## INITIAL GROWTH OF YELLOW PASSION FRUIT SEEDLINGS IN SUBSTRATE COMPOSED OF PULP MILL SLUDGE AND CATTLE MANURE

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**ABSTRACT** - In the last years, as interest in recycling and waste use has increased, many researchers have studied a wide range of potential substrates alternatives. Therefore, is possible that pulp mill sludge can be used as a substrate to seedlings production, while reducing costs. The aim was to evaluate the potential of using pulp mill sludge combined or not with cattle manure as an alternative substrate component for production of seedlings of yellow passion fruit. The experimental design was randomized blocks, in factorial 2 x 3 x 5 design, begin two cultivars (Yellow Maguary FB 100 and FB Master ® 200), three doses of N-P-K (0, 21.4 and 42.8 g plant<sup>-1</sup>) and five combinations of pulp mill sludge (PMS) and cattle manure (CM). Were used the following combinations: CM 100%, 25% PMS + 75% CM, 50% PMS + 50% CM, 75% PMS + 25% CM and PMS 100%, with four replicates. After 48 days were evaluated shoot and root length, number of leaves, stem diameter, shoot fresh and dry weight and root fresh and dry weight. Observed that increasing concentration of waste in the substrate caused significant damage to seedlings in both cultivars, probably due to nutrient imbalances, therefore is detrimental to the development of seedlings; the application of N-P-K, in the tested doses, is not sufficient to avoid adverse effects in the seedlings or to correct the balance nutritional of waste; and the cattle manure is the best option for the production of quality seedlings, in the conditions studied.

Keywords: Passiflora edulis f. flavicarpa. Organic waste. Nutrient imbalance. Paper and cellulose industry.

#### CRESCIMENTO INICIAL DE MUDAS DE MARACUJAZEIRO AMARELO EM SUBSTRATO COMPOSTO POR RESÍDUO DE CELULOSE E ESTERCO BOVINO

**RESUMO** – Nos últimos anos, o interesse na reciclagem e utilização de resíduos tem aumentado, muitos pesquisadores têm estudado uma grande variedade de substratos alternativos. Portanto, é possível que o resíduo de celulose possa ser utilizado como substrato na produção de mudas, reduzindo custos. Objetivou-se avaliar o potencial do resíduo de celulose, combinado ou não com esterco bovino como um componente alternativo no substrato para produção de mudas de maracujá amarelo. O delineamento experimental foi em blocos casualizados, em esquema fatorial 2 x 3 x 5, sendo duas cultivares (Yellow Maguary FB 100 e FB Master ® 200), três doses de N-P-K (0, 21,4 e 42,8 g planta-1) e cinco combinações entre resíduo de celulose (PMS) e de esterco bovino (CM). Foram utilizadas as seguintes combinações: CM 100%, 25% PMS + 75% CM, 50% PMS + 50% CM, 75% PMS + 25% CM e PMS 100%, com quatro repetições. Após 48 dias foram avaliados: comprimento da parte aérea, comprimento da raiz, número de folhas, diâmetro do caule, massa fresca e seca da parte e massa fresca e seca da raiz. Observou que o aumento da concentração de resíduo no substrato causou danos significativos em ambas as cultivares, provavelmente devido a desequilíbrios nutricionais, sendo prejudicial para o desenvolvimento das mudas; a aplicação de fertilizante, nas doses testadas, não é suficiente para evitar os efeitos negativos nas mudas ou para corrigir o equilíbrio nutricional do resíduo; e o esterco bovino é a melhor opção para produção de mudas de qualidade, nas condições estudadas.

**Palavras-chave**: *Passiflora edulis f. flavicarpa*. Resíduo orgânico. Desbalanço nutricional. Indústria de papel e celulose.

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## **INTRODUCTION**

Worldwide more than 300 million tons of wood fiber products are produced per year (BLANCO et al., 2004). The management of solid organic waste associated with this industry is a matter of scale and global significance (FRASER; O'HALLORAN; Van Den HEUVEL, 2009). In the Brazil, paper production reached 9.4 million tons in 2008 and the country was the fourth largest producer of pulp, with approximately 12.7 million tons (BRACELPA, 2009).

To achieve these outputs, the paper and cellulose industries generate large quantities of daily solid waste and liquid effluents (GUIMARÃES et al., 2010), which are a major economic and environmental concern (CHAUDHARI et al., 2010). When discarded in unsuitable environments, these wastes are potential sources of soil, water and air pollution, causing imbalance in different ecosystems. Among the options for disposal, waste recycling is interesting from the socioeconomic and environmental standpoint (PIRES; MATTIAZZO, 2008).

However, in the Brazil, the main wastes disposals are still landfills, which in most cases do not meet the environmental standards of quality (PIRES; MATTIAZZO, 2008). Furthermore, landfill disposal costs are increasing and it is becoming more difficult to acquire new sites for disposal (DEMEYER; VOUNDI NIKANA; VERLOO, 2001; MAH-MOOD; ELLIOT, 2006).

Growing substrates constitute one of the largest costs to growers in the seedlings industries. In the last few decades, as interest in recycling and waste use has increased, researchers have studied a wide range of potential substrates alternatives, including many agricultural, industrial, and consumer waste byproducts (BI; EVANS, 2010). Many of these materials have demonstrated the potential to replace commercial products or serve as substrate amendments. These include substrate components made from tree or wood residues (FREITAS et al., 2010; MURAISHI et al., 2010), coconut fiber (FREITAS et al., 2010; CARRIJO; LIZ; MAKISHIMA, 2002), vermicompost (SANTOS et al., 2010; STEFFEN et al., 2011), municipal waste compost (SCHEER; CARNEIRO; SANTOS, 2010; CORRÊA; BENTO, 2010), pulp mill boiler ash (BI; EVANS, 2010), and many other waste byproducts.

It is possible that pulp mill sludge can be used as a substrate amendment in greenhouse and nursery production while reducing substrate costs and alleviating some problems pulp mill operators confront when using current methods of disposal. However, the use of organic wastes based greenhouse and nursery substrates differ significantly from field soils, and plants may respond differently to wastes applications.

Brazil has a good soil and climate condition to yellow passion fruit plants (*Passiflora edulis f.* 

*flavicarpa*). Currently it is the largest producer and consumer of this fruit worldwide with estimated area of this crop of 47,032 ha (IBGE, 2007) and it is economically and socially important due to the increasing demand. It is estimated that over 60% of Brazilian fruit production is consumed without industrial processing and the remainder is used for pulp and juice production (ROSSI et al, 2001).

Using cultivars of yellow passion fruit as test plant, the objective of this study was to evaluate the potential of using pulp mill sludge combined or not with cattle manure as an alternative substrate component for yellow passion fruit seedlings production.

## MATERIAL AND METHODS

The study was conducted in a greenhouse in Governador Valadares, Minas Gerais State; Latitude 18°51'S, longitude 41°56'W and altitude of 148 m. According to Köppen Classification, the climate of this region is tropical, warm and dry, the average annual temperature is 25 °C, ranging from 22 °C (June) to 27 °C (February).

The pulp mill sludge (PMS) was collected in the discharge area of the Santa Therezinha S/A (Santher) paper mill. The raw material for this industry is newspaper, books, bank and office composts and other paper that can be recycled; these row material is characterized by being rich in short cellulose fibers (fibrillose). The cattle manure was collected in a cattle farm. A sample of each material was taken for chemical analysis (EMBRAPA, 1999). The chemical characteristics of organic wastes are shown in Table 1.

The study was arranged in a randomized complete block design, with four replications. Treatments were arranged in a factorial arrangement 2 x 3 x 5, begin two cultivars of yellow passion fruit recommended for the region (Maguary FB 100 and FB Yellow Master<sup>®</sup> 200), three proportions of N-P-K fertilizer (8-28-16) to test a possible response to fertilization used by farms in the region, D0 - control (no fertilizer ); D1 - 21.4 g plant<sup>-1</sup>, equivalent to 1.7 g of N, 6 g of P<sub>2</sub>O<sub>5</sub> and 3.4 g of K<sub>2</sub>O and D2 - 42.8 g plant<sup>-1</sup> and five substrates formed from the pulp mill (PMS) and cattle manure (CM) combination, being: 100CM = 100% CM; 25PMS = 25% PMS + 75% CM; 50PMS = 50% PMS + 50% CM; 75PMS = 75% PMS + 25% CM and 100PMS = 100% PMS.

The experimental units consisted of polyethylene bags,  $15 \times 10$  cm with a volume of approximately 1.8 L. The seeds were soaked in water for 3 hours and then it was sowed three of then per experimental unit. The seedlings were thinned 15 days after planting, leaving only the most vigorous one per container.

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Chemical attributes	Unit	PMS	CM
pH water (1:2.5)	-	8.30	8.20
Р	g dm <sup>-3</sup>	0.28	17.80
Ca	cmol <sub>c</sub> dm <sup>-3</sup>	190.23	155.18
Mg	cmol <sub>c</sub> dm <sup>-3</sup>	16.61	75.68
K	cmol <sub>c</sub> dm <sup>-3</sup>	1.20	83.12
Na	cmol <sub>c</sub> dm <sup>-3</sup>	3.26	35.65
Organic carbon	g kg <sup>-1</sup>	236.40	571.00
Organic matter	g kg <sup>-1</sup>	407.55	984.30
N-Ammonium	mg kg <sup>-1</sup>	263.20	nd
N-Nitrate	mg kg <sup>-1</sup>	171.08	nd
N-Kjeldahl	g kg <sup>-1</sup>	3.72	23.10
Cu	mg kg <sup>-1</sup>	162.2	33.1
Fe	mg kg <sup>-1</sup>	3,241	985.2
Zn	mg kg <sup>-1</sup>	327	329
Mn	mg kg <sup>-1</sup>	50.34	261
C:N ratio	-	63.55	32.0
PMS: pulp mill sludge; CM: cattle manure; nd: not determined			

**Table 1.** Chemical characterization (dry basis) of samples of pulp mill sludge and cattle manure used in the experiment.

x 5, begin two cultivars of yellow passion fruit recommended for the region (Maguary FB 100 and FB Yellow Master<sup>®</sup> 200), three proportions of N-P-K fertilizer (8-28-16) to test a possible response to fertilization used by farms in the region, D0 - control (no fertilizer ); D1 - 21.4 g plant<sup>-1</sup>, equivalent to 1.7 g of N, 6 g of P<sub>2</sub>O<sub>5</sub> and 3.4 g of K<sub>2</sub>O and D2 - 42.8 g plant<sup>-1</sup> and five substrates formed from the pulp mill (PMS) and cattle manure (CM) combination, being: 100CM = 100% CM; 25PMS = 25% PMS + 75% CM; 50PMS = 50% PMS + 50% CM; 75PMS = 75% PMS + 25% CM and 100PMS = 100% PMS.

The experimental units consisted of polyethylene bags,  $15 \ge 10$  cm with a volume of approximately 1.8 L. The seeds were soaked in water for 3 hours and then it was sowed three of then per experimental unit. The seedlings were thinned 15 days after planting, leaving only the most vigorous one per container.

At 25 days were applied mineral fertilizer and the seedlings were watered three times a day, with approximately 100 ml of distillated water during the first 15 days and then were reduced to once a day in order to keep the necessary moisture of substrate, for 24 hours. The experiment lasted 48 days, at that time the seedlings growed to a height suitable for transplanting to the field.

After harvesting, the seedlings were separated in shoots and root. The roots were washed under running water to remove the substrate bonded. The seedlings were evaluated for their growth from the following factors: SL - shoot length (cm), distance from the cervix to the apex of the apical meristem; RL - root length (cm), distance from the main root apex to the base of the plant according to Zanella et al. (2006); NL - number of leaves (unit); SD - stem diameter (mm); SFW - shoot fresh weight (g); SDW - shoot dry weight (g); RFW - root fresh weight (g) and RDW - root dry weight (g). The dry masses of roots and shoots were obtained after drying in an oven with forced air at 65 °C for 72 hours.

Data were subjected to analysis of variance (ANOVA) and regression. The percentage of residual cellulose in the mixture was related to the parameter estimated by regression equations. Analyses were performed using the statistical program SAS (SAS INSTITUTE, 2004).

### **RESULTS AND DISCUSSION**

Regarding the growth variables of passion fruit seedlings, there were no significant interactions between cultivars x doses of N-P-K fertilizer and in the triple interaction between cultivars x substrate x N-P-K fertilizer. The pH values of both wastes were very close, characterizing them as high alkali according to chemical classification and very high according to agronomic classification (RIBEIRO; GUIMARÃES; ALVAREZ, 1999).

The maximum shoot length (SL) in the FB100 cultivar (11.76 cm) was observed when the mineral fertilizer (D1) was used on the 100CM substrate (Figure 1A). The mineral fertilizer provided readily available nutrients associated with the chemical properties of cattle manure (Figure 1A), which also has an excellent water retention capacity and a high rate of mineralization. In other treatments the presence of sludge in the substrates resulted in linear reductions of the variable SL values (Figure 1A), the greatest reduction in this variable (4.09 cm) observed in the treatment without mineral fertilizer associated with the 100PMS substrate (Figure 1A).

Plants growing in a high pH substrate are subject to nutrient imbalances as a result of changes in nutrient availability as pH increases in the substrate (GUIHONG; WILLIAMS, 2009). This effect may be related to the mainly low availability of micronutrients related to vegetative growth, such as Cu which has an important role in photosynthesis, respiration, fixation and reduction of nitrogen, such as Fe, when deficiency occurs, severe inhibition of cell division and consequently reduction in leaf growth; many Zn-dependent enzymes are involved in particular in the metabolism of leaves and the Mn that is very important for the formation of chlorophyll and participates of the energetic respiratory metabolism (DECHEN; NACHTIGALL, 2006).

Although the pH values of the substrates being nearly equal, the differences in SL results may be related to differences in the Mg and K concentration, which do not suffer interference in absorption when increasing pH of the substrate (EPSTEIN; BLOOM, 2005). The Mg is involved in the biosynthesis of chlorophyll, protein synthesis, enzyme activation, phosphorylation and photosynthesis. The K is involved in enzyme activation, protein synthesis, photosynthesis, carbohydrate transport, osmoregulation and in this case, mainly the cell expansion

#### (EPSTEIN; BLOOM, 2005).

Moreover, the N content of the pulp mill sludge is very low, making it, another limiting for plant growth. The nitrogen is the most nutrient mineral required by plants, have a structural function in higher plants, as part of amino acid molecules, and proteins, besides being a constituent of nitrogenous bases and nucleic acids. Also participates in processes such as ion uptake, photosynthesis, respiration, cell proliferation and differentiation (MALAVOLTA; VITTI; OLIVEIRA, 1989).

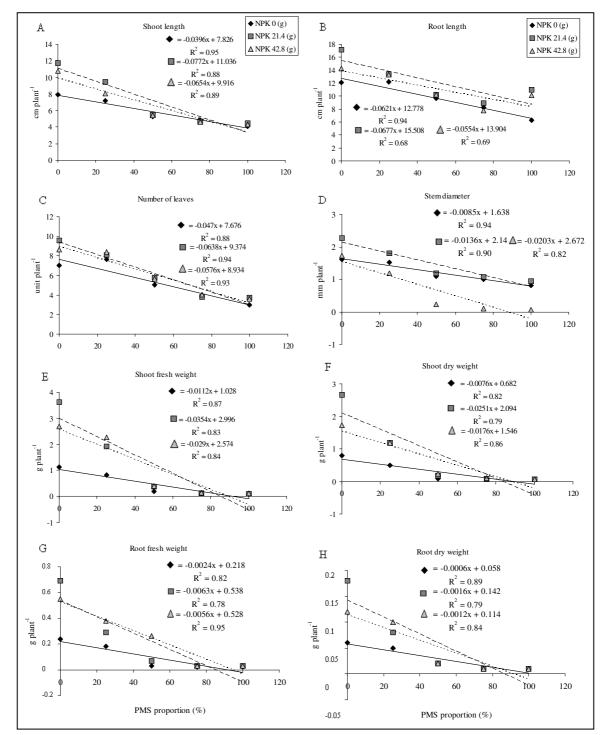


Figure 1. Physiological traits of FB100 cultivar on different substrates made from mixed pulp mill sludge and cattle manure receiving increased amounts of fertilizer.

The best root length (17.14 cm) for FB100 cultivar was achieved in the 100CM substrate, with both fertilizer rates. The fertilizer rates D1 and D2 reduced the negative effect of the pure sludge, with increases in the observed values for variable RL (Figure 1B). This caused a linear negative effect (Figure 1B). However, element Ca is a nutrient that plays an important role in root growth of plants. According to Quaggio (2000) the Ca uptake occurs only in the newer, not yet rubberized parts of roots, so there is need for continued absorption of this nutrient in order for the root system to develop system, which implies that Ca must be properly distributed. According to Miranda et al. (2010) who evaluated the nutritional deficiency in seedlings, they found that the absence of Ca was the most limiting factor in the initial development of seedlings. Pulp mill solids produced stunting and deformities in radish and cress seedlings. Both toxic constituents and nutrient imbalances may be responsible for the growth-inhibiting effects of these amendments (LEVY; TAYLOR, 2003).

The variable SD had the highest values for FB100 in the 100CM substrate (2.28 and 3.07 mm) at both D1 and D2 rates, respectively (Figure 1D). Increasing the concentration of sludge in the substrate caused a linear reduction in all rates of mineral fertilizer and a lower curve due to the values observed in the 100PMS and 75PMS substrates (Figure 1D). Levy and Taylor (2003) observed that pulp mill sludge were strongly inhibitory and produced stunting and deformities in radish and cress seedlings. However, fresh pulp mill sludge has a higher Ca concentration though other features should be considered to explain the negative effect as an unavailability of iron, essential for energy metabolism, active to nitrogen fixation and development of the trunk and roots.

For the NL variable, the best results obtained by FB100 cultivar (9.57 and 8.63) were observed in substrate 100CM, with D1 and D2 rates. However increasing the sludge concentration has a direct effect, causing linear reductions in the values observed for this variable (Figure 1C). This effect can be directly related to the dynamics of N in the substrate and its availability to seedlings, besides unavailability of micronutrients due to the high pH causing a nutritional imbalance.

For variables involving seedling weight in the FB100 cultivar, the behavior was the same, the best weights being observed in the substrate 100CM using mineral fertilizer rate (D1) (Figures 1E to 1H). There was a marked negative effect when the proportion of sludge in the substrates studied was increased, with differences of approximately 36 and 18 times between the highest (100CM) and lowest concentration (100PMS) in the variables as SDW and RDW, respectively (Figures 1F and 1G). This is in accordance with São José (1994) and Souza and Meletti (1997), who performed research testing different types of substrates in tubes and found that cattle manure had

an excellent effect on the initial development of seedlings. Application of pulp mill sludge to agricultural soil without composting may lead to deleterious effects on vegetable crops (LEVY; TAYLOR, 2003), due to the chemical characteristics unfavorable to the development of seedlings of yellow passion fruit.

Was found a decreasing behavior for the SL variable in FB200 cultivar, according to the concentration increased pulp mill sludge in the substrate independent of the mineral fertilizer rates applied. However, the D1 in the substrate with pure cattle manure (100 cm) obtained the best response to treatment with the changes for this variable (23.19 cm) (Figure 2A), exceeding the observed value for cultivation FB100 (11.76 cm) in the same treatment (Figure 1A). In all treatments the increased sludge concentration caused reduction of the values observed for this variable (Figure 2A). The same effects were observed for the FB100, due probably to the high pH of the pulp mill sludge and its nutritional imbalance, causing deficiencies in the seedlings by the lack of micronutrients, as Cu, Fe, Zn and Mn, directly connected to many metabolic processes in the plants.

For variable RL in the FB200 cultivar the trend of responses to the increased sludge was similar to that observed for FB100, with the best result in the D1 rate in the substrate with pure cattle manure (Figure 2B). At the highest sludge concentration (100PMS), a significant stimulus to root elongation was observed. This was not reflected in increased root biomass weight (Figure 2B). Fraser; O'Halloran; Van Den Heuvel (2009) studied the toxic constituents of the fresh pulp mill sludge and observed the same behavior in oats seedlings. They found that pulp mill sludge generally causes a high firing rate that eventually generates a suppressive effect on seedling growth. Both toxic constituents and nutrient imbalances may Be Responsible for the growth-Inhibiting effects of this amendment, as the low nitrogen content of this waste.

The plants are able to perceive the N content, both internally and externally, and modify their metabolism (SAKAKIBARA, 2006). Some of the responses to N are local, restricted only to the roots, directly exposed to nutritional signal, while others are systemic, resulting in intricate routes of perception and signaling of N (FORDE, 2002). In the case of N, owing to the great mobility and, therefore, easily be lost by leaching, the growth of the primary root is maintained, since this can reach the N that had leached, resulting in a disproportional elongation of the root system, with fewer lateral roots; (SILVA; DELATORRE, 2009, OLIVEIRA et al., 2007). Or, there may be morphological adaptations related to the availability of N, one being the suppression of lateral root initiation by high C:N ratio of the plant (ZHANG; RONG; PILBEAM, 2007).

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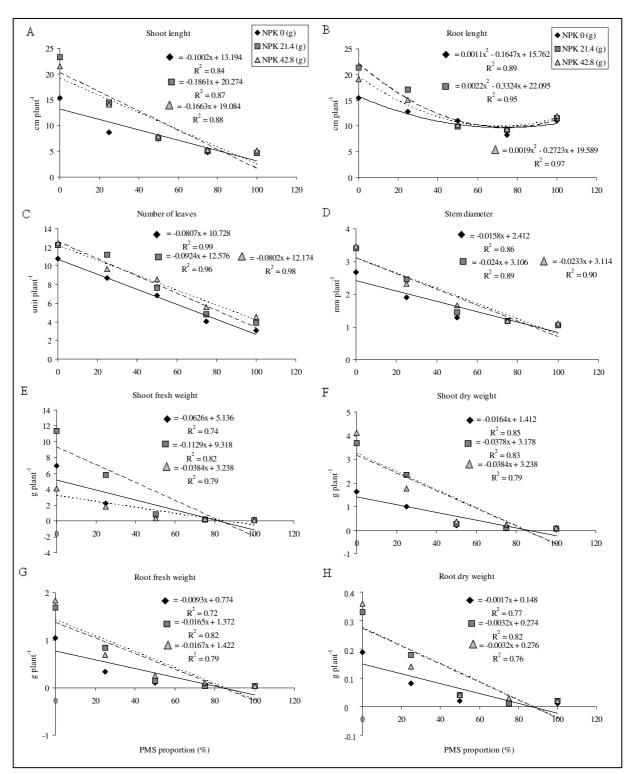


Figure 2. Physiological traits of FB200 cultivar on different substrates made from mixed pulp mill sludge and cattle manure receiving increased amounts of fertilizer.

The 100CM substrate was highest among the treatments that received D2 rate to NL and SD (Figures 2C and 2D). This behavior is similar to that observed for cultivar FB100 (Figures 1C and 1D). The observed values in this variable exceeded those found for cultivar FB100. The 25% increase in the amount of sludge in the substrate generated losses of more than 50% to SFW and RFW (Figures 2E and

2G), but the values found in the 25PMS substrate were higher than those observed in the 100CM substrate in the FB100 cultivar (Figures 1E and 1G).

As for the other substrates we observed reductions in SFW weight (Figure 2E) between 87.1 and 92.6% (50PMS), 94.9 and 98.6% (75PMS) and, 98.0 and 98.8% (100PMS) and reductions in RFW weights between 86.6 and 91.8% (50PMS), 94.3 and

97.9% (75PMS) and, 96.3 and 98.0% (100PMS). These results differ from those observed by Costa et al. (2007) who studied the use of pulp mill sludge in the early development of maize, concluding that no major reductions occurred in the production of root dry matter concentrations up to 40% PMS + 60% of cattle manure.

According to Levy; Taylor (2003) application of pulp mill solids to agricultural soil without composting may lead to deleterious effects on vegetable crops. Same results were found for Bi and Evans (2009), evaluating pulp mill ash as a substrate component in the production of greenhouse-grown French marigold (*Tagetes patula L. 'Janie Deep Orange'*) observed that plants grown in substrates containing 30% to 50% ash had lower shoot dry weights or root quality ratings than plants grown in commercial substrate. Plant growth index, shoot dry weight and root quality rating decreased with increasing ash volume.

These results reflect the nutritional imbalance caused to the seedlings of yellow passion fruit by pulp mill sludge pure or mixed with cattle manure, because the chemical characteristics intrinsic to this material that makes it undesirable for use as a substrate, resulting in seedlings of low quality.

## CONCLUSION

The fresh pulp mill sludge without composting, pure or mixed with cattle manure have growth inhibitory effects, probably due to nutrient imbalances, therefore is detrimental to the development of seedlings; the high pH of pulp mill sludge associated to nutritional deficiencies, cause delayed growth and development; the application of N-P-K fertilizer is not sufficient to avoid adverse effects in the seedlings or to correct the balance nutritional of pulp mill sludge; the cattle manure is the best option for the production of high-quality seedlings in the conditions studied.

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