IN VITRO EVALUATION OF BACTERIAL STRAIN ISOLATES FOR THE BIOLOGICAL CONTROL OF COLLETOTRICHUM SP RESPONSIBLE FOR CASHEW ANTHRACNOSE IN COTE D'IVOIRE

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ABSTRACT

Anthracnose is a fungal disease of cashew (Anacardium occidentale L.) caused by Colletotrichum sp. This disease causes several damages in cashew orchards in Côte d'Ivoire. The objective of this study was to isolate presumptive isolates of soil-borne bacteria and to evaluate their effects on the growth of Colletotrichum sp causing cashew anthracnose in Côte d'Ivoire. From soil samples, bacteria were isolated and their antifungal activities were evaluated in vitro. These activities were then evaluated on the mycelial growth of Colletotrichum sp in the presence of bacteria and two synthetic fungicides (Propiconazole and Prochloraz). Bacterial counts on agar showed an average load of 1,3.106 to 2,5.106 CFU/g of soil. The bacterial isolates and the synthetic fungicide Prochloraze showed an in vitro antagonistic activity of more than 50% against Colletotrichum sp compared to the fungicide Propiconazole. Two strains showed the highest average inhibition rates of 86,6±2,7 and 79,95±5,1% respectively. These two strains once determined can be used in the biological control of cashew anthracnose in Côte d'Ivoire.

Key words : Cashew tree, Bacterial isolates, Colletotrichum sp, Biological control, Côte d'Ivoire.

RESUME

EVALUATION IN VITRO DES ISOLATS DE SOUCHES BACTERIENNES POUR LE CONTROLE BIOLOGIQUE DE COLLETOTRICHUM SP RESPONSABLE DE L'ANTHRACNOSE DE L'ANACARDIER EN COTE D'IVOIRE

L'anthracnose est une maladie fongique de l'anacardier (Anacardium occidentale L.) causée par Colletotrichum sp. Cette maladie cause plusieurs dégâts dans les vergers d'anacardier en Côte d'Ivoire. L'objectif de cette étude a été d'isoler les isolats de bactéries telluriques de la rhizosphère de l'anacardier et d'évaluer leurs effets sur la croissance de Colletotrichum sp responsable de l'anthracnose de l'anacardier en Côte d'Ivoire. A partir d'échantillons de sols, des bactéries ont été isolés puis leurs activités antifongiques ont été évaluées in vitro. Ces activités ont été par la suite évaluées sur la croissance mycélienne de Colletotrichum sp en présence bactéries et deux fongicides

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de synthèse (Propiconazole et Prochloraze). Le dénombrement des bactéries sur gélose a présenté une charge moyenne de 1,3.106 à 2,5.106 UFC /g de sol. Les isolats de bactéries et le fongicide de synthèse Prochloraze ont montré une activité antagoniste in vitro supérieur à 50 % contre Colletotrichum sp comparé au fongicide Propiconazole. Deux souches de bactérie ont présenté les taux moyens d'inhibition les plus élevés, respectivement de 86,6±2,7 et 79,95±5,1%. Ces deux souches une fois déterminées peuvent être utilisées dans la lutte biologique contre l'anthracnose de l'anacardier en Côte d'Ivoire.

Mots clés : Anacardier, Isolats bactériens, Colletotrichum sp, Lutte Biologique, Côte d'Ivoire.

INTRODUCTION

The cashew tree (Anacardium occidentale L.) is a member of the Anacardiaceae family. It is a tree that occupies an important place in several countries of the world (Bezerra *et al.,* 2007). It originates from Brazil and was introduced to West and East Africa in the 18th century by the Portuguese. It is cultivated for its fruit, which is composed of two parts:the apple and the cashew nut, which is an important industrial and export commodity worldwide (Azam-Alli and Judge, 2001). Cashew cultivation was introduced in Côte d'Ivoire in the 1960s as part of a soil conservation and reforestation programme in the northern savannah zone. Gradually, this cashew forest plantation was transformed into a fruit plantation for the marketing of fruit. Indeed, thanks to the growing demand for cashew nuts on the international market, the cashew sector in Côte d'Ivoire has developed rapidly, with an increase in national production of raw nuts. This production increased from 19,000 tonnes in 1990 to more than 635,000 tonnes in 2015. Since 2015 until today, Côte d'Ivoire is the world's leading cashew nut producer with an annual production of 738,000 tonnes in 2018 (FIRCA, 2018). The cashew sector appears today as one of the main drivers of economic and social development in the Northern, Central and Eastern zones of Côte d'Ivoire. It is clearly an interesting source of economic growth with the advantage of having been developed in the poorest regions of the country and having the potential to generate significant rural employment through its cultivation and rural industrialisation (Rico *et al.*, 2016).

However, this sector is confronted with several fungal diseases namely rust, powdery mildew, pestalotiosis and anthracnose which lead to huge post-harvest losses. Anthracnose is a fungal disease caused by the fungus Colletotrichum gloeosporioides (Silué *et al.*, 2018). It is one of the most widespread diseases in all cashew orchards in Côte d'Ivoire. This disease develops on all organs of the plant, namely leaves, flowers and fruits, thus reducing fruit quality (Silué *et al*., 2017). It causes a significant decrease in cashew nut yield in cashew orchards in Côte d'Ivoire and could be the cause of the low yield of cashew trees, i.e. 350 and 524 kg/ha (CNRA, 2011; FIRCA, 2018).

To limit cashew yield losses caused by anthracnose, conventional control methods such as cultural control and the application of synthetic fungicides have been deployed by producers. However, these methods are ineffective and pose many environmental constraints. Indeed, the synthetic fungicides used can contaminate the environment due to their high toxicity, and can be found on fruit products (cashew nuts). These products can also induce long-lasting resistance of the pathogen (Bastard *et al.* 2015).

Because of the problems in controlling anthracnose, research is being conducted to identify alternative methods for protecting cashew against anthracnose that are less dependent on chemicals and more environmentally friendly. Biological control through the use of bacterial biopesticides could be one of the alternative solutions. This control consists of the use of bacteria that have either an inhibitory potential of the pathogen or the ability to increase the defence mechanism of the plant. Among these bacteria, rhizospheric bacteria of the genera Pseudomonas and Bacillus are the most commonly used. They are known for their capacity to produce bioactive metabolites

and their ability to colonise the rhizosphere, plant roots and to control phytopathogenic microorganisms (Pérez-Garcia *et al.*, 2011).

The objective of this study was to isolate presumptive isolates of telluric bacteria and to evaluate their effects on the growth of Colletotrichum sp responsible for cashew anthracnose in Côte d'Ivoire.

MATERIALS AND METHODS

STUDY SITE

This study was conducted in 18 cashews producing regions. These regions were grouped into five major production zones, namely Bondoukou, Bouaké, Korhogo, Odienné and Séguéla. The different soil samples were taken from the rhizosphere of cashew trees in 270 plantations (Figure 1).

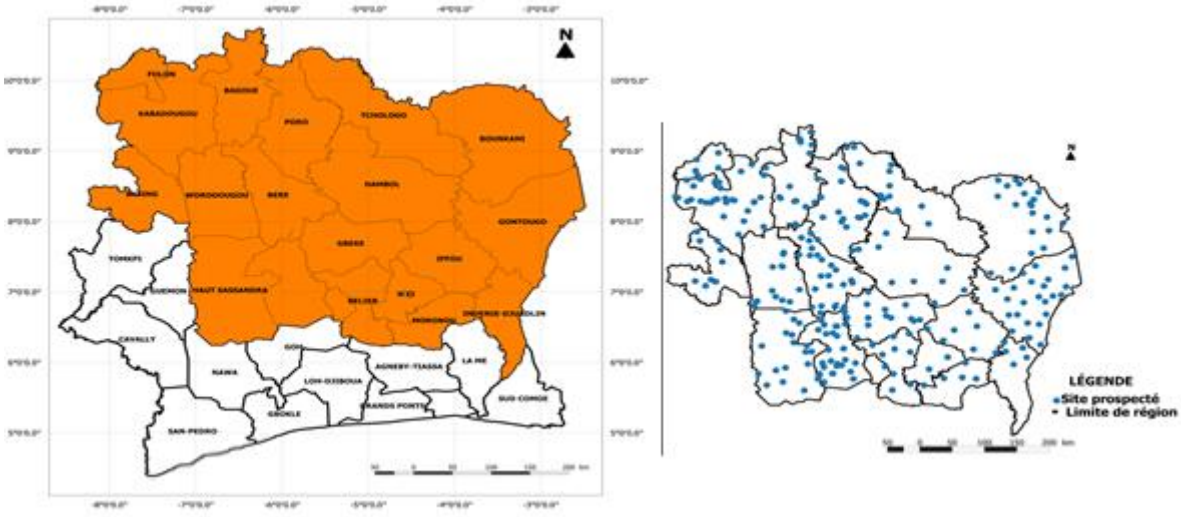

Figure 1 : Soil sampling site.

Site de prélèvement d'échantillons de sol.

MATERIAL

The material used consisted of soil samples from the rhizosphere of cashew trees from the five (5) cashew nut production zones in Côte d'Ivoire, namely Bouaké, Korogho, Seguela, Odienne and Bondoukou. These soil samples were used for the isolation of bacteria. Pathogenic Colletotrichum isolates isolated from cashew organs in Côte d'Ivoire were used for in vitro antagonistic tests. The synthetic fungicides Propiconazole and Prochloraz were used as positive controls for the in vitro confrontation tests to compare their effects with those of the soil-borne bacterial isolates isolated from the rhizosphere of cashew.

METHODS

Soil sampling, isolation and identification of bacteria

Collection of soil samples

Soil sampling was carried out during the period 01 October to 22 December 2019, in 270 cashews plantations at a rate of 15 plantations per cashew producing region. In each plantation, three apparently healthy cashew plants were randomly selected. Then, using a sterile spatula, three (03) 200g samples of soil were taken from the rhizosphere of each plant at a depth of 0 to 30 cm. A total of 810 soil samples were collected and transported to the laboratory for isolation and characterisation of bacteria (Dellal and Halitim, 1992).

Isolation of soil bacteria

The bacteria investigated belonged to the genus Pseudomonas and to the species fluorescens. This bacterium is characterised by the production of fluorescent pigment on King B agar. Isolation was carried out by the suspension-dilution method and by spreading on King B agar (Vidhyasekaran *et al.*, 1997). For this purpose, 1 g of soil sample was taken with a sterile spatula and suspended in 9 ml of sterile distilled water. After stirring the suspension for 10 minutes with a vortex, decimal dilutions from 10-1 to 10-8 were made. Then 0.1 ml of each suspension was spread on Petri dishes each containing 20 ml of King B agar medium. The plates were incubated at 30°C in the dark. After 48 hours of incubation, the bacteria were counted and the bacterial load

per soil sample location was determined by the following formula :

$$N\ (UFC/g) = \frac{\Sigma C}{V\ (n1 + 0{,}1\ n2)\ d}$$

ΣC = Sum of colonies

V = Volume of plating

d = Dilution considered

n1 = Number of plates at the first dilution considered

n2 = Number of plates at the second dilution considered

N = Bacterial load

CFU /g = Colony forming unit per gram of soil

After enumeration, bacterial isolates that produced fluorescent pigments were re-cultured on King B agar until pure colonies were obtained. The purified colonies were stored in 50% glycerol in cryotubes at -20°C and in tubes containing King B agar sloped at 4°C for determination of their belonging to the species P. fluorescens (Bossis *et al.*, 2000).

Characterisation of presumptive isolates of Pseudomonas fluorescens

The characterisation of P. fluorescens isolates was carried out by different methods based on morphological and physiological criteria. Indeed, after purification of the bacteria on King B medium, the colonies are observed with the naked eye. The macroscopic study was carried out taking into account the size, shape and brightness of the colonies (small, round, shiny and invasive, superimposed etc) (Vidhyasekaran *et al.*, 1997). The physiological study concerns the production of yellow-green pigment on King B agar at 4°C. The microscopic study was based on the Gram stain, which consisted of making a bacterial smear on a slide and fixing it with heat. The smear was then examined under a microscope with a 40X objective (Bossis *et al.*, 2000). The P. fluorescens were found to be pink coloured bacilli, i.e. Gram-negative bacilli. (Bossis *et al.*, 2000).

Testing for antagonistic activity in vitro

The test was carried out to verify the existence of a possible inhibitory action of Pseudomonas fluorescens towards pathogenic fungal isolates of Colletotrichum sp.

Preparation of the media and culture of the two antagonists

First, Pseudomonas fluorescens were purified 48 hours before the cultures were performed. A YPGA (Yeast extract Peptone Glucose Agar) medium was prepared for the cultures. Thus, these purified antagonistic bacteria were inoculated onto the previously prepared YPGA agar in the form of a straight streak that divides the Petri dish into two equal parts. Subsequently, two mycelial discs of the pathogenic fungi were placed on either side of the streak at 1 cm from the edge of the Petri dish (Korsten and Jager, 1995). The synthetic fungicides Propiconazole and Prochloraz were used to compare their effects with those of the bacteria. For this purpose, a 1000 ppm solution of each fungicide was prepared and incorporated into a supercooled YPGA medium. The medium containing the synthetic fungicides was dispensed into the Petri dishes at a rate of 20 ml per dish. Once the medium had solidified, a mycelial fragment of the pathogenic fungus was deposited in the centre of the Petri dish (Silué *et al.*, 2018). Control dishescontaining no control agent (fungicide and bacteria) were made by seeding a mycelial fragment of the pathogenic fungus in the centre of the Petri dish.

Evaluation of the cultures

Cultures are evaluated by direct measurement of the growth diameter of the pathogenic fungus (Colletotrichum sp) using a graduated ruler in each of the Petri dishes constituting the treatments. This measurement is taken every day until the pathogen fills the control dishes. At the end of these measurements, the rate of inhibition of mycelial growth of the pathogen is calculated according to the formula of Korsten and Jager. (1995).

Inhibition rate = [(R-r) /R] *100

r : The radial growth of the fungus against the antagonist (bacteria)

R: The radial growth of the fungus without the antagonist

STATISTICALANALYSIS

The Statistical Analysis System software, version 7.1 was used for all statistical analyses.

For the microorganism isolations, analyses of variance (ANOVA) were performed on the mean number of bacterial colonies counted on the culture medium and on the mean susceptibility score of Colletotrichum sp isolates in the presence of the bacteria. Comparisons between means were made using the Newman-Keuls test at the 5% threshold.

RESULTS

ISOLATION AND ENUMERATION

Isolation and enumeration of soil-borne bacteria by the soil suspension-dilution method followed by plating on King B agar showed an average load of bacteria ranging from 1,3.106 to 2,5.106 CFU/g of soil respectively for the Korhogo and Bouaké production zones. The highest bacterial loads were observed in the Bouaké and Séguéla production zones with values of respectives de 2,1. 106 and 1,9.106 UFC /g of soil. However, no significant differences in loadings were observed in these two zones above. The production zones of Korhogo and Odienné recorded the lowest average bacteria loads (Figure 2).

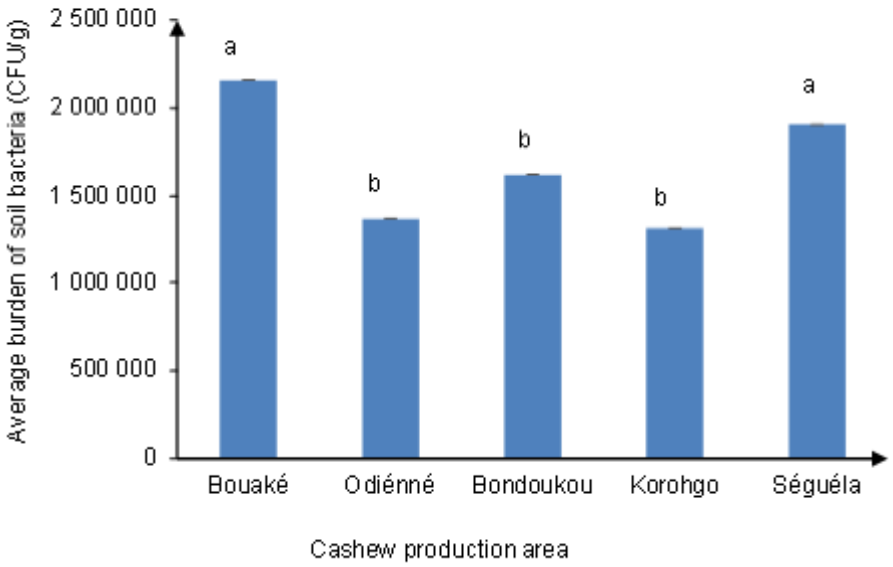

Figure 2 : Average load of soil-borne bacteria in CFU/g according to cashew production areas.

Charge moyenne en bactéries telluriques en UFC/ g selon les zones de production de la noix de cajou.

Phenotypic characterisation of Pseudomonas fluorescens

Characterisation of presumptive Pseudomonas fluorescens isolates using macroscopic characters after purification on King B agar showed the appearance of distinct colonies visible to the naked eye with the specific morphological criteria of the genus Pseudomonas fluorescens. On King B agar, Pseudomonas fluorescens isolates are characterised by rapid growth resulting in small, creamy, overlapping, circular, smooth, regular-looking, invasive colonies with yellow-green pigment production in the agar (Figure 3A). Microscopic observation reveals straight or slightly curved bacilli with rounded tips. The cells are isolated or in pairs. They are asporulated. After Gram staining, Pseudomonas fluorescens isolates appear as rods with a pink colour, characteristic of Gram-negative bacilli (Figure 3B).

Indeed, bacteria belonging to the genus Pseudomonas are small rods with a polar ciliate, exhibiting a Gram-negative stain. Pseudomonas fluorescens was able to optimise pigment production at 4°C over 30 days of incubation in the refrigerator (Figure 3C).

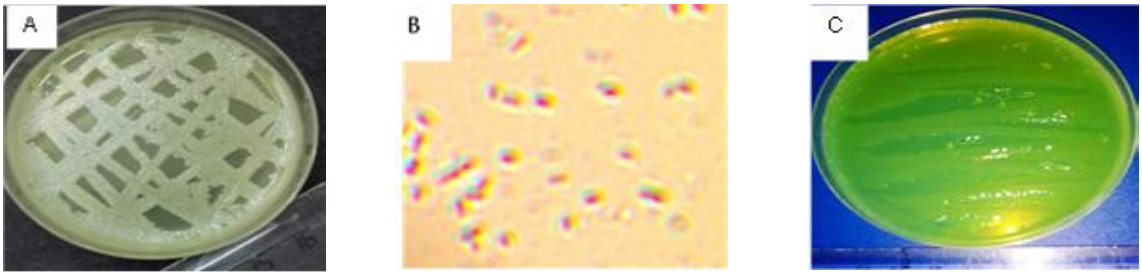

Figure 3 : Macroscopic and microscopic aspects of Pseudomonas fluorescens (A-Macroscopic appearance; B-Microscopic appearance; C-Optimisation of pigment production at 4°C).

Aspects macroscopiques et microscopiques des Pseudomonas fluorescens (A-Aspect macroscopique ; B-Aspect microscopique ; C-Optimisation de la production de pigment à 4°C).

In vitro antagonism activity

The in vitro antagonistic activity test performed according to the method of Korsten and Jager. (1995) showed a better inhibitory activity of the tested bacterial isolates on the mycelial growth of the pathogenic fungus Colletotrichum sp (Figure 5). These inhibition rates varied depending on the Pseudomonas fluorescens isolate tested. In general, no significant difference was observed between the antagonistic effect of the bacterial isolates and the synthetic fungicide prochloraz (p d» 0,05) (Figure 4). The maximum inhibition values were 86,6±2,7, 79,95±5,1 and 78,7±1% for Pseudomonas fluorescens isolates KOROO9094, SEPIBD1081 and GBO1 respectively. The fungicide prochloraz gave an average inhibition of 68,47±4,5% in contrast to propiconazole which showed an inhibition rate of 47,1±1,5% (Figure 4).

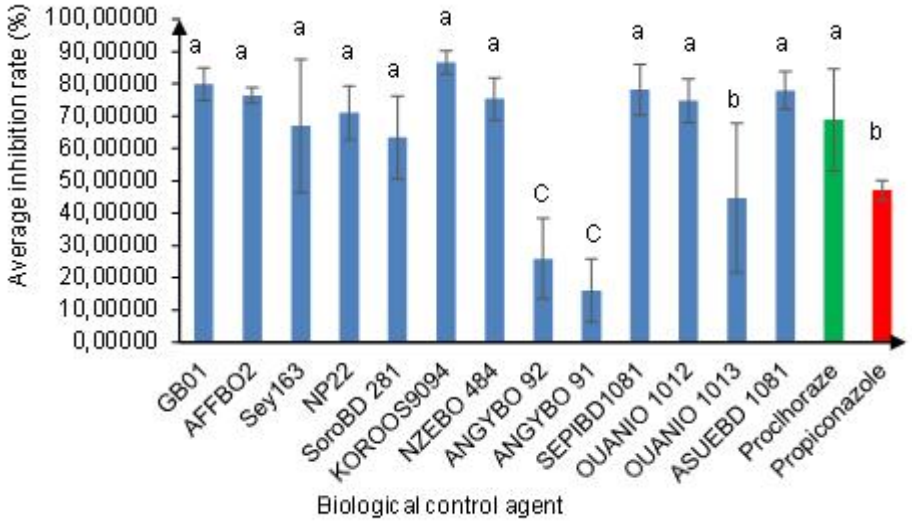

Figure 4 : Antagonistic effect of bacterial isolates and synthetic fungicides on mycelial growth of Colletotrichum sp isolates. (A- Blue band : Inhibition rate of bacteria, B-Green band : Inhibition rate of the fungicide prochloraz and C-Inhibition rate of the fungicide propiconazole).

Effet antagoniste des isolats bactériens et des fongicides de synthèses sur la croissance mycélienne des isolats de Colletotrichum sp. (A- Bande bleu : Taux d'inhibition des bactéries, B-Bande verte : Taux d'inhibition du fongicide prochloraze et C-Taux d'inhibition du fongicide propiconazole).

Mean values followed by the same alphabetical letter are not statistically different ($p \leq 0.05$) (Newman-Keuls).

Les valeurs moyennes suivies d'une même lettre alphabétique ne sont pas statistiquement différentes ($p \leq 0,05$) (Newman-Keuls).

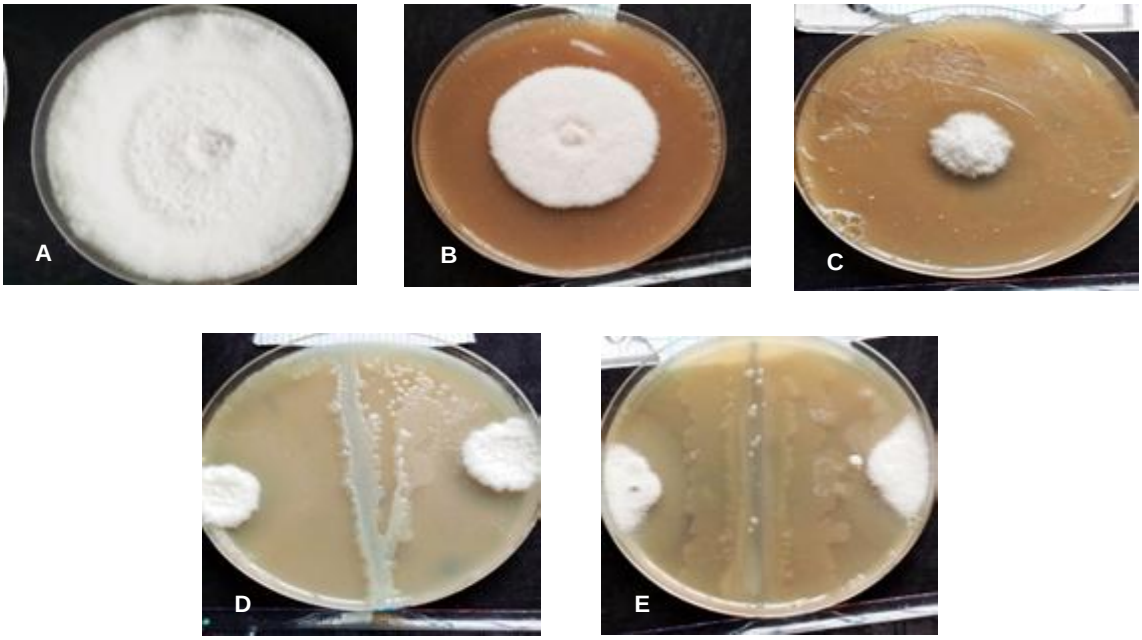

Figure 5 : Inhibition of mycelial growth of Colletotrichum in the presence of the control agents : (A-Temoin; B-Propiconazole; C- Prochloraz and D-Bacterial isolate KORO9094 and E-Bacterial isolate ASSUEBD1081).

Inhibition de la croissance mycélienne de Colletotrichum en présence des agents de lutte : (A-Témoin ; B-Propiconazole ; C- Prochloraze et D-Isolat bactérien KORO9094 et E-Isolat bactérien ASSUEBD1081).

DISCUSSION

Isolation and enumeration of telluric bacteria on King B agar showed a difference in the average bacterial load in the cashew rhizosphere of the five cashew production areas surveyed. These loads ranged from 1,3.106 to 2,5.106 CFU /g soil. However, the highest loads were observed in the Bouaké and Séguéla areas. This could be explained by the fact that these soils are rich in organic matter and other nutrients such as nitrogen and many others. Indeed, the abundance and microbial activity of a soil can be influenced by several factors, including environmental factors. Also, the organic matter that constitutes a source of carbon for the microbial populations of a soil can greatly influence the composition of this microflora (Bertrand *et al.*, 2000). Furthermore, the survival of bacterial isolates in a soil can vary depending on the proximity of the roots of each plant (Weyens *et al.*, 2010). Strains must therefore have the required characteristics to adapt to the various environmental constraints (Cacciari *et al.*, 2003).

Characterisation of Pseudomonas fluorescens isolates based on the presence of criteria such as macroscopic colony appearance (round), bacillary form with Gram-negative staining, pigment production on King B agar, growth at 4°C with optimisation of pigment production and positive catalase reaction revealed their membership to the genus P. fluorescens. Similar results were observed with the work of Benzina *et al.* (2016) who after identifying P. fluorescens isolates from the rhizosphere of tomato in Algeria obtained 15 genera of P. fluorescens.

The ability of P. fluorescens isolates isolated from cashew rhizosphere to inhibit the growth of Colletotrichum sp (causal agent of cashew anthracnose) was demonstrated by in vitro antagonism tests. The results showed that P. fluorescens isolates inhibited the mycelial growth of fungal isolates of Colletotrichum sp. These in vitro tests showed inhibition rates above 50% for most of the bacterial isolates tested. The highest inhibition rates were 91,87±1,6 and 88,33±3.3% for isolates KOROO9094 and SEPIBD1081 respectively. This would be due to the fact that P. fluorescens isolates would

produce antifungal substances that would inhibit the growth of pathogenic fungi. Indeed, P. fluorescens strains possess broad-spectrum antagonistic activity against phytopathogens. These activities include direct antibiosis, spatial or nutritional competition and siderophore production (Showkat *et al.*, 2012). Thus, in the stationary phase of growth, P. fluorescens produce several metabolites with high antifungal properties such as phenazines, pyrrolnitrin, pyoluteorin and 2,4-diacetylphloroglucinol (2,4-DAPG), which are thought to inhibit the growth of pathogenic fungi in vitro (haass and Defago, 2005). These metabolites act by altering the germination, growth and/or sporulation of the pathogen, distorting the hyphae of the pathogen, modifying the appearance of the colonies and producing specific forms such as pseudo-parenchyma (Campbell, 1989). These findings may also result from the secretion of lytic enzymes such as β-1,3-glucanase and chitinase, which have parasitism as their mechanism of action through the degradation of the walls of the fungal pathogen (Ban Koffi *et al.*, 2015). Work by Aké and colleagues in 2019 showed that the CI bacterium P. fluorescens isolated from tomato can inhibit the growth of post-harvest cashew spoilage fungi in vitro in Côte d'Ivoire. These results showed inhibition rates of 80 ± 1,76 and 82,5 ± 0% respectively on the mycelial growth of Fusarium sp and Absidia sp. These results are in agreement with the work of Koffi (2018) who revealed that Pseudomonas fluorescens CI inhibited in vitro the growth of pineapple fruit moulds of variety MD-2 in Côte d'Ivoire such as Geotrichum. candidum, Rhizopus oryzae and Fusarium sp. These activities include, among others, direct antibiosis and siderophore production (Koffi, 2016).

CONCLUSION

This study aimed at isolating isolates of soil-borne bacteria and evaluating their effects on the growth of Colletotrichum sp responsible for cashew anthracnose in Côte d'Ivoire, allowed the identification of presumptive isolates of Pseudomonas fluorescens for the control of anthracnose. The bacterial isolates KORO9094, GB01, ASSUEBD1081 and SEPIBD108 considerably reduced the in vitro mycelial growth of Colletotrichum sp. In view of the various results obtained in vitro, these bacterial strains should be tested in the field to evaluate their effectiveness at this level. They could then be used in the biological control of cashew anthracnose in Côte d'Ivoire.

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REFERENCES

Ake M. D. F., Tehua A. A., Koffi Y. F., Sanogo Y. M. et Alloue-Boraud W. A M. 2019. Phenotypical identification of moisture associated with cashew nuts (anacardium occidentale l.) in Côte d'Ivoire and control of Pseudomonas Fluorescens CI effect. Int. J. of Inov. and App. Res. 9 : 1-8.

Azam-Alli S. H et Judge E.C. 2001. Small scale cashew nut processing. ITDG. FAO, Rome, 70 p.

Bezerra M. A. 2007. Physiology of cashew plants grown under adverse conditions. Braz. J. Plant Physiol. 19 : 449-461.

Ban Koffi L, Alloue-Boraud WA, Assamoi J, Koussemon M et Ongenas M. (2015). Study of protective effect of Pseudomonas fluorescens F19 against microorganisms responsible of tomato fruits (Lycopersicon esculentum Mill) spoilage in Côte d'Ivoire. International Journal of Applied Science and Research. 9 : 9-16.

Belzile L et Grondines H. (2015). Les essais de fongicides foliaires pour lutter contre la fusariose dans les céréales à paille. Perspectives en économie de l'agroenvironnement. 1 : 3-4.

Benzina, F., Sahir-Halouane, F. et Hamed K. ((2016). Algerian isolates of fluorescent Pseudomonas spp. as potential biological control against wilt pathogen (Verticillium dahliae). Plant Omics journal. 9 : 48-60.

Bezerra M. A., Lacerda C. F., Filho E. G., Abreu C. E. B. et Prisco J. T. (2007). Physiology of

cashew plants grown under adverse conditions. J. Plant Physiol. 19 : 449-461.

Bertrand H., Plassard C., Pinoche X t., Touraine B., Normand P et Cleyet-Marel J.C. (2000). Stimulation of the ionic transport system in Brassica napus by a plant growthpromoting rhizobium (Achromobacter sp.). Can. J. Microbiol. 46 : 229-36.

Bossis E., Lemanceau P., Latour X. et Gardan L. (2000). The taxonomy of Pseudomonas fluorescens and Pseudomonas putida. current status and need for revision Agronomie. 20 : 51-63.

Cacciari I., Dimattia E., Quatrini P., Moscatelli M.C., S. Lippi Grego, D et De Paolis M.R. 2003. Réponses adaptatives des isolats de Rhizobium aux stress, p. 231-248. In M.

Campbell R. 1989. Biological Control of Microbial Plant Pathogens. 1st Edn., Cambridge University Press, Cambridge, ISBN : 0 521 34900 1.

CNRA (Centre National de Recherche Agronomique). 2011. Amélioration variétale de l'anacardier en Côte d'Ivoire. Rapport final. Décembre 2011, 16 p.

Dellal A et Halitim A. 1992. Activités microbiologiques en conditions salines : cas de quelques sols salés de la région de Relizane (Algérie). Cahiers Agric. 1 : 335-340

Haas D et Defago G. 2005. Biological control of soil-borne pathogens by fluorescent pseudomonads. Nature Reviews Microbiology. 3 : 307-319.

Koffi Y F (2018). Identification phénotypique et moléculaire des germes d'altération de l'ananas (Ananas comosus L.) et test de conservation à l'aide de biopesticides bactériens. Thèse unique de Doctorat, Université Nangui Abrogoua (Côte d'Ivoire), 162 p.

Koffi Y F., Ban Koffi L., Alloue-Boraud W A., Fabrice A A., Dje MK., et Ongenas M. 2016. Highlighting Bacillus subtilis GA1 antifungi potentialities for pineapple (Ananas comosus) conservation in Côte d'Ivoire. International Journal of Agricultural Research. 9 : 100-108.

Korsten L., et Jager E. S. D. 1995. Mode of action of Bacillus subtilis for control of avocado postharvest pathogens. South African Avocado Grower's Association Yearbook. 18 : 124-130.

Pérez-García A., Romero D. et De Vicente A. 2011. Plant protection and growth stimulation by microorganisms: biotechnological applications of Bacilli in agriculture. Current Opinion in Biotechnology. 22 : 187-193.

Rico R., Bulló M et Salas-Salvadó J. 2016. Nutritional composition of raw fresh cashew (Anacardium occidentale L.) kernels from different origin. Food Science and Nutrition. 4 : 29-338.

Showkat S., Murtaza I., Laila O., et Ali A. 2012. Biological Control of Fusarium Oxysporum and Aspergillus sp. By Pseudomonas Fluorescens Isolated From Wheat Rhizosphere Soil Of Kashmir. IOSR Journal of Pharmacy and Biological Sciences. 1 : 24-32.

Silué N., Abo K., Johnson F., Camara B., Koné M et Koné D. 2018. Evaluation in vitro et in vivo de trois fongicides de synthèse et d'un fongicide biologique sur la croissance et la sévérité de colletotrichum gloeosporioides et de pestalotia heterocornis, champignons responsables de maladies foliaires de l'anacardier (Anacardium occidentale L.) en Côte d'Ivoire. Agronomie Africaine. 30 : 107 - 122.

Silue N., Soro S., Koné M., et Daouda K. 2017. Parasitical fungi in Cashew (Anacardium occidentale L.) Orchard of Côte d'Ivoire. Plant Pathol. J. 16 : 82-88.

Weyens N., Monchy S., Vangronsveld J., Taghavi S., et Vander L. D. 2010. Plant Microbe Partnerships, Handbook of hydrocarbon and lipid microbiology, Springer-Verlag, Berlin Heidelberg. In K.N. Timmis (ed.), p. 254-2564.