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ARTICLE

Characterization of Mycoflora Associated with *Catharanthus roseus* Collected from Gardens in Kenitra City, Morocco

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ABSTRACT

This paper reports on the composition of fungal communities occurring on diseased tissues of *Catharanthus roseus*, which differed between organs. In total, ten different filamentous fungi were isolated, and the percentage of isolation varied significantly among the organs. *Botrytis cinerea* was the most prevalent fungus found on the plant's aboveground parts, with a frequency exceeding 50%. On twigs, the occurrence rate was 95.6%. It was isolated from leaves with a frequency of 88%, followed by *Aspergillus niger* (71.66%), *Alternaria alternata* (67.33%), *Cladosporium herbarum* (61%), *Fusarium oxysporum* (50.66%), *Epicoccum nigrum* (57.66%), *Curvularia lunata* (49.66%), *Trichoderma harzianum* (40%), and *Penicillium* sp. (27%). Whereas, Fusarium genus was more represented and six species were recorded: *F. subglutinans* (26%), *F. chlamydosporium* (20%), *F. vertillioides* (15.66%), *F. solani* (10%), *F. oxysporum* and *F. nivale*. Results highlighted dissimilar distribution of Fusarium species was noted on Catharanthus tissues on which *F. subglutinas*, *F. chlamydosporium* and *F. oxysporum* coexist on leaves and roots while *F. solani* was retrieved from leaves against *F. nivale* from roots. The floral buds and pods harbored opportunist fungi such as *B. cinerea, Alternaria alternata* and *E.*

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nigrum. On roots, *Aspergillus*, *Penicillium* and *Fusarium* were the main genera occurring with the frequencies of 26% (*F. chlamydosporium)*, 40% (*F. nivale*), 72.33% (*A. niger*), 47.66% (*A. flavus*) and 37.66% (*A. fumigatus*). But no fungal species were detected on seeds of *Catharanthus roseus*. This is the first study to describe and enumerate the fungal complex associated with various symptoms on the aerial parts of *Catharanthus roseus*.

Keywords: Isolation; Identification; *Catharanthus roseus*; Mycoflora; Morocco

1. Introduction

Catharanthus roseus known as Madagascar periwinkle, is a perennial plant belonging to the Apocynaceae family, widely distributed ornamental plant and holds significant medicinal value[1]. This species thrives in dry, warm areas and can grow in full sun or partial shade. There are two varieties of this plant which can be distinguished by the color of the flower viz, the rosea with pink flowers and the alba with white flowers[2]

Catharanthus roseus is a valued commercial plant due to many essential alkaloids used to treat high blood pressure and circulatory disorders[3, 4]*. It also possesses anti-cancer, anti-diabetic, anti-Alzheimer's properties in addition to be effective for wound-treatment*[5, 6]*. Periwinkle is also known to yield a variety of tertpenoid indole alkaloids in the form of vincristine, vinblastine, catharanthine, ajmalicin, vindoline, etc., which are of great significance in the medical industry*[7]*. The potential medicinal benefits include antiviral, antidiabetic, antihypertensive, antibacterial, antifungal and antioxidant activities*[8, 9]*. But, its value as an ornamental*[2] *also promises an increasing interest for use*.

Catharanthus roseus (L.) G. is the host of hundreds of endophytic bacteria and fungal organismes that inhabiting different areas of plants[10]*. The latter exhibit different types of association with their host organisms, such as a combination of symbiosis, pathogenesis, antagonistic relationships, etc., beside being able to promote plant growth, counteract plant stress conditions, and even elicit plant defense against any pathogenic attack*[11, 12]*. C. roseus endophytic fungi have also been reported to produce certain compounds with potential cytotoxic activities*[13] *and antioxidant activity*[14]*. Dhayanithy*[15] *have shown that Chaetomium nigricolor found among twenty endophytic isolates obtained from different parts of the plant, had significant cytotoxic, apoptotic and antioxidant potential. Various species of the Nigrospora genus, such as Nigrospora zimmermanii and N. sphaerica, isolated from Catharanthus plants, have the capacity to produce the anticancer agent vincristine and hydrolytic enzymes*[16, 17].

On the other hand, there are several pathogens documented worldwide causing many different forms of disease in *C. roseus*[18–21]. Diseases include root rot and seedling damping-off, caused by a wide range of fungi, such as *Rhizoctonia solani*, *Fusarium oxysporum*, *F. solani*, *Thielaviopsis* sp., *Sclerotium rolfsii* and *Pythium aphanidermatum* or Foliar blight caused by *Colletotrichum dematium* and *Botrytis cinerea*[22]. Others fungi such as *Sclerotinia sclerotiorum* are regarded as a plant pathogen on Periwinkle[23] and *Macrophomina phaseolina* (Tassi) Goid that was proved to be pathogenic to *C. roseus* by causing leaf blight[24]. Madagascar periwinkle is also affected by *Phytophthora tropicalis* responsible for leaf blight disease symptoms[25]. Sharma[21] have recoded twig blight on *C. roseus* induced by *Colletotrichum gloeosporioides*. Garibaldi[26] described leaf blight symptoms on *Catharanthus roseus* plant caused by *Rhizoctonia solani*. Baysal-Gurel[27] have identified the causal agent for Phytophthora aerial blight symptom on annual vinca as *Phytophthora nicotiana*. Also, Intaparn[28] have reported root and stem rot induced by *Phytopythium* sp.

Generally, when inhabiting host plant, endophytes and pathogens can be related via a various interactions which probably determine survival, pathologies appearance and growth performance of plants. In Morocco, no study was carried out to determine the fungal community in association with *Catharanthus roseus*.

Against this background, the aim was to describe and identify the fungal organisms involved in the necrotic damage observed on this plant species.

2. Materiel and Methods

2.1. Samples Collection

Symptomatic *Catharanthus roseus* leaves, tiges, pods and seeds showing necrotic lesions were detached from twenty Catharanthus plant samples which were collected from gardens in Kenitra city, Morocco. Sterilized polythene bags were used to collect the samples. They were brought out to the laboratory, and they were processed within twenty-four hours of being collected.

2.2. Isolation and Identification

Fungi were isolated from aerial parts of *C. roseus* as described by [29]. Different tissues surface was first disinfected by immersing them in 70% ethanol for 60 seconds, then 90 seconds in 70% sodium hypochlorite, and finally in sterile distilled water for 60 seconds before air-drying for 5 minutes on filter paper (150 mm) under sterilization conditions. Disinfected leaf tissues were cut into 1cm × 1cm segments and ten pieces were placed on petri dishes. At room temperature, the dishes were incubated for 14 days then repeatedly observed for fungal growth. To obtain a pure culture, an inoculum of each new colony was continuously transferred onto a fresh PSA medium (Potato Sucrose Agar: 200 g potato, 20 g sucrose, 15 g Agar-agar, 1000 ml distilled water) added with 100 mg/L of chloramphenicol.

Diseased seedlings were rooted out and cut at a height of 5 cm from crown. The roots were then rinsed under running water and cut into pieces 0.5 cm long. Samples were surface disinfected with sodium hydochlorite (0.5% chlorine) for three minutes, then rinsed twice with distilled water for a further two minutes and dried with sterile filter paper. Root fragments were then placed in 9 cm Petri dishes containing potato sucrose agar (PSA). Plates were kept in incubation at 25 ± 1 °C for 4 days, after which the fungi were purified by cutting out a bit of mycelia taken from the edge of colony and cultured in the center of a petri dish containing the PDA and incubated for 7 days [30]. Subsequently, the purified fungal collections were identified using fungal recognition keys derived from phenotypic hallmarks, such as fungal mycelial structure and spore morphology, fungal colony color, branching nature of the mycelium, conidiophore shape and size, spores formed and ability to form sclerotia [31–34]. Cultivation set-ups on slides were used to study the arrangement of conidia under the light microscope. Periodic examination of the slides was performed using the lactophenol cotton blue stain when sufficient growth had been achieved. Fungal types and species were selected by light microscopy in accordance with the key determinations of [35–40]. The isolation percentage and/or contamination, which defines the incidence of isolation of the different fungi from 100 lesions, was then calculated using Ponchet's formula [41]: PC = (NLI/NTL) × 100 PC: Percent of infection and/or contamination; NLI: The number of lesions containing the fungal species. NTL: Total number of segments with lesions used for fungus isolation.

2.3. Statistical Analysis

Analysis was performed using a one-way (ANOVA) and an LSD test at the 5% test level. Per cent were transformed into Arcsin √P (where P is the isolation percentage).

3. Results and Discussion

Infected plants showed symptoms mainly on the leaves. Lesions are circular (approx. 0.1 to 0.3 cm long), brownish-yellow to blackish. These spots lengthen and become elliptical or rectangular (2 cm long), with a dark brown center. Occasionally, the attacked leaves become twisted, deformed, and desiccated. Leaves then rot and become covered with a velvety, grayish felt (**Figure 1**).

Figure 1. Various symptoms on *Catharanthus roseus* plants. (**a**) Leaf brown spots and yellowing; (**b**) desiccation symptoms of leaves detached from infected plants of *Catharanthus roseus* (L.) G. Don. in the gardens of the city of Kenitra.

It has been observed that infection generally starts at the upper end of flower buds and pods, and subsequently, the whole pod may be attacked (**Figure 2**). On the stems, infected parts are covered with a conidiation greyish felting (**Figure 3**).

A diverse fungus complex was found on the leaves, stems, flower buds, pods, and roots. As a result, *C. roseus*

recorded a large and varied fungal community (**Table 1**). From the host's leaf, stem, and root tissues, a total of 17 species, representing 13 fungal groups, were identified (**Table 2**). The most dominant fungal organisms were *B. cinerea* and *Alternaria alternata*. *B. cinerea* was found on leaves, floral buds and pods with respective percentage of contamination of 88%, 53% and 49% compared to 67,3%; 36,66% and 47,33% for *A. alternata* undetectable at twigs level of infected *Catharanthus roseus*. The appearance of a fungus on a tissue did not express a close and specific relationship with *Catharanthus* tissues. Lesions in one organ can contain many species that also appear in other organs, despite having different frequencies. A complex of species is transmitted regardless the microenvironment within each organ. In general, 13 species were isolated from leaves, 4 from flower buds, 3 from pods, 1 from stems, 10 from roots, and no species from seeds.

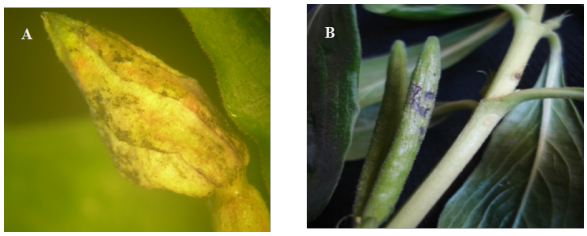

Figure 2. Necrosis on surface of floral buds. (**a**) pods; (**b**) of *Catharanthus roseus* plants samples.

Figure 3. Infected plants of Catharanthus roseus showing greyish to black mycelia and fruiting bodies covering stems.

Table 1. Fungal isolates recovered from leaves, stems, pods, seeds, and roots of symptomatic *Catharanthus roseus* Cv *rosea* planted in gardens area, Kenitra city.

Fungal Species	Stems	Leaves	Floral Buds	Pods	Roots	Seeds
Botrytis cinerea	BcS1, BcS1′, BcS1″	BcS2, BcS2′, BcS2″	BcS3, BcS3′, BcS3″	BcS4	_	_
Aspergillus niger	_	An fS1, AnfS2	_	_	An rS1, An rS2	_
Alternaria alternata	_	Aa fS1, AafS2, AafS3	Aa bS1, AabS2, Aab S3	Aa gS1, AagS2	_	_
Cladosporium herbarum	_	ChS1, ChS2, ChS3, Ch4	_	_	_	_
Fusarium oxysporum	_	Fo f	_	_	Fo r	_
Curvularia lunata	_	Cl f	Cl b	_	Cl r	_
Trichoderma harzianum	_	ThS1, Th S2, ThS3, ThS4	_	_	_	_
Penicillium sp	_	Pn S1sp, PnS2sp	_	_	_	_
Fusarium chlamydosporum	_	Fch f	_	_	Fch r	_
Fusarium vertillioides,	_	Fv f	_	_	Fv r	_
Epicoccum nigrum	_	EnfS1, Enf S2, EnfS3	En bS1, EnbS2	EngS1, EngS2	_	_
Penicillium frequetans	_	_	_	_	PfS1, PS2	_
Fusarium solani	_	FsS1, FsS2	_	_	_	_
Fusarium nivale	_	_	_	_	Fn	_
Fusarium subglutinans	_	Fsb f	_	_	Fsb r	_
Aspergillus flavus	_	_	_	_	Af	_
Aspergillus fumigatus	_	_	_	_	Afu	_

(-): non-isolated species; (S): strains. *BC: Botrytis cineria*, Af: *Aspergillus flavus*, An*: Aspergillus niger*, Afg: *Aspergillus fumigatus*, Pn sp.: *Penicellium* sp., Cl: *Curvularia lunata*, Th: *Trichoderma harzianum*, Fc: *Fusarium chlamydosporium*, Aa: *Alternaria alternata*, Ch: *Cladosporium herbarum,* Fn: *Fusarium nivale*, Fo: *Fusarium oxysporum*, Fv: *Fusarium verticillioides*, Fs: *Fusarium solani*, Fsb: *Fusarium subglutinans*, Pf: *Penicllium frequetan s*, En: *Epicoccum nigrum.*

Table 2. Percentage of contamination of *Catharanthus roseus* by different fungal species isolated using Blotter method.

Fungal Species	Twigs (%)	Leaves (%)	Floral Buds (%)	Pods (%)	Roots (%)
Botrytis cinerea	95 [a]	88 [a]	53 [cd]	49 [cd]	_
*Aspergillus niger**	_	71,66 [b]	_	_	72,33 [b]
Alternaria alternata	_	67,3 [b]	36,66 [ef]	47,33 [d]	_
Cladosporium herbarum	_	61 [bc]	_	_	_
*Fusarium oxysporum**	_	50,66 [cd]	_	_	67,3 [b]
*Curvularia lunata**	_	49,66 [cd]	18,33 [fg]	_	49,66 [cd]
*Trichoderma harzianum**	_	40 [e]	_	_	70 [b]
Penicillium sp.	_	27 [f]	_	_	_
*Fusarium chlamydosporum**	_	20 [fg]	_	_	26 [f]
*Fusarium verticillioides **	_	15,66 [fg]	_	_	35 [ef]
Epicoccum nigrum	_	57,66 [c]	27 [f]	20,66 [fg]	_
Penicillium frequetans	_	_	_	_	47,33 [d]
Fusarium solani	_	10 [g]	_	_	_
Fusarium nivale	_	_	_	_	40 [e]
Fusarium subglutinans	_	26 [f]	_	_	_
Aspergillus flavus	_	_	_	_	47,66 [d]
Aspergillus fumigatus	_	_	_	_	37,66 [ef]

* At the 5% level, there was no significant difference between two results that were read on the same column.

At the root level, out of 10 retrieved fungal species, the genus Fusarium was represented by three species: *Fusarium oxysporum* (67.33%), *F. nivale*, and *F. verticillioides* (35%), with *Fusarium chlamydosporium* (26%). The highest frequency was found in *A. niger* (72.33%), followed by *Trichoderma harzianum* (70%) and 49.6% for *C. lunata*, which significantly displayed an equal isolation frequency to those of *P. frequetans* and *A. flavus*. Both macroscopic and microscopic morphological characteristics have been described for fungal species grown on Potato sucrose agar culture medium.

The morphological examination of *Alternaria alternata* grown on PSA showed brownish olivaceous colony. Conidiophores were usually short being either ramified or simple, solitary or grouped mostly curved but some are straight. Conidia produced usually in long branched strings in diverse shapes; oval, obclavate, obpyriform or ellipsoidal with average size 4–38 × 7–14 µm with 1 to 8 septa. They possessed 1 to 8 transverse septa with a short apical beak (**Figure 4a,b**).

Epicoccum nigrum colony is fast growing, suede-like to downy, with a strong yellow to orange-brown diffusible pigment. Conidiophores vary in size and occur in dense, pale-colored clusters measuring 12–14 × 5–4 µm. They are grouped into aggregates (sporodochia). The conidia, 15–25 µm in diameter, are globe-shaped, pyriform, and lightly pigmented with a bright yellow to orange-brown color (**Figure 4c**).

Botrytis cinerea had fluffy colony, with gray-white aerial mycelia on PSA, then turned gray and formed ovate to elliptical conidia on the mycelia after 7 days of incubation. It develops tufts of upright, greyish (brownish grey or ash grey) conidiophores with branches at the top. The conidia and conidiophores appear grayish in mass when young, turning a light to medium brown on maturation with size ranging from 12–18 to 13.59 mm in length by 5–12 µm in width (**Figure 4d**). The fungus generates black sclerotia. These structures ensure the survival of the fungus during unfavourable periods.

Curvularia lunata forms black colonies. Conidiophores are straight or flexible, geniculate, usually smooth and brown in color. Conidia are solitary, simple, usually curved, elliptical, fusiform, ovoid, measuring 17–30 × 4–13 µm with 3 transverse septa, pale to dark brown, smooth or verruculose (**Figure 4e**).

Culture of *Fusarium verticillioides* has white mycelia, cottony, ranging in color from white to salmon. The short conidiophores (phialides), which taper towards the tip with rough collars, are solitary or in branched clusters. Microconidia are oval, 0-septate borne from monophialides measuring 7–10 × 2,5 à 3,2 µm. This fungus produces oval or club-shaped macroconidia, 31 à 58 × 2,7 à 3,6 µm, usually 3–7 septate sometimes forming chains. No chlamydospores were observed (**Figure 4f**).

The olivaceous-brown, slow-growing *Cladosporium herbarum* colony turns powdery as a result of the creation of

a lot of conidia. It is olivaceous-black on the back. Conidiophores can be upright, straight or flexuose, unbranched or just branching in the apical region, and in certain species, they have geniculate sympodial elongation. Conidia are ovoid to cylindrical, 4–11 µm in size, and formed in branched chains with a noticeable black hilum (**Figure 4g**).

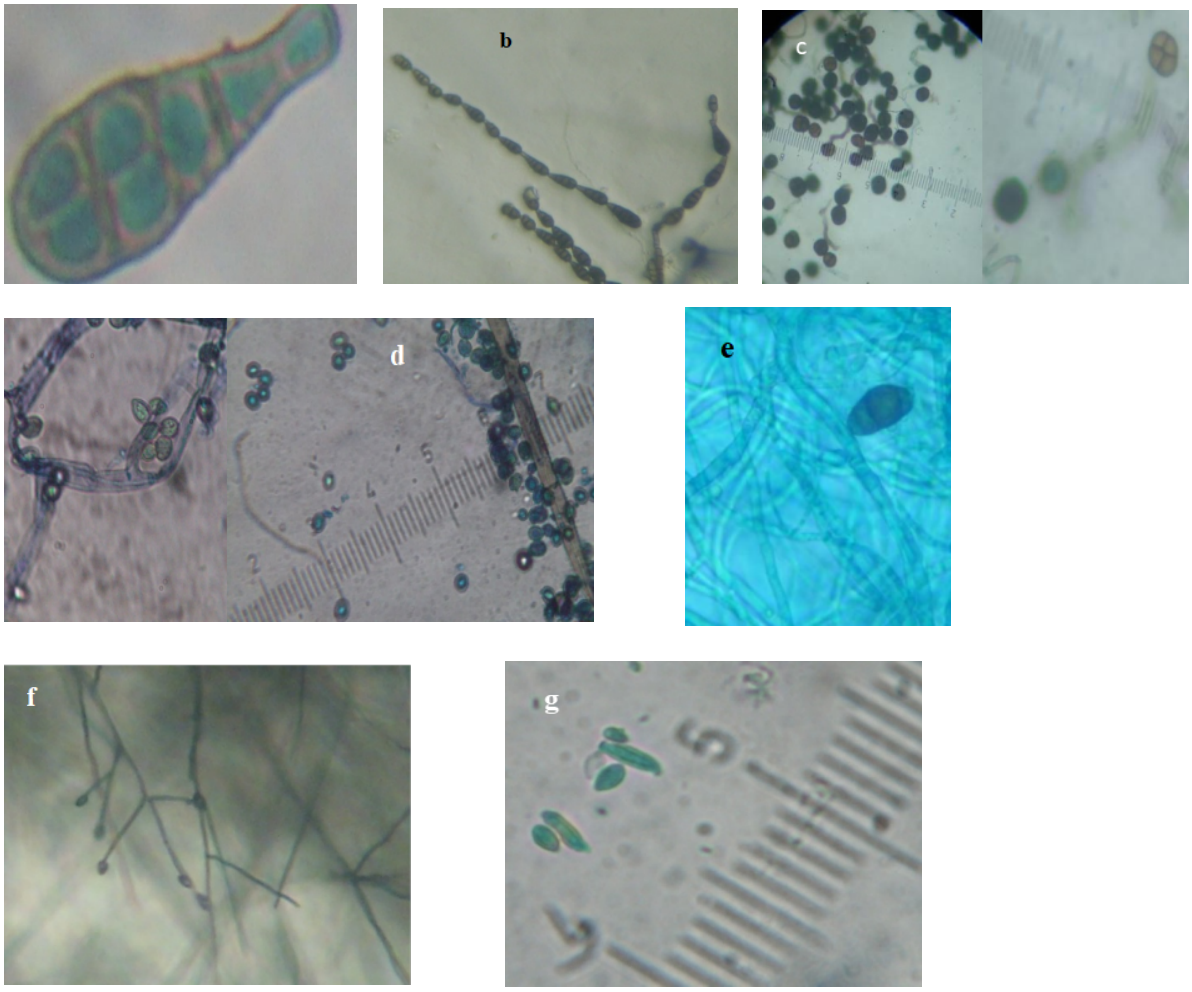

Figure 4. Conidia and conidiophore morphology of fungal species isolated from *Catharanthus roseus*. (**a**) conidia; (**b**) chain of *Alternaria alternata* conidia; (**c**) conidiophore and conidia of *Epicoccum nigrum*; (**d**) conidiophore and conidia of *Botrytis cinerea*; (**e**) conidia of *Curvularia lunata;* (**f**) conidiophores and conidia of *Fusarium verticillioides*; (**g**) conidia of *Cladosporium herbarum* observed under light microscope at (×400) magnification, Mounting liquide: Bleu coton.

The *Trichoderma harzianum* colony grows quickly. Initially, the mycelium is white, but it later turns green and powdery. The conidiophore is branched and produces phialides, which bear conidia at the top that group together into bundles. The conidia are unicellular, and they are globular or ovoid in shape (**Figure 5A$_1$, A2**).

The aerial mycelium of *Fusarium oxysporum* is white and dense. The mycelium turns violet or blue when there are a lot of chlamydospores, but it turns yellowish brown when there are a lot of sporodochia. Usually abundant, microconidia (3–15 × 3–5 µm) are carried on branched conidiophores or short, simple phialides. They are typically unicellular, ellipsoidal to cylindrical, straight, or curved, and they are never generated in chains. Macroconidia are fusiform, somewhat curved, pointed at the ends, and frequently triseptate (20–35 × 3–5µm). On the mycelium, chlamydospores can be terminal or intercalary, spherical, solitary, or in clusters of two to three, and hyaline with smooth or rough walls (**Figure 5B**).

The colony of *Fusarium solani* grows rapidly with abundant mycelium. Microconidia are sparse, with a single cell. The macroconidia are large, with 3 to 4 septa. The spore -wall are parallel and the apical and basal cells are

slightly domed (Figure). Chlamydospores are solitary (7.3 µm) or in pairs 8 to 10.6 µm long, abundant, with smooth walls (**Figure 5C**).

Fusarium nivale (Fr.) Sorauer 1901; has a yellowish-white colony with an orange underside. Aerial mycelium is sparse. Microconidia are absent. Macroconidia are present, small, curved with 1 to 3 septa (**Figure 5D_1, D_2**).

The *Fusarium subglutinans* culture appears white with a dark purple underside. It contains abundant oval microconidia, most of which are formed by a single cell. The macroconidia are also abundant, almost straight, thin-walled, and with the two surfaces of the spore almost parallel (**Figure 5E_1, E_2**).

The *Fusarium chlamydosporium* culture grows quickly on PDA. The mycelium is flaky, fairly dense, off-white in color, and turns violet in the older parts of the colony, with a reverse of red to brown shades. Microconidia are abundant straight to comma-shaped, usually without any septation. Macroconidia were sparse, moderately curved with 2 to 3 septa. Chlamydospores were found in large number and were single, in chains and intercalary. Conidiophores are short and branched with mono- and polyphialides (**Figure 5F_1, F_2**).

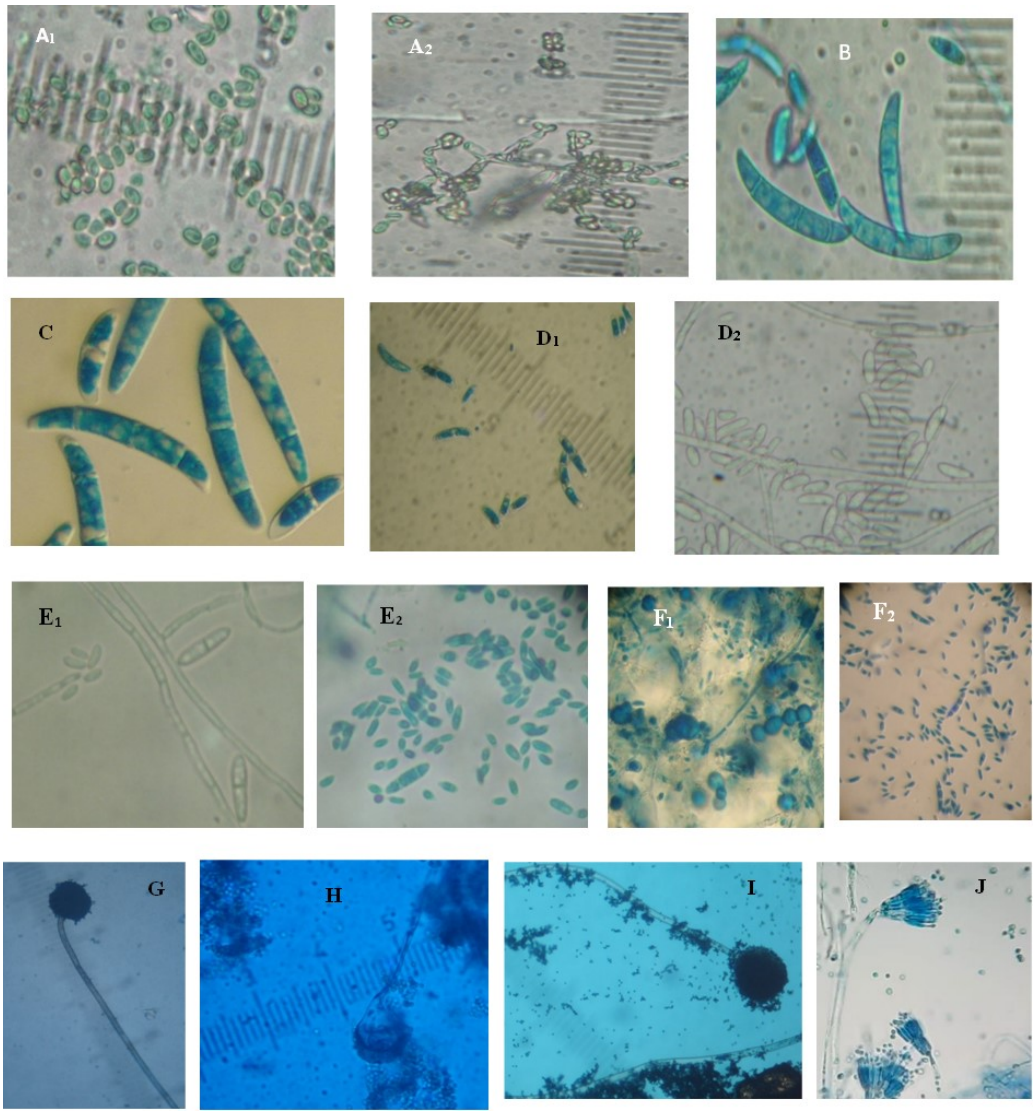

Figure 5. Micromorphology (after methylene blue staining) of fungal species isolated from *Catharanthus roseus* plants. (**A_1,A_2**) Conidia and conidiophores of *Trichoderma harzianum*; (**B**) conidia of *Fusarium oxysporum*; (**C**) conidia of *Fusarium solani*; (**D_1,D_2**) Conidia and conidiophores of *Fusarium nivale*; (**E_1,E_2**) conidiophore and conidia of *F. subglutinas*; (**F_1,F_2**) Chlamydospores and maco-microconidia of *Fusarium Chlamydosporium*; (**G**):conidia and conidiophore of *Aspergillus niger*; (**H**): conidiophore of *Aspergillus flavus*; (**I**) conidiophore and conidia of *Aspergillus fumigatus*; (**J**) conidiophore and conidia of *Penicillium* sp. obtained in PSA medium at (×400) magnification.

The colony of *Aspergillus niger* starts off as white and translucent, then turns black as it produces spores. Aspergillus species are identified by a conidiophore with a swollen end (or vesicle). This end is spherical (40 µm in diameter) and has bottle-shaped phialides arranged all around it. The conidiophores are smooth, clear, brownish at the top, and very long (1 to 3 mm). The conidia are single-celled and single-nucleated, round, 2 to 3 µm in diameter, black or dark brown, and covered with spines and projections (echinulate form). They are often found in chains and spread by the wind (**Figure 5G**).

The colony of *Aspergillus flavus* Link, 1809 grows quickly, displaying sparse, greenish-yellow aerial mycelium with a light yellow reverside. The conidiophore gradually enlarges upwards to form a vesicle 10–40 µ in diameter. The conidia are almost round, greenish-yellow in color, and vary greatly in size from 2 × 3.3 × 4.6 µ to 5 × 6 µ in diameter (**Figure 5H**).

The colony of *Aspergillus fumigatus* has an olive-green color with a yellowish underside. The conidiophore is short and septate, gradually enlarging towards the apex to form a vesicle 20–30 µ in diameter. The conidia are blackish-green, globose, and 2 to 3.5 µ in diameter (**Figure 5I**).

The *Penicillium* sp. colony is green. The mycelium hyphae are septate. The straight conidiophore is generally unbranched, with whorls at the top. The conidia are globular in shape, smooth-walled, and arranged in chains (**Figure 5J**).

4. Discussion and Conclusion

No research on fungal infections of *Catharanthus roseus* has been done in Morocco. The acquired data demonstrated how the fungal community's makeup affects the health of these attractive plants. The presence of a varied fungal complex on the plant organs under study can be explained by the fact that a lesion might be started by a single infection and then colonized by others. Ellis[37] asserts that each species' frequency is determined by the temperature and humidity levels that are conducive to its growth[42]. There are fewer species detected on roots than leaf lesions, which is consistent with findings by[43]. As per the hypothesis of[15], the *Catharanthus* plant's bark has the highest species variety and richness, followed by the leaf and stem tissues.

The obtained results confirmed previous studies that have already reported some of our isolated fungal species while others were encountered for the first time on *Catharanthus roseus* in Morocco. The lesions observed on the aerial parts were mainly dominated by *B. cinerea*. In fact, this species has a wide range of plant hosts such as vegetables, grapevine and fruit trees[44–46], strawberry plants[47, 48], tomatoes[49, 50], lettuce (Beneden et al., 2009)[51], ornamentals[52], including *Rosa hybrida*[53], and *Hibiscus* twigs and brunches[54]. *B. cinerea* caused necrosis on the stems of *Catharanthus roseus*, which later spread to the leaf stalks and, finally, the entire leaf. On the afflicted tissues, the fungus subsequently produce a thin, fragile, gray mycelium, which was especially dispersed on the stems[55]. Generally, *B. cinerea* infection symptoms include leaf spot; blight; stem canker or rot, and damping-off of young seedlings. Blight is common symptoms begin as small water-soaked lesions or areas[56, 57] on flower petals, leaves, or stems. Similar to this, numerous crops and ornamental plants are seriously threatened by *Alternaria alternata*, a pathogen that is well-known for its cosmopolitan nature and broad host range. rice seeds[58], tomato, potato[59], saffron[60, 61]. This fungus can also cause latent infections, which can result in post-harvest spoilage on citrus[62] or pears[63]. *A. alternata* targets its hosts' leaves, stems, and fruits. On the leaves, lesions manifest as roughly round patches with distinct black patches[64]. Also, it was reported that this fungus caused different types of rose periwinkle leaf blight in Iraq[65]. This author claims that the symptoms began as tiny, irregular, round, light brown dots that eventually grew larger and darker to cover the majority of the leaf surface. The brown or black spots on *C. roseus* leaves does not necessarily indicate that *B. cinerea* or *A. alternata* are responsible for these troubles, because other species such as *Curvularia lunata* can also induce the same symptoms. *Curvularia lunata* has been retrieved from leaves of *C. roseus*[66, 67]. This species has been recorded among the recoverable endophytic fungi colonizing the roots, leaves and stems of *C. roseus*[68]. Pandey[69] have reported *Curvularia* sp. among 7 fungal endophytes isolated from the leaves of this plant species. In Morocco, it was detected on aerial parts of *Hibiscus rosa-sinentis*[54] . Lamrani[58] noted *C. lunata* as a rice-seed contaminant. The infected tissues manifested elongated or circular brown-to-black lesions on the leaves and stems of certain varieties of rice, which later expand and become merged[70].

The genus Fusarium was well represented in this study, with six identified species. Of them, *Fusarium oxysporum* was the most prevalent fungus. *F. oxysporum* is common in soil and is widely distributed in nature, where it act as a saprophyte or parasite, causing root rot in attractive plants such as *Tulipa gesneriana* L., *Cyclamen persicum*, *Gladiolus callianthus*, *Acacia dealbata* [71] , but it was also recovered as endophytic fungus in *C. roseus* [72]. Symptoms infection induced by this fungus often manifest as browning of the vessels and yellowing of the leaves, which can result in complete desiccation of the plants [40]. It results in reddish-brown rot with a wet appearance on the stems, where clusters of spores, often pink in color, emerge in various spots [73].

Fusarium oxysporum has been reported to affect strawberry in a specialized form known as *F. oxysporum* f. sp. *fragariae* [74]. It has also been found on banana plants (*Musa accuminata* L.) [75] and on the stems and branches of *Hibiscus rosa-sinensis*, where it appears as orange-colored domes [54]. For *Fusarium solani,* it was isolated from *Catharanthus*'s bark in India [15] and earlier in Malaysia from roots [16]. Yasir and Almaliky [76] also stated the presence of *F. solani* alongside *F. equiseti* and *Rhizoctonia solani* when diagnosing the cause of root rot and wilt disease observed on *C. roseus*. This species also attacks crops such as cotton [77] and has been found in the mycoflora associated with banana plants (*Musa accuminata* L.) [76]. In Morocco, Rieuf [78] reported this species as responsible of Fusarium attacks in *Geranium rosa*.

However, *Catharanthus roseus* has never been mentioned as a host plant for *F. subglutinans* and *F. nivale* in addition to *Fusarium verticillioides* and *F. chlamydosporium* showing moderate isolation frequencies. This latter fungus could damage seed and affect vigour of rice seedling [79, 80]. *F. verticillioides* can infect all plant parts of Maize [81], roots [82], stems [83], and ears [84] during every growth stage.

We can suggest that different pathogens can cause similar symptoms, and the same pathogens can lead to different symptoms depending on the conditions when the disease starts. Many observations pointed out that lesions can be inhabited by both pathogens and saprophytes, such as *Trichoderma harzianum* et Fusarium species.

T. harzianum is known as a biological control agent [85]. In some cases, the antagonistic effect of *T. harzianum* is simply associated with parasitic behavior manifested by the hyphae wrapping around the filaments of the parasitic fungus [86]. Numerous studies have demonstrated that Trichoderma has interesting potential for controlling various pathogens [87].

Aspergillus niger was strongly represented with high frequencies on roots and on leaves but was exclusively found on the leaves and roots of the plant under investigation. This particular species is known to cause black crown rot on groundnuts, leading to plant death [88]. For *Aspergillus flavus*, it was reported in Brazil by [89] on *Musa* sp., rice (*Oryza sativa*), and coconut (*Cocos nucifera*). This fungus can affect several banana species such as *M. textilis* in the Philippines [90] and *M. accuminata* in Thailand (Lumyong et al., 2003) [91]. The third Aspergillus species viz, *A. fumigatus* retrieved from vegetative parts, is commonly encountered in air and soil [92]. The genus Penicillium was found on the leaves and roots but recently [68] have isolated *P. citrinum* associated with endophytic fungi of leaves, stems and roots of *C. roseus*.

Epicoccum nigrum is known to cause discoloration in rice seeds and can also affect their germination capacity [93].

Although a variety of fungal communities may inhabit in medicinal plants [94] who have reported a total of 153 fungal isolates which were isolated from thirty leaves of the *C. roseus* plant, including 11 endophytic and 142 exophytic isolates. Other studies revealed the occurrence of endophytic group including *Lasiodiplodia theobromae*, *Cophinforma mamane*, *Phoma putamium* with *F. solani*, *Colletotrichum* sp., *Alternaria longipes* [15]. However, the expression, the activity and the impact of these organismes vis a vis *C. roseus* are still a concern for debate.

The study indicated that the lesions found on *Catharanthus roseus* are not monospecific, but are instead colonized by a diverse fungal complex. This complex includes several fungi known to be pathogenic while others were categorized as non-pathogenic endophytes. The different organs yielded variable results vis-à-vis the genera of fungal isolates. This study represents the first report of fungal diversity from diseased *Catharanthus roseus* plants in Morocco. However, our findings are insufficient to determine the specific causal agent responsible for the symptomatic areas on the organ surface. Therefore, it is necessary to conduct a precise and adequate characterization of the biology, physiology, and pathogenicity of isolates of these fungi. Examining the evo-

lution of the fungal flora and analyzing the plants before and after plant transplanting could help determine the origin of infections in ornamental plants and facilitate the adoption of more appropriate and timely control methods.

Furthermore, there are many chances to find novel metabolites with potential bioactivity by investigating endophytic fungi that live inside plant species that have therapeutic qualities.

Author Contributions

Conceptualization, A.O.T. and A.D.; methodology, R.B.; software, J.M.; validation, N.A. and M.E.; writing—original draft preparation, N.M.; writing—review and editing, A.D. and A.O.T. All authors have read and agreed to the published version of the manuscript.

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References

[1] Naeem, M., Ansari, A.A., Aftab, T., et al., 2019. Application of triacontanol modulates plant growth and physiological activities of *Catharanthus roseus* (L.). International Journal of Botany Studies. 4(2), 131–135.

[2] Jaleel, C.A., Panneerselvam, R., 2007. Variations in the antioxidative and indole alkaloid status in different parts of two varieties of *Catharanthus roseus*. Chinese Journal of Pharmacology and Toxicology. 21(6), 487–494.

[3] Karthikeyan, B., Jaleel, C.A., Gopi, R., et al., 2007. Alterations in seedling vigour and antioxidant enzyme activities in *Catharanthus roseus* under seed priming with native diazotrophs. Journal of Zhejiang University Science B. 8, 453–457.

[4] Das, S., Sharangi, A.B., 2017. Madagascar periwinkle (*Catharanthus roseus* L.): diverse medicinal and therapeutic benefits to humankind. Journal of Pharmacognosy and Phytochemistry. 6(5), 1695–701.

[5] Pham, H.N.T., Vuong, Q.V., Bowyer, M.C., et al., 2020. Phytochemicals derived from *Catharanthus roseus* and their health benefits. Technologies. 8(4), 80. DOI: https://doi.org/10.3390/technologies8040080

[6] Ayob, F.W., Simarani, K., Zainal Abidin, N., et al., 2017. First report on a novel *Nigrospora sphaerica* isolated from *Catharanthus roseus* plant with anticarcinogenic properties. Microbial Biotechnology. 10(4), 926–932.

[7] Almagro, L., Fernández-Pérez, F., Pedreño, M.A., 2015. Indole alkaloids from *Catharanthus roseus*: bioproduction and their effect on human health. Molecules. 20(2), 2973–3000.

[8] Balaabirami, S., Patharajan, S., 2012. *In vitro* antimicrobial and antifungal activity of *Catharanthus roseus* leaves extract against important pathogenic organisms. International Journal of Pharmacy and Pharmaceutical Sciences. 4 (3), 487–490.

[9] Gajalakshmi, S., Vijayalakshmi, S., Devi, R.V., 2013. Pharmacological activities of *Catharanthus roseus*: a perspective review. International Journal of Pharma and Bio Sciences. 4(2), 431–439.

[10] Wilson, D., 1995. Endophyte: The evolution of a term and clarification of its use and definition. Oikos. 73(2), 274–276.

[11] Musetti, R., Polizzotto, R., Grisan, S., et al., 2007. Effects induced by fungal endophytes in *Catharanthus roseus* tissues infected by phytoplasmas. Bulletin of Insectology. 60(2), 293.

[12] Pathak, P., Rai, V.K., Can, H., et al., 2022. Plant-endophyte interaction during biotic stress management. Plants. 11(17), 2203.

[13] Asai, T., Luo, D., Yamashita, K., et al., 2013. Structures and biomimetic synthesis of novel α-Pyrone polyketides of an endophytic *Penicillium* sp. in *Catharanthus roseus*. Organic Letters. 15(5), 1020–1023.

[14] Jasmine, D.J., Agastian, P., 2013. *In vitro* antioxidant activity and in vivo alpha glucosidase activity of endophytic actinomycetes isolated from *Catharanthus roseus* (l.) G. Don. Journal of pharmacy research. 6(6), 674–678.

[15] Dhayanithy, G., Subban, K., Chelliah, J.. 2019. Diversity and biological activities of endophytic fungi associated with *Catharanthus roseus*. BMC Microbiology.

19(22), 1–14.

[16] Ayob, F.W., Simarani, K., 2016. Endophytic filamentous fungi from a *Catharanthus roseus*: Identification and its hydrolytic enzymes. Saudi Pharmaceutical Journal. 24(3), 273–278.

[17] Birat, K., Binsuwaidan, R., Siddiqi, T.O., et al., 2022. Report on vincristine-producing endophytic fungus *Nigrospora zimmermanii* from leaves of *Catharanthus roseus*. Metabolites, 12(11), 1119.

[18] Holcomb, G.E., 1998. First report of Choanephora flower spot and blight of periwinkle. Plant Disease. 82(4), 447.

[19] Holcomb, G.E., 2000. First report of *Sclerotium rolfsii* on *Catharanthus roseus*. Plant disease. 84(2), 200.

[20] Montano, H.G., Dally, E.L., Davis, R.E., 2001. First report of natural infection by "Candidatus phytoplasma brasiliense" in *Catharanthus roseus*. Plant Disease. 85(11), 1209.

[21] Sharma, P., Meena, P.D., Singh Y.P., 2013. New record of twig blight on *Catharanthus roseus* in India. African Journal of Microbiology Research. 7(38), 4680–4682.

[22] Ou-Yang, W., Wu, W.S., 1998. Survey of periwinkle diseases in Taiwan. Plant Pathology Bulletin. 7(3), 147–149

[23] Sun, Z.X., Hsiang, T., 2016. First report of *Sclerotinia sclerotiorum* on periwinkle (*Catharanthus roseus*) in Ontario, Canada. Plant Disease. 100(8), 1789.

[24] Mridha, M.A.U., Rahman, M.M., 2015. Leaf blight of *Catharanthus roseus* (L). G. Don caused by *Macrophomina phaseolina* (TASSI) Goid and its *In vitro* control through bio-pesticides. Pakistan Journal of Botany. 47(2), 741–745.

[25] Hao, W., Richardson, P.A., Hong, C.X., 2010. Foliar blight of annual vinca (*Catharanthus roseus*) caused by *Phytophthora tropicalis* in Virginia. Plant Disease. 94(2), 274–274.

[26] Garibaldi, A., Bertetti, D., Gullino, M.L., 2006. First report of leaf blight caused by *Rhizoctonia solani* AG 1B on Madagascar periwinkle (*Catharanthus roseus*) in Italy. Plant Disease. 90 (10), 1361–1361.

[27] Baysal-Gurel, F., Bika, R., Simmons, T., et al., 2022. Identification and management of Phytophthora aerial blight caused by *Phytophthora nicotianae* on *Catharanthus roseus*. Plant Disease. 106(4), 1271–1277.

[28] Intaparn, P., Noireung, P., Maumoon, R., et al., 2020. First record of Phytopythium sp. causing root and stem rot on *Catharanthus roseus* in Thailand. Plant Pathology & Quarantine. 10(1), 10–20. DOI: https://doi.org/10.5943/ppq/10/1/2

[29] Benkirane, R., 1995. Contribution à l'étude des maladies du riz au Maroc. Cas de la pyriculariose due à *Pyricularia oryzae*, 3rd cycle [Doctoral thesis]. Ibn Tofail University, Faculty of Science: Kenitra, Marocco. 189p. (in French)

[30] Okayo, R.O., Andika, D.O., Dida, M.M., et al., 2020. Morphological and molecular characterization of toxigenic *Aspergillus flavus* from groundnut kernels in Kenya. International Journal of Microbiology. 2020(1), 8854718. DOI: https://doi.org/10.1155/2020/8854718

[31] Samson, R.A., Visagie, C.M., Houbraken, J., et al., 2014. Phylogeny, identification and nomenclature of the genus *Aspergillus*. Studies in mycology. 78(1), 141–173. DOI: https://doi.org/10.1016/j.simyco.2014.07.004

[32] Visagie, C.M., Houbraken, J., Frisvad, J.C., 2014. Identification and nomenclature of the genus *Penicillium*. Studies in mycology. 78(1), 343–371.

[33] Frisvad, J.C., Hubka, V., Ezekiel, C.N., et al., 2019. Taxonomy of *Aspergillus* section Flavi and their production of aflatoxins, ochratoxins, and other mycotoxins. Studies in mycology. 93(1), 1–63. DOI: https://doi.org/10.1016/J.SIMYCO.2018.06.001

[34] Qostal, S., Kribel, S., Chliyeh, M., et al., 2019. *Curvularia spicifera*, a parasite of the fungal complex of root rot of wheat and barley in Morocco. Plant Cell Biotechnology and Molecular Biology. 20(9&10), 354–365.

[35] Gilman, C.J., 1957. A manual of soil fungi, 2nd ed. The Iowa State College Press: Ames, IA, USA. p. 452.

[36] Tarr, S., 1962. Diseases of Sorghum, Sudan Grass and Broom Corn. The Commonwealth Mycological Institute: Kew, Surrey, UK. p. 380.

[37] Ellis, M.B., 1971. Dematiaceous Hyphomycetes. Commonwealth Mycological Institute: Kew, Surrey, UK. pp. 287 –608.

[38] Chidambaram, P., Mathur, S.B., Neergaard, P., 1974. Identification of seed-borne *Drechslera* species. Handbook on Seed Health Testing, series 2 B. 10(3), 165–207.

[39] Domsch, K.H., Gams, W., Anderson, T.H., 1980. Compendium of soil fungi, Volume 1. Academic Press: London, UK. 859p.

[40] Champion, R., 1997. Identifier les champignons transmis par les semences. INRA: Editions Quae, Paris. pp. 1–400. (in French)

[41] Ponchet, A., 1966. Etude des contaminations mycopéricarpiques du caryopse du blé. Crop Research (Hisar). 7(3), 554–460. (in French)

[42] Anahosur, K.H., 1992. Sorgum diseases in India: Knowledge and research needs. In: de Milliano, W.A.J., Frederiksen, R.A., Bengston, G.D. (Eds.). Sorghum and millets diseases: a second world review. International crops research institute for the semi-arid tropics: Patancheru, IN, USA. pp. 45–56.

[43] Singh, S.D., Bandyopadhyay, R., 2000. Grain mold. In: Frederiksen, R.A., Odvody, G.N. (Eds.). Compendium of Sorghum Diseases. American Phytopathological Society: St. Paul, MN, USA. pp. 38–40.

[44] MacKenzie, S.J., Xiao, C.L., Mertely, J.C., et al., 2003. Uniformity of strawberry yield and incidence of *Botrytis cinerea* fruit rot in an annual production system.

Plant Disease. 87(8), 991–998.

[45] Gabriolotto, C., Monchiero, M., Gre, M.L., et al., 2009. Effectiveness of control strategies against *Botrytis cinerea* in vineyard and evaluation of the residual fungicide concentrations. Journal of Environmental Science and Health Part B. 44(4), 389–396.

[46] Monteiro, E., Gonçalves, B., Cortez, I., et al., 2022. The Role of biostimulants as alleviators of biotic and abiotic stresses in Grapevine: A Review. Plants. 11(3), 396.

[47] Schmid, A., Daniel, C., Weibel, F., 2005. Effect of cultural methods on leaf spot (*Mycosphaerella fragariaegray*) and gray mold (*Botrytis cinerea*) damage in strawberries. BioControl. 50(1), 179–194.

[48] Mouden, N., Al Batnan, A., Benkirane, R., et al., 2016. Diversity and distribution of fungi from strawberry plants grown in Gharb-Loukkos (Morocco). International Journal of Recent Scientific Research. 7(10), 13630–13641.

[49] O'Neill, T.M., Shtienberg, D., Elad, Y., 1997. Effect of some host and microclimate factors on infection of tomato stems by *Botrytis cinerea*. Plant Disease. 81(1), 36–40.

[50] Mouria, B., 2009. Contribution à la lutte biologique contre la pourriture grise et la verticilliose de la tomate cultivée sous serre par utilisation du compost et les *Trichoderma* spp. seul ou en combinaison. Ibn Tofail University: Kenitra, Marocco. p. 295. (in French)

[51] Van Beneden, S., Pannecoucque, J., Debode, J., et al., 2009. Characterization of fungal pathogens causing basal rot of lettuce in Belgian greenhouses. European Journal of Plant Pathology. 124(1), 9–19.

[52] Bika, R., Baysal-Gurel, F., Jennings, C., 2021. *Botrytis cinerea* management in ornamental production: a continuous battle, Canadian Journal of Plant Pathology. 43(3), 345–365. DOI: https://doi.org/10.1080/07060661.2020.1807409

[53] Araújo, A.E., Maffia, L.A., Mizubuti, E.S.G., et al., 2005. Survival of *Botrytis cinerea* as mycelium in rose crop debris and as sclerotia in soil. Fitopatologia Brasileira. 30, 516–521.

[54] Meddah, N., Ouazzani Touhami, A., Benkirane, R., et al., 2006. Caractéristation de la mycoflore pathogène d'*Hibiscus rosa sinensis* L. et d'*Acalypha wilkesiana* J. Mueller de la ville de Kénitra (Maroc). Bulletin de l'Institut scientifique, Rabat, section sciences de la vie. 28, 7–11. (in French)

[55] Garibaldi, A., Bertetti, D., Gullino, M.L., 2009. First report of *Botrytis* blight caused by *Botrytis cinerea* on Periwinkle (*Catharanthus roseus*) in Italy. Plant Disease. 93(5), 554. DOI: https://doi.org/10.1094/PDIS-93-5-0554B

[56] Pikovskyi, M.Y., Kolesnichenko, O.V., Melnyk, V.I., et al., 2018. Flower-ornamental plant- the host of *Botrytis cinerea*. Bioresources and Nature Management. 10, 5–10.

[57] Mouden, N., Ouazzani Touhami, A., Benkirane, R., et al., 2023. Comparative pathogenic capacity of three fungal species onto detached leaves of sabrina strawberry variety. In Proceedings of the 5th International Food, Agriculture and Veterinary Sciences Congress; Kafkas University, Kars, Türkiye, 17–19 March 2023.

[58] Lamrani, N., Elabdellaoui, F., Touhami, A.O., et al., 2013. Etude De La Mycoflore Des Grains De Trois Variétés De Riz Et Effet d'*Alternaria Padwickii* (Ganguly) Mb Ellis Sur Les Grains Pré-Germés. Bulletin De L'Institut Scientifique, Rabat. 35, 1–7. (in French)

[59] Park, J., Kim, S., Jo, M., et al., 2024. Isolation and Identification of *Alternaria alternata* from Potato Plants affected by leaf spot disease in Korea: Selection of Effective Fungicides. Journal of Fungi. 10(1), 53. DOI: https://doi.org/10.3390/jof10010053

[60] El Aymani, I., Qostal, S., Mouden, N., et al., 2019. Fungi associated with saffron (*Crocus sativus*) in Morocco. Plant cell biotechnology and Molecular Biology. 20, 1180–1188.

[61] Ourras, S., Aymani, I.E., Mouden, N., et al., 2022. Mycoflora of Dormant *Crocus Sativus* Corms in Morocco. In International Conference on Advanced Intelligent Systems for Sustainable Development. Springer: Cham, Switzerland. pp. 479–489.

[62] Masunaka, A., Ohtani, K., Peever, T.L., et al., 2005. An isolate of *Alternaria alternata* that is pathogenic to both tangerines and rough lemon and produces two host selective toxins, ACT- and ACR-toxins. Phytopathology. 95(3), 241–247.

[63] Maouni, A., Lamarti, A., Aidoun, A., et al., 2007. Effect of benzimidazole fungicides and calcium chloride on *Alternaria alternata* and *Penicillium expansum* rot during storage of pears. African Journal of Biotechnology. 6(11), 1289–1292.

[64] Bigre, J.-P., Morand, J.-C., Tharaud, M., 1987. Pathologie des cultures florales et ornementales. Ed. Lavoisier: Paris, France. 233p. (in French)

[65] Lahuf, A.A., 2019. *Alternaria alternata* causes leaf blight of rosy periwinkle (*Catharanthus roseus*) in Iraq. Australasian Plant Disease Notes. 14(4), 1–3. DOI: https://doi.org/10.1007/s13314-019-0334-9

[66] Krishnamurthy, Y.L., Naik, S.B., Jayaram, S., 2008. Fungal communities in herbaceous medicinal plants from the Malnad region, Southern India. Microbes and environments. 23(1), 24–28.

[67] Ramalingam, P., Muthukrishnan, S. 2013. Screening and characterization of antimicrobial compound from endophytic fungus *Curvularia lunata*, isolated from *Catharanthus roseus*. World Journal of Pharmaceutical research. 2(6), 3078–3086.

[68] Kharwar, R.N., Verma, V.C., Strobel, G., et al., 2008. The endophytic fungal complex of *Catharanthus roseus* (L.) G. Don. Current science. 95(2), 228–233.

[69] Pandey, S.S., Singh, S., Babu, C.V., et al., 2016. Fungal endophytes of *Catharanthus roseus* enhance vindoline content by modulating structural and regulatory genes related to terpenoid indole alkaloid biosynthesis. Scientific reports. 6(1), 26583.

[70] Hassikou, K., Hassikou, R., Douira, A., 1997. Behaviour of some rice cultivars in relation to *Curvularia lunata*. Phytopathologia Mediterranea. 36(3), 163–164.

[71] Knox, G.W., Klingeman, W.E., Paret, M., et al., 2012. Management of pests, plant diseases and abiotic disorders of *Magnolia* species in the Southeastern United States: a review. Journal of Environmental Horticulture. 30(4), 223–234.

[72] Kafur, A., Basheer Khan, A., 2011. Isolation of endophytic actinomycetes from *Catharanthus roseus* (L.) G. Don leaves and their antimicrobial activity. Iranian Journal of Biotechnology. 9(4), 302–306.

[73] Lambert, L., 2004. Pourritures des jeunes plants en culture ornementale. Cultures en serres. 1–4. (in French)

[74] Hyeon, N.M., Park, M.S., Kim, H.G., et al., 2009. Biological control of strawberry Fusarium wilt caused by *Fusarium oxysporum* f. sp. fragariae using *Bacillus velezensis* BS87 and RK1 formulation. Journal of Microbiology Biotechnology. 19(5), 520–524.

[75] Meddah, N., Ouazzani Touhami, A., Douira, A., 2010. Mycoflore associée au bananier (*Musa accuminata* L.), variété Grande naine, cultivé sous serre dans la région du Gharb (Maroc). Bulletin de l'Institut Scientifique, Rabat, section Sciences de la Vie. 32(1), 1–11.

[76] Yasir, L.B., Almaliky, B.S., 2023. Integrated control of root rot and wilt disease on *Catharanthus roseus* using biological and chemical control. Iraqi Journal of Market Research and Consumer Protection. 15(1), 132–146.

[77] Abd-Elsalam, K.A., Omar, M.R., El-Samawaty, A.R., et al., 2007. Response of commercial cotton cultivars to *Fusarium solani*. The Plant Pathology Journal. 23(2), 62–69.

[78] Rieuf, P., 1965. Champignons observés sur *Geranium rosat* (*Pelargonium capitatum* AIT.) au Maroc. Al Awamia. 16, 43–98.

[79] Subramanyam, P., 1991. Control of seedling disease of groundnut. Nigerian Tropical Pest Management. 37, 118–119.

[80] Islam, N.F., Borthakur, S.K., 2012. Screening of the mycota associated with *Aijung* rice seed and their effects on seed germination and seedling vigour. Plant Pathology & Quarantine. 2(1), 75–85.

[81] Vigier, B., Reid, L.M., Seifert, K.A., et al., 1997. Distribution and prediction of *Fusarium* species associated with maize ear rot in Ontario. Canadian Journal of Plant Pathology. 19(1), 60–65.

[82] El-Meleigi, M.A., Claflin, L.E., Raney, R.J., 1983. Effect of seedborn *Fusarium moniliforme* and irrigation scheduling on colonization of root and stalk tissue, stalk rot incidence and grain yield. Crop Science. 23(6), 1025–1028.

[83] Cardwell, K.F., Kling, J.G., Maziya-Dixon, B., et al., 2000. Interactions between *Fusarium verticillioides*, *Aspergillus flavus*, and insect infestation in four maize genotypes in lowland Africa. Phytopathology. 90(3), 276–284.

[84] Farrar, J.J., Davis, R.M., 1991. Relationship among ear morphology, western flower thrips, and *Fusarium* ear rot of corn. Phytopathology. 81(6), 661–666.

[85] Fraval, A., Silvy, C., 1999. La lutte biologique (II). Dossiers de l'environnement de l'INRA. 19, 274. (in French)

[86] Elad, Y., Chet, I., Boyle, P., et al., 1983. Parasitism of *Trichoderma* spp. on *Rhizoctonia solani* and *Sclerotium rolfsii*. Scanning electron microscopy and fluorescence microscopy. Phytopathology. 73(1), 85–88.

[87] Wells, H.D., Bell, D.K., Jaworski, C.A., 1972. Efficiency of *Trichoderma harzianum* as a biocontrol for *Sclerotium rolfsii*. Phytopathology. 62(4), 442–447.

[88] Ezzahiri, B., Sekkat, A., 2001. Maladies et ravageurs de l'arachide identification et moyens de lutte. Bulletin Mensuel d'Information et de Liaison du PNTTA. Transfert de Technologie en Agriculture. 86, 1–4. (In French)

[89] Mendes, M.A.S., da Silva, V.L., Dianese, J.C., 1998. Fungos em Plants no Brasil. Embrapa-SPI/Embrapa-Cenargen, Brasilia, Brazil. p. 555. (in Portuguese)

[90] Teodoro, N.G., 1937. An enumeration of Philippine fungi. Technical bulletin 4, Philippines Department of Agriculture and Commerce. 4, 585.

[91] Lumyong, P., Photita, W., McKenzie, E.H.C., et al., 2003. Saprobic fungi on dead wild banana. Mycotaxon. 85, 345–346.

[92] Zhuang, W.Y., 2001. Higher fungi of tropical China. Mycotaxon, Ltd.: Ithaca, NY, USA, p. 485.

[93] Gnancadja-André, L., Ouazzani Touhami, A., Badoc, A., et al., 2004. Test de détection des contaminants fongiques des grains de riz en fin de cycle végétatif. Bulletin de la Société de pharmacie de Bordeaux. 143(1–4), 39–50. (in French)

[94] Alsubaie, S., Bokhari, F., Najjar, A., 2023. Diversity of endophytic and exophytic fungi isolated from *Catharanthus roseus* LG Don. leaves in Saudi Arabia. AGBIR. 39(3), 572–577.