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EVALUATION OF THE EFFICACY OF TWO AROMATIC PLANT EXTRACTS AGAINST PHYTOPATHOGENIC TOMATO FUNGI IN BURKINA FASO

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ABSTRACT

Tomato production is faced with many fungal diseases including alternariosis and anthracnose caused by *Alternaria* solani and *Colletotrichum* sp. respectively, which cause major losses in production. Identify effective biofungicidal formulations based on extracts of *Eucalyptus camaldulensis* Dehnh. and *Cymbopogon citratus* (DC.) Stapf on the radial growth of *Alternaria solani* and *Colletotrichum* sp. The efficacy of aqueous or ethanolic extracts of *Eucalyptus camaldulensis* or *Cymbopogon citratus* as well as combinations of aqueous extracts (4/1; 3/1 and 2/1) on the radial growth and sporulation of *Alternaria solani* and *Colletotrichum* sp. was studied in vitro. Then incubated at 25°C under an alternating 12/12 cycle of near ultra-violet light and darkness. The inhibition rate was measured on days 2, 4 and 7 after incubation (DAI) and for sporulation on day 14. All the extracts of the two plants, aqueous or ethanolic, showed the maximum inhibition rate (100%) on the radial growth of *Alternaria solani* and *Colletotrichum* sp. at concentrations of 20% and 30%. On the other hand, the combination of aqueous extracts was only effective at the 30% dose with inhibition rates of 49% on *Alternaria solani* and 21% on *Colletotrichum* sp. At doses of 20% and 30%, aqueous or ethanolic extracts of *Eucalyptus camaldulensis* or *Cymbopogon citratus* completely inhibit the mycelial growth of *Alternaria solani* and *Colletotrichum* sp.

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INTRODUCTION

Tomatoes are the 2nd most important vegetable crop in Burkina Faso after bulb onions, with production of more than 21 tonnes per hectare in 2018 (DGESS/MAAH, 2018). This crop faces many problems due to biotic and abiotic constraints. Bio-aggressors such as bacteria, viruses and pests include *Alternaria solani* and *Colletotrichum* sp. which cause alternariosis and anthracnose respectively. These fungi cause major losses to farmers both in terms of yield and production quality. To combat these pests, farmers opt for synthetic chemical pesticides. However, despite their effectiveness, the consequences for humans and the environment of this conventional agricultural practice

are highlighted: intoxication of producers and consumers, environmental pollution and the appearance of new strains of bio-aggressors that are more resistant to pesticides (Gnankine *et al.*, 2013).

One of the alternatives being promoted to synthetic chemical pesticides is the use of biopesticides, including those of plant origin, which have the advantage of being derived from available and renewable resources. Despite the antifungal efficacy of various plants, including *Cymbopogon citratus* and *Eucalyptus camaldulensis* (Bonzi, 2007) a standardised, marketable formulation is not available. In order to produce a more stable, cost-effective and environmentally friendly biofungicide formulation, efficacy tests of *Cymbopogon citratus* and *Eucalyptus camaldulensis* extracts compared with their mixture need to be carried out for validated scientific evidence. The aim of this study was to find the most effective combination of extracts from the two plants against *Alternaria solani* and *Colletotrichum* sp., two fungal pathogens of tomato.

MATERIALS AND METHODS

Experimental Site

The Laboratory of Analytical, Environmental and Organic Chemistry (LCAEBiO) of the Training and Research Unit in Exact and Applied Sciences (UFR/SEA) of the Joseph KI-ZERBO University located in Ouagadougou served as the setting for the preparation of the extracts.

The Phytopathology/Mycology Laboratory of the Rice and Rice-growing Programme of INERA Farako-Bâ located 10 km from Bobo-Dioulasso on the Bobo-Banfora road, with GPS coordinates of longitude 4°20 West, latitude 11°06 North and an altitude of 40 m, was used for mycological manipulations and antifungal tests. The climate at the laboratory site is South Sudanian, with annual rainfall varying between 950 mm and 1100 mm.

Plant Material

Tomato plants from three vegetable-growing sites in Bobo Dioulasso, including one in the village of Kodeni and two in the village of Yenneta, and *Eucalyptus camaldulensis* and *Cymbopogon citratus* leaves collected in Ouagadougou were used.

Collection and Isolation of Fungal Strains

Samples of tomato leaves naturally infected by alternariosis or anthracnose were collected in Bobo Dioulasso from three collection sites, one in Kodeni and two in Yenneta. The leaves were placed in sterile paper envelopes and transported to the laboratory. Each leaf, washed in clean drinking water, was cut into fragments of at least 1 cm and then placed in Petri dishes containing sterilized filter paper moistened with sterile distilled water (Bouakaz and Oussaid, 2013). After three days of incubation in a humid chamber at 25°C room temperature with alternating 12 hours of near-Ultra-Violet (UV) light and 12 hours of darkness, the fragments were examined under a binocular magnifying glass to detect the presence or absence of conidia of *Alternaria solani* and *Colletotrichum* sp (Mathur and Kongsdal, 2003). Fragments with conidia were used for the isolation and culture of *A. solani* and *Colletotrichum* sp. Conidia were transferred to the culture medium under a magnifying glass using a drawn and flame-sterilized glass capillary (INRA, 2014), obtaining a pure culture.

Synthetic Chemical Fungicide

The synthetic chemical fungicide used as a reference control is Coga 80 WP; Mancozeb (800g/kg), broad-spectrum for vegetable, fruit, food and flower crops. It is in wettable powder form.

Preparation of Aqueous and Ethanolic Extracts

Treatment of the leaves

The leaves of *Eucalyptus camaldulensis* and *Cymbopogon citratus* (lemongrass) were picked and dried in the shade under ventilation in order to avoid any further chemical transformation that might reduce the content of active molecules. After drying, the leaves were ground into a fine powder. This powder was used to prepare the extracts.

Extraction process

The principle of extraction is based on the property of solvents to be able to solubilize some organic molecules contained in the plant material due to the polarity of the solvent. Two types of extracts were prepared for each plant: aqueous and ethanolic.

For the aqueous extraction, a mass of 200 g of leaf powder from each sample was extracted with 2000 mL of distilled water (1:10 ratio) by maceration and stirring for 24 hours. The mixture was then filtered through a linen cloth and then a second time through cotton wool before being centrifuged. The resulting aqueous extract was frozen at -20°C and dried using a LABCONCO FreeZone 2.5 Plus freeze-dryer to provide the dry aqueous extract. These aqueous extracts were subjected to antifungal tests, as were their combinations in *E. camaldulensis* - *C. citratus* weight ratios (2:1), (3:1) and (4:1), at concentrations of 300 mg/mL (30%), 200

mg/mL (20%) and 100 mg/mL (10%) respectively.

The ethanol extracts were obtained by maceration of the powders with 70% ethanol, with stirring in an Erlenmeyer flask for 24 h in the same 1:10 (m/v) ratio. After separation of the solid residue (pomace) from the liquid phase (macerate), the ethanolic phase was concentrated using a rotavapor (BUCHI Rotavapor R-100; bath temperature 40°C, pressure 175 mbar), then the concentrate recovered with a minimum of water, frozen at -20°C and finally dried using a freeze-dryer (LABCONCO FreeZone 2.5 Plus) to form the ethanolic extract. Ethanol extracts were tested at concentrations of 30%, 20% and 10%.

Efficiency Test *in vitro* Extracts

The antifungal test consisted in evaluating the inhibitory action of plant extracts on the growth radial of the mycelium and on the sporulation of fungi. The chemical fungicide Coga Mancozeb has was used as a reference control. Each fungal strain was tested using the method of diffusion of the extracts in the culture medium. PDA (Potato Dextrose Agar) medium was prepared with each extract (in triplicate) according to three increasing concentrations respectively 10%, 20% and 30% (Table 1), then sterilized at 120°C for 10 min. After cooling, the culture medium was distributed in Petri dishes. For each fungus, mycelial explants of 5 mm in diameter and seven days old, are each placed in the center of a Petri dish containing the medium of culture to be tested. Sealed and inoculated Petri dishes were incubated at room temperature. 22-25°C under an alternating cycle of near ultraviolet light and darkness (12/12) for 7 days. The diameter (in mm) of the colonies was measured in each dish at 7thday after incubation (JAI) then sporulation was checked on 14thI HAVE. The data collected was processed using the formula used by Bonzi (2007).

$$\text{Inhib (\%)} = \frac{\text{DMCt} - \text{Ctt}}{\text{DMCt}} \times 100$$

Inhib (%) = percentage inhibition; DMCt = mean diameter of the colonies of the negative control; Ctt = mean colony diameter of each treatment.

Table 1. Summary of the different biofungicide treatments according to concentrations.

Codes	Treatment according to the biofungicide formulations used	Concentrations
Te:	Water control	
Tco:	Fungicide control	
Tca1 :	*Cymbopogon citratus* aqueous	10 %
Tca2 :	*Cymbopogon citratus* aaqueous	20 %
Tca3 :	*Cymbopogon citratus* aqueous	30 %
Tea1 :	*Eucalyptus camaldulensis* aqueous	10 %
Tea2 :	*Eucalyptus camaldulensis* aqueous	20 %
Tea3 :	*Eucalyptus camaldulensis* aqueux	30 %
Tce1 :	*Cymbopogon citratus* ethanolic	10 %
Tce2 :	*Cymbopogon citratus* ethanolic	20 %
Tce3 :	*Cymbopogon citratus* ethanolic	30 %
Tee1 :	*Eucalyptus camaldulensis* ethanolic	10 %
Tee2 :	*Eucalyptus camaldulensis* ethanolic	20 %
Tee3 :	*Eucalyptus camaldulensis* ethanolic	30 %

Data Analysis and Presentation of Results

Variance analyzes were performed with XLSTAT-pro7.5.2 (2016) software. The test of Student-Newman-Keuls (SNK) was used to compare mean diameters of growth radial of each fungus as well as the percentages of inhibition during the different periods evaluation according to the multiple comparison test. As for the calculation of yield (r), the yield means of extraction was calculated according to the formula of r used by Ouattara *et al.* (2011):

$$r = \frac{M}{m} \times 100$$

RESULTS

Extraction Yield

Aqueous extracts

The table 2 reports the yields of dry extracts obtained from the raw mass of the powders of *E. camaldulensis* and *C. citratus* leaf samples.

Ethanolic extracts

The different yields of the extracts obtained from the raw mass of leaf powder are contained in the table 3.

Tables 2 and 3 indicate that for each plant, the yield of

the aqueous extract is higher than that of the ethanolic extract; the species *Eucalyptus camaldulensis* gave the highest extraction yields, ie 25.24% for the aqueous extract and 18.45% for the ethanol extract.

Table 2. Yield of aqueous extracts.

Powdered extract	Mass of plant powder (g)	Water distilled volume (mL)	Mass of dry extracts after freeze-drying (g)	Yield (%)
E. camaldulensis	200.40	2000	50.60	25.24
C. citratus	200.20	2000	30.58	15.27

Table 3. Mass of ethanol extracts.

Powdered extract	Mass of plant powder (g)	Ethanol volume (mL)	Mass of dry extract (g)	Yield (%)
E. camaldulensis	100	1000	18.45	18.45
C. citratus	100	1000	8.50	8.50

Antifungal Efficacy of Extracts and their Combinations on the Radial Growth of *A. solani*

The results of the analysis of variance at the three observation dates (2 JAI, 4 JAI, 7 JAI) indicate very highly significant differences between the different treatments (Pr < 0.0001). As shown in Table 4, the best results were obtained with the application of 20% and 30% of the aqueous and ethanolic extracts of *C. citratus, E. camaldulensis* with inhibition percentages of 100% for Tca2, Tca3, Tea2, Tea3, Tce2, Tce3, Tee2 and Tee3. Low inhibition percentages were observed at the 10% concentration of aqueous extracts of *C. citratus, E. camaldulensis* and the combination of aqueous extracts with respective inhibition percentages of between 23% and 45% (Tca1) ,45% and 64% (Tea1) ,49% and 52% (Tec1) which, however, differed significantly from the water control (Te = 0%). In addition, the 10% concentration of ethanolic extract of *Eucalyptus camaldulensis* (Tee1) gave a high inhibition percentage of between 90% and 100%.

Table 4. Inhibitory effect of different treatments of *Cymbopogon citratus* and *Eucalyptus camaldulensis* on mycelial growth of *Alternaria solani* according to different concentrations and observation dates.

Treatments	Avg 2 JAI in %	Avg 4 JAI in %	Avg 7 JAI in %
Tca1	44.043^{c}	38.573^{f}	23.990^{f}
Tca2	100.000^{a}	100.000^{a}	100.000^{a}
Tca3	100.000^{a}	100.000^{a}	100.000^{a}
Tce1	62.203^{b}	70.677^{c}	77.617^{c}
Tce2	100.000^{a}	100.000^{a}	100.000^{a}
Tce3	100.000^{a}	100.000^{a}	100.000^{a}
Tco	100.000^{a}	100.000^{a}	100.000^{a}
Te	0.000^{d}	0.000^{g}	0.000^{g}
Tea1	45.743^{c}	60.077^{d}	63.810^{d}
Tea2	84.397^{a}	67.287^{c}	67.570^{d}
Tea3	100.000^{a}	100.000^{a}	100.000^{a}
Tec1	51.047^{c}	52.403^{e}	49.410^{e}
Tec2	100.000^{a}	75.817^{b}	74.517^{c}
Tec3	100.000^{a}	79.683^{b}	85.877^{b}
Tee1	100.000^{a}	100.000^{a}	90.900^{ab}
Tee2	100.000^{a}	100.000^{a}	100.000^{a}
Tee3	100.000^{a}	100.000^{a}	100.000^{a}
Fisher's F	60.691	272.201	221.425
Pr > F	< 0.0001	< 0.0001	< 0.0001
Meaning	****	****	****

Effectiveness of Combinations of Aqueous Extracts of *Cymbopogon citratus* and of *Eucalyptus camaldulensis* on the Radial Growth of *A. solani*

The application of the combination of aqueous extracts at 2 DAI, 4 DAI, 7 DAI gives very appreciable with the three concentrations of 10%, 20% and 30%. In fact, the average inhibition rate is greater than 50%. The best rate is obtained with the combination of aqueous extracts at 30% close to those of the synthetic fungicide control (Tco = 100%). It should be noted that the inhibition rate of the radial growth of *Alternaria solani* increases with the content of the Eucalyptus *camaldulensis* in the combination of the aqueous extracts and with the concentration (Figure 1).

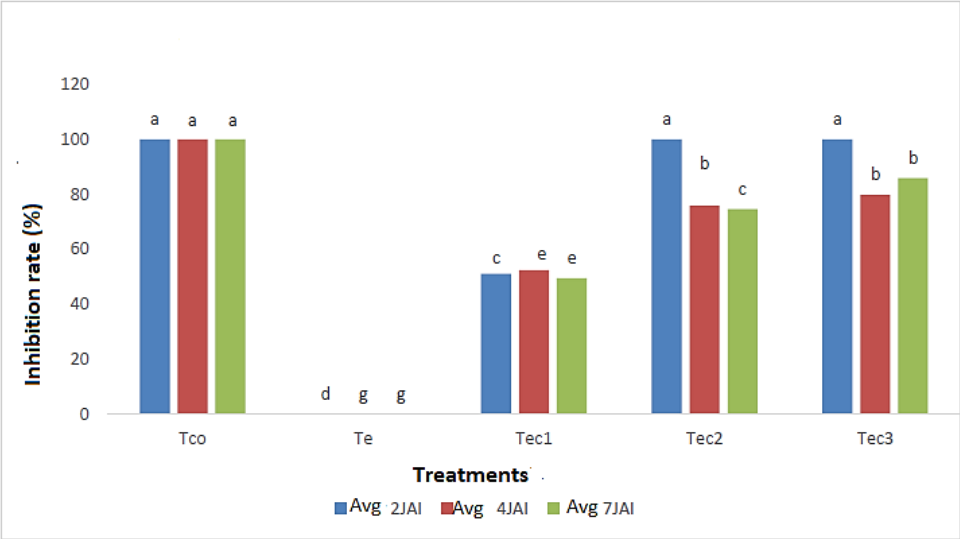

Figure 1. Inhibitory action of the combination of aqueous extracts of *Cymbopogon citratus* and *Eucalyptus camaldulensis* on the mycelial growth of *Alternaria solani* according to different concentrations**.** NB: Histograms assigned the same alphabetical letter do not differ significantly at the 5% threshold (Student-Newman-Keuls test).

Efficacy of Extracts on Mycelial Growth of *Colletotrichum* sp.

The results of the analysis of variance of the efficacy of the different extracts on the mycelial growth of *Colletotrichum* sp. during the three evaluation dates (2 JAI, 4 JAI, 7 JAI) showed very highly significant differences between the different treatments (Pr < 0.0001). The best results were obtained with the application of 20% and 30% of the aqueous and ethanolic extracts of *C. citratus* and *E. camaldulensis*, with inhibition percentages of 100% for Tca2, Tca3 and Tea3 (Table 5). The combination of aqueous extracts of Tec 2 and Tec 3 had an inhibition percentage of 46% and 100% respectively.

The lowest inhibition percentages were observed with the 10% concentration of the aqueous extracts of *C. citratus* and *E. camaldulensis* and were respectively: 34% and 80% (Tca 1), 36% and 40% (Tea 1), 35% and 41% (Tec 1), 20% and 28% (Tec 1). However, these rates differed significantly from the water control (Te = 0%). The ethanolic extract of *Eucalyptus camaldulensis* at 10% gave the best inhibition percentage (100% for Tee 1). In short, ethanolic extracts were more effective than aqueous extracts. A comparison of the aqueous extracts showed that the aqueous extracts of *C. citratus* were more effective than those of *E. camaldulensis*. On the other hand, a comparison of the ethanolic extracts showed that the ethanolic extract of *E. camaldulensis* was more effective than that of *C. citratus*. Finally, it was found that the efficacy of the combination of the different extracts (aqueous and ethanolic) increased with concentration, but that the trend decreased with the observation periods (2 JAI, 4 JAI and 7 JAI).

Percentage inhibition of the combination of aqueous extracts of *Cymbopogon citratus* and *Eucalyptus camaldulensis* on *Colletotrichum* sp. as a function of different concentrations and days after incubation

Analysis of Figure 2 shows that application of the combination of aqueous extracts at the three evaluation dates of 2, 4 and 7 days after incubation gave average results for concentrations of 10%, 20% and 30%, which were below (< 30% to 100%) those observed for *A.*

solani. Optimum efficacy (Tec= 100%) was obtained with the combination of Tec extract at 2 JIA at a concentration of 30%, which is close to the synthetic fungicide control (Tco = 100%). In short, the inhibition rate of the combination of aqueous extracts increased with the m/v concentration.

Table 5. Inhibitory effect of different treatments of *Cymbopogon citratus* and *Eucalyptus camaldulensis* on mycelial growth of *Colletotrichum* sp. according to different concentrations and observation dates.

Treatments	Avg 2 JAI en %	Avg 4 JAI in %	Avg 7 JAI in %
Tca1	79.630^{a}	55.397^{bcd}	34.453^{bcd}
Tca2	100.000^{a}	100.000^{a}	100.000^{a}
Tca3	100.000^{a}	100.000^{a}	100.000^{a}
Tce1	54.377^{b}	40.860^{bcd}	31.240^{bcd}
Tce2	100.000^{a}	100.000^{a}	100.000^{a}
Tce3	100.000^{a}	100.000^{a}	100.000^{a}
Tco	100.000^{a}	100.000^{a}	100.000^{a}
Te	0.000^{d}	0.000^{e}	0.000^{d}
Tea1	40.610^{bc}	38.067^{cd}	35.720^{bcd}
Tea2	100.000^{a}	72.230^{ab}	66.507^{ab}
Tea3	100.000^{a}	100.000^{a}	100.000^{a}
Tec1	27.853^{c}	29.110^{d}	20.593^{cd}
Tec2	52.390^{b}	47.817^{bcd}	46.133^{bc}
Tec3	100.000^{a}	65.947^{abc}	62.530^{ab}
Tee1	100.000^{a}	100.000^{a}	100.000^{a}
Tee2	100.000^{a}	100.000^{a}	100.000^{a}
Tee3	100.000^{a}	100.000^{a}	100.000^{a}
Fisher's F	29.703	17.292	15.416
Pr > F	< 0.0001	< 0.0001	< 0.0001
Meaning	****	****	****

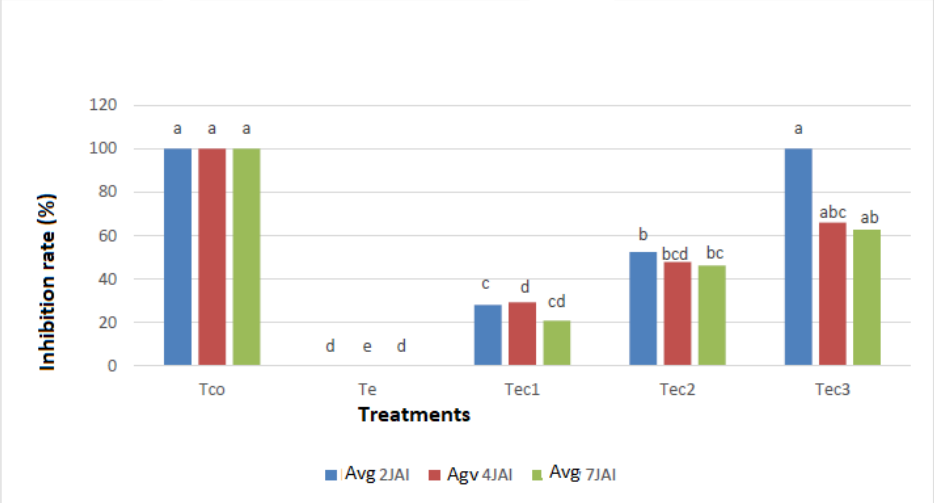

Figure 2. Inhibitory action of the combination of aqueous extracts of *Cymbopogon citratus* and *Eucalyptus camaldulensis* on the mycelial growth of *Colletotrichum* sp. according to different concentrations.

Comparison of the inhibitory action of the combination of aqueous extracts of *Cymbopogon citratus* and *Eucalyptus camaldulensis* on the mycelial growth of *Alternaria solani* and *Colletotrichum* sp. on the seventh day of incubation

Analysis of Figure 3 shows that application of the combination of aqueous extracts at concentrations of 10%, 20% and 30% on day 7 of the incubation gave convincing results. The inhibitory action was greater on *A. solani* than on *Colletotrichum* sp., with a minimum inhibition rate of around 49% for *A. solani* and 21% for *Colletotrichum* sp. The best results were always obtained

with the combination of Tec extracts at a concentration of 30% on *Alternaria solani*.

Checking Sporulation on the Fourteenth Day after Incubation

Binocular microscopy of fragments of mycelial explants taken from Petri dishes (figure 4) and visualized between slide and coverslip showed that application of the different extracts at concentrations of 10%, 20% and 30% respectively prevented sporulation of *Alternaria solani* mycelium (figure 5A). On the other hand, conidia (spores) were observed on *Colletotrichum* sp. mycelium at all concentrations. This means that even the 30% concentration of the extracts did not significantly inhibit the mycelial growth of *Colletotrichum* sp., leading to sporulation at 14 days old (figure 5B).

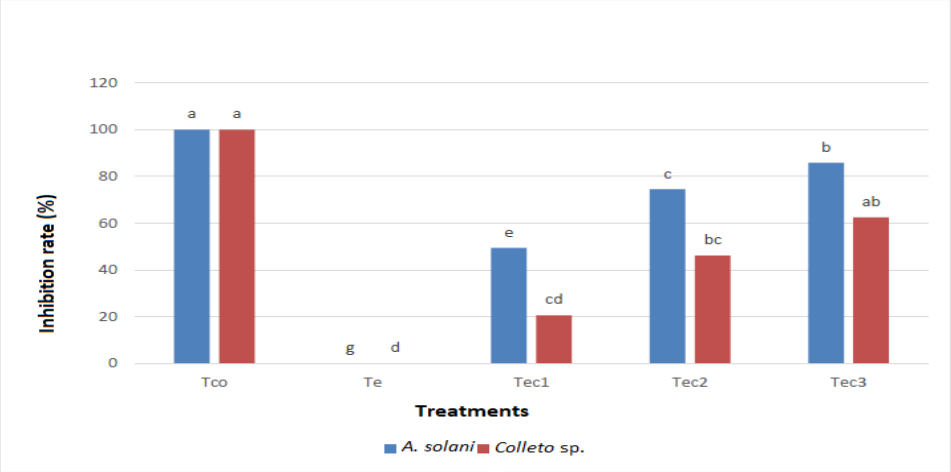

Figure 3. Inhibitory action of the combination of aqueous extracts of *Cymbopogon citratus* and *Eucalyptus camaldulensis* on the mycelial growth of *Alternaria solani* compared to that of *Colletotrichum* sp.

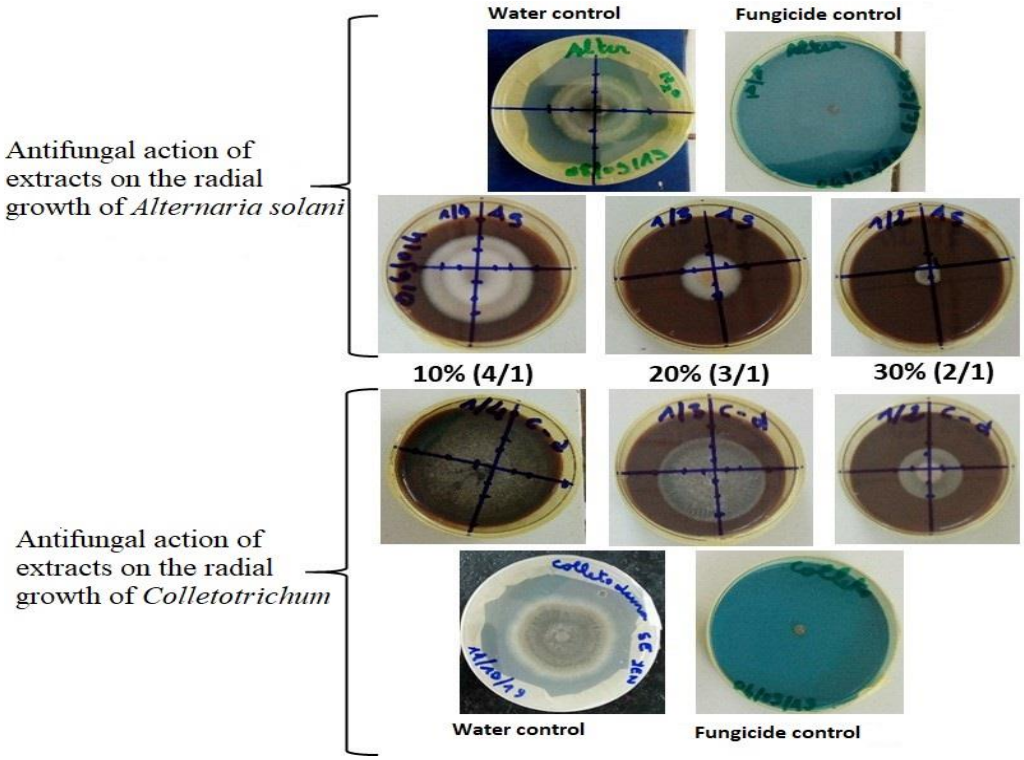

Figure 4. Inhibition of mycelial growth of *A. solani and Colletotrichum* sp. with the combination of extracts as a function of concentrations.

Figures 5 A & B. Microscopic observation of fungal spores from the culture media of 30% of the plant extracts.
A. Illustration of the absence of spores for *A. solani* with G = 16X40; **B**. Illustration of the presence of spores or conidia for *Colletotrichum* sp. with G = 16 X 100.

DISCUSSION

The different yields of aqueous and ethanolic extracts obtained were 25.24% and 18.45% respectively for *Eucalyptus camaldulensis* and 15.27% and 8.50% for *Cymbopogon citratus*. The richness of *Eucalyptus camaldulensis* in water-soluble compounds such as phenolic acids, flavonoids and many other phenolic compounds explains its high yields. Extraction yields can be influenced by the harvesting period, the age of the plant material and/or the type of solvent used, the type of plant or the part of the plant used for extraction (Tiendrebeogo, 2011).

Extracts (aqueous or ethanolic) of *Cymbopogon citratus* and *Eucalyptus camaldulensis* applied at concentrations of 20% and 30% completely inhibited the mycelial growth of *Alternaria solani* and around 60% to 90% that of *Colletotrichum* sp. We can therefore report that, from a concentration of 20% upwards, all the plant extracts taken alone or in combination inhibited the mycelial growth of both pathogens, but to different degrees. The effectiveness of these extracts could be explained by the presence in them of secondary metabolites such as terpenoids, alkaloids, steroids and phenolic compounds, which constitute a plant defence weapon. These metabolites give them antifungal properties that enable them to totally or partially inhibit the mycelial growth of pathogenic fungi. These findings of biological efficacy against agricultural production diseases have been made by several authors. (Kassankogno *et al.*, 2015) showed that the 20% aqueous extract of *C. citratus* inhibited the mycelial growth and sporulation of *Pyricularia oryzae* conidia.

On the other hand, (Dao, 2008) showed that the 30% aqueous extract of *Cymbopogon citratus* inhibited the development of *Fusarium moniliforme*. In addition, (Dabiré, 2004) pointed out that the aqueous extract of *Eucalyptus camaldulensis* at a concentration of 20% significantly reduced the infection index of *Curvularia* spp. on sorghum compared with the untreated control; ethanolic extracts were also the most effective of all the aqueous extracts. In our study, ethanolic extracts of *Eucalyptus camaldulensis* were more effective than those of *Cymbopogon citratus*. Ethanol (70%) is known to be more effective for extracting polyphenols than water. (Sankara *et al.*, 2020) evaluated the polyphenol content and antioxidant activity of ethanolic extracts obtained from solid residues after hydro-distillation. Generally speaking, these extracts are richer in total phenolic compounds and flavonoids, which justify the anti-tremite and antifungal activities. A concentration of only 10% is therefore an effective concentration for ethanolic extracts of *Eucalyptus camaldulensis*. Similar results were obtained by (Sankara *et al.*, 2020) and (Bonzi-Coulibaly *et al.*, 1997), according to whom ethanolic extracts obtained from solid residues after hydro-distillation are generally richer in total phenolic compounds and flavonoids with antifungal activity. In all cases, mycelial growth decreased as the concentration of extract in the medium increased. This suggests that the higher the concentration of the extract in the medium, the higher its concentration of antifungal active ingredient. Indeed, (Nasr *et al.*, 2019) believe that the biological activity of a substance could be a function of its proportion of active principle.

Generally speaking, analysis of the results obtained over the three evaluation dates (2 JAI, 4 JAI, 7 JAI) reveals a drop in the inhibition percentages. This finding could be explained by the instability of the extract below a certain concentration threshold or by the resistance abilities of the micro-organisms, resulting in a drop in antifungal efficacy. As ethanol extracts can contain volatile molecules (constituent of essential oils), there is a long-

term risk of isotability.

Comparison of the inhibition effect of the combination of aqueous extracts revealed that *Alternaria solani* was more sensitive than *Colletrichum* sp. The combination of extracts follows a certain law of increasing proportionality with the increase in the proportion of *Eucalyptus camaldulensis* in the three concentrations with the ratios (4/1) for 10% or 8% of *Eucalyptus camaldulensis* and 2% *Cymbopogon citratus* (3/1) for 20% or 15% *Eucalyptus camaldulensis* and 5% *Cymbopogon citratus* and (2/1) for 30% or 20% *Eucalyptus camaldulensis* and 10% *Cymbopogon citratus*. The combination respecting the (2/1) ratio corresponding to 30% concentration was the most effective, with a high inhibition rate, but no more so than with the aqueous extracts taken individually, whose inhibition is total for the same concentration. The higher the proportion of *Eucalyptus camaldulensis* in the combination, the lower the antifungal activity. This suggests an antagonism between the aqueous extract of *Eucalyptus camaldulensis* and the aqueous extract of *Cymbopogon citratus*. This could be linked to a neutrality in the interaction of the active molecules contained in each of the two plants. Consequently, combining these extracts is not beneficial. However, reversing these ratios with a higher proportion of *Cymbopogon citratus* than *Eucalyptus camaldulensis* could change our results in favour of synergy between these extracts. Combining an odd number of plants could also produce the desired results.

CONCLUSION

Aqueous and ethanolic extracts of *Eucalyptus camaldulensis* and *Cymbopogon citratus* leaves were effective against *Alternaria solani* and *Colletotrichum* sp., tomato pests in Burkina Faso. Ethanolic extracts of *Eucalyptus camaldulensis* proved more effective than those of *Cymbopogon citratus*. The combination (2/1) of *Eucalyptus camaldulensis* and *Cymbopogon citratus* corresponding to the 30% concentration was better than the other ratios (3/1) and (4/1) with a percentage inhibition of more than 85% on *Alternaria solani* and 62% on *Colletotrichum* sp. after seven days of incubation. *Alternaria solani* was more sensitive to the different treatments than *Colletotrichum* sp. However, a comparison of the aqueous extracts taken individually did not reveal any synergy between these extracts in the fight against the parasites. These results could contribute to the production of more effective and stable biopesticides as an alternative to chemical pesticides, which are too expensive and toxic for the environment.

CONFLICT OF INTEREST

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

AUTHORS CONTRIBUTION

Kibsa J. E. Sedego, the primary investigator and interested party in the work, set up and monitored the trials, overseeing data collection and processing. Jean C. W. Ouedraogo assisted with the extraction of essential oils in the laboratory and their application on crops. Elise Sanon Abalo and I. Kassankogno provided ongoing support and assistance throughout the research. Kounbo Dabiré was responsible for data analysis. Philippe Sankara and Yvonne L. Bonzi-Coulibaly contributed to the reading and correction of the manuscript, ensuring accuracy in both form and content.

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