IN VITRO EVALUATION OF HOMEOPATHIC REMEDIES AGAINST *COLLETOTRICHUM MUSAE* OF BANANA

A THESIS

BY

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STUDENT ID: MS-230821

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KHULNA-9208

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DEDICATED

TO

MY

BELOVED PARENTS

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ABSTRACT

Banana (*Musa paradisiaca*) is one of the most economically and socially significant plants in tropical and subtropical regions across the world. However, its production is severely affected by *Colletotrichum musae*, the pathogen responsible for anthracnose disease in banana. This study focused on evaluating the *in vitro* effectiveness of various homeopathic remedies against *C. musae*. The experiment was performed in Plant Pathology Laboratory of Agrotechnology Discipline, Khulna University, Khulna. Seventeen homeopathic medicines (Arsenicum album, Calcaria sulp, Selenium, Cina, Nux vomica, Silicea, Arnica montana, Calcarea carbonicum, Carbo vegetabilis, Ferrum metallicum, Natrum muriaticum, Phosphorus, Sulphur, Sepia officinale, Belladonna, Kali iodatum, and Thuja occidentalis) at three potencies (6CH, 30CH, 200CH) were tested for their effects on the sporulation and mycelial growth of *C. musae*. A Completely Randomized Design (CRD) was employed, using untreated controls as references. Among the treatments, the least radial mycelial growth (38.02 mm) was recorded in plates treated with Arsenicum album at 6CH potency, followed by Selenium at 6CH (40.03 mm) and Arsenicum album at 30CH (41.44 mm). The highest inhibition of mycelial growth (49.66%) was also observed with Arsenicum album at 6CH, followed by Selenium at 6CH (46.52%) and Arsenicum album at 30CH (44.65%), while Silicea at 200CH exhibited the lowest inhibition (30.23%) and the highest mycelial growth (52.62 mm). Spore production was most significantly suppressed by Thuja occidentalis at 30CH (66.68%), followed by Sepia officinale at 6CH (63.13%) and Sulphur at 200CH (62.99%) as well as the lowest inhibition was recorded with Arnica montana at 30CH (30.41%). Overall, the result of this study suggest that homeopathic medicines could be an effective and sustainable approach to managing *C. musae* in banana plants.

Keywords: Banana, Anthracnose, *In-vitro*, Homeopathic remedies, Potency

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CHAPTER I

INTRODUCTION

Banana (*Musa paradisiaca*) is an important fruit crop under the family Musaceae, grown in tropical and subtropical regions of the world (Mohapatra et al., 2010). It is considered as the world's oldest grown plants (Kumar et al., 2012). Many tropical areas, it is used as a staple food and dietary supplements (Assani et al., 2001). Banana is mainly cultivated for it's ripen fruits and also cooked as vegetables in Bangladesh (Khanum et al., 2000). It occupies an important position among the fruits of the country not only for its wide cultivation all over the Bangladesh but also from the standpoint of food value and availability throughout the year. Banana is cultivated almost everywhere in Bangladesh round the year. The foremost banana growing areas in Bangladesh are Narsingdi, Gazipur, Tangail, Rangpur, Bogura, Natore, Pabna, Noakhali, Faridpur and Khulna Sylhet, Moulvibazar, Netrokona; Rangamati, Khagrachhari as well as Bandarban are considered as the wild grown banana producing area in Bangladesh. The fruit is considered to be a poor man's food due to it's availability throughout the year. It is a rich source of human nutrition bearing carbohydrate, potassium and vitamins, including A, C and B6. As a diet, banana is an affluent source of carbohydrate with calorific value of 67 calories per 100g fruit and is one of the most popular and widely traded fruits across the world (Kumar et al., 2012). They provide fat-free dietary fiber and are commonly introduced as the first solid food for infants. In 2023-2024, the total production of banana in Bangladesh was 774286 metric tons and the cultivated area was about 119325 acres (BBS, 2024).

Banana is affected by numerous diseases throughout their growth stages. Among these, anthracnose, caused by *Colletotrichum musae*, is recognized as one of the most significant diseases affecting banana in the global level. It poses a major challenge to banana production by compromising fruit quality, rendering them unsuitable for both consumption and sale, and causing substantial losses to farmers and traders. It is the most important postharvest disease of banana that can result in 30-40% losses of marketable fruits (Bazie et al., 2014). *Colletotrichum musae* primarily targets

wounded green and ripe banana but can also infect the bracts, flowers, petioles, and leaves of banana plants (Stover and Simmonds, 1987). Occasionally, the fungus attacks the necks of green fingers damaged by flexing, forming sunken lesions covered with salmon-colored acervuli. Infections accelerate fruit ripening, and lesions expand as the fruit ripens. On ripe banana, sunken brown spots with orange acervuli appear. The fungal colonies are initially loose with white aerial mycelium, which eventually turns orange (Jinyoung et al., 2002).

To control this disease proper control measures should be used. Due to the harmful effect for human health, the postharvest use of chemicals as fungicides is restricted in most countries (Serrano et al., 2005). Consumers have a strong preference for agricultural products free from pesticide residues. (Serrano et al., 2005; Cutler and Cutler, 1999). In this situation, a new preservation technologies are needed, which have to be considered as human-safe and environmentally friendly (Duru et al., 2003). In this context, homeopathic medicine emerges as potential alternatives, as they activate the vital energy of plants by promoting environmental balance. This approach adopts a systemic mechanism to mitigate diseases, offering the advantage of being free from side effects (Baumgartner et al., 2000).

Homeopathic medicines are formulated using a diverse array of substances, including plant-based materials such as herbs, flowers, roots, leaves, and bark. They are also produced by mineral-based components like salts, metals, and naturally occurring compounds, alongside animal-derived products obtained from animals or their secretions. Additionally, chemical compounds, whether synthetic or naturally occurring as well as pathological specimens, diseased tissues, and microorganisms are used as homeopathic medicine. Furthermore, extracts from healthy animal tissues or organs serve as sources for these remedies. However, currently the homeopathy is being used in various segments of agriculture such as pest and plant disease control (Bonato et al., 2006). The use of homeopathic preparation on plants demonstrate their effectiveness in enhancing plant growth, regulating behavior, and promoting the production of secondary defense compounds and influence on plant physiology (Bonato and Silva, 2003). These also improve fruit quality, increase leaf abundance, and aid in controlling or reducing the prevalence of disease (Ileana et al., 2017). Moreover, homeopathy is a low cost alternative to chemical fungicides, easy to use by farmers (Toledo et al., 2011).

The use of homeopathic medicines to control anthracnose is highly effective and safe for banana fruit and environment. Inhibitory effect of homoeopathic drugs such as Lycopodium, Thuja, Kali, Arsenicum, Zincum etc. against *Alternaria alternata, Fusarium moniliforme, Gloeosporium psidii, Colletotrichum gloeospoioides* and *Pestalotia sp.* and certain fruit rot pathogens have been reported (Baviskar and Suryawanshi, 2015). But in our country, the research on homeopathic medicine for controlling plant pathogens are very scanty. Considering the above facts, the objective of the present study is:

To evaluate the effect of homeopathic medicine on growth and sporulation of *Colletotrichum musae*

CHAPTER II

REVIEW OF LITERATURE

2.1 Effect of Homeopathic Medicine on mycelial growth

Kumar et al. (2023) explored the potential of homeopathic medicines as a novel approach for managing plant pathogenic fungi. Their study evaluated the efficacy of Arnica montana, Thuja occidentalis, Sulfur, and Silicea against fungal pathogens, including *Fusarium oxysporum f. sp. lycopersici*, *Ascochyta rabiei, Alternaria solani, Septoria lycopersici,* and *Phomopsis vexans.* Under in vitro conditions, increasing concentrations of these homeopathic treatments significantly suppressed the radial growth of fungal mycelia. At 8 days post-inoculation, the highest mycelial growth inhibition was recorded for T. occidentalis at 200 ppm against *F. oxysporum f. sp. lycopersici* (73.73%) and *A. solani* (76.73%), followed by A. montana at 200 ppm for *S. lycopersici* (64.22%), silicea at 200 ppm for *A. rabiei* (59.77%), and sulfur at 200 ppm for *P. vexans* (53.82%). Correspondingly, the minimum radial growths observed were 22.80 mm, 18.10 mm, 31.66 mm, 34.91 mm, and 38.12 mm for the respective fungal species.

Toledo et al. (2016) investigated the fungitoxic potential of homeopathic medicines against *Alternaria solani*, the causative agent of black spot disease, a significant threat to tomato (*Lycopersicon esculentum Mill.*) cultivation. The study assessed the in vitro fungitoxicity of various homeopathic remedies, including Propolis, isotherapics derived from *A. solani* and ash, at dynamizations of 6, 12, 30, and 60CH (Hahnemannian centesimal scale), alongside Sulphur, Silicea terra, Staphysagria, Phosphorus, Ferrum sulphuricum, and Kali iodatum, tested at 6, 12, 30, and 100CH. Controls consisted of distilled water and a 30% hydroalcoholic solution at corresponding dynamizations (12, 30, 60, and 100CH). Parameters such as mycelial growth, sporulation, and conidial germination of *A. solani* were analyzed. Their findings revealed that mycelial growth was notably inhibited by Sulphur and Staphysagria at 100CH, demonstrating significant suppression compared to the controls. In terms of sporulation, Propolis at 6, 30, and 60CH, as well as Ferrum sulphuricum at 6 and 30CH, exhibited inhibitory effects, distinctly outperforming the

controls. Furthermore, isotherapics of *A. solani* and ash at 6CH, along with Ferrum sulphuricum at 30CH, effectively reduced conidial germination of the pathogen.

Suya et al. (2023) conducted a study to assess the antifungal efficacy of homeopathic medicines through both in vitro and on-farm trials. The experiment involved the application of Sulphur 30CH, Ocimum basilicum 30CH, and Cuprum metallicum 30CH. Among the tested treatments, Ocimum basilicum 30CH exhibited the highest inhibition of mycelial growth, achieving suppression rates of 62.15%, 62.64%, and 71.14% at concentrations of 20 µL, 60 µL, and 80 µL, respectively. This was followed by Sulphur 30CH, which recorded inhibition rates of 58.07%, 61.53%, and 69.61%, and Cuprum metallicum 30CH, with rates of 57.33%, 60.59%, and 68.45%. All the tested homeopathic formulations displayed notable antifungal activity, with Sulphur 30CH demonstrating superior efficacy against *Alternaria solani*, underscoring its potential utility in sustainable agricultural practices.

Alam and Mistry (2022) conducted a study to evaluate the efficacy of selected homeopathic medicines in suppressing the growth of *Colletotrichum musae*, the causal agent of anthracnose in bananas. Their research aimed to contribute valuable insights into the antifungal properties of homeopathic remedies. The study involved testing ten homeopathic medicines such as Sepia officinale, Belladonna, Sulphur, Kali iodatum, Thuja occidentalis, Bryonia, Arsenicum album, Calcarea sulphurica, Selenium, and Phosphorus, at a concentration of 3000 ppm, with three potencies (Mother Tincture Q, 30X, and 200X) for each treatment. Remarkably, all the tested medicines exhibited inhibitory effects on the mycelial growth of *C. musae*. Among these, the most significant growth inhibition was observed with Arsenicum album at Q potency (48.64%), followed by Kali iodatum at 30X (21.80%) and both Calcarea sulphurica and Phosphorus at 200X potency (19.92%). Overall, Arsenicum album at Q potency emerged as the most effective treatment for suppressing the pathogen's mycelial growth.

Gama et al. (2015) conducted a study to investigate the use of homeopathic remedies for controlling red rot disease in sisal plants, which is caused by *Aspergillus niger*. They tested a range of homeopathic drugs—Carbo vegetabilis, Ferrum metallicum, Natrum muriaticum, Phosphorus, and Sulphur—at various centesimal Hahnemannian (CH) dynamizations (3CH, 5CH, 7CH, 9CH, and 12CH) on *Aspergillus niger*. The

researchers assessed in vitro growth inhibition, sporulation, and germination of the fungus over a 12-day period. Their findings revealed that the homeopathic treatments inhibited fungal growth in a dynamization-dependent manner. Among the treatments, Natrum muriaticum at 5CH demonstrated the highest growth inhibition, achieving 66%, while Sulphur at 5CH exhibited the lowest inhibition at 6.4%. Additionally, all the homeopathic drugs stimulated spore production across all dynamization levels, but they resulted in a reduction in spore germination.

Rissato et al. (2018) conducted an experiment to assess the fungitoxic effects of Phosphorus and Calcarea carbonica against *Sclerotinia sclerotiorum,* aiming to control white mold in common bean (*Phaseolus vulgaris*) through the application of highly diluted aqueous solutions. The study sought to evaluate the efficacy of homeopathic solutions, including. Phosphorus and Calcarea carbonica at dynamizations of 6CH, 12CH, 24CH, 36CH, and 48CH (centesimal Hahnemannian dilutions), in controlling white mold and suppressing the growth of *S. sclerotiorum*. A completely randomized design with 10 treatments and 5 replicates per treatment, using water as the control, was employed. The results revealed that Phosphorus 12CH, Phosphorus 48CH, Calcarea carbonica 12CH, and Calcarea carbonica 48CH demonstrated resistance-inducing effects, reducing disease progression by up to 83% and decreasing plant mortality by as much as 90%. Furthermore, in vitro experiments showed that Phosphorus 12CH, Phosphorus 48CH, and Calcarea carbonica 48CH effectively inhibited mycelial growth.

Subhani (2015) conducted a study on the isolation and effectiveness of both fungicides and homeopathic fungicides against chili anthracnose, caused by *Colletotrichum capsici.* This disease severely impacts chili fruits during both pre- and postharvest stages, with even minor anthracnose lesions significantly reducing the market value of the crop. The study evaluated the fungitoxic effects of eight different fungicides and homeopathic fungicides using the poisoned food technique in vitro. The results showed a notable reduction in mycelial growth of the fungus as the concentration of both fungicides and homeo-fungicides increased compared to the control. Regarding homeopathic fungicides, Vampire was the most efficient, achieving an 83% control rate at the highest concentration (1000 µg/ml), followed by Rigrous with 81% control. Vantage, however, was the least effective, providing only 76% control at the same concentration. These findings suggest that homeopathic

fungicides could serve as viable alternatives to synthetic fungicides, offering a more environmentally friendly solution for managing chili anthracnose.

Reis and Ottoni (2021) conducted an investigation into the antifungal effects of homeopathic medicines against *Sclerotinia sclerotiorum,* the pathogen responsible for white mold. They assessed the in vitro antifungal activity of three homeopathic remedies such as Sulphur, fungal sclerotium Nosode, and Calcarea carbonica, at dynamizations of 30CH, 200CH, and 1000CH, by evaluating their impact on the mycelial growth of the fungus. The experiment followed by completely randomized design with four replications, and the control group received no treatment. All tested homeopathic remedies and their respective dynamizations demonstrated an inhibition of fungal growth. Notably, Calcarea carbonica at the 1000CH dynamization exhibited the most significant inhibitory effect, producing a mycelial halo 40% smaller than that observed in the control group.

Losch et al. (2021) investigated the effects of homeopathic medicines on the phenological development and management of insects and diseases affecting sweet pepper (*Capsicum annuum L.*). The researchers evaluated the influence of Sulphur 30CH and Calcarea carbonica 30CH on plant growth and disease control. Sulphur demonstrated a positive impact on plant development, fruit production, and fruit diameter under field conditions, while Calcarea carbonica yielded notable improvements in plant height when grown in a greenhouse setting. Additionally, Calcarea carbonica was associated with a reduction in the number of fruits showing signs of anthracnose. The effects of these homeopathic preparations varied significantly depending on the cultivation environment, highlighting their differing impacts on sweet pepper growth and disease management.

Patil and Suryawanshi (2014) identified *Alternaria alternata* as a highly destructive pathogen responsible for fruit rot in strawberry (*Fragaria ananassa* Dutch.) in India. They collected 25 isolates from infected strawberries across various regions of Maharashtra. To manage the mancozeb-resistant mutant (Aa-EMS-2) of *A. alternata,* the researchers tested nineteen homeopathic remedies. Among these, Nux vomica exhibited the highest percent control efficiency (PCE) of 50% when applied alone. However, Sulphur 30CH demonstrated the greatest efficacy, achieving a PCE of 84.45% when combined with mancozeb. Other effective remedies included

Arsenicum album, Cina, Rhus toxicodendron, Arnica montana, Sanguinaria canadensis, Tarentula hispana, and Selenium, in decreasing order of effectiveness.

Damin et al. (2015) conducted an in vitro study to evaluate the effects of various homeopathic solutions on the biological parameters of *Metarhizium anisopliae* (Metsch). The homeopathic remedies tested included Arsenicum Album 24CH, Calcarea carbonica 30CH, Kali iodatum 100CH, Phosphorus 3CH, Silicea 30CH, Staphysagria 6, 30, and 100CH, Spodoptera frugiperda 30CH, Sulphur 100 and 200CH, and Thuja occidentalis 200CH. The study assessed several aspects of fungal activity, including germination, colony-forming units, vegetative growth, conidial production, and the insecticidal effects of the fungus against *Diatraea saccharalis* larvae (Lepidoptera: Crambidae). The results showed that none of the homeopathic treatments had a negative impact on the evaluated parameters. Consequently, all homeopathic solutions were deemed compatible with *M. anisopliae.*

Babli et al. (2022) conducted an in vitro study to assess the efficacy of nine homeopathic remedies such as Cina, Borex, Selenium, Arsenicum album, Nux Vomica, Silicea, Phosphorus, Arnica montana, and Calcarea carbonicum against *Alternaria brassicae* (Berk.) Sacc., the causative agent of Alternaria blight in Indian mustard (*Brassica juncea*). The study evaluated the effects of these homeopathic drugs at two concentrations (500 and 1000 ppm), measuring mycelial growth seven days post-inoculation. All tested remedies significantly inhibited *A. brassicae* mycelial growth at both concentrations. At 500 ppm, Arsenicum album was the most effective, reducing mycelial growth to 43.67 mm, followed by Silicea (46.00 mm), Cina (51.00 mm), Nux vomica (54.67 mm), Selenium (57.67 mm), Phosphorus (59.33 mm), Borex (62.33 mm), Arnica montana (64.33 mm), and Calcarea carbonicum (67.00 mm), with the control group showing the highest growth (89.00 mm). At 1000 ppm, Arsenicum album remained the most effective, reducing mycelial growth to 13.33 mm, followed by Silicea (15.00 mm), Cina (21.00 mm), Nux vomica (23.67 mm), Selenium (26.67 mm), Phosphorus (28.00 mm), Borex (30.33 mm), Arnica montana (34.67 mm), and Calcarea carbonicum (36.67 mm), while the control group exhibited the greatest mycelial growth at 90.00 mm.

Hanif and Dawar (2015) conducted an experiment to examine the fungicidal efficacy of homeopathic drugs in managing root rot fungi and promoting the growth of both

leguminous and non-leguminous crops. Homeopathic drugs due to fungicidal potential were used as substitute technique in reducing the incidence of root rot fungi like *Rhizoctonia solani, Fusarium spp* and *Macrophomina phaseolina.* The investigation employed Arnica montana and Thuja occidentalis at concentrations of 100%, 75%, and 50% (v/v) through soil drenching and seed treatment techniques. The findings revealed that the highest concentration (100% v/v) not only significantly enhanced plant growth but also completely suppressed the incidence of root rot fungi. Lower concentrations (75% and 50% v/v) also contributed to notable improvements in plant growth and demonstrated substantial reductions in root colonization. These effects were observed in leguminous crops such as mung (*Vigna radiata* (L.) R. Wilczek) and mash (*Vigna mungo* (L.) Hepper) as well as non-leguminous crops, including sunflower (*Helianthus annuus* L.) and okra (*Abelmoschus esculentus* (L.) Moench).

Peerzada et al. (2018) conducted an in-vitro investigation to evaluate the antifungal efficacy of various homoeopathic medicines against the plant pathogen *Aspergillus niger.* The study examined five homoeopathic treatments—Calcarea Carbonica, Lachesis Mutus, Arsenicum album, Silicea, and Thuja occidentalis—in a range of potencies (6C, 12C, 30C, 200C, 1M, 10M, and CM). Antifungal assays revealed zones of inhibition in specific potencies, such as Thuja occidentalis (6C, 1M, 10M), Silicea (6C, 12C, 30C, 10M, CM), Calcarea Carbonica (6C, 12C, 30C, 200C, CM), Lachesis Mutus (12C, 200C, 1M, CM), and Arsenicum album (30C, 200C, 1M, 10M). The results demonstrated that specific potencies of Thuja occidentalis (6C, 12C, 30C, 1M, 10M, CM), Arsenicum album (6C, 12C, 200C, 1M, 10M), and Lachesis Mutus (12C, 30C) significantly regulated fungal growth. Overall, the study provided conclusive evidence of the potent inhibitory effects of these homoeopathic treatments against *Aspergillus niger.*

Hanif et al. (2015) conducted a study to assess the fungicidal potential of homeopathic pellets in mitigating root rot fungi and enhancing crop productivity. The investigation focused on the efficacy of potentised homeopathic pellets (30C) of Arsenicum album and Thuja occidentalis in promoting seed germination, plant growth, crop yield, and suppressing root rot pathogens, specifically *Fusarium spp., Rhizoctonia solani,* and *Macrophomina phaseolina.* In vitro experiments demonstrated that the homeopathic pellets effectively inhibited the mycelial growth of the test fungi. Furthermore, in vivo

field trials revealed that A. album and T. occidentalis pellets, applied at a concentration of 75% v/w (derived from 30C potency), significantly controlled pathogenic fungi and enhanced crop growth and yield. The 50% v/w concentration also showed notable effectiveness but to a lesser extent compared to the 75% concentration. The findings conclusively highlighted the pellets' dual role in reducing disease intensity caused by root rot pathogens and improving crop plant growth parameters.

Prajapati et al. (2021) investigated the antimicrobial efficacy of various homeopathic drugs and their potencies against *Aspergillus niger* under in vitro conditions. The study aimed to evaluate the biological activity of 15 homeopathic mother tinctures and their potencies (3x, 6x, and 12x). To ensure reproducibility, the experiment was conducted twice. The results revealed significant antifungal activity, particularly with the mother tincture of *Zingiber officinale*, which exhibited the highest zone of inhibition (15.4 ± 2.88 mm), followed by *Holarrhena antidysenterica* (13.2 ± 1.09 mm) and *Terminalia chebula* (10.6 ± 1.14 mm). Furthermore, potencies also demonstrated notable antifungal activity, with *Allium cepa* 6x (10.4 ± 0.89 mm), *Caesalpinia bonducella* 6x and 12x (12.8 ± 0.54 mm and 10.4 ± 1.14 mm, respectively), *Eucalyptus globulus* 12x (11.3 ± 1.94 mm), *Ruta graveolens* 12x (15.0 ± 2.23 mm), *Thuja occidentalis* 6x (10.8 ± 0.83 mm), and *Zingiber officinale* 3x and 6x (13.0 ± 2.73 mm and 11.4 ± 2.30 mm, respectively), outperforming the control.

Patil et al. (2018) investigated the in-vitro antifungal activity of various potencies of homeopathic medicines against the plant pathogen *Fusarium moniliforme.* The study utilized Silicea, Mercurius Solubilis, and Lachesis in different potencies, employing both the agar well diffusion assay and minimum inhibitory concentration (MIC) techniques. Positive controls, including Bavistin (Carbendazim at 0.1 mg/ml) and Terbinafine (0.01 mg/ml), along with a vehicle control of 90% ethyl alcohol, were used for comparison. The agar well diffusion assay revealed significant inhibitory effects, with Silicea 12C producing a zone of inhibition measuring 13.5 ± 3.5 mm, Mercurius Solubilis 6C, 12C, and 1M exhibiting zones of 12.5 ± 5.5 mm, 15 ± 2.0 mm, and 11.5 ± 1.5 mm respectively, and Lachesis 6C, 30C, 200C, and 1M yielding zones of 15 ± 4.5 mm, 17.5 ± 0.5 mm, 15 ± 3.0 mm, and 16 ± 4.0 mm, respectively. Among the tested potencies, Silicea (12C), Mercurius Solubilis (6C, 12C, 1M), and

Lachesis (6C, 30C, 200C, 1M) exhibited the highest levels of growth inhibition against *Fusarium moniliforme,* highlighting their potential antifungal efficacy.

Shinde et al. (2018) conducted an in-vitro study to evaluate the anti-Candida activity of various homeopathic medicines against *Candida albicans*. The research aimed to screen multiple homeopathic remedies and their potencies for their inhibitory effects on *C. albicans*. The study tested Azadirachta indica, Cinchona officinalis, Zincum metallicum, Iodium, Selenium, Sulphur, Acidum Benzoicum, Phosphorus, Acidum Sulphuricum, and Zingiber officinale at potencies of 6C, 12C, 30C, 200C, and 1M. Results indicated that specific potencies—Iodium 6C, Zincum metallicum 30C, Selenium 6C, Zingiber officinale 6C, Cinchona officinalis 12C, Acidum Benzoicum 12C, Phosphorus 6C, Azadirachta indica 6C, Acidum Sulphuricum 6C, and Sulphur 6C—exhibited significant inhibitory activity against *C. albicans.* Additionally, these medicines demonstrated the ability to inhibit germ tube formation in *C. albicans*, underscoring their potential antifungal properties.

Chinche et al. (2018) conducted a study investigating the in vitro antifungal efficacy of homoeopathic medicines against *Ashbya gossypii,* a fungal pathogen responsible for stigmatomycosis in cotton and various other crops worldwide. The primary objective of their research was to evaluate the inhibitory potential of different homoeopathic remedies and their potencies against *A. gossypii.* Using the agar well diffusion method and minimum inhibitory concentration (MIC) assay, the researchers tested Calcarea Carbonica, Lachesis Mutus, Mercurius Solubilis, Silicea, and Thuja occidentalis across potencies of 6C, 12C, 30C, 200C, 1M, 10M, and CM. The findings revealed notable antifungal activity, particularly with Calcarea Carbonica at 12C, 30C, and 1M; Mercurius Solubilis at 30C; and Thuja occidentalis at 1M. Additionally, most of the potencies of these homoeopathic medicines exhibited inhibition in the MIC assay against *A. gossypii.*

2.2 Effect of Homeopathic Medicine on fungal spore production

Toledo et al. (2016) investigated the potential of homeopathic formulations to control fungal pathogens and reported significant reductions in sporulation rates. Their study demonstrated that Propolis, when applied at potencies of 6CH, 30CH, and 60CH, inhibited sporulation by 65.5%, 58.5%, and 52.1%, respectively. Similarly, the remedies IAS and ICF were effective in reducing spore production, achieving

sporulation inhibition of up to 52.52% and 56.9%. These findings underscore the potential of homeopathy as a viable alternative for reducing fungal sporulation, thereby limiting the spread of fungal pathogens.

Balbi-Peña et al. (2016) examined the effects of homeopathic treatments on *Alternaria solani*, a significant fungal pathogen in crops. Their study revealed that Sulphur inhibited sporulation by approximately 50%, effectively curbing the disease's progression. This suggests that homeopathic remedies can play an essential role in managing fungal diseases by restricting spore production and preventing pathogen dissemination.

Franzener et al. (2017) studied the impact of homeopathic treatments on the development of *Alternaria brassicae*. They found that Cina, Calcarea carbonicum and Nux vomica reduced sporulation by 62.34%, 55.71% and 46.89%, respectively, further validating the efficacy of homeopathic remedies in suppressing fungal reproduction across different pathogen species.

Gondim et al. (2014) evaluated the effectiveness of homeopathic remedies in controlling *Colletotrichum musae*. They observed that treatments such as Arsenicum album, Sulphur, and Thuja occidentalis significantly inhibited spore production in vitro. Their research supported the hypothesis that homeopathy could serve as a sustainable and eco-friendly alternative to synthetic fungicides for managing anthracnose in bananas.

Patil and Pawar (2018) tested homeopathic treatments against *Colletotrichum* species and reported that Arsenicum album and Sulphur inhibited sporulation by 58% and 53%, respectively. This highlights the potential of these remedies in controlling fungal pathogens, particularly in horticultural crops.

Raj et al. (2017) focused on the biological control of *Colletotrichum gloeosporioides* using homeopathic remedies. Their study identified Calcarea carbonica as a promising treatment, achieving 55% sporulation inhibition.

Katoch et al. (2015) conducted an experiment on evaluation of homeopathic remedies in controlling fungal pathogens affecting horticultural crops. They highlighted the effectiveness of Belladonna in controlling *Colletotrichum gloeosporioides*, where it reduced sporulation and spore germination by 64% at 30CH potency.

Sharma et al. (2020) investigated the effect of ten homeopathic medicine with two concentration (500 ppm and 1000 ppm) against *Fusarium oxysporum* and found that it inhibited the sporulation of *Fusarium oxysporum* by 62.7% when applying thuja occidentalis at 1000 ppm, followed by Kali iodatum (59.3% at 1000 ppm), Arsenicum album at same concentration (58.6%).

CHAPTER III

MATERIALS AND METHODS

3.1 Experimental site

This experiment was conducted in the Plant Pathology Laboratory at Agrotechnology Discipline in Khulna University, Khulna.

3.2 Sample collection

Banana (*Musa paradisiaca)* having anthracnose symptoms were collected from Gollamari local market, Gollamari, Khulna. Additionally, Selected seventeen homeopathic medicines with three distinct dynamization were collected from different homeopathic dispensaries across Khulna city, Bangladesh.

3.3 Isolation and Identification of fungi

Banana exhibiting anthracnose symptoms, characterized by sunken lesions, were collected and processed for analysis. The samples were initially rinsed with tap water to eliminate dust and surface contaminants. Diseased sections of the fruit, including adjacent healthy tissue, were excised into small pieces using a sterilized blade. These sample were then sterilized by immersing them in a 0.1% sodium hypochlorite solution for 20-30 seconds with sterilized forceps. To ensure the removal of any remaining trace of sodium hypochlorite solution, the pieces were thoroughly rinsed three times in sterilized water. Excess moisture was carefully blotted using pre-sterilized blotting paper. Sterilization of petri dishes were performed in a hot air oven at 160-180°C for two hours. Potato Dextrose Agar (PDA) was prepared as the culture medium, and 20 ml of the sterilized medium was poured into the petri dishes under a laminar airflow hood. The prepared samples were transferred onto the PDA medium using sterilized forceps. Each dish was labeled with the date of isolation using a marker and sealed with electrical tape. The cultures were incubated at 25-28°C and monitored for fungal growth at 24-hour intervals. After five days of incubation, fungal hyphal tips were subcultured onto fresh PDA medium using a sterilized 2.5 mm mycelial block cutter for further purification, identification, and maintenance of cultures. The purified fungal cultures were examined under a compound microscope at 10X magnification. Observations revealed oval-shaped, hyaline conidia with rounded ends, which were consistent with the morphological characteristics of

Colletotrichum musae (Barnett and Barry, 2002). The confirmed fungal cultures were subsequently stored at 4°C in a refrigerator for preservation.

Figure 3.3.1: Five days old pure culture of *C. musae* on PDA media

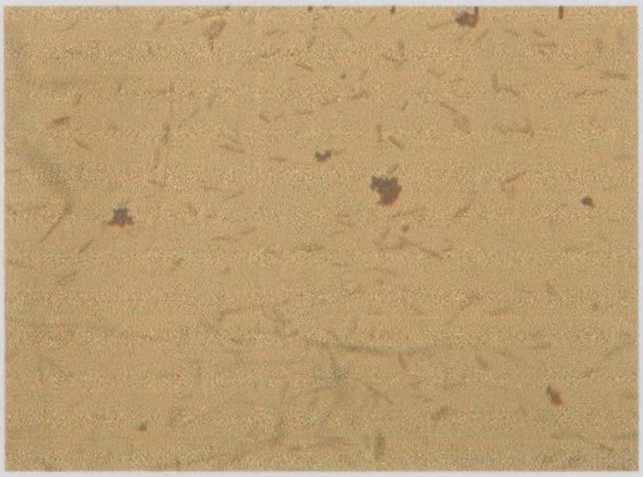


Figure 3.3.2: Conidia of *C. musae* under compound microscope with 10X magnification

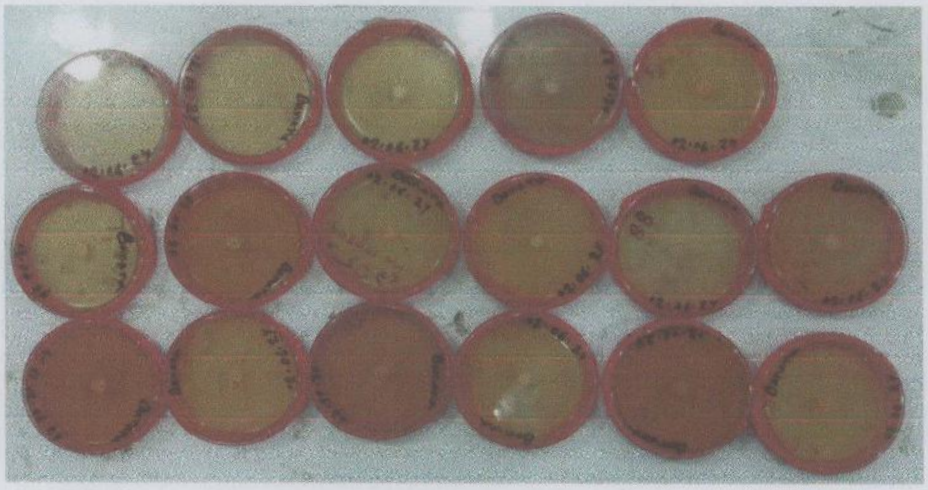


Figure 3.3.3: Subculture of *Colletotrichum musae*

3.4 Pathogenecity test

The pathogen was identified through an analysis of its morphological and cultural traits, alongside its pathogenic interactions with the host. To conduct the pathogenicity test, healthy bananas were surface-sterilized by immersing them in 70% ethanol for 30 seconds, followed by the removal of excess ethanol using sterilized cotton. Using flame-sterilized needles, minor injuries were gently inflicted on specific areas of the banana samples. A conidial suspension, prepared from the pure culture of *Colletotrichum musae,* was then applied by immersing the samples into the suspension. Subsequently, the treated bananas were placed in separate sterilized polybags and incubated at 25 ± 1°C. The samples were monitored regularly for the appearance of characteristic symptoms.

3.5 Selection of homeopathic medicines

Seventeen homeopathic medicines, each prepared in three potencies (Potency is the strength of a homeopathic remedy), 6CH, 30CH, and 200CH were selected based on their antifungal activities observed in various experiments (Chaurasia and Vyas, 2020).

Figure 3.5.1: Homeopathic medicine used in this study

The abbreviation "CH" represents "Centesimal Hahnemannian," a dilution technique devised by Samuel Hahnemann, the founder of homeopathy.

6CH: The potency 6CH means, the substance has been diluted six times in the 1:100 ratio.

30CH: The substance has been diluted thirty times in the 1:100 ratio.

200CH: The substance has been diluted two hundred times in the 1:100 ratio.

Table 3.5.1: List of homeopathic medicine used in this study

Name of homeopathic medicines	**Potencies**	**Concentration**
Arsenicum album	6CH, 30CH and 200CH	3000ppm
Calcaria sulp		
Selenium		
Cina		
Nux vomica		
Siliciea		
Arnica montana		
Calcarea carbonicum		
Carbo vegetabilis		
Ferrum metallicum		
Natrum muriaticum		

Phosphorus		
Sulphur		
Sepia officinale		
Belladona		
Kali iodatum		
Thuja occidentalis		

3.6 Preparation of concentration of homeopathic medicines and mixing with PDA medium

3000 ppm concentration was prepared for each potency of every homeopathic medicine. Subsequently, 60 µL of each medicine was individually added into petridishes containing 20 mL of PDA medium (where 1 ppm equals 1 µL/L) under sterile conditions within a laminar airflow cabinet. The mixture was thoroughly mixed through manual shaking before being dispensed into sterilized petridishes and allowed to solidify. Each plate was considered as a replication. For the control treatment, no medicine was added to the medium. All procedures were conducted under aseptic conditions, with three replications performed for each potency.

3.7 Evaluation of homeopathic medicines against *C. musae*

3.7.1 Inoculation and incubation

The evaluation of homeopathic medicine was conducted using the Poison Food Technique (Baviskar and Suryawanshi, 2015). Following the incorporation of the medicines, 2.5 mm mycelial disc derived from a five days old pure culture of *Colletotrichum musae* was inoculated at the center of the plates. These plates were then incubated at a controlled temperature of 28±1°C. After an incubation period of eight days, data on mycelial growth rate (measured in mm) were recorded.

Figure 3.7.1.1: Incubation of *C. musae* in growth chamber

3.7.2 Measurement of radial growth and calculation of percent inhibition

The radial growth of mycelium was monitored at two-day intervals. Once the mycelial growth reached the edge of the control plates, two crosswise radial measurements were taken for each plate. The average of these two diameters was then calculated and analyzed.

Percentage inhibition of mycelial growth was calculated by using the following formula (Alam et al., 2016):

$$\text{Percent of inhibition} = \frac{X-Y}{X} \times 100$$

Where,

X= Average growth of *Colletotrichum musae* in control petridishes

Y= Average growth of *Colletotrichum musae* in each medicine treated petridishes

3.7.3 Measurement of sporulation in *Colletotrichum musae*

Conidia were harvested 15 days after inoculation by suspending the spores in 20 mL of sterile distilled water added to the culture medium. The surface of the medium was gently scraped with a sterilized slide to release the spores into the suspension. The resulting spore suspension was filtered through filter paper to eliminate debris. Subsequently, lactophenol blue and Tween 80 were added to the suspension. Then 50 µL of suspension was taken with micropipette and placed on slide. Sporulation was assessed by quantifying the conidia produced per Petri dish, which included

treatments with various homeopathic drugs at different dynamizations as well as a control. The spores were counted under a microscope at 10X magnification. Then, the spores were converted to spores per milliliter (spores/mL).

Percentage inhibition of sporulation was calculated by using the following formula (Alam et al., 2016):

$$\text{Percent of inhibition} = \frac{A-B}{A} \times 100$$

Where,

A= Average sporulation of *Colletotrichum musae* in control petridishes

B= Average sporulation of *Colletotrichum musae* in each medicine treated petridishes

3.8 Experimental design and data analysis

The experiment was conducted following Completely Randomized Design (CRD) with three replications. Statistical analysis of the data was performed using Statistix software version 10, and the means were compared using Tukey's HSD test at a 95% confidence level.

CHAPTER IV

RESULT AND DISCUSSION

4.1 Effect of different homeopathic medicine for mycelial growth of *Colletotrichum musae*

Table 4.1.1: Effect of homeopathic remedies at different potencies on radial mycelial growth

Treatments	Potency	Mycelial Growth (mm)			
		2 days	4 days	6 days	8 days
Arsenicum album	6CH	14.56±0.34 j-q	24.07±0.33 xy	32.58±0.18 v	38.02±0.26 z
	30CH	11.65±0.06 wx	24.75±0.11 xy	36.53±0.23 r-u	41.44±0.11 x
	200CH	14.62±0.38j-q	30.82±0.11 l-p	41.73±0.38 i-l	48.63±0.26 g-j
Calcaria sulp	6CH	15.22±0.09 e-l	28.65±0.35 r-v	37.67±0.28 q-s	42.78±0.31 r-u
	30CH	10.91±0.24 x	22.51±0.12 z	39.32±0.26 op	43.67±0.09 q-s
	200CH	12.48±0.11u-w	29.56±0.22 o-s	35.66±0.13 u	43.47±0.25 q-s
Selenium	6CH	15.51±0.06 e-k	26.32±0.20 x	33.59±0.08 v	40.03±0.34 y
	30CH	13.95±0.08 m-s	29.44±0.18 p-t	39.19±0.23 op	42.69±0.21 r-w
	200CH	16.06±0.07 c-g	36.06±0.20 b	40.62±0.18 l-n	47.59±0.11 i-m
Cina	6CH	14.16±0.16 l-r	31.21±0.19 klm	42.76±0.22 g-i	47.01±0.40 lm
	30CH	13.68±0.14 o-t	29.58±0.36 o-s	38.16±0.13 pq	41.52±0.15 wx
	200CH	15.23±0.29 e-l	27.48±0.15 vw	43.54±0.16 d-g	48.49±0.21 h-k
Nux vomica	6CH	17.61±0.43 b	31.95±0.12 g-m	41.69±0.33 i-l	46.46±0.13 m-o
	30CH	14.71±0.11 i-q	31.65±0.19 i-m	36.71±0.19 r-u	42.73±0.25 r-v
	200CH	15.79±0.44 c-i	31.05±0.14 l-n	40.14±0.11 m-o	46.65±0.09 mn
Siliciea	6CH	15.02±0.13 f-m	30.83±0.10 l-o	39.56±0.21 no	44.64±0.15 pq
	30CH	14.89±0.17 h-n	27.57±0.31 u-w	37.84±0.18 qr	43.06±0.11 r-t
	200CH	16.23±0.26 c-e	33.22±0.24 e-g	45.71±0.27 b	52.62±0.07 b
Arnica montana	6CH	14.84±0.27 i-o	32.14±0.32 g-l	41.04±0.25 k-m	45.30±0.12 op
	30CH	15.71±0.13 c-j	26.34±0.25 x	41.03±0.17 k-m	46.56±0.09 l-n
	200CH	14.95±0.24 g-m	26.99±0.28 w	43.17±0.34 f-h	49.39±0.19 e-h
Calcarea carbonicum	6CH	16.10±0.04 c-g	33.02±0.39 f-i	44.77±0.11 b-d	50.34±0.14 de
	30CH	14.70±0.11 i-q	32.53±0.28 g-k	42.81±0.21 g-i	46.75±0.18 lm

	200CH	13.78±0.12 n-t	27.00±0.21 w	45.68±0.11 b	51.70±0.20 bc
Carbo vegetabilis	6CH	14.55±0.13 j-q	29.45±0.34 o-t	45.35±0.13 bc	52.12±0.07 bc
	30CH	12.78±0.08 t-w	35.78±0.28 bc	44.23±0.12 c-f	49.34±0.05 e-h
	200CH	16.76±0.04 b-d	34.77±0.15 b-d	45.41±0.13 bc	52.25±0.22 bc
Ferrum metallicum	6CH	16.01±0.15 c-h	30.56±0.51 m-q	37.63±0.15 q-s	43.79±0.24 qr
	30CH	10.90±0.26 x	27.56±0.21 u-w	36.98±0.16 q-t	47.55±0.18 i-m
	200CH	14.36±0.07 k-r	28.30±0.20 s-v	39.35±0.11 n-p	47.65±0.56 i-l
Natrum muriaticum	6CH	13.68±0.16 p-t	29.15±0.54 r-t	36.89±0.22 q-u	42.91±0.07 r-u
	30CH	12.69±0.04 t-w	36.13±0.24 b	43.71±0.20 d-g	49.86±0.32 ef
	200CH	14.46±0.27 k-q	34.11±0.25 def	44.59±0.15 b-e	49.82±0.09 ef
Phosphorus	6CH	16.03±0.11 c-h	28.35±0.14 r-v	36.32±0.18 tu	42.54±0.13 s-x
	30CH	13.22±0.26 r-v	31.62±0.16 j-m	39.66±0.19 no	45.34±0.10 op
	200CH	16.04±0.11 c-h	31.69±0.13 h-m	41.93±0.31 h-k	48.71±0.12 f-i
Sulphur	6CH	17.58±0.19 b	31.84±0.20 g-m	41.39±0.19 j-m	47.42±0.05 k-m
	30CH	14.59±0.09 j-q	32.91±0.42 f-j	45.34±0.26 bc	51.21±0.16 cd
	200CH	14.43±0.09 k-q	25.23±0.05 xy	42.47±0.13 g-j	47.14±0.07 lm
Sepia officinale	6CH	12.94±0.23 s-v	28.41±0.16 r-v	36.57±0.10 r-u	42.24±0.33 t-x
	30CH	14.16±0.05 l-r	36.11±0.32 b	43.51±0.21 d-g	52.14±0.02 bc
	200CH	14.78±0.14 i-p	29.72±0.09 n-r	43.35±0.28 e-g	49.71±0.12 e-g
Belladonna	6CH	15.64±0.07 d-j	33.06±0.25 f-h	41.41±0.11 j-m	47.40±0.23 k-m
	30CH	12.47±0.13 uvw	28.09±0.38 t-v	40.29±0.58 m-o	47.51±0.09 j-m
	200CH	16.82±0.22 bc	31.24±0.23 k-m	45.72±0.25 b	51.42±0.05 cd
Kali iodatum	6CH	16.18±0.16 c-f	29.14±0.19 q-t	39.32±0.26 op	45.48±0.16 n-p
	30CH	10.78±0.06 x	23.59±0.11 z	35.63±0.16 u	41.75±0.14 u-x
	200CH	13.57±0.45 q-u	27.28±0.14 vw	35.50±0.27 s-u	45.23±0.25 p
Thuja occidentalis	6CH	14.44±0.07 k-q	28.94±0.37 r-u	36.49±0.17 s-u	41.60±0.16 v-x
	30CH	12.22±0.15 vw	34.47±0.16 c-e	41.66±0.18 i-l	45.56±0.32 n-p
	200CH	14.67±0.43 i-q	26.24±0.20 x	37.54±0.08 q-t	47.29±0.46 lm
Control	-	23.77±0.17 a	41.32±0.13 a	59.41±0.18 a	74.87±0.38 a
CV%		2.35	1.47	0.74	1.36

Values with different letters are significantly differed at 5% level of significance according to Tukey's HSD Test

Table 4.1.1 revealed the efficacy of seventeen homeopathic medicines at three potencies (6 CH, 30 CH, and 200 CH) under *in vitro* conditions. The results demonstrated a significant inhibition of the mycelial growth of *Colletotrichum musae* across all tested potencies compared to the control.

Among all treatments, the least radial mycelial growth, measuring 10.78 mm, was observed in plates treated with Kali iodatum at 30 CH, whereas Nux vomica at 6 CH recorded the highest radial growth of 17.61 mm at 2 days after inoculation (DAI).

At 4 days after inoculation (DAI), the lowest mycelial growth was achieved with Calcaria sulp at 30 CH, measuring 22.51 mm, while Natrum muriaticum at 6 CH demonstrated the largest mycelial growth of 36.13 mm.

Arsenicum album at 6 CH exhibited the minimal radial growth of mycelium at 32.58 mm, in contrast to Silicea at 200 CH, which showed highest value of 45.71 mm at 6 days after inoculation (DAI).

In Arsenicum album, the maximum mycelial growth was observed at 200 CH (48.63 mm), followed by 30 CH (41.44 mm) and 6 CH (38.02 mm) at 8 days after inoculation (DAI). For Calcarea sulp, the highest mycelial growth was recorded at 6 CH (42.78 mm), while the lowest was at 200 CH (43.47 mm), with 30 CH showing a growth of 43.67 mm. The minimum mycelial growth in Selenium was observed at 6 CH (40.03 mm), whereas at 30 CH and 200 CH, the values were 42.69 mm and 47.59 mm, respectively. Cina exhibited the lowest mycelial growth at 30 CH (41.52 mm), whereas growth at 6 CH and 200 CH was slightly higher, measuring 47.00 mm and 48.49 mm, respectively, which was significantly different from each treatments.

In Nux vomica, the minimum radial growth of the mycelium was observed at 30 CH (42.73 mm), while 6 CH and 200 CH exhibited nearly similar growth values of 46.46 mm and 46.65 mm, respectively. Silicea displayed maximal mycelial growth at 200 CH (52.62 mm), while 6 CH and 30 CH showed 44.64 mm and 43.06 mm, respectively. The radial growth of Arnica montana was recorded as 45.30 mm, 46.56 mm, and 49.39 mm at 6 CH, 30 CH, and 200 CH, respectively. For Calcarea carbonica, the maximum mycelial growth was 51.70 mm at 200 CH, while the minimum was observed at 30 CH (46.75 mm), and 6 CH showed a growth of 50.34 mm, which was significantly different from other treatments.

The radial growth of Carbo vegetabilis at 6 CH, 30 CH, and 200 CH was measured at 52.10 mm, 49.34 mm, and 52.25 mm, respectively. In Ferrum metallicum, the highest mycelial growth was observed at 200 CH (47.65 mm), with the lowest at 6 CH (43.79 mm), while 30 CH showed a growth of 47.55 mm. For Natrum muriaticum, the radial growth of the mycelium was 42.91 mm, 49.86 mm, and 49.81 mm at 6 CH, 30 CH, and 200 CH, respectively. Phosphorus exhibited the highest radial growth at 200 CH (48.71 mm) and the lowest at 6 CH (42.54 mm). The mycelial growth observed in Sulphur was 47.70 mm, 51.21 mm, and 47.14 mm at 6CH, 30CH, and 200CH potencies, respectively, each treatments were significantly different.

In case of Sepia officinalis, the highest mycelial expansion of 52.14 mm was recorded at 30CH, followed by 49.71 mm at 200CH and 42.24 mm at 6CH. In the case of Belladonna, the maximum radial growth of 51.42 mm was noted, with nearly identical results of 47.40 mm and 47.51 mm at 6CH and 30CH, respectively. The mycelial development in Kali iodatum varied, measuring 45.48 mm, 41.75 mm, and 45.23 mm across 6CH, 30CH, and 200CH, respectively. A notable result for Thuja occidentalis was the highest growth of 47.29 mm at 200CH, while the lowest measurement, 41.60 mm, was observed at 6CH; the intermediate value at 30CH reached 45.56 mm. The mycelial growth was 74.87 mm in control plate.

Among all treatments, the least radial growth of 38.02 mm was recorded in plates treated with Arsenicum album at 6CH potency, followed by Selenium at 6CH (40.03 mm), Arsenicum album at 30CH (41.44 mm), Cina at 30CH (41.52 mm) and Thuja occidentalis at 6CH (41.60 mm). Conversely, the highest mycelial growth, 52.62 mm, was achieved with Silicea at 200CH.

Previous studies had highlighted the effectiveness of various homeopathic treatments in controlling fungal pathogens and enhancing plant health. This present study supported the findings of Babli et al. (2022) and reported that the minimum mycelial growth was observed with Arsenicum album (43.67 mm), followed by Calcarea carbonicum (46.00 mm), Cina (51.00 mm), Nux vomica (54.67 mm), Selenium (57.67 mm), Phosphorus (59.33 mm), Borex (62.33 mm), Arnica montana (64.33 mm), and Siliciea (67.00 mm) at a concentration of 500 ppm. Similarly, Toledo et al. (2016) also demonstrated the suppression of mycelial growth in Sulphur and Phosphorus 100 CH compared to both untreated and treated controls.

For *Ascochyta blight* of chickpea caused by *Ascochyta rabiei,* the recorded mycelial growths at 50 ppm were 57.73 mm for sulfur, 28.06 mm for T. occidentalis, 27.87 mm for A. montana, and 38.87 mm for Silicea (Kumar et al., 2023). Hanif and Dawar (2016) evaluated the fungicidal activity of homoeopathic formulations of Thuja occidentalis, Arsenicum album and Arnica montana in 30 CH potencies against root rot disease in non-leguminous plants. Their results, derived from both *in vitro* and *in vivo* trials, demonstrated significant antifungal efficacy.

Reis and Ottoni (2021) also reported mycelial growth measurements of 53.5 mm for Calcarea carbonicum and 48.0 mm for Arsenicum album at a dynamization level of 6 CH which supported the current study. In the case of Phomopsis blight of brinjal caused by *Phomopsis vexans*, sulfur, T. occidentalis, A. montana, and Silicea at 50 ppm resulted in mycelial growths of 46.77 mm, 47.50 mm, 48.20 mm, and 50.50 mm, respectively (Kumar et al., 2023). Ikram and Dawar (2015) also demonstrated the antiviral efficacy of homoeopathic drugs against both animal and plant viruses.

Arsenicum album (6CH and 200CH) exhibited the highest inhibitory activity against *Phomopsis vexans* under *in vitro* condition (Sheikh et al., 2006) that is similar with the current research. Gupta et al. (2015) observed significant suppression of Fusarium wilt in tomato plants following treatment with Arsenicum album. Homeopathic medicine not only inhibit the fungal growth but also minimizes economic losses by mitigating the impact of pathogenic fungi (Martha et al., 2003).

Damin et al. (2015) investigated the radial growth of mycelium under the influence of various homoeopathic treatments and reported that Siliciea exhibited a growth of 52.6 mm at 24 CH, Calcarea carbonicum reached 50.8 mm at 30 CH, Kali iodatum recorded 44.2 mm at 100 CH, Phosphorus achieved 54.0 mm at 3 CH, Arsenicum album measured 41.4 mm at 200 CH, Sulphur reached 51.6 mm at 100 CH, and Thuja occidentalis demonstrated 50.8 mm at 200 CH. In a study by Subhani (2015), reported the application of homeo-fungicides such as Rigorous, Ventage, and Vampire resulted in radial mycelial growth measurements of 39 mm, 23 mm, and 44 mm, respectively, at a concentration of 20 ppm.

The antifungal efficacy of Arsenicum album, Cina, Thuja occidentalis were further corroborated by Singh et al. (2000). Singh and Gupta (2005), who also demonstrated the antiviral activity of homeopathic treatments against animal and plant viruses.

Saxena et al. (2007) reported the inhibition of 22 fungal genera on okra seeds treated with T. occidentalis. Hanif and Dawar (2015) found that seed treatment and soil drenching with Arnica montana (30CH) significantly suppressed *R. solani, Fusarium spp.,* and *M. phaseolina* while promoting plant growth at 100%, followed by 75% and 50% v/v concentrations. Arnica montana at potencies of 3, 6, and 12 CH was shown to enhance plant growth (Bonfim et al., 2010).

The efficacy of sulfur, Thuja occidentalis, Silicea and Arnica montana was evaluated at 50 ppm against *Fusarium oxysporum f. sp. lycopersici.* The recorded radial mycelial growths were 43.5 mm, 45.61 mm, 61.12 mm, and 28.42 mm, respectively (Kumar et al., 2023). This aligns with the present study, where Silicea exhibited the highest mycelial growth. Similarly, when tested against early blight disease of tomato caused by *Alternaria solani,* the mycelial growths for sulfur, T. occidentalis, A. montana, and Silicea at 50 ppm were found to be 36.0 mm, 26.66 mm, 32.44 mm, and 47.13 mm, respectively.

Gupta et al. (2015) reported that arsenicum album in 30CH and 200CH potencies was effective against *Aspergillus flavus*, the causative agent of cutaneous aspergillosis. Asha et al. (2014) demonstrated that all potencies of Arsenicum album (Q, 30C, 200C, 1M, 10M, 50M) significantly inhibited the growth of *Bipolaris spp.,* followed by *Curvularia spp., Exserohilum spp.,* and *A. flavus.* Cina, Phosphorus, kali iodatum, Thuja occidentalis and Natrum muriaticum were found to be highly effective against *Fusarium spp.* (Hussain et al., 2000).

Against Septoria leaf spot of tomato caused by *Septoria lycopersici,* the mycelial growths at 50 ppm concentration were 43.60 mm for sulfur, 45.61 mm for T. occidentalis, 61.12 mm for A. montana, and 28.42 mm for Silicea (Kumar et al., 2023) Khanna and Chandra (1992) observed that Kali iodatum (149CH) and Thuja occidentalis (87CH), when applied under pre- and post-harvest conditions, significantly controlled tomato fruit rot caused by *Fusarium roseum.*

Arsenicum album demonstrated antifungal activity against *Aspergillus parasiticus, Saccharomyces cerevisiae, Macrophomina phaseolina, Fusarium solani, Candida albicans,* and *Trichophyton rubrum* (Jahan et al., 2010). In 6CH and 30CH potencies were effective against *F. solani, Rhizoctonia solani,* and *M. phaseolina* (Dawar et al., 2007). Similarly, Thuja occidentalis and Kali iodatum at the same concentration

significantly reduced the growth of these pathogens. Gupta and Srivastava (2002) noted that T. occidentalis in 30CH and 2000CH potencies were effective against *A. flavus*, which was similar with the present study.

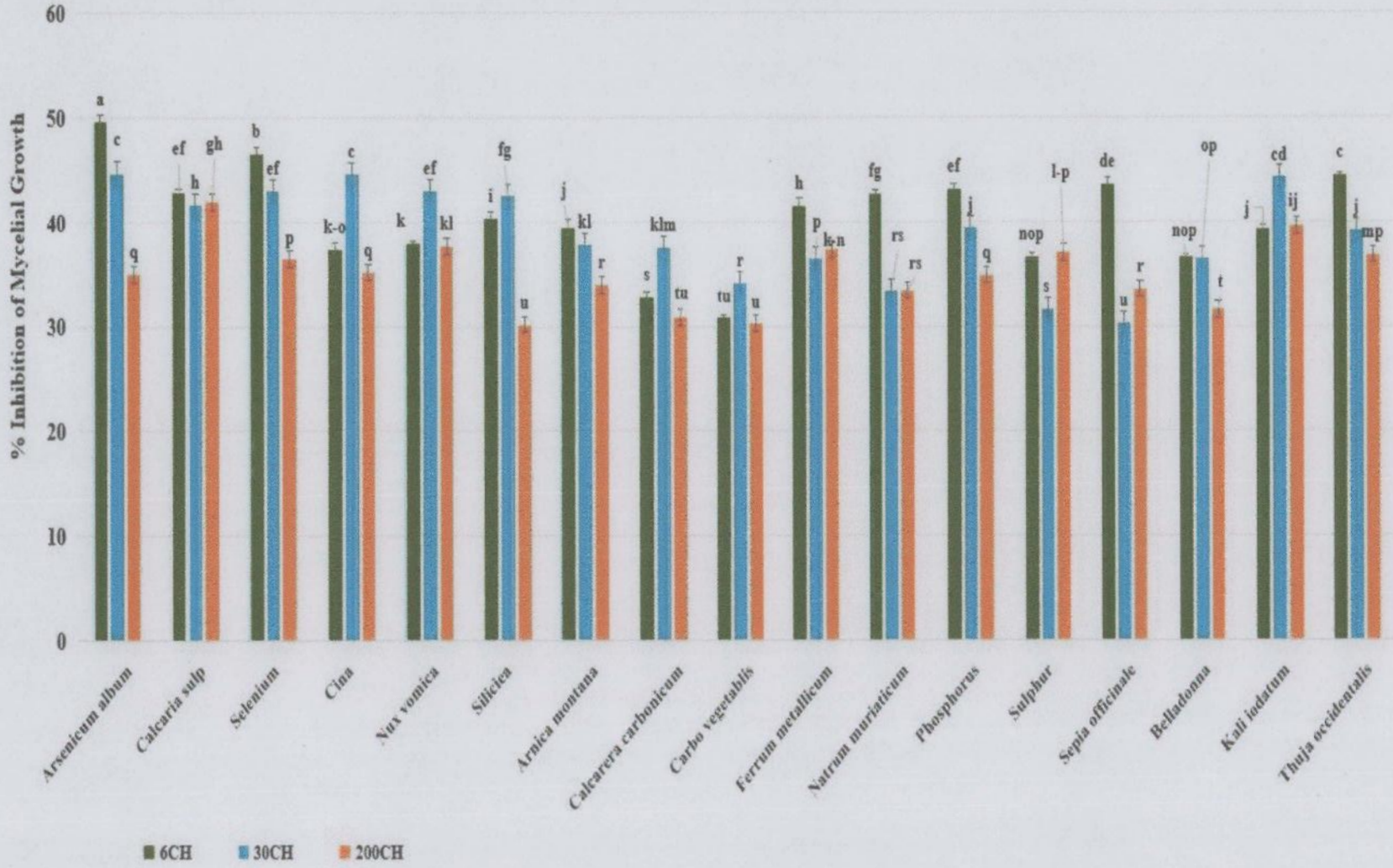


Figure 4.1.1: Effect of different homeopathic medicines on growth inhibition (%)

This figure revealed that Arsenicum album exhibited the highest mycelial growth inhibition at 6 CH (49.66%), followed by 30 CH (44.65%) and 200 CH (35.04%) at 8 days after inoculation (DAI). For Calcarea sulp, the maximum inhibition was observed at 6 CH (42.86%), with the lowest inhibition at 30 CH (41.67%) and 200 CH showing an intermediate value of 41.93%. Similarly, Selenium demonstrated the greatest inhibitory effect at 6 CH (46.52%), while lower values were recorded at 30 CH (42.97%) and 200 CH (36.44%).

Cina showed the highest inhibition at 30 CH (44.54%), with reduced effects observed at 6 CH (37.33%) and 200 CH (35.23%). Nux vomica achieved maximum mycelial growth suppression at 30 CH (42.92%), whereas similar values were recorded at 6 CH (37.94%) and 200 CH (37.69%). Silicea displayed its greatest inhibitory potential at 30 CH (42.48%), with reduced suppression at 6 CH (40.37%) and the lowest at 200 CH (30.23%). For Arnica montana, the inhibition rates were 39.49%, 37.81%, and 34.03% at 6 CH, 30 CH, and 200 CH, respectively, which was significantly different from other treatments.

The highest mycelial growth inhibition for Calcarea carbonica was observed at 30 CH (37.55%), while the lowest occurred at 200 CH (30.94%), and a moderate level of inhibition (32.76%) was noted at 6 CH. The inhibition rates for Carbo vegetabilis were recorded as 30.83% at 6 CH, 34.097% at 30 CH, and 30.25% at 200 CH.

For Ferrum metallicum, the maximum inhibition occurred at 6 CH (41.51%), while the lowest was observed at 30 CH (36.49%), with an intermediate value of 37.35% at 200 CH. Similarly, Natrum muriaticum exhibited inhibition rates of 42.68%, 33.41%, and 33.45% at 6 CH, 30 CH, and 200 CH, respectively.

Phosphorus displayed its highest inhibition at 6 CH (43.17%), while the lowest was observed at 200 CH (34.94%). For Sulphur, the recorded inhibition rates were 36.68%, 31.59%, and 37.03% at 6 CH, 30 CH, and 200 CH, respectively. In the case of Sepia officinalis, the lowest mycelial inhibition was observed at 30 CH (30.36%), followed by 33.60% at 200 CH, and the highest suppression was recorded at 6 CH (43.58%). All treatments were significantly different in case of Sepia officinalis.

Belladonna exhibited maximum inhibition of radial growth at 6 CH (36.68%), with comparable values at 30 CH (36.54%) and 200 CH (31.61%). Kali iodatum achieved

inhibition rates of 39.25%, 44.23%, and 39.53% at 6 CH, 30 CH, and 200 CH, respectively. A notable result was observed for Thuja occidentalis, with the highest inhibition rate recorded at 6 CH (44.44%), while the lowest was noted at 200 CH (36.83%), and an intermediate value of 39.14% was achieved at 30 CH, these treatments were significantly different from each other.

Among all treatments, the highest inhibition rate of radial mycelial growth (49.66%) was recorded in plates treated with Arsenicum album at 6 CH, followed by Selenium at 6CH (46.52%), Arsenicum album at 30CH (44.65%), Cina at 30CH (44.54%) and Thuja occidentalis at 6CH (44.44%). Conversely, the lowest inhibition rate (30.23%) was observed with Silicea at 200 CH.

According to Babli et al. (2022), the inhibition of mycelial growth varied across different homeopathic treatments, with Cina, Silicea, Borex, Selenium, Nux vomica, Arsenicum album, Phosphorus, Arnica montana, and Calcarea carbonicum showing inhibition rates of 42.70%, 29.96%, 35.21%, 38.58%, 48.31%, 50.94%, 33.33%, 27.72%, and 24.72%, respectively that supported the current research. Patil and Suryawanshi (2014) reported that the inhibition percentages of Thuja occidentalis, Sulphur, Kali iodatum and Selenium were 48.89%, 26.67%, 21.12%, and 46.67%, respectively, at 30CH potency. Furthermore, Nux vomica and Arnica montana exhibited inhibition rates of 50.0% and 37.78%, respectively, at 200CH potency.

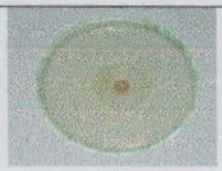
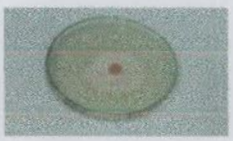
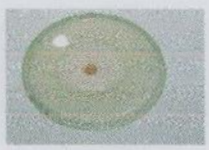
Lorenzetti et al. (2016) demonstrated that the application of *Thuja occidentalis* medicine effectively controlled *Macrophomina phaseolina*, the pathogen responsible for grey stem in soybean, reducing the area under the mycelial growth curve by 14% and 15% at 12CH and 48CH dynamizations, respectively. Leonel and Barros (2013) observed that Arsenicum album at 6CH significantly reduced coffee rust caused by *Hemileia vastatrix* for up to three consecutive months. Toledo et al. (2009) evaluated the efficacy of Arsenicum album against *Alternaria solani*, the causal agent of early blight in tomatoes, and reported a reduction in disease severity by 48.82% and 56.47% at 12CH and 60CH dynamizations, respectively. Sinha and Singh (2003) highlighted Sulphur's ability to inhibit the growth of *Aspergillus parasiticus*, a key aflatoxin producer, by 100% at 200CH.

Promising results were also observed for selenium, with Barkund et al. (2018), reporting its effectiveness in inhibiting *Gibberella fujikuroi* at 30CH and 200CH

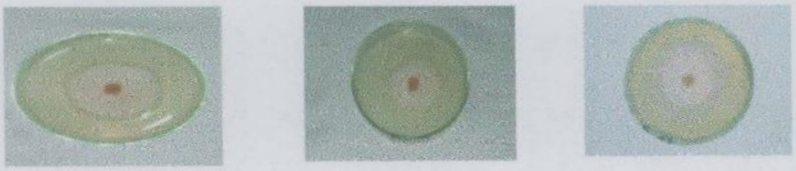
potencies. Chinche et al. (2018) demonstrated selenium's inhibitory activity against *Eremothecium gossypii* at 12CH, 30CH, and 1M dynamizations. Gama et al. (2015) reported varying inhibition percentages for other homeopathic treatments, including Sulphur (6.4% at 5CH and 36% at 9CH), Phosphorus (14% at 9CH and 21% at 3CH), Carbo vegetabilis (27% at 12CH and 34% at 7CH), Ferrum metallicum (44% at 9CH), and Natrum muriaticum (66% at 5CH).

Phosphorus was found effective against other *Aspergillus* species, such as *A. parasiticus*, with inhibition rates of 63% and 70% at 6CH and 12CH, respectively (Bee and Atri, 2013). In contrast, Ferrum metallicum dynamizations (3CH and 5CH) stimulated the growth of *Aspergillus niger* by 17% and 20%, respectively, emphasizing the potential for long-term effects of homeopathic treatments.

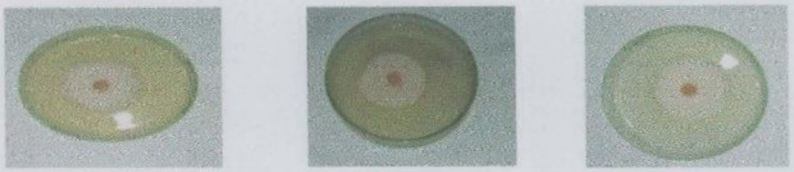
Arsenicum album exhibited strong antifungal properties, achieving a 100% inhibition rate against *A. parasiticus* at 200CH (Sinha and Singh, 2003), similar to Thuja occidentalis, which reduced aflatoxin production by 100% in stored products. Phosphorus inhibited the mycelial growth of Dulcamara fungus by 50% while reducing toxin production by 90%. Gama et al. (2015) also observed inhibition of mycelial growth with certain treatments, including Carbo vegetabilis (30%), Phosphorus (64%), Sulphur (97%), Natrum muriaticum (136%), and Ferrum metallicum (140%) that supported the current study.

Homeopathic medicine	6CH	30 CH	200CH
Arsenicum album			
Calcaria sulp			
Selenium			

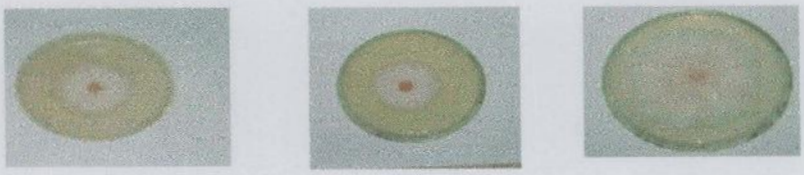
Cina



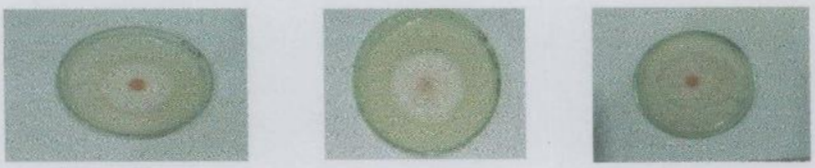
Nux vomica



Siliciea

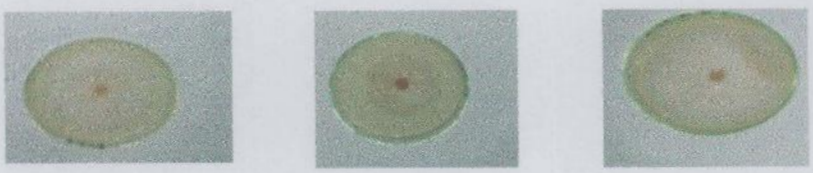


Arnica montana



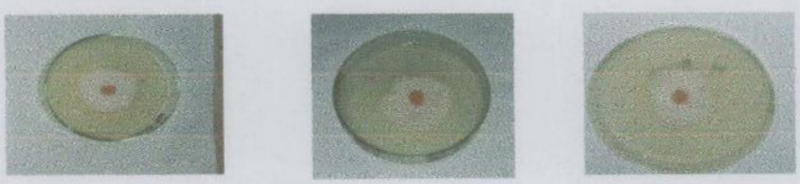
Calcarea carbonicum

Carbo vegetabilis

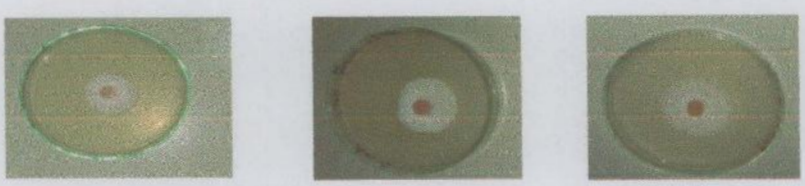


Ferrum metallicum

Natrum muriaticum



Phosphorus



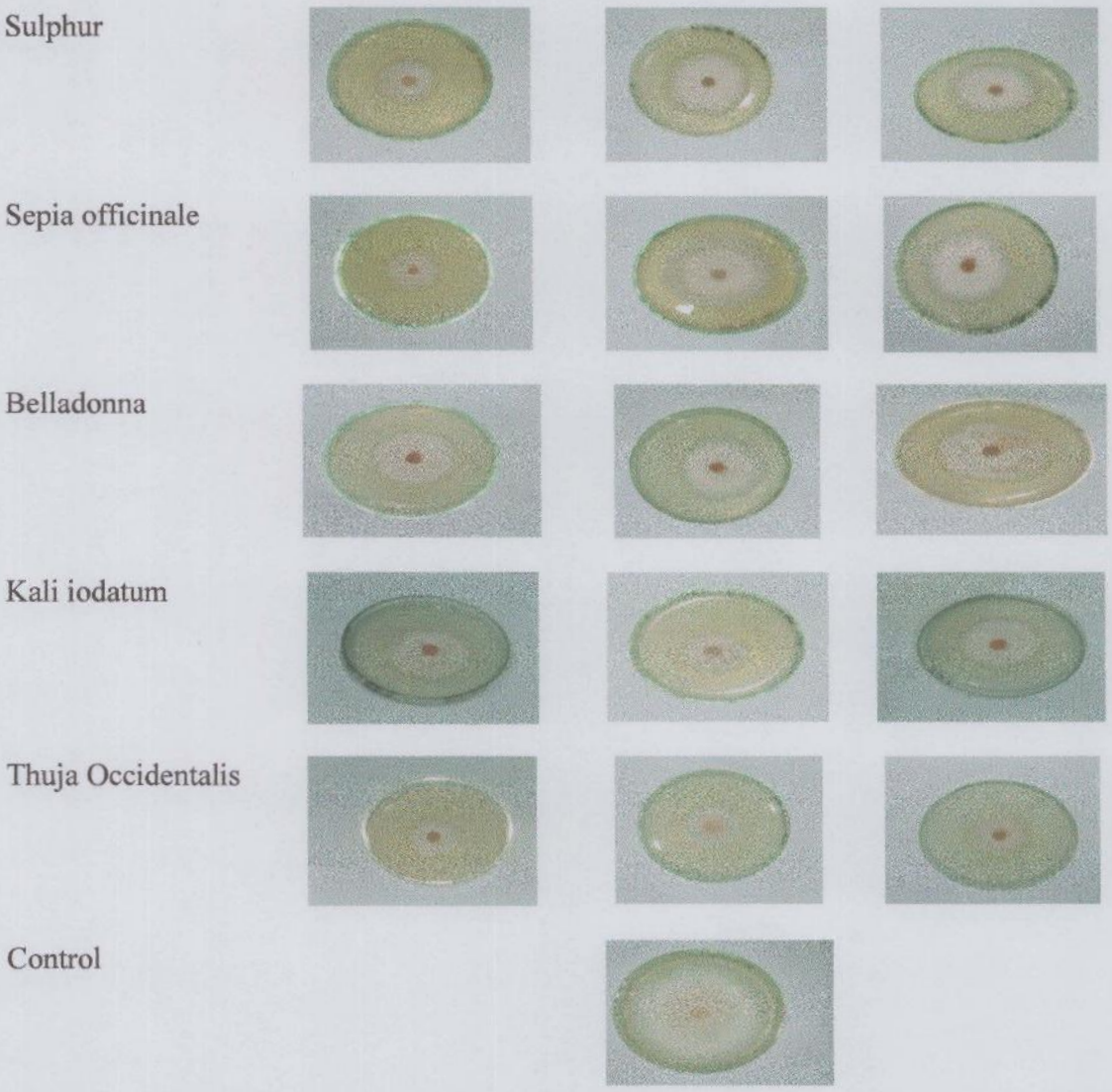


Figure 4.1.2: Effect of homeopathic remedies at different potencies on mycelial growth

4.2 Effect of different homeopathic medicine on sporulation of *Colletotrichum musae*

Table 4.2.1: Effect of homeopathic remediefs at different potencies on spore production

Treatments	Potency	Sporulation (spores/mL)	Sporulation % inhibition
Arsenicum album	6CH	1613.33 xy	62.83 cd
	30CH	2006.7 qr	53.78 g
	200CH	1746.67 vw	59.77 de
Calcaria sulp	6CH	2080.0 op	52.07 gh
	30CH	2166.7 lmn	50.09 hi

	200CH	1660.0 xy	61.75 cd
Selenium	6CH	1740.0 vw	59.91 de
	30CH	1886.67 st	56.54 f
	200CH	2066.7 pq	52.39 gh
Cina	6CH	2280.0 jk	47.46 jk
	30CH	1733.33 vw	60.07 de
	200CH	1926.67 rs	55.62 fg
Nux vomica	6CH	2180.0 k-n	49.77 i
	30CH	1933.33 rs	55.46 fg
	200CH	2386.7 hi	45.02 l
Siliciea	6CH	2520.0 fg	41.93 n
	30CH	2786.7 c	35.81 pq
	200CH	2973.3 b	31.49 r
Arnica montana	6CH	22333 klm	48.55 j
	30CH	3020.0 b	30.41 s
	200CH	2460.0 fgh	43.32 lmn
Calcarea carbonicum	6CH	2660.0 de	38.71 o
	30CH	1706.67 wx	60.69 de
	200CH	2340.0 ij	46.09 jkl
Carbo vegetabilis	6CH	2426.7 ghi	44.10 lmn
	30CH	2133.33 mn	50.85 hi
	200CH	2560.0 ef	41.01 n
Ferrum metallicum	6CH	2113.3 no	51.31 h
	30CH	2213.3 klm	49.00 ij
	200CH	2126.7 no	51.01 h
Natrum muriaticum	6CH	1793.33 tu	58.68 def
	30CH	1733.33 vw	60.07 de
	200CH	2233.3 klm	48.54 j
Phosphorus	6CH	1946.67 rs	55.16 fg
	30CH	2753.3 cd	36.56 p
	200CH	2720.0 cd	37.33 op
Sulphur	6CH	2393.3 hi	44.86 l

	30CH	2026.7 qr	53.32 g
	200CH	1606.67 y	62.99 cd
Sepia officinale	6CH	1600.0 y	63.13 c
	30CH	1746.67 vw	59.77 de
	200CH	2926.7 b	32.58 q
Belladonna	6CH	2653.3 de	38.87 o
	30CH	2193.3 klm	49.47 i
	200CH	2746.7 cd	36.73 p
Kali iodatum	6CH	2240.0 jkl	48.39 j
	30CH	1713.33 vwx	60.53 de
	200CH	1853.33 stu	57.30 ef
Thuja occidentalis	6CH	1780.0 uvw	58.98 def
	30CH	1446.67 z	66.68 b
	200CH	1946.67 rs	55.16 fg
Control	=	4340.0 a	-
CV%		2.36	2.39

Values with different letters are significantly differed at 5% level of significance according to Tukey's HSD Test

Table 4.2.1 investigated the effects of different homeopathic remedies at varying potencies (6CH, 30CH, and 200CH) on the sporulation of *Colletotrichum musae*, with untreated control serving as the baseline. Sporulation was measured by number of spores per mL under microscope and percentage inhibition was calculated relative to the control.

In Arsenicum album the highest inhibition (62.83%) was observed at 6CH potency, producing 1613.33 spores/mL, while 30CH showed reduced inhibition (53.78%). 200CH inhibited sporulation by 59.77%. The potency 200CH in Calcarea sulp exhibited the highest inhibition (61.75%), producing 1660 spores. 6CH and 30CH showed lower inhibition rates of 52.07% and 50.09%, respectively. In Selenium, 6CH resulted in the highest inhibition (59.91%) with 1740 spores, while 200CH showed the lowest inhibition (52.39%) that were significantly different from the other treatments.

Maximum sporulation inhibition in cina (60.07%) occurred at 30CH potency, producing 1733.33 spores where 6CH showed the least inhibition (47.46%). In the case of Nux vomica, 30CH exhibited an inhibition of 55.46%, whereas 6CH and 200CH showed lesser inhibition rates of 49.77% and 45.02%, respectively. Minimum sporulation inhibition was observed for Silicea across all potencies. 6CH and 30CH showed only 41.93% and 35.81% inhibition, respectively, with 200CH being the least effective at 31.49%. The highest sporulation (3020 spores) and the lowest inhibition (30.41%) were observed at 30CH in Arnica montana. 6CH and 200CH achieved moderate inhibition levels of 48.55% and 43.32%, respectively.

In Calcarea carbonica, 30CH demonstrated the highest inhibition (60.69%) with 1706.67 spores. 6CH and 200CH showed lower inhibition at 38.71% and 46.09%, respectively. Carbo vegetabilis displayed conidial densities of 2426.7, 2133.3, and 2560 spores at 6 CH, 30 CH, and 200 CH, respectively. In Ferrum metallicum, spore production peaked at 30 CH (2213.3 spores per mL, corresponding to 49.0 %), with almost similar outputs recorded at 6 CH (2113.3) and 200 CH (2126.7/mL). Potencies at 6CH and 200CH were not significantly different but the value of 30CH in Ferrum metallicum was significantly different.

Natrum muriaticum exhibited conidial levels of 1793.33 spores and the inhibition percentage is 58.68% at 6 CH, 1733.33 spores at 30 CH, and 2233.3 spores at 200 CH. For Phosphorus, the highest spore production was recorded at 30 CH (2753.3 spores, corresponding to 36.56%), followed closely by 200 CH (2720 spores), with the lowest production observed at 6 CH (1946.67 spores/mL). In case of Sulphur, the highest inhibition (62.99%) occurred at 200CH potency, producing 1606.67 spores. On the other hand, 6CH and 30CH showed lesser inhibition of 44.86% and 53.32%, respectively.

6CH in Sepia officinale exhibited the greatest inhibition (63.13%) with 1600 spores/mL, whereas 200CH showed the least inhibition (32.58%). For Belladonna, the maximum conidial density occurred at 200 CH (2746.7 spores, corresponding to 36.73%), followed by 30 CH (2653.3 spores), with the lowest output at 6 CH (2193.3 spores), the values were significantly different from each other.

In Kali iodatum, conidial production levels were 2240 spores/mL at 6 CH, 1713.33 at 30 CH, and 1853.33 spores at 200 CH. In Thuja occidentalis exhibited the highest

inhibition rate (66.68%) at 30CH producing only 1446.67 spores. Potencies 6CH and 200CH showed inhibition levels of 58.98% and 55.16%, respectively. The untreated control recorded the highest sporulation (4340 spores), indicating no inhibition.

Among all of the treatments, Thuja occidentalis showed highest percentage of spore inhibition (66.68%) at 30 CH, followed by Sepia officinale at 6CH (63.13%), Sulphur at 200 CH (62.99%), Arsenicum album at 6CH (62.83%) and Calcarea sulp at 200CH (61.75%). In constant, Arnica montana was observed only 30.41 % at 30CH potency.

The findings of this study align with and extend the results of prior research investigating the use of homeopathic remedies to manage fungal pathogens, particularly through the inhibition of sporulation. According to Alam and Mistry (2022), homeopathic medicines, including Arsenicum album, Thuja occidentalis, and Sulpher, demonstrated significant inhibitory effects on spore formation. Thuja Occidentalis, at dynamizations of 6 CH and 30 CH, exhibited a suppressive impact on sporulation by 63.5% and 59.5%, respectively (Alam and Mistry, 2022), similarly, Sulpher inhibited sporulation by 32.90% and 52.52% compared to the control. Sporulation inhibition by Arsenicum album was reported as 60.19% and 56.9% for the 3CH and 6CH potencies (Alam and Mistry, 2022). This outcome aligns closely with the findings of the present study.

The sporulation of *Alternaria solani* was effectively suppressed by various homeopathic medicines, including Thuja occidentalis, Kali iodatum, Arsenicum album, Sulphur, Silicea, Staphysagria, Phosphorus, Arnica montana, and Natrum muriaticum, administered at 6CH, 12CH, 30CH, and 100CH (Bonato, 2007). Kali iodatum inhibited sporulation by 45.0% and 30.2% at 6CH and 30CH, respectively, while Arnica montana achieved inhibition rates of 36.36% and 21.86% at the same potencies. Arsenicum album demonstrated inhibitory effects of 59.4% and 53.1% at 6CH and 30CH, respectively. Among these medicines, they found Thuja occidentalis at 30CH achieved the greatest suppression of sporulation, with an inhibition rate of 63.1% compared to the other treatments (Bonato et al., 2006).

Khanna and Chandra (1992) observed that homeopathic treatments suppressed the spore production of several fungi, including *Alternaria alternata, Colletotrichum gloeosporioides, Fusarium roseum,* and *Gloeosporium psidii.* Balbi-Peña et al. (2016) reported that Sulphur reduced sporulation by approximately 50% in *A. solani.*

Likewise, Franzener et al. (2017) observed 46% inhibition of *Alternaria brassicae* sporulation with the use of Nux vomica. These findings supported the current study, underscoring the potential of homeopathic medicines to control *Colletotrichum musae* in banana cultivation.

The significant inhibition of sporulation by Thuja occidentalis (66.68% at 30CH) was consistent with the findings of Leite and Stangarlin (2008), who reported the efficacy of Thuja occidentalis in suppressing spore germination and reducing pathogen penetration. Similarly, Arun et al. (2018) demonstrated the effectiveness of Thuja occidentalis in reducing sporulation and disease incidence in tropical crops. The high efficacy of Sulphur in this study (62.99% inhibition at 200CH) supported the results of Gondim et al. (2014) and Balbi-Peña et al. (2016), who observed substantial reductions in spore production when using Sulphur against fungal pathogens such as *Alternaria solani* and *Colletotrichum musae.*

The performance of Sepia officinale (63.13% inhibition at 6CH) aligned with the findings of Bonato (2007), who noted variability in sporulation inhibition based on the potency of homeopathic treatments. Potencies 6CH and 30CH consistently showed higher inhibition compared to 200CH in most treatments. This supports the observations of Toledo et al. (2016), who reported that lower potencies such as 6CH and 30CH were more effective in inhibiting sporulation. Bonato (2007) also highlighted that different potencies of the same remedy can yield varying levels of efficacy, necessitating careful selection based on pathogen type and environmental conditions.

The observed efficacy of Selenium (59.91% inhibition at 6CH) and Nux vomica (55.46% inhibition at 30CH) extended the findings of Franzener et al. (2017), who demonstrated the suppression of sporulation in other fungi like *Alternaria brassicae.* The substantial reduction in sporulation achieved with several homeopathic treatments supported their use as eco-friendly alternatives to synthetic fungicides. Gondim et al. (2014) emphasized the potential of homeopathy for sustainable disease management in banana cultivation, echoing the findings of this study.

Several studies, including Dawar et al. (2008), had suggested that homeopathic treatments might target physiological processes such as respiration, enzymatic activity, or nutrient uptake in fungi, leading to reduced sporulation. The significant inhibition

observed in this study corroborates this hypothesis, particularly for Arsenicum album, Sulphur, and Thuja occidentalis. The high sporulation inhibition of Natrum muriaticum and Phosphorus (up to 60.07% at 30CH for Natrum muriaticum) aligned with Rodrigues et al. (2020), who found that these remedies effectively inhibited sporulation in various fungal pathogens. These findings were similar with the present study.

CHAPTER V

SUMMARY AND CONCLUSION

5.1 Summary

Homeopathic medicines are cost-effective and free from side effects, making them a promising alternative for managing plant diseases. This study evaluated their effectiveness against *Colletotrichum musae,* the causative agent of anthracnose in bananas. Seventeen homeopathic medicines (Arsenicum album, Calcaria sulp, Selenium, Cina, Nux vomica, Silicea, Arnica montana, Calcarea carbonicum, Carbo vegetabilis, Ferrum metallicum, Natrum muriaticum, Phosphorus, Sulphur, Sepia officinale, Belladonna, Kali iodatum, and Thuja occidentalis) at three potencies (6CH, 30CH, 200CH) were tested. The in vitro results demonstrated significant potential of these remedies in inhibiting the pathogen's mycelial growth and sporulation.

Among all treatments, the least radial growth of 38.02 mm was recorded in plates treated with Arsenicum album at 6CH potency, followed by Selenium at 6CH (40.03 mm), Arsenicum album at 30CH (41.44 mm), Cina at 30CH (41.52 mm) and Thuja occidentalis at 6CH (41.6 mm). Conversely, the highest mycelial growth, 52.62 mm, was achieved with Silicea at 200CH.

The highest inhibition of mycelial growth was observed in Arsenicum album at 6CH, achieving 49.66% suppression. This was closely followed by Selenium at 6CH (46.53%), Arsenicum album at 30CH (44.65%), Cina at 30CH (44.55%) and Thuja occidentalis at 6CH (44.44%). Conversely, the lowest growth inhibition was recorded with Silicea at 200CH (30.23%) at 8 DAI.

For spore production, Thuja occidentalis at 30CH showed the highest inhibition (66.68%), followed by Sepia officinale at 6CH (63.13%), Sulphur at 200 CH (62.99%), Arsenicum album at 6CH (62.83%) and Calcarea sulp at 200CH (61.75%), while the lowest was observed with Arnica montana at 30CH potency (30.41%).

5.2 Conclusion

Homeopathy has demonstrated promising potential as an alternative and sustainable approach to managing *Colletotrichum musae.* In this study, homeopathic remedies such as Arsenicum album at 6 CH showed the highest inhibition of mycelial growth,

while Thuja occidentalis at 30 CH effectively suppressed spore production. These findings suggest that homeopathic treatments can replace hazardous chemicals and pesticides in plant disease management. Since these remedies are affordable, widely available, safe, and environmentally friendly, their large-scale application is strongly recommended.

CHAPTER VI

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