Allelopathic Potential of Rice Cultivars Sowing in Santa Catarina State on Weedy Rice

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Abstract

Weedy rice belongs to the same species as grown rice, and because it has similar morphophysiological characteristics, its control is difficult, bringing negative effects to the crop. Thus, the work aimed to evaluate the allelopathic potential of the main paddy rice cultivars sowing in Santa Catarina on weedy rice. In the laboratory, with treatments organized in a 4 x 5 factorial scheme, the aqueous extract of four paddy rice cultivars (SCSBRS Tio Taka, SCS 116 Satoru, SCS 121 CL and SCS 122 Miura) was evaluated in five concentrations (0, 25, 50, 75 and 100%). In a greenhouse, with treatments in a 4 x 2 factorial scheme, the straw allelopathy capacity of the same rice cultivars in two quantities (0.0 and 3.0 t ha⁻¹) was evaluated to suppress germination and weedy rice initial growth. All experiments were carried out in a completely randomized design. In the first assay, the aqueous extract of the paddy rice cultivars used in the state of Santa Catarina did not have an allelopathic effect on weedy rice. In the second assay, it was found that the presence of rice straw reduced the density of weedy rice plants. However, the presence of straw on the soil favored the initial growth of the weed, significantly increasing the plant height, number of leaves per plant and shoot dry biomass.

Keywords: allelopathy, aqueous extract, *Oryza sativa*, straw, weed management

1. Introduction

Weedy rice is one of the most frequent weeds and causes the greatest damage to paddy rice fields in Rio Grande do Sul (RS) and Santa Catarina (SC) State, with reduced yield and quality of the harvested product (Nunes et al., 2018). That is why it became important to search for agronomical alternatives that are efficient to suppress or eliminate this problem, and, consequently, maximize grain yield and preserve the industrial and culinary quality of the harvested product.

The term allelochemical is used to designate the chemical substances that act in the plant x plant interaction, including also microorganisms that can result in inhibitory or stimulating effects, the allelochemicals are released into the environment by root exudation, leaching, volatilization and decomposition of the plant straw (Souza Filho, 2014). Among allelochemicals commonly cited as responsible for causing direct and indirect effects are phenolic compounds, terpenes and nitrogen compounds (Macías et al., 2019). A series of secondary metabolites synthesized by rice can suppress the growth of weeds, including fatty acids, indols, momylactones, phenolics and terpenes (Khanh et al., 2005). One of the most
active allelochemicals synthesized by rice is momilactone B, which has a high capacity to inhibit plant growth (Sultana et al., 2019). Momilactone B is a diterpene synthesized from the mevalonic acid and methylyerythritol phosphate pathway (Macias et al., 2019).

All plants synthesize secondary metabolites, which change in quality and quantity from species to species, even in the amount of the metabolite from one place of occurrence or growing cycle to another, since many of them have their synthesis triggered by eventual environmental condition to which the plants are exposed. The synthesis of allelochemicals is also dependent on several conditions such as genotype, stage of development, part of the plant, genetic influence, environmental conditions, biotic and abiotic factors (Ferreira and Aquila, 2000).

Research that assesses the allelopathic potential of different paddy rice genotypes is scarce in Brazil, with the work of Brunes et al. (2016) one of the pioneers to report the allelopathic potential of high concentrations of aqueous extract of rice leaves. The authors evaluated the allelopathic potential of aqueous extract from paddy rice genotypes used mainly in the State of Rio Grande Sul and the weeds assessed were southern crabgrass (Digitaria ciliaris) and joint-vetch (Aeschynomene denticulata). These species are weeds with less importance in the paddy rice fields in Santa Catarina State.

Much is known about compounds with allelopathic activity synthesized by paddy rice, the differences between cultivars and their impact on interspecific competition. However, few was researched about the ability of these allelochemicals to mitigate intraspecific competition, which occurs when weedy rice grows in co-culture to paddy rice.

Likewise, the initial hypothesis of this work was that the paddy rice cultivars sowing in Santa Catarina could have allelopathic potential over weedy rice, and that this could be used as a tool for the integrated management of weeds. Thus, the objective of the work was to evaluate the allelopathic potential of the main paddy rice cultivars used in Santa Catarina on weedy rice.

2. Material and Methods

2.1 Allelopathic Potential of Aqueous Extract From Paddy Rice Cultivars on Weedy Rice Germination

The assay was carried out in the seed laboratory of the Catarinense Federal Institute (IFC) in the city of Rio do Sul, SC, Brazil. A completely randomized design with treatments organized in a 4 x 5 factorial scheme was used, evaluating the aqueous extract of four paddy rice cultivars (SCSBRS Tio Taka, SCS 116 Satoru, SCS 121 CL and SCS 122 Miura) in five concentrations (0, 25, 50, 75 and 100%). The weed assessed was the weedy rice specie (Oryza sativa).

The shoot aqueous extract (SAE) was obtained in a similar way to that described by Prates et al. (2000), Magiero et al. (2009) and Brunes et al. (2016). For this, 200 g of fresh rice leaves were used, macerated in 1 liter of distilled water (20% p v⁻¹), in a blender for 1 minute. The macerate was filtered through filter paper, being called 100% concentration. SAE was used
concentrated (100%) and diluted with distilled water in the concentrations of 75, 50 and 25%, the concentration 0% consisted only of distilled water. The leaves of rice cultivars were collected at the R1 stage, that is, in panicle differentiation (Counce et al., 2000).

The seeds of weedy rice were submitted to the germination test, using four replicates, with 50 seeds each. The seeds were placed in a gerbox with germitest paper substrate, which were previously moistened with the substrate in a proportion of three times the dry paper mass. After packing the seeds, the gerboxes were sent to a germination chamber at a temperature of 25 °C.

At four days, the first germination count was performed and at seven days, the final germination test, counting the number of normal seedlings. A normal seedling was one that showed coleoptile and radicle development without any alteration or deformity.

The data were subjected to analysis of variance and the means compared by confidence interval, the treatments were considered different when the confidence intervals did not overlap (Cumming et al., 2004; Concenço et al., 2018).

2.2 Effect of Straw Suppression From Different Paddy Rice Cultivars on Weedy Rice

The assay was carried out in a greenhouse at IFC in Rio do Sul, SC, Brazil. Initially, rice plants of the cultivars SCSBRS Tio Taka, SCS 116 Satoru, SCS 122 Miura and SCS 121 CL were grown in plastic pots up to the R1 stage (Counce et al., 2000), at this moment the shoot was collected, which was later triturated and dried in an oven with forced air circulation at 40 °C, to obtain the straw used in the assay.

The experiment was carried out in a completely randomized design organized in a 4 x 2 factorial scheme, with four replicates. The interaction of straw from four rice cultivars (SCSBRS Tio Taka, SCS 116 Satoru and SCS 121 CL SCS 122 Miura) was evaluated in two quantities (0 and 3.0 t ha⁻¹) on germination and initial development of weedy rice.

The experimental units used in the assay consisted of plastic pots with a capacity of 1 dm⁻³ of soil. In each experimental unit, 10 weedy rice seeds were sown, then the amount of straw equivalent to the treatment and irrigation was accommodated. Irrigation was performed daily using a sprinkler irrigation system, without flooding the soil.

The variables assessed were number of plants emerged per pot, number of fully expanded leaves, plant height and shoot dry mass. The variables plant density, plant height and number of leaves per plant were evaluated at 7, 14 and 28 days after emergence (DAE), evaluating all plants in the experimental unit. The shoot collects to determine the dry mass occurred at 28 DAE.

The data were subjected to analysis of variance by the F test. When the anova showed difference, the means were tested by the Tukey test. The level of significance adopted was 5% (p <0.05).
3. Results

3.1 Allelopathic Potential of Aqueous Extract From Paddy Rice Cultivars on Weedy Rice Germination

No significant difference was observed for the aqueous extract concentration factor and for the rice cultivar interaction and aqueous extract concentration on the germination percentage in the first and final count (Table 1). For the rice cultivar factor, there was a significant difference only for the final count (p < 0.05) (Table 1).

Table 1. Variance analysis summary. Rio do Sul, Brazil, SC, 2018

| Variation Source     | p value | First count | Final count |
|----------------------|---------|-------------|-------------|
| Cultivars (C)        | 0.10 ns | 0.01 *      |
| Extract concentration (EC) | 0.87 ns | 0.42 ns     |
| C x EC              | 0.28 ns | 0.33 ns     |

ns = not significative (p>0.05) and * = significative (p<0.05).

When splitting the aqueous extract concentration factor for each cultivar, it was observed that the polynomial regression models were not significant (Table 2). In addition, the regression deviation was not significant in all analyzes, indicating that the variation in the concentration of the aqueous extract did not influence the germination results, thus not allowing mathematical modeling (Table 2). For this reason, was chosen to present the means followed by the confidence interval (p <0.05 and n = 4).

Table 2. p value to polynomial regression analysis. Rio do Sul, Brazil, 2018

| Cultivars         | First count | Linear | Quadratic | Regression deviation |
|-------------------|-------------|--------|-----------|----------------------|
|                   |             |        |           |                      |
| SCSBRS Tio Taka   | 0.99 ns     | 0.13 ns| 0.86 ns   |                      |
| SCS 116 Satoru    | 0.93 ns     | 0.73 ns| 0.25 ns   |                      |
| SCS 121 CL        | 0.12 ns     | 0.44 ns| 0.08 ns   |                      |
| SCS 122 Miura     | 0.28 ns     | 0.44 ns| 0.88 ns   |                      |

| Cultivars         | Final count | Linear | Quadratic | Regression deviation |
|-------------------|-------------|--------|-----------|----------------------|
|                   |             |        |           |                      |
| SCSBRS Tio Taka   | 0.72 ns     | 0.07 ns| 0.71 ns   |                      |
| SCS 116 Satoru    | 0.72 ns     | 0.76 ns| 0.15 ns   |                      |
| SCS 121 CL        | 0.21 ns     | 0.88 ns| 0.14 ns   |                      |
| SCS 122 Miura     | 0.13 ns     | 0.32 ns| 0.84 ns   |                      |

ns = not significative (p>0.05).

For the first count, it was observed that the germination rate of weedy rice was lower in cultivar SCSBRS Tio Taka than in cultivar SCS 121 CL, only in the control with distilled water (Figure 1A). In the other concentrations (25, 50, 75 and 100%) there was no difference
in the germination of weedy rice between rice cultivars. This result is indicative of the absence of allelopathic effect of the aqueous extract from the shoot of rice cultivars on weedy rice.

Figure 1. weedy rice germination (%) in function to concentration to aqueous extract of paddy rice cultivars to first (A) and final count (B). Bars indicate the confidence intervals (p < 0.05 and n = 4). Rio do Sul, Brazil, 2018

In the final count, it was observed that the germination of the weedy rice was affected only by the rice cultivar factor (Figure 1B). The germination of weedy rice seeds treated with extract of the cultivar SCSBRS Tio Taka was significantly less to the cultivar SCS 116 Satoru. Despite the significant difference, it cannot be said that the cultivar SCSBRS Tio Taka inhibited the germination of weedy rice, due to the lack of response from the aqueous extract concentration factor.
3.2 Effect of Straw Suppression From Different Paddy Rice Cultivars on Weedy Rice

The interaction cultivar and straw quantity, as well as the isolated factor cultivar were not significant for the variable density of plants per pot. Significance was observed only for the straw quantity factor (Table 3). The interaction cultivar and straw were not significant for the plant height variable (Table 3). However, the isolated factors significantly affected this variable. As for the factor number of leaves per plant, it was noted that both the isolated factors, the interaction between cultivars and straw were significant (Table 3). As for the shoot dry mass variable, there was significance only for the straw quantity factor (Table 3).

Table 3. Variance analysis summary. Rio do Sul, Brazil, SC, 2018

|                          | Plant density | p value          | 7 DAE | 14 DAE | 28 DAE |
|--------------------------|--------------|-----------------|-------|-------|-------|
|                          | Cultivars (C)|                 | 0.76ns| 0.89ns| 0.89ns|
|                          | Straw (S)    |                 | 0.02* | 0.01* | 0.01* |
|                          | C x S        |                 | 0.69ns| 0.22ns| 0.22ns|

|                          | Plants height | p value           | 7 DAE | 14 DAE | 28 DAE |
|--------------------------|--------------|-----------------|-------|-------|-------|
|                          | Cultivars (C)|                 | 0.01* | 0.64ns| <0.00*|
|                          | Straw (S)    |                 | 0.01* | <0.00*| <0.00*|
|                          | C x S        |                 | 0.26ns| 0.70ns| 0.18ns|

|                          | Leaves number | p value          | 7 DAE | 14 DAE | 28 DAE |
|--------------------------|--------------|-----------------|-------|-------|-------|
|                          | Cultivars (C)|                 | -     | 0.31ns| <0.00*|
|                          | Straw (S)    |                 | -     | <0.00*| <0.00*|
|                          | C x S        |                 | -     | 0.86ns| <0.00*|

|                          | Shoot dry mass | p value         | 7 DAE | 14 DAE | 28 DAE |
|--------------------------|---------------|-----------------|-------|-------|-------|
|                          | Cultivars (C) |                 | -     | -     | 0.08ns|
|                          | Straw (S)     |                 | -     | -     | <0.00*|
|                          | C x S         |                 | -     | -     | 0.14ns|
The presence of 3.0 t ha\(^{-1}\) of rice straw reduced significantly the number of weedy rice plants per pot, in all evaluations (Table 4). This result was already expected, since the physical effect of soil cover on suppressing the emergence of weeds is widespread. The weedy rice plants that developed in the presence of the straw of the SCSBRS Tio Taka cultivar had plant height lower than the other cultivars (Table 4). It is worth remembering that this cultivar also influenced the germination percentage of weedy rice in the previous assay, being a strong indication that this cultivar is the only one that did not benefit the initial growth of weedy rice. Regarding the straw quantity factor, the results were consistent in all evaluations and indicated that weedy rice plants had significantly higher height when they developed in the presence of straw (Table 4).

Table 4. Weedy rice density (plants pot\(^{-1}\)) and height (cm) at 7, 14 and 28 DAE. Rio do Sul, Brazil, 2018

| Cultivars          | Plant density (plants pot\(^{-1}\)) | Plant height (cm) |
|--------------------|------------------------------------|------------------|
|                    | 7 DAE | 14 DAE | 28 DAE | 7 DAE | 14 DAE | 28 DAE |
| SCSBRS Tio Taka    | 5.2 a  | 8.1 a  | 8.1 a  | 8.8 b | 17.8 a | 21.2 b |
| SCS 116 Satoru     | 7.5 a  | 7.8 a  | 7.8 a  | 9.8   | 18.3 a | 24.0 a |
| SCS 121 CL         | 7.8 a  | 7.8 a  | 7.8 a  | 9.8 a | 18.9 a | 24.8 a |
| SCS 122 Miura      | 8.1 a  | 8.2 a  | 8.2 a  | 9.8 ab| 19.1 a | 25.6 a |
| Straw              |        |        |        |       |        |        |
| 0 t ha\(^{-1}\)    | 8.5 a  | 8.5 a  | 8.5 a  | 9.2 b | 16.1 b | 20.9 b |
| 3 t ha\(^{-1}\)    | 7.3 b  | 7.5 b  | 7.5 b  | 9.8 a | 20.9 a | 26.9 a |
| VC (%)             | 16.8   | 14.5   | 14.5   | 7.8   | 12.2   | 6.6    |

Means followed by the same letter do not differ by Tukey's test (p>0.05).

The presence of straw on the soil also raised the number of leaves per plant, mainly at 14 and 28 DAE (Tables 5 and 6). At 14 DAE, it was observed that weedy rice plants that grew in the presence of straw had an average of 2.8 leaves, while plants in treatments without straw had 2.1 leaves. At 28 DAE, the interaction cultivar and amount of straw was significant (Table 6), and the only treatment in which the presence of straw did not increase the number of weedy rice leaves was in the SCSBRS Tio Taka cultivar. The results obtained are consistent and demonstrate that the cultivar SCSBRS Tio Taka was the only one that did not benefit the initial growth of weedy rice.

The weedy rice that developed in the presence of straw showed a higher accumulation of biomass in the shoot (Table 5), which corroborates with that described in the other growth variables (plant height and number of leaves per plant). In summary, the presence of rice...
straw on the soil reduced the density of emerged weedy rice plants, but significantly favored their initial growth, increasing plant height, number of leaves per plant and accumulated dry biomass in the shoot).

Table 5. Weedy rice leaves number (LN in n°) at 7 and 14 DAE and shoot dry mass (SDM in mg pot⁻¹) at 28 DAE. Rio do Sul, Brazil, 2018

| Cultivars            | LN-7DAE | LN-14DAE | SDM-28DAE |
|----------------------|---------|----------|-----------|
| SCSBRS Tio Taka      | 2.0     | 2.3 a    | 442.6 a   |
| SCS 116 Satoru       | 2.0     | 2.5 a    | 506.7 a   |
| SCS 121 CL           | 2.0     | 2.2 a    | 531.7 a   |
| SCS 122 Miura        | 2.0     | 2.6 a    | 518.9 a   |

| Straw                | LN-7DAE | LN-14DAE | SDM-28DAE |
|----------------------|---------|----------|-----------|
| 0 t ha⁻¹             | 2.0     | 2.1 b    | 434.2 b   |
| 3 t ha⁻¹             | 2.0     | 2.8 a    | 565.7 a   |

VC (%) 16.7 14.0

Means followed by the same letter do not differ by Tukey's test (p>0.05).

Table 6. Weedy rice leaves number (n°) at 28 DAE. Rio do Sul, Brazil, 2018.

| Cultivars            | Straw                  |
|----------------------|------------------------|
|                      | 0 t ha⁻¹ | 3 t ha⁻¹ |
| SCSBRS Tio Taka      | 3.0 aA | 3.0 bA         |
| SCS 116 Satoru       | 3.0 aB | 3.7 aA         |
| SCS 121 CL           | 3.0 aB | 3.7 aA         |
| SCS 122 Miura        | 3.0 Ab | 4.0 aA         |

VC (%) 7.5

Means followed by the same letter lowercase in column (cultivars) and uppercase do not differ by Tukey's test (p>0.05).
4. Discussion

4.1 Allelopathic Potential of Aqueous Extract From Paddy Rice Cultivars on Weedy Rice Germination

The aqueous extract of the cultivars SCSBRS Tio Taka, SCS 116 Satoru, SCS 121 CL and SCS 122 Miura did not suppress weedy rice germination, indicating absence of autotoxicity effect. Previous research that evaluated the allelopathic potential of rice genotypes has shown promising results regarding the allelopathic effect, however these studies have explored the effect of heterotoxicity. Dilday et al. (1994) demonstrated the allelopathic effect of rice genotypes on ducksalad (*Heteranthera limosa*). Other research has reported that some rice cultivars have been promising in suppressing the germination or development of barnyardgrass (*Echinochloa crus-galli*) (Olofsdotter et al., 1995, Chung et al., 1997, Ahn and Chung, 2000, Chung et al., 2001).

However, it is noteworthy that these results are applied to the experimental conditions adopted, that is, using aqueous extract from the shoot of plants that were grown in a greenhouse, free of interspecific competition and collected at the R1 stage. Chang et al. (2001) evaluated the allelopathic effect of rice cultivars by three methods, through germination in aqueous extract, incorporation of cultural residues in the soil and through the coexistence of the crop with the weed in the field. The authors demonstrated that the allelopathic effect of rice depends on the methodology used in the assay, so that the aqueous extract of the Gin shun cultivar was the one that most suppressed barnyardgrass. When the cultural residue was incorporated into the soil, Philippine 2 and Juma 10 cultivars showed greater allelopathic potential. In the research carried out in field conditions, with rice cultivars living exclusively with barnyardgrass, it was observed that cultivars Yuan Hsing I and Juma 10 were the most effective in suppressing the development of the weed.

According to Olofsdotter et al. (1995) the greatest allelopathic potential of rice cultural residues was observed with the incorporation of straw into the soil, after harvesting the crop. The influence of incorporating cultural residues of rice into the soil on the inhibition of weed growth and development suggests that allelopathic compounds are produced during the process of decomposition of cultural residues, in short the interaction between cultural residues and the activity of microorganisms of the soil is essential for the toxic effect (Chung et al., 2001).

The growing of rice cultivars in a greenhouse, in an environment free from stresses and living without weeds, may have contributed to the absence of allelopathic effect. Dayan (2006) demonstrated that factors such as plant age, temperature, light quality, soil pH and water availability modulate the synthesis of sorgoleone in roots of *Sorghum bicolor* (cultivar SX17). This author also demonstrated that when *S. bicolor* developed in the presence of the velvetleaf weed (*Abutilon theophrasti*), the synthesis of the allelochemical sorgoleone was greater, indicating that interspecific competition has an important role in modulating the synthesis of allelochemicals. For rice crop, Sultana et al. (2019) demonstrated that the expression of five genes related to the synthesis of allelochemicals in barnyardgrass (*E. crus-galli*) was higher when the weed developed in co-culture with rice.
The biotic stress that occurs in field conditions plays a fundamental role in the allelopathic capacity of rice crop. Bi et al. (2007) demonstrated that the exogenous application of methyl jasmonate (MeJA) and methyl salicylate (MeSA) increased the allelopathic activity of rice due to the accumulation of phenolic compounds and increased activity and expression of the enzymes phenylalanine ammonia-lyase (PAL) and cinnamate 4-hydroxylase (C4H), which are two key enzymes in the phenylpropanoid biosynthesis pathway. The synthesis of MeJa and MeSa is usually induced in response to attack by insects and pathogens.

4.2 Effect of Straw Suppression From Different Paddy Rice Cultivars on Weedy Rice

The presence of straw from rice cultivars on the soil surface reduced the density of weedy rice. This effect was expected because the soil cover plays an important role in the weed control, primarily due to the physical effect, which limits the passage of light, creating difficulties for the germination of the seeds (Pires et al., 2008), and through the barrier it forms, hindering the initial growth of seedlings, which is one of the main control methods (Guerra et al., 2015). Seeds of reduced size, such as weedy rice (Poaceae), are more susceptible to the physical effect of soil cover with straw (Monqueiro et al., 2011, Silva Jr. et al., 2016).

There was a positive effect of rice straw on the height of weedy rice plants. This effect may be associated with the etiolation of plants in response to soil shading. It is known that plants adapted to the sun and intolerant to the limitation of light have efficient mechanisms to avoid shade. These, when shaded, allocate their reserves for elongation of internodes, accelerating the longitudinal growth of the stem. This condition constitutes extra reserve expenditure, which normally leads to a decrease in the leaf area, the root system and the inhibition of the development of the lateral buds (Vidal et al., 2012). In addition, the increase in plant height as a result of shading is the result of altering physiological processes related to hormonal dynamics and cell division, in addition, the plant's ability to absorb nutrients and water and perform photosynthesis can be impaired (Merotto Jr. et al., 2009).

The straw of the paddy rice cultivars provided a positive effect on the number of leaves and shoot dry mass of the weedy rice. A positive response was not expected, as normally the plant etiolation resulting from the shading of the soil reduces the leaf area and the dry mass of the plants (Merotto Jr et al., 2009, Lamego et al., 2015). In this way, the increase in the height of weedy rice plants that developed in the presence of straw may not be associated with etiolation, but with a positive allelopathic effect of rice straw on the initial growth of weedy rice. Chung et al. (2001) reported that barnyardgrass plants that developed in coexistence with the rice cultivar Rexmont had their growth stimulated, presenting an increase in leaf area, dry leaf mass, stem dry mass and shoot dry mass of 95%, 54%, 20% and 37%, respectively.

It is noteworthy that both assays were carried out in a controlled environment, free from environmental restrictions and competition. Whereas in field conditions, where the availability of resources is limited and weed interference is present, the results may be different, as it is expected that the field conditions improve the allelochemicals production of paddy rice.
The management of weedy rice in paddy rice production systems in southern Brazil is a major challenge, therefore, the use of rice cultivars with high allelopathic potential would be a sustainable strategy. However, with the results of this research it was evident that the main paddy rice cultivars used in the state of Santa Catarina do not have allelopathic activity capable of suppressing the initial growth of weedy rice, on the contrary, the presence of the straw of most cultivars on the surface of the soil stimulated the initial growth of weedy rice.

5. Conclusion

The aqueous extract of the main paddy rice cultivars used in the state of Santa Catarina has no allelopathic effect on weedy rice. The presence of rice straw on the soil reduced the density of emerged weedy rice plants. The weedy rice that developed in the straw of the cultivars SCS 116 Satoru, SCS 121 CL and SCS 122 Miura had the initial growth raised. The SCSBRS Tio Taka cultivar was the only one that did not stimulate the initial growth of weedy rice. Nowadays weedy rice management is a great challenge to rice growers, so researches to identify rice cultivar with high allelopathic potential is a sustainable option. However, little is known about autotoxicity potential of rice genotypes.

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