Australian Journal of Basic and Applied Sciences, 10(14) September 2016, Pages: 115-123

AUSTRALIAN JOURNAL OF BASIC AND APPLIED SCIENCES

ISSN:1991-8178 EISSN: 2309-8414
Journal home page: www.ajbasweb.com

Nitrogen sources on the physiological quality of proso millet seeds

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ARTICLE INFO

Article history:
Received 26 July 2016
Accepted 21 September 2016
Published 30 September 2016

Keywords:
Organic fertilizer, Germination, Vigor.

ABSTRACT

Background: Nitrogen fertilization can influence the quality of seeds of proso millet (*Panicum miliaceum L.)* and the effects vary with environmental conditions.Pig and poultry activities are important economically and socially for Brazil, mainly for the southern region, as it generates economic development and sustainability for small and medium producers.These activities are undergoing a continuous process of technological inclusion, with increasing concentration of animals in small areas. Generating large volumes of waste with nitrogen and most of the producers cannot meet the requirements of environmental legislation, so the fate of this waste is the application in agricultural crops. However, there is little information on the influence of nitrogen fertilization with these sources as proso millet crop seeds.Objective: To evaluate the effect of different nitrogen sources on the physiological and sanitary quality of proso millet seeds cv. AL Tibagi. The experiment was conducted under field conditions in the period from August to November 2014, and laboratory evaluations conducted from March to June 2015. The experiment was conducted at the Federal University of Santa Maria, Campus Frederico Westphalen under Rhodic. The experimental design was a randomized block design with four treatments and four replications, the experimental plots of 4.0 x 3.6 meters. The treatments were three sources of nitrogen fertilizer, poultry litter, pig slurry composting, mineral fertilizer (NPK) and control. Results: The poultry litter gave negative results on the physiological quality of millet seeds. The health of proso millet seed was influenced by N sources, which provided lower incidences of pathogens with the use of nitrogen fertilizer and higher rates found when used organic fertilizers. Conclusion: The pig slurry composting and chemical fertilizers provide the best results in the quality of seeds. Sanity millet seeds showed a higher incidence of pathogens with the poultry litter and lower incidence of pathogens with chemical fertilizer.

INTRODUCTION

The proso millet (*Panicum miliaceum L.*) is one of the oldest crops domesticated by man (Lu *et al.*, 2009), is currently cultivated on a larger scale in Eastern Europe, Russia, China, India, Canada and North America (Lu*et al*., 2009). In Brazil it is still little expressive culture when compared to more traditional crops such as wheat, oats and sorghum.

ToCite ThisArticle: Dionei Schmidt Muraro, Alieze Nascimento da Silva, Claudir José Basso, Stela Maris Kulczynski, Vanessa Graciela Kirsch, Andressa Calderan, Marcia Gabriel, Vanessa Alba da Silva., Nitrogen sources on the physiological quality of proso millet seeds. ***Aust. J. Basic & Appl. Sci.,*** *10(14): 115-123, 2016*

Over the years proso millet has aroused the interest of producers and gaining ground in some regions of São Paulo, MatoGrosso do Sul and Rio Grande do Sul where the goal is the production of grains for use in animal feed, especially captive birds, replacing the birdseed (Abrantes el et., 2010), in breweries mixed in minor proportion with barley (Zarnkow *et al*., 2010).

In addition, the proso millet has been used as an alternative crop in autumn / winter period, for ground cover and production of straw in no-till system, with the advantage of its low cost of implementation and the rapid and abundant formation of straw (Basso *et al*., 2013). Another advantage of proso millet is the possibility of being seeded for almost every year since it has availability of water and there is no risk of frost.

With increasing proso millet economic interest, so does the demand for quality seeds.The quality of seed is used mainly by companies as a competitive differentiator. Seed quality is characterized by genetic, physical, sanitary and physiological is of fundamental importance in the production process of any plant species, to influence the development and crop quality.Seed quality is characterized by genetic, physical, physiological and health aspects having fundamental importance in the production process, by directly influence the development and crop yields (Carvalhoand Nakagawa, 2012). The highest quality is achieved at the time of physiological maturity, which from that moment begin to occur degenerative processes, physical, physiological or biochemical characterizing the deterioration.

Productivity and quality seeds are directly related to plant nutrition which makes fertilization important factor in the search for better quality seed. Among the necessary elements should highlight the nitrogen fertilizer due to the important role of nitrogen in the plant (Carvalhoand Nakagawa, 2012), mainly because it is a grassy and generally responsive to nitrogen fertilization.

The organic fertilizer has been used since ancient times in agriculture to meet the nutritional needs of plants. Attention has been given the poultry litter and the pig slurry composting used primarily as a source of nitrogen in total replacement and / or partial mineral fertilizer. The organic fertilizer, using waste from animal production is a common way to conduct small farms or even large farmers (Barrena *et al*., 2011, Lyimo *et al*., 2012).Sustainable solid waste management required a good policy together with comprehensive strategies and plans (Wee and Abas, *et al*., 2016). Agricultural residues can cause various environmental problems when accumulated, if not disposed of properly cause contamination of soil and groundwater due to decomposition of organic matter (Veiga *et al*., 2016).

By studying the nitrogen fertilization on proso millet crop, Basso *et al*. (2015) and Abrantes *et al*., (2010) observed increases in plant height, dry weight, thousand grain weight and millet crop yield by applying mineral nitrogen in coverage under field conditions. However, the effects of nitrogen fertilization on physiological and sanitary quality of seeds are somewhat controversial, even in crops such as corn, rice, beans and wheat, evidencing the need for more studies on the relationship between fertilization / nutrition and production high quality seed.

There is little information on the use of different sources of nitrogen in the productivity and quality of proso millet seeds, especially when it comes to the use of organic manure as a source of N. Therefore, the objective of this study was to evaluate the effect of different sources nitrogen on physiological and sanitary quality of proso millet seeds.

MATERIAL AND METHODS

The experiment was conducted in the experimental area of the Federal University of Santa Maria campus of Frederico Westphalen in the state of Rio Grande do Sul, whose coordinates are 27 23'47.58 "S and 53 25'41.24" W, at an altitude of 566 meters.

The soil is classified as Rhodic Hapludox soil (USDA 2003), or Latossolo vermelhodistróficotípico in the Brazilian System of Soil Classification (Embrapa 2013). At the time of the experiment, showed the following characteristics in 0-20 cm: pH in H_2O (1:1) 4.6; SMP index 5.1; clay 650 g kg^{-1}; organic matter 2.7 g kg^{-1}; P-mehlich 7.2 mg dm^{-3}; potassium 137 mg.dm^{-3}; calcium 2.1 $cmol_c dm^{-3}$ (extracted by KCl 1 mol L^{-1}); magnesium 1.2 $cmol_c dm^{-3}$; H + Al 8.0 $cmolcdm^{-3}$; Al 2.1 $cmolcdm^{-3}$ (extracted by KCl 1 mol L^{-1}),; CTC 11.6 $cmolcdm^{-3}$; and a percentage of CTC with bases and Al of 31.3 and 36.5%, respectively. Six months before the experiment, lime was applied to the soil in order to raise the pH to 6.0 following the recommendation of CQFS-RS / SC (2004).

According to Köppen classification, the climate is Cfa, with annual average temperature around 18 ° C, with highs in the summer can reach 41 ° C and minimum in winter reaching values below 0 ° C. The average annual rainfall is high, usually between 1,800 and 2,100 mm, well distributed throughout the year (Moreno, 1961). In Figure 1 it can be seen that the distribution of rainfall was favorable to the development of culture. Throughout the conduct of the experiment were recorded 677.8 mm of precipitation, these being evenly distributed throughout cycle without jeopardizing the development of culture. Average temperatures recorded for the months of August, September, October and November, minimum 15.9, 18.2, 21.0 and 21.9 ° C and maximum 17.5, 19.3, 22,4e 22,2ºC, respectively.

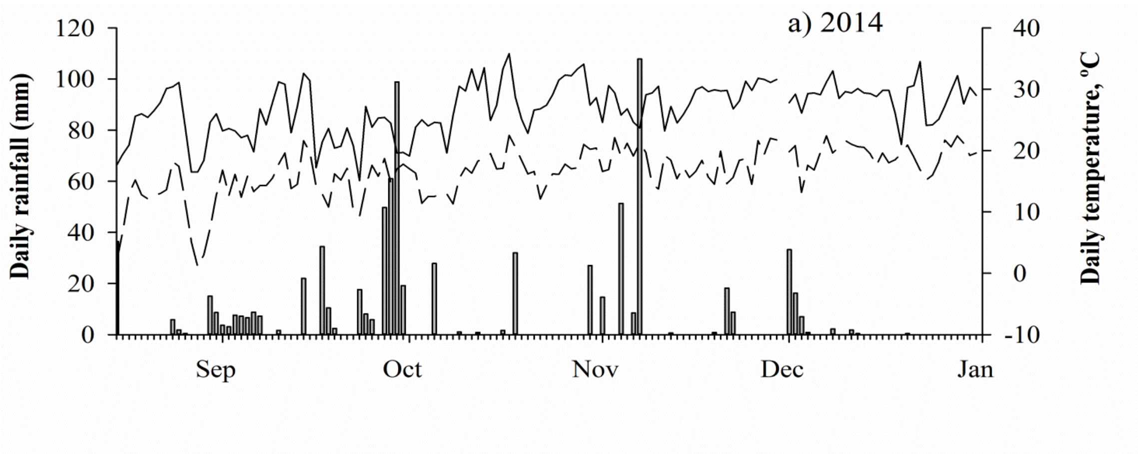

Fig. 1: Average daily temperature and daily rainfall which occurred during the experiment. The rainfall data were obtained from the Meteorological Station in Frederico Westphalen – RS

The experimental design was randomized blocks, with four treatments and four replications, and the experimental plots measured 4.0 x 3.6 meters, totaling 14,4m².The treatments were three sources of nitrogen fertilization: mineral fertilizer (NPK), pig slurry, control and poultry litter.

The amount of nitrogen required to be applied in culture was determined according to the soil analysis, and following the recommendation of CQFS-RS / CS (2004). Phosphorus and potassium fertilization was performed haul after sowing to standardize treatments, with the use of triple superphosphate and potassium chloride, respectively.

The experiment was conducted in soybean stubble, harvest in late April remaining area set aside for about 3 months. At the end of August, the desiccation was carried out of the area for later installation of the experiment. Sowing was done manually on 22/08/14, using cv. AL Tibagi, spaced 0.45m between lines and density of 120 seeds per meter. Harvest occurred on 11.12.14, for a total cycle of 65 days for the crop.

The following analyzes were performed:

Thousand seed weight*:*held from the weighing of eight repetitions of 100 seeds for each treatment (Brazil, 2009).

Germination:four replications were used with four replicates of 100 seeds were sown in towel paper, moistened with water volume 2,5 times the dry mass of substrate (paper), and kept in chamber at 25 ° C and 12 photoperiod hours. The counts were performed at three and seven days after sowing (Brazil, 2009) and the results expressed as a percentage of normal plants.

First count:held together with the germination test for evaluation of seed vigor. the percentage of normal seedlings, three days after sowing was evaluated.

Accelerated aging:The test was conducted as described by Mark Son (1999) modified. Plastic boxes were used gerbox containing a blade 40 ml of water, the seeds were spread on a screen positioned above the water depth, kept in an incubator at 42 ° C for 36 hours. After this period, the germination test was conducted with four replications with four sub samples of 100 seeds per treatment, and the count was performed seven days after sowing (Brazil, 2009).

Electrical conductivity: Were performed using four replications of 50 seeds for each treatment. Samples were weighed on precision balance and then soaked in water for plastic container containing 75 ml of distilled water, and kept at a temperature of 25 ° C for 24 h (Krzyżanowski *et al.*, 1999). After this period, we proceeded to read the conductivity in the soaking solution, using a digital conductivity meter (Conductivity Meter, Model CD-4303), the results were expressed in µS cm-1.g-1.

Cold test: it was used the methodology described by Krzyzanowski *et al*. (1999) adapted for proso millet culture. They used four repetitions of 100 seeds for each treatment, which were distributed in germitest paper, moistened with an amount of water equivalent to 2.5 times the mass of the paper. After seeding the rolls were placed inside plastic bags that once sealed with adhesive tape were kept in chamber at 5 ° C for seven days. Elapsed this period, the rollers inside the plastic bags were transferred to B.O.D type chamber at 20 ° C, where they remained for seven days. The interpretation was done by computing the percentage of normal seedlings.

Emergency speed index: the test was carried out from the sowing of four repetitions of 100 seeds per treatment, in soil mixture and substrate covered with a thin layer of substrate, counting daily from the start of the emergency, number of seedlings until the process was stabilized. The calculation of the emergency speed index was performed by Maguire formula (1962) IVG = (G1 / N1) + (G2 / N2) + ... + (Gn / Nn). And for each repetition, he calculated the IVE, using the formula, where the arithmetic mean of four replications was the index.

Seedling emergence in the field: was conducted jointly with the emergency speed index test. The evaluation was performed 21 days after sowing, determining the seedling emergence rates (Nakagawa, 1994).

Sanity: The sanitary quality of proso millet seeds was determined by the method of "Blottertest" without aseptic. For this, four replications of 25 seeds were placed in gerboxes containing three sheets of paper germitest previously dampened with deionized water and incubated in a growth chamber for seven days at a temperature of 25 ° C (± 2) under 12 hours photoperiod. After this incubation period the seeds were examined individually under stereoscopic microscope and / or optical microscope, by computing the fungi incidence percentage. The identification was based on its morphological characteristics and with the aid of specific literature.

The results were submitted to analysis of variance and when significant for the variables, the averages were compared by Tukey test 5.0% probability. For these analyzes, we used the statistical program Assistat 7.7 beta.

RESULTS AND DISCUSSION

The weight of one thousand seeds is an important production component, however the present work it was found that the different N sources did not affect this variable (Table 1) numerically, the treatments involving the addition of nitrogen were higher than those in control.Working with doses of chemical fertilizers and N application times of the proso millet crop, Soratto *et al*., (2007), Abrates *et al*. (2010) and Basso *et al*. (2015) corroborate the results presented, which also found no effect of nitrogen in the grain mass.This lack of proso millet response weight of a thousand seeds in this study, may be associated with soybean residue with the predecessor culture that is rich in nitrogen and own relatively short culture cycle.

The evaluation of the physiological quality of proso millet seeds was determined by the germination and vigor tests. The germination values expressed by the percentage of normal seedlings, except for the chicken litter did not differ significantly between the other treatments (Table 1). The pig slurry composting was the treatment that provides a higher percentage of 63% germination, followed by chemical fertilizer 57% and 52% witness.

Already the poultry litter showed lower germination percentage 30%, differing significantly from other treatments. This poor seed germination under treatment with chicken litter, it will against seen in Figure 1 where it is observed that the proso millet seeds derived from the treatment poultry litter showed the highest percentage of contaminated seeds. Moraes and Lie (2006) observed drop in bean seed germination in the presence of some isolates of Alternaria which meets observed for the chicken litter in Figure 1 where he presented the mior percentage of contaminated seeds. Abrantes *et al*. (2010) in proso millet cultivation observed that the highest germination rates were found when obtained lower percentages of Cladosporium. For Yorinori (1982) and Balardin& Loch, (1987) the high percentage of infected seeds are associated with decreased quality of seeds, more specifically the germinating power.

Table 1: Thousand seed weight (TSW), germination (G), first count (FC), accelerated aging (AA), cold test (CT), electrical conductivity (CE), emergence speed index (ESI) e field emergence (FE) due on the application of different nitrogen sources.

Treatments	TSW (g)		G (%)		FC (%)		AA (%)		CT (%)		CE (uS cm g^{-1})		ESI		FE (%)	
Control	2.7	a	52	a	55	a	73	a	62	a	77.4	a	6.8	a	48	a
Poultry litter	3.8	a	30	b	28	b	56	b	50.3	b	133.7	b	4.3	c	18	b
Pigslurrycomposting	3.8	a	63	a	59	a	76	a	70.3	a	65.0	a	6.7	a	56	a
Mineral fertilizaer (NPK)	3.7	a	57	a	58	a	77	a	64	a	82.4	a	5.7	b	50	a
CV - %	11.9		9.6		12.2		2.9		12.9		7.5		2.1		6.5	

[*] Means followed by the same letter in the column do not differ by Tukey test at 5% probability.

In work done with proso millet (Abrantes*et al*., 2010), beans (Crusciol*et al*., 2003) and wheat (Prando *et al*., 2012), assessing levels of nitrogen found no influence on germination by the applied doses. Otherwise, with beans, Farinelli *et al*. (2006), evaluating different soil tillage system and nitrogen fertilization, they found a linear increase of germination by the aumentodas doses applied.

The proso millet (Panicum miliaceum L.) and grass (Panicum maximum Jacq.) Are grasses that belong to the same genus, and the pattern of germination for this species is 50% (Brazil, 2010). For proso millet the lowest value was 30% using the bed as poultry manure (Table 1), this shows that the use of this fertilizer germination is 20% below the standard set for outraespécie the same gender.

The vigor testing aims to check the physiological potential of seeds, ie determine the seed's ability to provide adequate performance in different environmental conditions. For the different sources of N the results were significant on the force expressed by the first count (Table 1), which was observed with the application of chicken litter a germinating only 28% compared to 59% observed for pig slurry composting a reduction of 51%. Aside from chicken litter there was no difference between the other treatments as the first count.

Working with application rates and times of N, Abrantes *et al*. (2010) found that for N doses there was no significant difference, but noted that the dose of 60 kg ha-1, had the highest percentage of 65% germination. Likewise, Prandus *et al*. (2012), no significant effects evaluating N rates in coverage on physiological quality of wheat seeds. However, Crusciol *et al*. (2003) observed reduction in bean seed vigor evaluated by the first count test with the use of nitrogen fertilizer, reducing the percentage of sprouted plants.

For the accelerated aging test, no significant differences between treatments (Table 1) was observed, and the chemical fertilizer was 77% higher, but not significantly differ from the solid pig manure and control. Already the poultry litter 56%, had lower germination percentages, observing a decrease of 26% compared to the best treatment, differing from the other treatments. Similar results were found by Suzana *et al*. (2012), which evaluated the influence of different sources of nitrogen on physiological quality of bean, found that the application of chemical fertilizer (urea) influenced superior to treatment with pig slurry composting, however, did not differ from the control. The accelerated aging test results showed higher germination, first count, cold test, before these results can be observed that proso millet may have numbness in their seeds, which exposed to heat and moisture provided better results.

The cold test evaluates the seed vigor through the exposure of the same will lower temperatures. Using data obtained (Table 1), it can be seen that the treatments with the use of solid manure of pigs, chemical fertilizer and control did not differ significantly from each other, unlike the poultry litter, where seeds had lower germination values. Working with doses of N, Abrantes *et al* (2010) found no differences in the results for the cold test in proso millet seeds, but found lower quality compared to the results obtained through the germination test, demonstrating crop susceptibility to low temperatures contrary to the results shown in this study, where the germination and first count values were lower than those obtained in the cold test.

The electrical conductivity is to assess the quality of the membrane of the seeds being measured leaching of electrolytes from seeds soaked in water. Seeds with high electrical conductivity and have high leaching capacity of electrolyte loss So, the higher the electrical conductivity smaller the seed quality, so in this work can be seen in treatment with use of chicken litter a conductivity of 133.73 µs.cm.g, this high value and statistically superior to the other treatments. The results obtained by Fields *et al*. (2014) corroborate the work where working with chicken litter doses in the culture of cabbage observed that the higher dose resulted in higher levels of electrical conductivity in seeds. Already Abrantes *et al* (2010) in proso millet, Farinelli *et al*. (2006) and Crusciol *et al*. (2003) in beans, revealed no significant differences in the use of nitrogen (urea and ammonium sulfate) between treatments in the electrical conductivity of bean seeds.

The emergence speed index and field emergence are among the most used force tests, of which are closer to the sowing conditions in the field. The emergence speed index (Table 1), measured every day until stabilization, revealed better rates in the control, manure solid pig and chemical fertilization, where they did not differ significantly from each other, but significantly differed frompoultry litter, with increases in germination of the order of 35.7, 35.0 and 23.4% respectively in comparison topoultry litter. Working with doses of N Abrantes coverage *et al*. (2010) also found no response in emergency speed being the most proso millet emergency speed index found in 0 kg ha-1doses Ha-1 (control). Already Campos *et al* (2014), working with 10 t ha-1 poultry litter found that lots of cabbage seeds with higher germination rate index, unlike what was previously mentioned and observed in this study.

The emergence in the field, evaluated at 21 DAE showed differences only for treatment with chicken litter (18%), which was obtained the smallest emergence rates compared to the other treatments. Works Nakagawa *et al*. (2000) with black oats and Day *et al*. (2008) with proso millet, showed no effects of N application in percentage of field emergence. Since Bono *et al*. (2008), to evaluate the physiological quality of corn seeds with the use of nitrogen fertilizer found in the increased percentage of field emergence, especially for the source of N with slow release.

The use of plant pathogens of free seeds for crop establishment is extremely important for good crop establishment, however, cultural practices, including the management of nitrogen fertilization, can influence the contamination / infection of seeds (ABRANTES *et al*., 2010).

In assessing the sanitary quality of the seeds, it was found the presence of various pathogens. The results presented in Figure 2 show a significant increase as the application of different N sources only for the Cladosporiumspp showed that higher incidence of contaminated seeds in the use of chicken litter and 22% lower incidence in the control 6%. As for the fungi Alternariaspp, Phomaspp, Bipolarisspp and Rhizopusspp, there were no significant responses among the treatments, but was observed that using the poultry litter as a source of N, resulted in an increase in relation to the seeds infected 43 5%, 5%, 1.5%, 1%, respectively. However, for the Penicillium, Aspergillussppig slurry compostingprovided higher infection rates and fungi Epicoccumspp, Fusariumspp, they showed higher rates in the control and chemical fertilization, respectively.

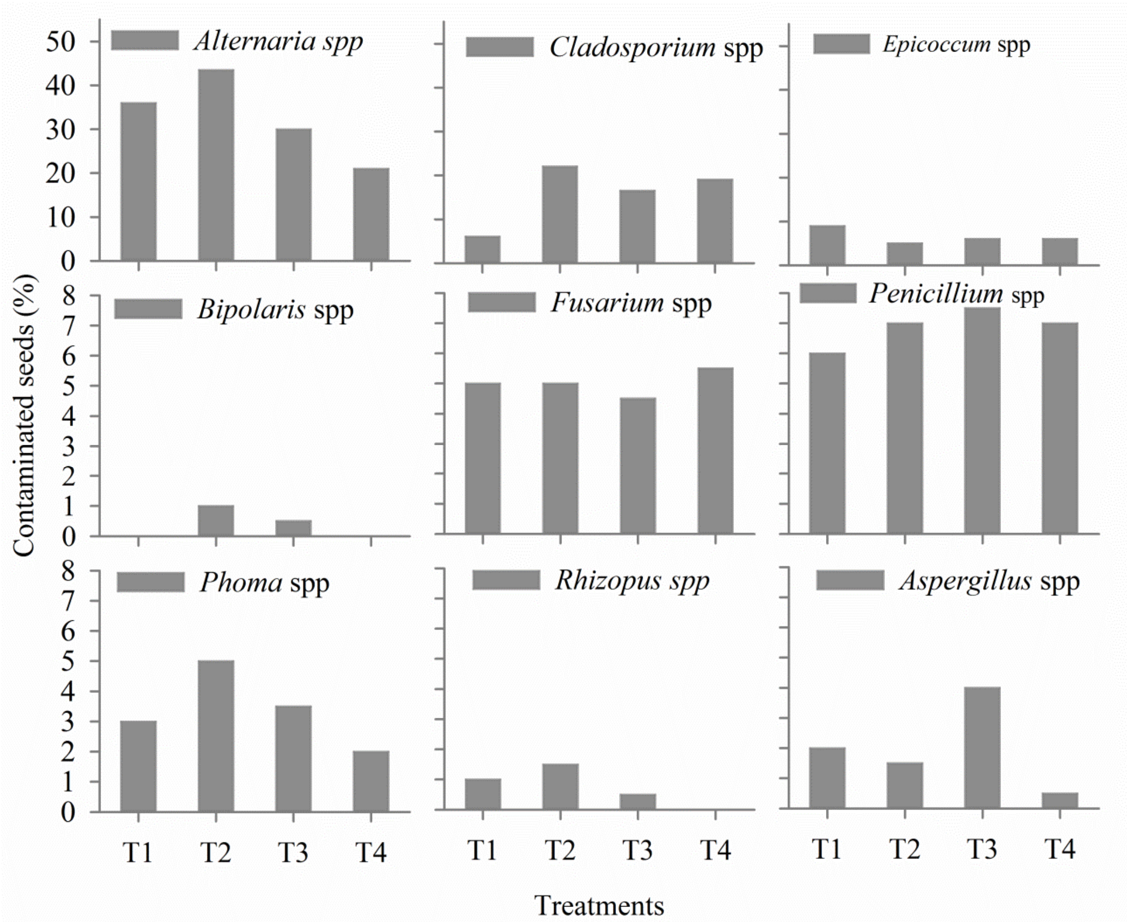

Fig. 2: Percentages of proso millet seeds contaminated with fungus due to the application of different sources of N, T1-Control; T2- poultry litter; T3- pig slurry composting; T4- mineral fertilizer (NPK).

Thus in (Figure 3) is observed full of healthy seeds and contaminated, strengthening the results shown above, and the poultry litter and pig slurry composting had the highest contamination levels 81 and 70%, respectively, subsequently lower rates of healthy seeds 19:30%, respectively, did not differ significantly from each other, but differing chemical fertilizer and control, which % provided lower contamination levels 57 and 60.

High concentrations of N second Marschner (1995) reduced the production of phenolic compounds, which possess fungi static and lignification of the sheets, decreasing the resistance required pathogens, in addition, N may increase the concentration of amino acids and amides cells which favor the development of fungi. Working with doses and N management in proso millet culture Abrantes *et al*., (2010) observed that sanity is not influenced by the levels and nitrogen fertilization times, but the main pathogens found were Cladosporium*spp*., Curvularia*spp*., Phomaspp ., *Bipolaris spp*., *Fusarium spp*. and *Alternaria spp*. In rice, excessive application of N (200 kg ha^{-1}) increases the incidence of Alternariapadwickii, and N levels exceeding 100 kg. ha^{-1} increase the incidence of *Curvularialunata* and *Phomasorghin*(Agarwal *et al*., 1975).

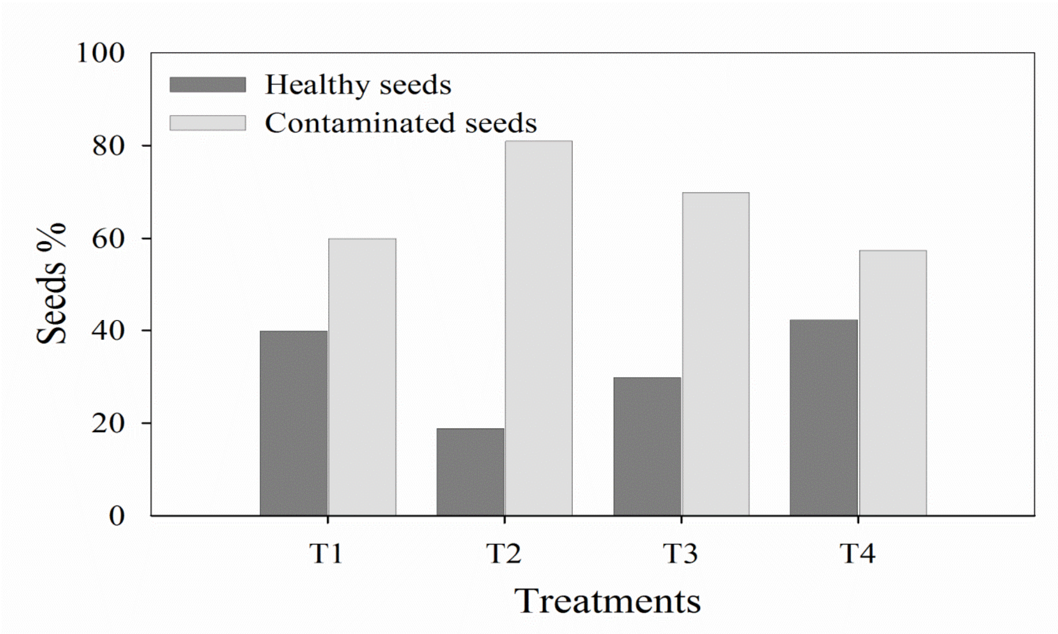

Fig. 3: Percentages of healthy seeds and proso millet contaminated due to the application of different sources of N, T1- control, T2- Poultry litter, T3- pig slurry composting, T4 - mineral fertilizer (NPK).

It is indisputable today, the importance of quality seeds for agricultural production. And in the future will increase this trend requiring further care processes to ensure the final product quality. Quality seed is synonymous with security and stability for sustainable agriculture. The results of this study demonstrated the potential of pig slurry composting and mineral fertilizer in improving the quality of proso millet seeds. Mineral fertilizer provides borne diseases incidence reduction in proso millet, suggesting the added benefit of better disease management for millet small-scale producers in southern Brazil. The poultry litter provided greater incidence of pathogens on proso millet seeds. It has been recommended the use of animal wastes and mineral fertilizer, to overcome the negative effects caused by adding composted manure (Nyamangara *et al*., 2003), and resistant varieties and local fertilizer inputs can lead to a production system more sustainable millet. However, more research is needed in different millet production systems, and in other areas with different climatic and environmental conditions.

Conclusion:

The pig slurry composting and chemical fertilizer provided the best results in seed quality.

The sanity of pros millet seed was influenced by the sources of N. chicken litter provides seeds with higher incidence of pathogens. Minor pathogens incidences are found with the use of chemical fertilizer.

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