 a	1 a	11 a	12 a	12 a
2,5-3,5	-	-	-	4 a	3 a	4 a
3,5-4,5	-	-	-	1 a	1 a	1 a

Different letters indicate significant differences according to Tukey test ($p \leq 0.05$).

CONCLUSIONS

• The bio-fertilizer Twin N increases yields compared to the control and represents a reliable partial replacement of urea alternative.

• The study demonstrates the feasibility of reducing pollution and an alternative for the sustainable development of agro-ecosystems and the commitment to develop clean technologies for agriculture and healthier environment.

• Increased performance Twin N is associated with a reduced colonization HMA.

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