Available online at https://asric.africa/agricultural-sciences
ASRIC Journal on Agricultural Sciences Vol.6(2)(2025) 17-22

Survival of *Pyricularia Oryzae* on Rice Residues used in Manure Processes in Madagascar

Raveloson Harinjaka[1,2], Ramanantenasoa Marie Hantaniriana Ravakiniaina Sidonie[3], Rakotonanahary Manda Natolotra[1], Rabakomanantsoa Landry Richard Gabriel[1,4], Tharreau Didier[4,5]

[1]Centre National de Recherche Appliquée au Développement Rural : FOFIFA, BP 230 and Institut d'Enseignement Supérieur d'Antsirabe Vakinankaratra (IESAV), BP 105, Antsirabe 110, Madagascar. raveloharinjaka@yahoo.fr, rakotonanahary.natolotra@yahoo.com
[3]Athénée Saint Joseph, BP 287, Antsirabe110, Madagascar. ravakiniainasaphyra@gmail.com
[4]Plant Health Institute of Montpellier (PHIM)/Centre de coopération Internationale en Recherche Agronomiques pour le Développement : CIRAD, Montpellier 34398, France. landryrgrabakomanantsoa@gmail.com
[5]Centre de coopération Internationale en Recherche Agronomiques pour le Développement : CIRAD, Montpellier 34398, France. didier.tharreau@cirad.fr

Received 21 July 2025; revised 22 August 2025; accepted 12 September 2025

Abstract

Manure constitutes the principal fertilization of rice field in the Vakinankaratra region. Adding rice residues on the cattle excretion was recommended to improve manure composition. However, adding rice residues infected by *Pyricularia oryzae*, could play a role on the onset of rice blast disease in the field. To evaluate this risk, the survival of *P. oryzae* on rice residues used in different scenario of manure processes was monitored. Six treatments were tested where infected panicles were placed inside cattle excretion (CE) under anaerobic condition (T1), on CE (T2), inside CE (T3), inside CE with watering every week (T5), on the soil (T6) and infected panicles and CE were returned every two weeks (T4). No sporulation of *P. oryzae* was observed in the T1 (panicles placed in CE with anaerobic condition). However, the panicles with sporulating *P. oryzae* and spore of it were observed in the 5 other treatments at least after seven months. These results show that infected rice residues used in manure processes could be a potential primary inoculum source of rice blast. To minimize the risk of blast epidemic, we propose that only uninfected rice residues should be added to manure.

Keywords: Manure, *Pyricularia oryzae*, rice blast, rice residues, survival

Introduction

The production of sufficient staple food is limited by many constraints in most African countries. One of them is the low use of fertilization for the crops. Mineral fertilization is an efficient way of increasing yield but is not financially sustainable for many farmers in Africa today. This situation is likely to persist since the price of mineral fertilization recently tripled and will continue to rise in the near future (Brunelle et al., 2015). Therefore, alternative solutions for fertilization should be considered. Livestock effluents are used very often in many African agricultural production systems (Chianu et al., 2012), and in particular in Madagascar. However, the quantity of some plant nutrient elements, such as nitrogen, are very low or even absent in traditional manure (Rasolofo, 2017).

Some recommendations were proposed to improve the quality of cattle manure: concrete paving of the cattle barn, watering and return the manure frequently and add plant residues in the cattle excretion. But, the use of rice residues to improve manure could generate a problem of disease management. It was previously shown that rice residues infected by a rice pathogen (*Pyricularia oryzae*), could be a potential source of inoculum and favor the onset of a major disease of rice, blast (Raveloson et al., 2018).

Several studies attempted to determine the role of potential inoculum sources of rice blast like seeds (Faivre-Rampant et al., 2013; Guerber and TeBeest, 2006; Long et al., 2001; Manandhar et al., 1998) and rice residues (Raveloson et al., 2018). However, the survival of *P. oryzae* on rice residues used in the manure processes was not documented. Since manure constitutes the principal fertilization of rice field in the Vakinankaratra region of Madagascar, it is important to evaluate the survival of *P. oryzae* in manure and to determine if adding infected rice residues in the manure is increasing the risk of blast epidemics.

This study aimed determining the survival capacity of the blast pathogen on infected rice residues in different scenario of manure processes, either traditional or improved.

Materials and Methods

Survey and Collect of Panicles infected by *Pyricularia oryzae*

During the cropping season 2020-2021, rice blast attack was monitored in rice fields in the Vakinankaratra region of the highlands of Madagascar. Samples of panicles attacked by rice blast pathogen were collected for conducting the trial.

Preparation of Blast Infected Panicles

Blast infected panicles were prepared in the Research Regional Center of FOFIFA (*Centre National de la Recherche Appliquée au Développement Rural*: National Agriculture Research Center) at Antsirabe. Ten panicles per batch were attached by fabric yarn. Then 10 batches were used for each replication of each treatment.

Set up of Infected Panicles in Different Cattle Manure Processes

The experiment was designed based on the recommendations for improving the cattle manure and on traditional manure processes. Fresh cattle excretion was put in a square basin. Six treatments were tested and differentiated by the positioning of the infected panicles and by the treatment : in the first treatment (T1), infected panicles were placed inside the cattle excretion (CE) i.e. under anaerobic conditions (basin covered by plastic bag); in the second treatment (T2), infected panicles were placed on the CE; in the third treatment (T3), infected panicles were placed inside the CE; in the fourth treatment (T4), infected panicles and CE were returned every two weeks; in the fifth treatment (T5), infected panicles were placed inside CE with watering every week; and in the sixth treatment (T6), infected panicles were placed on the soil without CE. A randomized complete block design with five replications were used for this trial.

Monitoring of *P. Oryzae* Survival in Infected Panicles Under Different Manure Processes

In May 2021, before placing the panicles in different conditions, fifty panicles were checked for the sporulation of the *Pyricularia oryzae* and for the number of produced spores on these panicles which were considered as the initial situation. The monitoring was planned from May 2021 to Mars 2022. One batch of panicle stems were removed from each treatment and each replication every month to assess the remaining panicles in each scenario of cattle manure process. Then the survival of *P. oryzae* was tested by observing the production of spores under a binocular microscope after placing panicles in Petri dishes with dampened filter paper to stimulate sporulation. Evaluation was based on the presence or absence of typical spores of *P. oryzae* on each panicle. After seven days of sporulation observation, the total number of spores produced by the 10 panicles in the batches were counted by using Malassez cell as described by (Gallet et al., 2014). The percentage of panicles with living *P. oryzae* was calculated as the number of panicles with visible sporulation of the rice blast fungus divided by the number of panicles remaining in the batches. The average number of remaining panicles, the percentage of panicles with *P. oryzae* sporulation and the number of spores produced were calculated for each of the five replicates.

Results

Due to natural degradation, the number of panicle stems observed are likely to decrease with time and may limit the observation of *P. oryzae* survival. The number of panicle stems observed (table 1) was stable and maximum all over the experiment for three treatments: panicles on the cattle excretion (T2), return of panicles and CE every two weeks (T4) and panicles on the soil (T6). The number of panicle stems observed started to significantly decrease at the 6th, 7th and 8th observation dates for the treatments panicle inside the CE with anaerobic condition (T1), panicles inside de CE (T3) and panicles inside CE with watering every week (T5), respectively.

Table 1. Variation over time of the average number of panicle stems observed per treatment. T0: initial situation before placing panicle stems in the manure.

Notation dates **Treatment**	T0	1st	2nd	3rd	4th	5th	6th	7th	8th	9th	10th
T1	10	10	10	10	10	9.8	7.4	5.8	0	0	0
T2	10	10	10	10	10	10	10	10	10	10	10
T3	10	10	10	10	10	10	8.8	9	6.6	5.8	5.2
T4	10	10	10	10	10	9.6	10	10	10	10	10
T5	10	10	10	10	10	9.4	9.6	8.2	3	1.2	2.6
T6	10	10	10	10	10	10	10	10	10	10	10

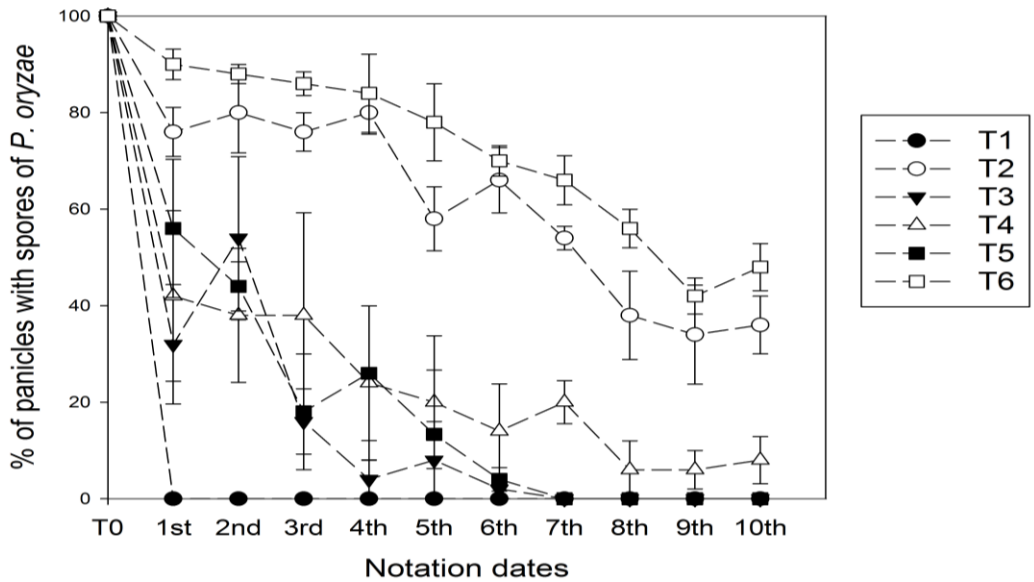

Figure 1: Variation over time of the percentage of panicle stems producing *Pyricularia oryzae* spores in the different treatments of cattle manure processes: infected panicles were placed inside cattle excretion (CE) under anaerobic condition (T1), on CE (T2), inside CE (T3), infected panicles and CE were returned every two weeks (T4), inside CE with watering every week (T5) and on the soil (T6). T0: initial situation; Vertical bars are standard errors of the means.

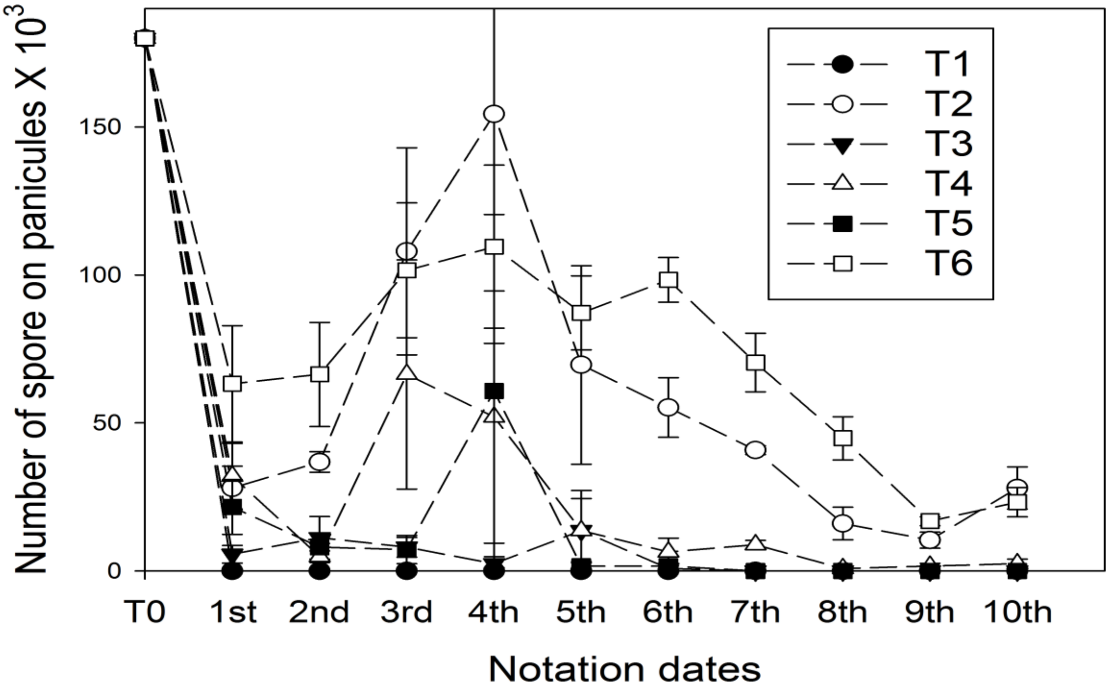

Figure 2. Variation over time of the number of spores produced on remaining panicles in a batch for different treatments of cattle manure processes: infected panicles were placed inside cattle excretion (CE) under anaerobic condition (T1), on CE (T2), inside CE (T3), infected panicles and CE were returned every two weeks (T4), inside CE with watering every week (T5) and on the soil (T6). T0: initial situation; Vertical bars are standard errors of the means.

A significant effect ($P < 0.05$) of treatments and observation dates was observed for the percentage of panicles with sporulation of *P. oryzae* (figure 1). In treatment T1, the rate of sporulating panicles decreased rapidly and no more sporulation was observed after the first observation. For treatments T3, T4 and T5, the rate of sporulating panicles decreased relatively at the first date of observation (50% of sporulating panicles). No sporulation was observed starting at the 7^{th} observation date for T3 and T5. An average rate of 8% was observed for the treatment T4 at the end of notation. For treatments T2 and T6, the sporulating rate decreased slowly and remained stable around 40% at the end of the notation.

In addition to a qualitative evaluation of sporulation, the number of spores produced by the remaining infected panicles in a batch was counted. The initial average number of spores produced was 1.8×10^5 per batch. The number of spores produced followed a similar trend as the percentage of panicles with sporulation of *P. oryzae* (figure 2). The difference of spore produced was significant ($P < 0.05$) for the six treatments. No spore was produced for the treatment T1 (panicles inside CE under anaerobic condition) from the first notation. Then, the spore number produced decreased rapidly for treatments T3 (panicles inside CE) and T5 (panicles in CE with watering every week) and no spores were observed from the 7^{th} notation dates. For the treatments T2 (panicles placed on cattle excretion) and T6 (panicles placed on the soil) remained high over the notation period. At the end of notation, 28 000 and 23 000 spores were counted for T2 and T6, respectively. For the treatment T4 (return of panicles and CE every two weeks, the spore number produced was less than T2 and T6 over the time but 2400 spores were observed at the end of notation in March 2022.

Discussion

Because of the importance of manure in fertilization of crops, several studies are interested in the survival of pathogens in the manure. According to Black et al. (2021) and Sharma and Reynnells (2016), some bacterial pathogens could survive in the manure. Understanding the survival of rice blast pathogen in the manure was a useful information in particular for rice culture in the Vakinankaratra Region of Madagascar. To the authors' knowledge, the present study is the first work to monitor the survival of *P. oryzae* in rice residues used in manure processes. the results showed that the *P. oryzae* could be survived in different manure processes. It is important to consider this finding in the rice blast epidemics problem. Because rice blast was considered as one of the major constraints of rice culture in this region (Sester et al., 2014).

The idea of this trial is also to compare the survival capacity of *P. oryzae* between the improvement of manure like : placing rice residues inside the cattle excretion, watering and return rice residues and manure frequently (Rasolofo, 2017 ; Fanjaniaina et al., 2022) and traditional manure like : placing rice residues on the cattle excretion. A part from the placing rice residues inside the cattle excretion under anaerobic condition where no sporulation of *P. oryzae* was observed from the first observation, the sporulation and number of spores produced were still observed for the other five treatments. Therefore, the recommended technic allowed to improve the plant nutrient elements in the manure but did not influence the survival of *P. oryzae*. Infected rice residues added in manure constitute a reservoir of rice blast pathogen.

Previous study conducting by Raveloson et al. (2018) showed that the *Pyricularia oryzae* survived on the infected rice residues and generated the rice blast epidemic for the next season. The monitoring of the *P. oryzae* survival in the current study support the risk of rice blast onset by infected rice residues used in manure processes. Whatever the treatments (on and inside with or without return and watering), sporulation of *P. oryzae* on the panicles at a relatively high rate was still observed after seven month (December) and up to 10 months (March) in some manure processes after scoring. Generally, the onset of foliar rice blast occurs at the end of December in the highland of Vakinankaratra region. So, the survival of the blast fungus in the manure is longer than the intercropping season. Thus, the addition of infected rice residues in the manure could be a potential source of primary inoculum for rice blast epidemics.

Placing the infected panicles inside the cattle excretion with anaerobic condition was the only way to rapidly and completely kill *P. oryzae* in the manure process. So, to minimize the risk of rice blast from the rice residues added in manure, a short-term solution is to use uninfected rice residues for manure and use infected rice residues to feed the cattle.

Acknowledgments

We acknowledge funding from the African Union and European Union through the ECOAFRICA Project coordinated by FIFAMANOR (AURG II-1-075-2016).

References

Black, Z., Balta, I., Black, L., Naughton, P. J., Dooley, J. S. G., and Corcionivoschi, N. (2021). The fate of foodborne pathogens in manure treated soil. *Frontiers in Microbiology*, 12 (781357): 1–21.

Brunelle, T., Dumas, P., Dorin, B. and Nadaud, F. (2015). Evaluating the impact of rising fertilizer prices on crop yields. *Agricultural Economics*, 46(March), 653–666.

Chianu, J. N., Chianu, J. N. and Mairura, F. (2012). Mineral fertilizers in the farming systems of sub-Saharan Africa. A review. *Agronomy for Sustainable Development*, 32(2): 545–566.

Faivre-Rampant, O., Geniès, L., Piffanelli, P. and Tharreau, D. (2013). Transmission of rice blast from seeds to adult plants in a non-systemic way. *Plant Pathology*, 62(4): 879–887.

Fanjaniaina, M. L., Stark, F., Ramarovahoaka, N. P., Rakotoharinaivo, J. F., Rafolisy, T., Salgado, P. and Becquer, T. (2022). Nutrient flows and balances in mixed farming systems in Madagascar. *Sustainability*, 14(2) : 1-21.

Gallet, R., Bonnot, F., Milazzo, J., Tertois, C., Adreit, H., Ravigné, V., Tharreau, D. and Fournier, E. (2014). The variety mixture strategy assessed in a G × G experiment with rice and the blast fungus *Magnaporthe oryzae*. *Frontiers in Genetics*, 4(312), 1–11.

Guerber, C. and TeBeest, D. O. (2006). Infection of rice seed grown in Arkansas by Pyricularia grisea and transmission to seedlings in the field. *Plant Disease*, 90(2): 170–176.

Long, D. H., Correll, J. C., Lee, F. N. and TeBeest, D. O. (2001). Rice Blast Epidemics Initiated by Infested Rice Grain on the Soil Surface. *Plant Disease*, 85(6): 612–616.

Manandhar, H. K., Jorgensen, H. J. L., Smedegaard-Petersen, V. and Mathur, S. B. (1998). Seedborne infection of rice by Pyricularia oryzae and its transmission to seedlings. *Plant Disease*, 82(10): 1093–1099.

Rasolofo, I. L. (2017). Impact des innovations agro-écologiques sur les flux de carbone et d'azote des cultures pluviales . Cas des Hautes Terres de Madagascar. In *These*. Université d'Antananarivo. 169p.

Raveloson, H., Ratsimiala Ramonta, I., Tharreau, D. and Sester, M. (2018). Long-term survival of blast pathogen in infected rice residues as major source of primary inoculum in high altitude upland ecology. *Plant Pathology*, 67(3), 610–618.

Sester, M., Raveloson, H., Tharreau, D. and Dusserre, J. (2014). Conservation agriculture cropping system to limit blast disease in upland rainfed rice. *Plant Pathology*, 63(2): 373–381.

Sharma, M. and Reynnells, R. (2016). Importance of soil amendments: survival of bacterial pathogens in manure and compost used as organic fertilizers. *Microbial Spectrum*, 4(4): 159–175.