Evaluationof the effectiveness of copper hydroxide $CU(OH)_2$against the main diseases of tomato(*Solanum Lycopersicum esculentum*) in Burkina Faso

ABSTRACT

Fungal and bacterial diseases cause a permanent threat to tomato production and can cause economic losses. This study aims to evaluate the biological effectiveness of copper hydroxide 65.6% in Burkina Faso. The evaluation was carried out in the Hauts-Bassins region in a farming environment during the dry and cold season, and the hot and humid season. The plant material used was the F1 Mongal tomato variety. The experimental design was a Fisher block comprising ten (10) repetitions and three (03) treatments. The treatments were distributed as follows: T0 (untreated), T1 (Mancozeb 800g/kg) and T2 (copper hydroxide 65.6%). Copper hydroxide 65.6% was applied at a dose of 4 kg/ha and Mancozeb 800g/kg at 3 kg/ha. The treatments were applied at intervals of one week over a period of one month. Tomato yield, disease prevalence and severity were the parameters studied. At the end of this study, the diseases identified were early blight, anthracnose, vascular fusarium wilt and bacterial wilt of tomatoes. Copper hydroxide 65.6% was found to be more effective than Mancozeb 800g/kg. It significantly reduced the prevalence and severity of early blight and anthracnose. The yield analysis showed that the treatment with copper hydroxide 65.6% recorded a yield gain of 3.16 tons compared to Mancozeb 800g/kg, and 4.87 tons compared to the non-control. processed. Given the biological effectiveness of copper hydroxide 65.6%, it would be desirable to recommend it to producers with a view to increasing tomato productivity.

Keywords:bacterial agents, biological effectiveness, Burkina Faso,copper hydroxide 65,6%, fungal and, tomato.

1. Introduction

In order to overcome a long periods of drought which constitute an obstacle to a quantity of food, the Burkinabe state is committed to promoting market gardening. At the national level, market gardening production is marked by wide specific and varietal diversification [1]. The tomato with 140,369 tons produced occupies 3rd place in total vegetable production after onion (216,780 tons) and cabbage (178,632 tons). The area exploited for tomato production is approximately 10,284 ha or 18.9% of the area total market gardening at the national level. Tomatoes remain the most profitable vegetable crop in the country. Indeed, the monetary value of the tomato is estimated at more than 48 billion (MAAH).

However, despite its economic importance and the opportunities it offers for Burkina Faso economy, tomato cultivation faces numerous biotic constraints including parasitic pressure which can reduce yields. Fungal, bacterial and viral diseases are frequently encountered in market gardening areas. Fusarium wilt (Fusarium oxysporum f. sp. lycopersici) is one of the fungal diseases that affects both adult tomato plants and seedlings in nurseries [3]. Also, early blight (Alternaria dauci f. sp. solani) is a fungal disease which considerably reduces the

quantity and quality of tomato production by causing chlorotic spots on leaflets, stems, sepals and fruits at all stages of development. of the plant [4]. Bacterial wilt of tomato caused by Ralstonia solanacearum Smith is responsible for necrosis, wilting and mortality of plants [4].
Although numerous studies have demonstrated non-compliance with phytosanitary practices by market gardeners, chemical control still remains the most widely used in the fight against tomato diseases and pests [5]. Indeed, out of 432 pesticides authorized by the Sahelian committee, commercial specialties such as TAMEGA, OXAMAX, JUMPE 75 WG are mainly intended for the fight against fungal diseases and tomato pests [1].
The objective of our study is to evaluate in a farming environment the fungicidal and bactericidal activities of copper hydroxide 65.6% against the main fungal and bacterial diseases of tomatoes (Lycopersicon esculentum Mill) in the dry and cold season; and in the hot and humid season in Burkina Faso.

2. MATERIALS AND METHODS

2.1. Site of experiment

The study was carried out at farming environment in two gardening production areas, namely Sakabi and Leguema. These areas are located near to the town of Bobo-Dioulasso, Houet province, Hauts Bassins region (Figure 1). They belong to the South Sudanese climate characterized by a rainy season of 5 to 6 months (mid-October), an annual rainfall exceeding 900 mm, a dry season from November to April, a relative humidity between 23.1% and 81.65 %, and a thermal amplitude of 5.4°C (Figure 2) (Meteo, 2019).

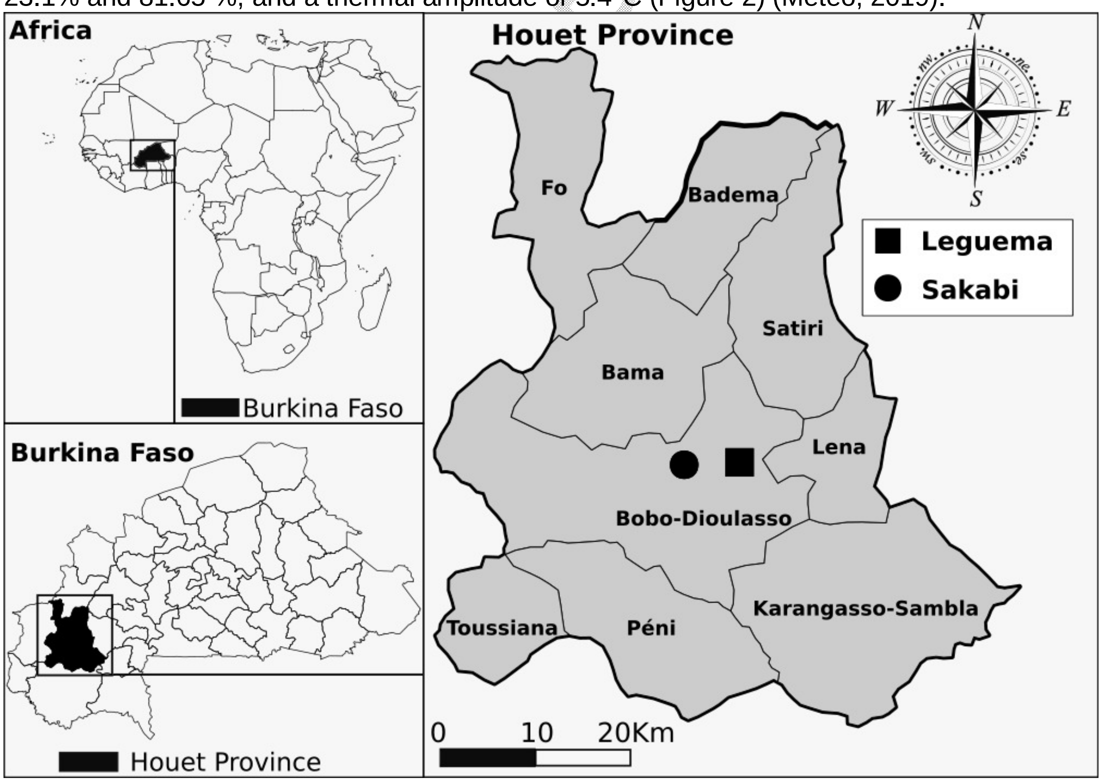


Figure 1: Representative map of the study area

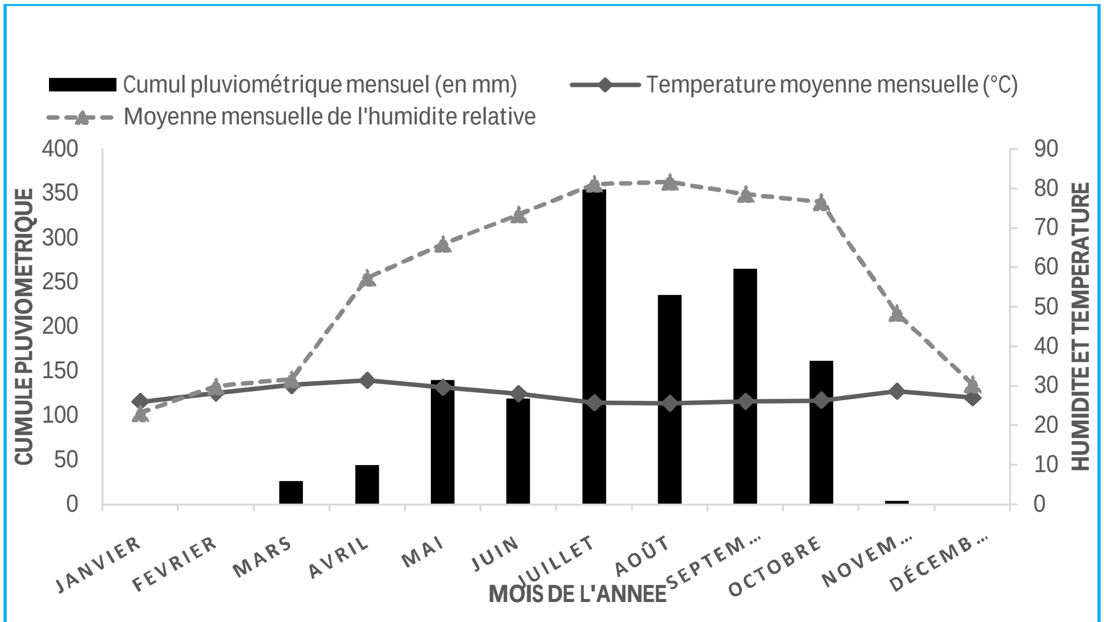


Figure 2: Climate diagram of Bobo-Dioulasso in 2019 (airport weather,2019).

2.2. Plant material

TheMongal tomato variety F1 was initially selected for the agronomic performance. It is a variety with determinate growth of 90 to 120g in size, the maturity period is between 60 and 65 days. This variety provides an average production of 20 tons per hectare in rainfall season and 15 tons in dry season.

2.3. Chemical material

Copper hydroxide 65.6%

The product used was the active ingredient copper hydroxide 65.6% and WP formulation was recommended in vegetable and fruit crops. It is a broad-spectrum bactericidal fungicide. It is used as a wettable powder at a quantity of 2 kg/ha. It is a contact product which inhibits the germination of spores through a multi-site action (at the level of respiratory processes, protein biosynthesis and membrane activities) and therefore cannot select resistant mutant strains. The treatments are renewed every 7 days, depending on weather conditions.

Mancozeb 800g/kg

The product used containing the active ingredient Mancozeb 800g/kg is a WP wettable powder fungicide approved by the Sahelian Pesticides Committee. Its active ingredient is Mancozeb 800g/kg, used at a dose of 2 to 3 kg/ha. It is used in vegetable and fruit crops as a preventative measure. The treatment is renewed every 14 days depending on climatic conditions. It is used in the fight against Downy Mildew, Anthracnose, Rust, Cercosporiosis, Pyriculariosis, Helminthosporiosis, and Cladosporiosis.

2.4. OTHER PRODUCTS USED

In addition to IDEFIX and MANGA PLUS, commercial specialties such as Deltacal 12.5 EC and Acarius 18 EC have been used against insects and mite pests of tomato (Table 1). Mineral fertilizer NPK (14-23-14) at 100 kg/ha and urea at 100 kg/ha were applied as fertilizers.

Table1 : Other products used

Commercialname	Active ingredients	Authorized use
Deltacal 12,5 EC	Deltamethrine 12.5 g/l1	Insecticide authorized against cartepillars*Helicoverpaarmigera*and whiteflies in tomato production
Acarius 18 EC	Abamectine 18g/l	Insecticide-Acaricide against the strains of mites resistant to organophosphates.

2.5. Field management

For the dry and cold season, the nursery was set up on December 18, 2018 in the plant protection station under a shade house to protect the plants against sunlight and against the attack of sucking biters which carry viruses. The soil was previously sterilized and the seed was coated with MONTAZ 45WS fungicide. The plants were transplanted onto the ridges on January 10, 2019 in the open field by 10 producers spread across two market gardening sites: Leguema and Sakabi. For the hot and humid season, the nursery was set up on July 5, 2019 and the transplanting in a farming environment on July 22, 2019. The weeding operation of each site was periodic, with an additional fertilizer, insecticide and acaricide treatment. The NPK 14-23-14 mineral fertilizer was added in three (03) steps: three (03) weeks after transplanting, the second addition took place during the bolting phase, and finally the last fraction was mixed with the 'Urea 46%.

2.6 Inventory of fungal and bacterial flora

Suspected leaf, twig and stem samples were soaked in 70% alcohol for 30 seconds then in 1% bleach for 1 minute and finally rinsed thoroughly with sterile distilled water. The samples were then placed in sterile Petri dishes containing a round of sterilized filter paper. The Petri dishes were incubated in a room at 25°C (Gaston et al., 2023). At the end of the incubation period, the fungal colonies were observed using a binocular microscope and an optical microscope in order to highlight the characteristics of the hyphae, conidiophores and conidia. Identification was made using the identification key of Mathur and Kongsdal (2003). On each box, the species of mushrooms observed were listed using an anti-reading marker by writing the first two or three letters of the name of the mushroom.

For the isolation of bacteria, the wilting symptoms on the stems and roots were previously cut out, cleaned with 70% alcohol then rinsed with sterile distilled water. They were then dilacerated in 1 ml of Tris buffer (pH 7.2). The ground material obtained is left to macerate for 30 minutes to allow the bacteria to spread. A volume of 50 µl of the supernatant was spread using the three (03) sector method on a semi-selective Kelman culture medium (Bacto Peptone 10 g, Glucose 5 g, Casamino acids 1 g, Yeast Extract 1 g, Agar 15 g, Distilled water 1000 ml). After 3 to 4 days in the oven at 30◦C, the typical colonies of Ralstonia solanacearum were selected.

2.7. Experimental design

The experimental design was ten (10) simple blocks with three (3) treatments, each block represented one farmer. In each block, we had three elementary plots corresponding to the three (03) treatments. Each elementary plot had an area of 400 m2 (40 m × 10 m) and was separated from the others by a distance of 2 m. The spacings were 80 cm between the lines and 50 cm between the pockets. The different treatments carried out were as follows: T0 = untreated control; T1 = the reference control (Mancozeb used at 3 Kg/ha); and T2 = product to be tested (Copper hydroxide used at 2 kg/ha).

2.8. Assessment of prevalence, severity, and yield

Prevalence was evaluated on 20 plants chosen randomly per treatment. This assessment was made visually by direct observation of leaf symptoms.
Prevalence is calculated by taking the ratio of the number of diseased leaves on a tomato plant in a given week.
The yieldof tomato was estimated by accumulating the production per treatment and per market garden site.

2.9. Data analysis

Observations on the prevalence and severity of wilt, early blight, and downy mildew was collected every week over a period of one month.
The experiment was conducted in dry, cold, hot and humid season. The data collected were analyzed with R software, version 3.4.1. Tukey's post-hoc multiple comparison test (multicomp package) of the severity and performance of all treatment pairs was used in the case where the analysis of variance (ANOVA) test was significant at 5%

3. RESULTS AND DISCUSSION

3.1. The main fungal diseases inventoried

Two main fungal diseases were observed in the plots during the experimental trial period. These are:

3.1.1. Alternariasis

3.1.1.1. Symptoms of disease

Symptoms of this disease have been recorded on leaves, stem and fruit. At the foliar level, the disease results in the appearance of black, circular and necrotic spots which enlarge into concentric circles (A). On the stems and fruits the spots are brown or gray, concentric and elliptical (B & C).

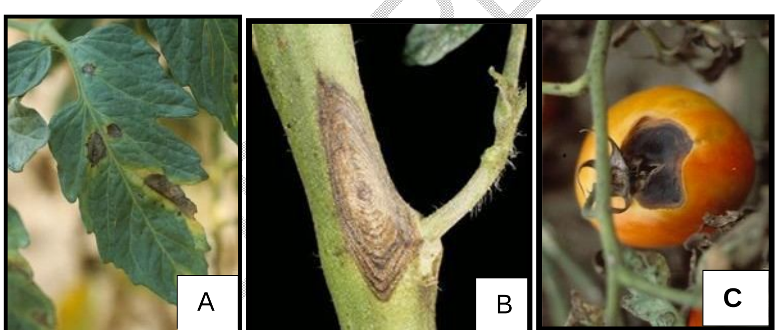


Figure 3: Symptoms of alternariasis (A: black spots, circular and necrotic; B and C: brown or gray spots,concentricandelliptical)

3.1.1.2.Species of mushrooms characterized

From the incubated samples, blackish filamentous structures were observed under a magnifying glass (Figure 4). Under the microscope, the presence of ovoid, sometimes elliptical conidia was noted. They often have a conical to cylindrical beak, brown and short. They are divided by transverse and/or longitudinal partitions (or septa) (Figure 4). The electrophoresis of seven (07) isolates and a control with primers from the ITS region show amplicons on agarose gel of sizes between 500 to 600 bp. These results are close to *Alternaria sp.*

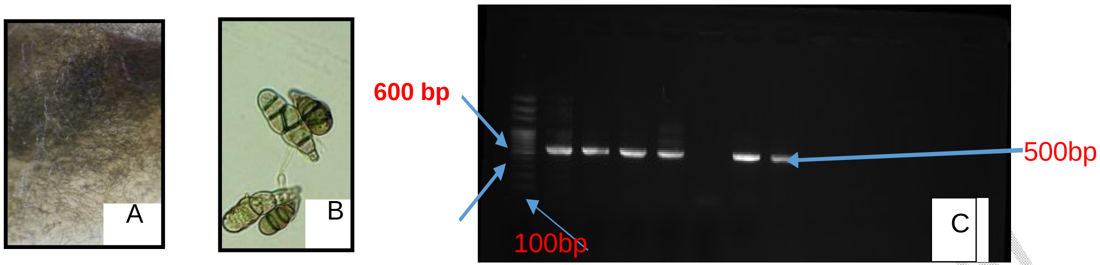


Figure 4: Characterization of Alternaria sp. Associated with alternariasis of tomato. A: Filamentous structure under glass; B: Ovoid to ellipsoid conidia; C: Electrophoresisof seven isolates tested.

3.1.2. Anthracnose

3.1.2.1 Symptoms of the disease

Symptoms of the disease
Small round, brown spots with a yellow halo were observed on the leaves and stem of tomato plants (A & B). Symptoms were less frequent on fruit in the field. Other symptoms have occasionally been observed. These are:

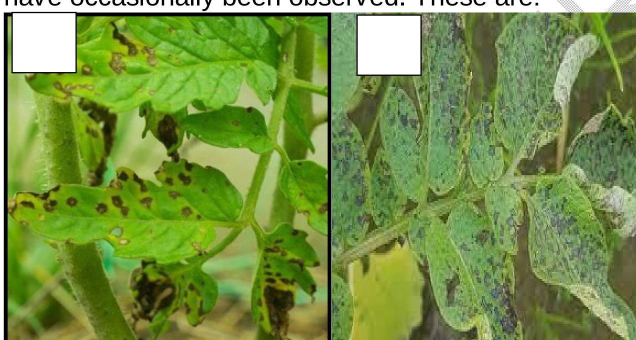
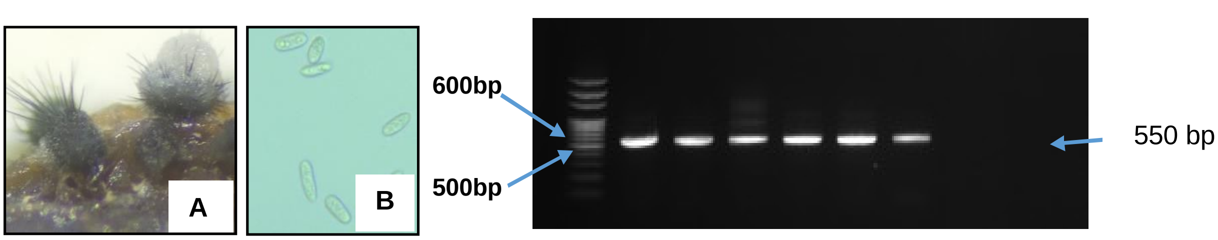


Figure 5: Anthracnose of tomato on the experimental site. A and B necrotic spots respectively on leaves.

3.1.2.2. Characterization of fungal species associated with disease

Under a magnifying glass, structures in the form of orange to whitish aggregates were observed from the incubated organs (photo A). Under the microscope, elongated hyaline, unicellular and generally rod-shaped conidia are noted. The electrophoresis of eight (08) isolates and a control with primers from the ITS region show amplicons on agarose gel of sizes between 500 to 600 bp. These results are close to *Colletotrichum spp.*



6

550bp C

Figure 6: Characterization of *Colletotrichum* sp. associated to anthracnose of tomato on the experimental site. A:filamentous structure under glass; B: Ovoid to ellipsoid in the form of sticks; C: Electrophoresis of eight isolates tested.

3.2 The main bacterial disease inventoried

3.1.3.Bacterial wilt of tomato

Bacterial wilt of tomatoes is manifested by slowed plant growth, leaf epinasty with yellowing of the lower leaves and the emission of adventitious root shoots on the stem.. These different symptoms can appear both on young seedlings in the nursery and on adult plants. A longitudinal section allows the destruction of the conductive vessels to be seen. The wilting of the plant is irreversible and the plant eventually dies (Figure 7, A). The pathogen is *Ralstonia solanacearum* presenting colonies with a mucous appearance, ovoid in shape and whitish in color changing to pink in the center. (Figure 7, B)

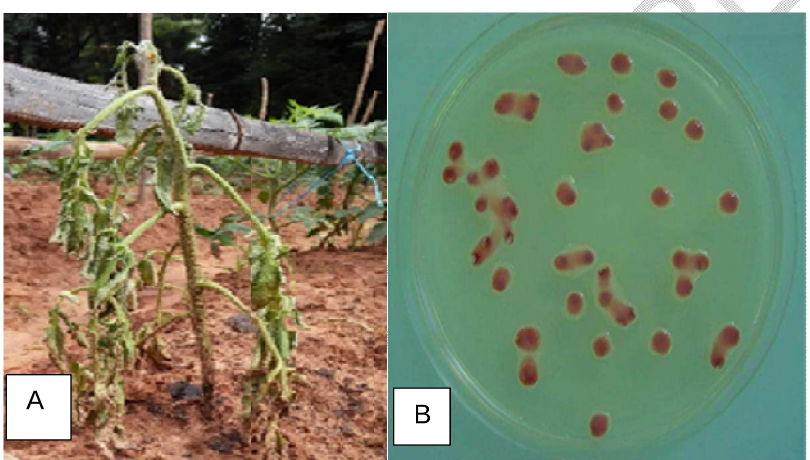


Figure 7: Bacterial wiltsymptoms (A), Colony of *Ralstonia solanacearum* on cultivation (B)

3.3diseases observed occasionally

3.3.1 Fusarium wilt: yellowing followed by drying of the leaves was noted on certain plants (figure 8, A & B). From the incubated samples, whitish to pinkish aggregates were observed under a magnifying glass (figure 8, C). Under the microscope the conidia are spindle-shaped with partitions (figure 8, D).

A B C D

Figure 8: Tomato fusarium wilt. A and B: on leaves; C: under glass D: under microscope

3.3.2 Mildew: Greenish-black, oily and irregular spots have been observed at the apex or margin of old leaves (Figure 9). In addition, brown areas on these organs, wilting and even death of plants have been observed. been recorded (Figure 9). Brown cankers were noted on the petiole and stem of the plants (Figure 9). On the fruits, greenish-brown, oily, bumpy spots have been recorded.

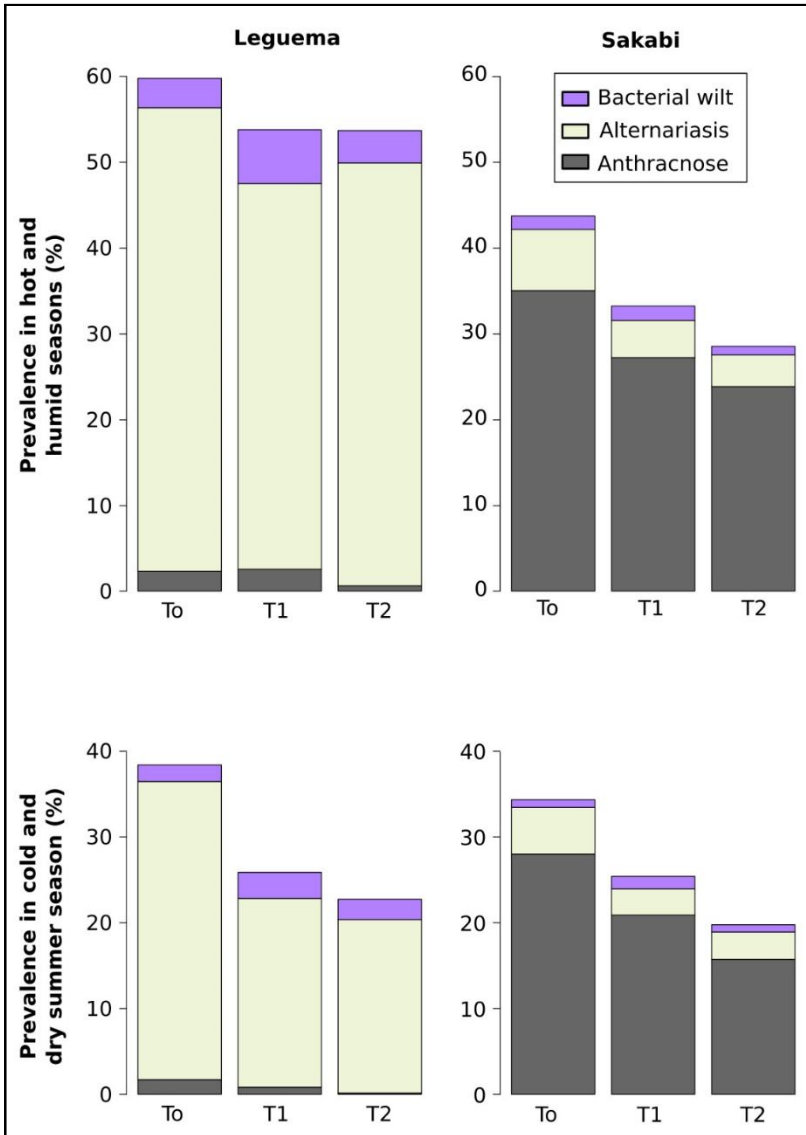
Figure 9: Characteristic Symptoms of tomato mildew

3.2. Disease prevalence in the two growing seasons

In the dry and cold season in the untreated plots, a high prevalence of alternaria was revealed in Leguema (34.77%) compared to Sakabi (5.44%).However, Sakabi was recorded the high prevalence of anthracnose (28%) compared to Sakabi (1.7%).Bacterial wilt was recorded the lowest prevalence in the two vegetables crops areas.

In the hot and humid season, it was observed in the untreated plots an increase in the prevalence of alternaria in Leguema of 19.21%;an increase in the prevalence of anthracnose of 7.05% in Sakabi and a slight increase in the prevalence of bacterial wilt of tomato compared to the dry and cold season.

Overall, treatments with Mancozeb 800 g/Kg (T1) and copper hydroxide 65.6% (T2) were recorded the lowest prevalence compared to untreated control.Treatment with Mancozeb 800 g/Kg (T1) and copper hydroxide 65.6% behaved similarly with however a slight decrease in prevalence for all diseases with treatment with copper hydroxide 65.6%..

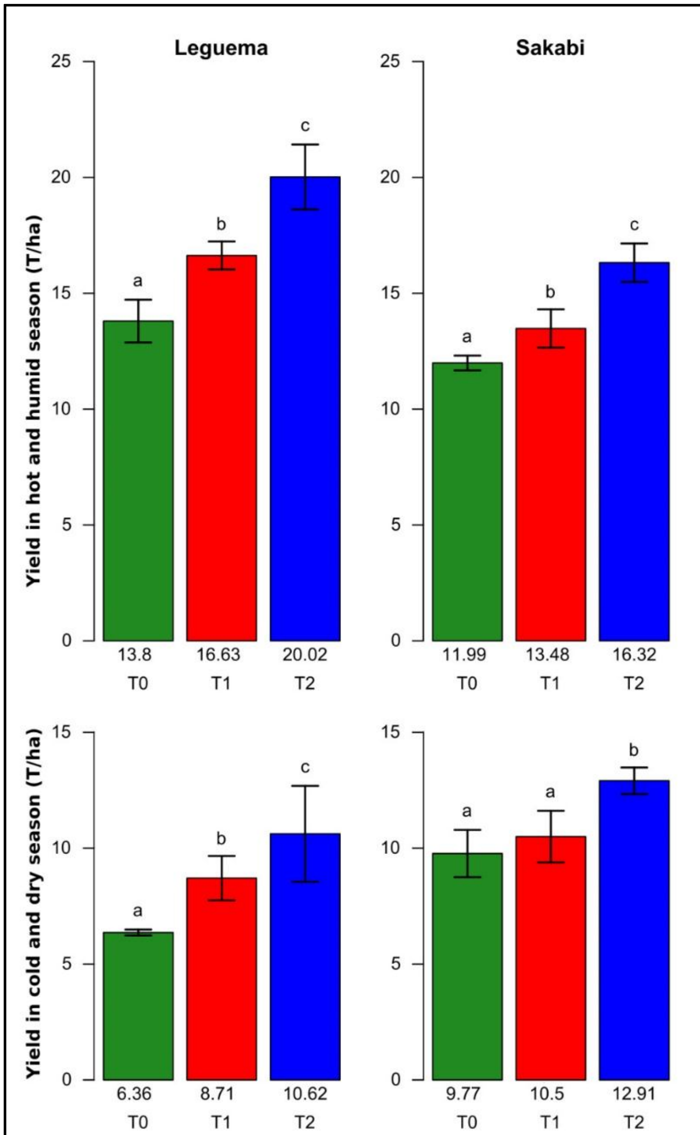


T_0 = untreated control; T_1 = Mancozeb 800 g/Kgused at 3 Kg/ha*(*reference control*)* andT_2 = Copper hydroxide 65.6% used at 2 kg /ha.

Figure 10: Prevalence of diseases in two growing seasons

3.3. Tomato Yield Analysis

The analysis shows that the yield obtained at copper hydroxide 65.6% (T2) is significantly different from the other two treatments. Copper hydroxide records the highest yield in the hot and humid season with 20.2 T/ha to leguema and 16.32 T/ha in Sakabi and in the cold and dry season with 10.62T/ha to leguema and 12.91 T/ha Sakabi whatever the experimental site.



T_0 = untreated control; T_1 = Mancozeb 800 g/Kgused at 3 Kg/ha *(*reference control*)* and T_2 = Copper hydroxide 65.6% used at 2 kg /ha.

Figure 11*:* Yield in two growing seasons depending on treatments

4. Discussion

Direct observations of symptoms and isolations and molecular characterizations of samples collected at each assessment revealed three main diseases in the vegetable perimeters of Leguema and Sakabi, namely alternaria, anthracnose, and bacterial wilt of tomato. Some diseases such as downy mildew and fusarium wilt were also encountered. Thirty-two fungal species had already been identified in Burkina Faso on samples of diseased plants in the field, the most widespread of which were *Fusarium oxysporum* (Schltdl.) (66.4%), *Colletotrichum* sp.(27.4%), *Alternaria solani* (Sorauer) (26.9%), *Alternaria* sp.(27.5%), *Botrytis cinerea* (Pers) (24.1%) and *Fusarium equiseti* (Sacc.) (19.5%) (Tiendrebeogo et al.,2023). These diseases compromise tomato production given their persistence from one production campaign to another. Studies were carried out in southern Togo and made it possible to list 22 species of mushrooms divided into 14 genera, including the genera Alternaria, Colletotrichum, and Fusarium (N'dri et al, 2020). In Ivory Coast, the work of [6] N'Guessan et al. (2019) revealed different tomato diseases including bacterial wilt of tomatoes related to Solanaceae.

The presence of these diseases could be explained by the exchange or use of contaminated seeds from one growing area to another, the cultivation of the crop from one production campaign to another without rotation and the hot, humid conditions conducive to disease exposure. Furthermore, the results showed that the prevalence rate of the disease is high during the hot and humid season (35%) than in the cold and dry season (28%).

This result is justified by the presence of copper molecules which reduced the development of phytopathogenic agents in the organs of the tomato. The same observations were made by [7] Haougui and Abdou (2013) who demonstrated that copper hydroxide significantly reduced the incidence of bacterial scab in tomato crops by more than 50% compared to mancozeb under the conditions climate of Niger. Our results also showed that the incidence and severity levels are significantly high during the hot (xx%) and humid season compared to the cold and dry season. This observation can be justified by the amounts of rain and the high temperatures recorded during the long rainy season compared to the short rainy season.

The high levels of incidence and severity of these tomato diseases obtained during our study during the hot and humid season are attributable to the constant humidity maintained by watering and the rainfall regime recorded. Our results confirm the work of [8]Latour et al. (2008), Platt (2008) then [9]Vicente and Holub (2012), according to which humidity is one of the primary factors in the development of bacterial and cryptogamic diseases. According to Carisse et al. (1999), thermal variations and ambient humidity are factors that influence tomato infection levels.

The yields obtained are much higher in the treatment of copper hydroxide compared to the other two treatments. This could be explained by the effect of copper hydroxide on fungal and bacterial diseases which reduces the incidence and severity of bacterial wilt, alternaria and downy mildew. According to [10]Hema (2016), the fungicide-bactericide copper hydroxide reduces the incidence of bacterial wilt and fungal diseases and increases tomato yields.

5. Conclusion

This study confirms that tomato diseases are caused by bacterial wilt, alternariasis and mildew in Burkina Faso. It also appears that the development of these diseases is favored by high humidity. Copper hydroxide effectively controls alternaria, anthracnose and bacterial wilt.

CONSENT

Not applicable

ETHICAL APPROVAL

Not applicable

Disclaimer (Artificial intelligence)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

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