STUDIES ON DERMATOPHYTES ISOLATED FROM PATIENTS IN TWO TERTIARY HEALTH INSTITUTIONS IN LAGOS STATE, NIGERIA

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A THESIS SUBMITTED FOR THE DEGREE OF DOCTOR OF PHILOSOPHY (BOTANY)

DEPARTMENT OF BOTANY,

UNIVERSITY OF LAGOS, NIGERIA

NOVEMBER, 2012

CERTIFICATION

We certify that the work embodied in this thesis for the award of the degree of Doctor of Philosophy (Botany) by **OGUNTADE, TEMITOPE OLUWASEUN** has been carried out under our supervision in the Department of Botany, University of Lagos.

PROF. A. A. ADEKUNLE	**DATE**
PROF. O. T. OGUNDIPE	**DATE**

DEDICATION

TO GOD BE THE GLORY

This work is dedicated to God Almighty and to the memory of my late mother, **Mrs. E.M.O. Oguntade**

ACKNOWLEDGEMENT

I give glory and praise to God Almighty, the all-knowing and life-giver for enabling me to carry out this work successfully. I remain indebted to my supervisor and mentor, Prof. A.A. Adekunle for giving everything and imprinting in my conscience a legacy of striving to attain excellence. Sir, may you live long to reap all you have sown. Also, to my co-supervisor, Prof. O.T. Ogundipe, whose help, stimulating suggestion, encouragement, constructive criticisms and contributions to this work has helped me during the period of this research. I say God bless you sir.

A special thanks to Prof. J.D. Olowokudejo for his encouragement and concern at every stage of this study. I wish to express my deep appreciation to all academic staff in the Department of Botany; Dr. A. Akinsoji, Dr.(Mrs). C.E. Umebese, Dr.(Mrs). O.E. Ade-Ademilua, Dr. V.J. Ojegba, Dr.(Mrs). O.O. Shonubi, Dr.(Mrs). T.A. Adesalu, Dr.(Mrs). E.M. Adongbede, Dr. A.B. Kadiri, Dr.(Mrs). H.O. Adekanmbi and Dr. A.P. Adeonipekun for their assistance and encouragement during the period of this research work. To all non-teaching staff of the department of Botany, I am grateful.

I also wish to acknowlegde Dr (Mrs) O. S. Odesanmi, Dr. K. Adekoya, Dr. L.A. Ogunkanmi and Dr. A. Omotayo for their invaluable criticism on my work and the patience of going through my manuscript. My immense gratitude goes to Dr. J.K. Saliu, Dr. A.B. Adeloye, Mr G. Adesokan (FIRS), Mr. E. Adegoke, Mrs. A. Ajibode (SPGS) and Mr. T. Akeju (SPGS) for the inspiration and encouragment provided by them.

My profound gratitude goes to the following people, who assisted me in various ways in the course of carrying out this research. Dr.(Mrs). A.M. Ebabhi, Dr. T.A. Adeyemi, Mr. Adeogun, Dr. Sanyaolu, Dr.(Mrs). Kanife, Mr. Iwuyanwu (LUTH), Mr. Ohakem (CPHL) Mr. Adeleke (Pharmacognosy), Mr. Adura Ogundele and Mr. Abiodun Anipole. I appreciate you all. I must say a BIG THANK YOU to my daddy and his wife, Mr. and Mrs. M.A. Oguntade, my siblings (Tonia, Tosin, Tunji, Toyin), In-laws, friends and members of my family, who have remained loyal and supportive throughout my studies; Sola, Bose Ugboke, the Olaoyes, the Oguntades, Mrs. M.O. Oresanya, Mrs B.O. Omaigho, the Samuels, the Seleres and the Ajadis. Words cannot describe the depth of my gratitude to you all.

I wish to particularly thank Prof. M. Arabatzis of School of Medicine, University of Athens, Greece. for his countless efforts towards the success of this study. Special appreciation to Biotechnology Centre, University of Agriculture, Abeokuta, Ogun state and Dept. of Plant and Biotechnology, University of Johannesburg, South Africa for the molecular training inculcated in me. I will not forget to thank University of Lagos for the Graduate Fellowship which was helpful in the course of this study.

Finally, to my best friend, companion, brother, better-half and lovely husband OLUSEGUN. B. SAMUEL. I will forever remain grateful to you for giving the best of peace, understanding, encouragement, love and support which i needed for the successful completion of this work. I will certainly not forget “the apple of my eyes” TEMIDAYO. B. SAMUEL, whose presence in my life motivated the success of this work. I cheerish your love.

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ABSTRACT

Studies on dermatophytes isolated from patients in tertiary health institutions in Lagos State Nigeria were carried out between August 2009 and January 2011. Collection, isolation, characterization and identification of the isolated fungi were achieved using both conventional laboratory methods (*In-vitro* culture and microscopy) and molecular methods (DNA extraction, PCR-RFLP and DNA sequencing). Sensitivity tests using some marketed antifungal drugs, antiseptic soaps, detergents and extracted oil from three Nigeria plants seeds (*Azadirachta indica, Detarium seneganlense* and *Treculia africana*) was investigated. Growth study using six different media was also carried out. Twenty- five different isolates were obtained, and based on molecular data, were classified into ten species and two strains of dermatophytes (*Epidermohyton fluccosum, Microsporum audouinii, Microsporum ferrugineum, Microsporum nanum, Trichophyton concentricum* strain A, *Trichophyton concentricum* strain B, *Trichophyton mentagrophytes var. quinckeanun, Trichophyton rubrum, Trichophyton soudanense, Trichophyton tonsurans* strain A, *Trichophyton tonsurans* strain B and *Trichophyton violaceum,*), two species of systemic mycoses agents (*Blastomyces dermatitidis* and *Exophiala dermatitidis*), seven opportunistic mycoses agents (*Absidia corymbifera, Aspergillus terreus, Emericlla nidulans, Mucor racemosus, Pencillium aschersonia, Pencillium citrinum, Pencillium* species) and four yet to be identified fungal species. The result of the BLAST DNA sequence database of the isolated dermatophytes revealed that no sequence data was 100% homologous with those in the Gene bank. The sensitivity test revealed that resistance to the antifungal drugs might be a result of probable emergence of new strains of dermatophytes in the populace and development of resistance. The results of the sensitivity tests showed that the various antiseptic soaps and detergents used could be

a mean of reducing the incidence of dermatomycoses in the population. The extracted oil from the seeds of *Detarium senegalense* was shown to possess the highest zone of inhibition among the three plant seeds used. The phytochemical test revealed that the three extracted seed oils (*Azadirachta indica*, *Detarium senegalense* and *Treculia africana*) contained alkaloid, tannins, cardiac glycosides and reducing compounds but in different percentage ratios. Growth studies showed that one percent (1%) peptone agar is the best medium for the cultivation *Trichophyton* and *Epidermaphyton* species, and *Microsporum* species attained their optimum growth on Sabouraud dextrose agar (SDA). The growth studies also revealed that nutrient constituents of each medium can interfere with phenotypic characteristics of dermatophytes. The findings from these studies will help in preventing wrong diagnosis of dermatomycoses in health institutions.

CHAPTER ONE

1.0 INTRODUCTION

1.1 BACKGROUND OF STUDY

Fungi are eukaryotic, heterotrophic, unicellular to filamentous, rigid cell walled, spore-bearing organisms that usually reproduce by both sexual and asexual means. They have cell walls that contain chitin and glucans as the skeletal components, which are embedded in a matrix of polysaccharides and glucoproteins (Agrios, 1997).

Human pathogenic fungi are of five categories; superficial mycoses, subcutaneous mycoses, cutaneous mycoses (dermatophytes); systemic and opportunistic mycoses (Uma, 2009). Dermatophytes are associated with diseases of the skin in man, animal or both. Some, if not all, appear to survive in the soil, thus becoming a source of infection (Uma, 2009). Dermatophytes are a highly specialized group of fungi that through long evolutionary processes became adapted to invade, colonize and nourish themselves on the keratinized tissues of human and animals (Ajello, 1974).

Dermatophytes are pathogenic fungi specialized in the infection of skin, hair and nails that utilize keratinous substrates as the Carbon, Nitrogen and Sulphur sources. They belong to three anamorphic (asexual or imperfect) genera, *Epidermophyton, Microsporum and Trichophyton*, and have long been classified as anthropophilic, zoophilic and geophilic species on the basis of their primary habitat associations. Anthropophilic dermatophytes are associated with humans and rarely infect other animals. Zoophilic dermatophytes usually infect animals or are associated with animals, but occasionally infect humans and geophilic dermatophytes are primarily associated with keratinous materials such as hair, feathers, hooves and horns. When

dermatophytes infect humans, they colonize the keratinized outermost layer of the skin, and usually do not invade the living tissue (Martinez-Rossi *et al*., 2008).

Human skin is a dynamic environment, hosting a range of organisms including bacteria, viruses, fungi, parasites, and even mites (MacLeod *et al.,* 2009). For most individuals, the skin barrier ensures that the majority of these microorganisms can be hosted or encountered without concern, although the frequency of these interactions inevitably increases the prevalence of cutaneous infection. Fungi can also be regular inhabitants of the normal human skin flora, although many of the species colonizing the skin under normal conditions are also responsible for infections in compromised host skin (MacLeod *et al.,* 2009). These three types of keratinophilic fungi are responsible for the majority of fungal skin infections in humans. The three genera, *Trichophyton*, *Microsporum*, and *Epidermophyton*, together comprising more than forty different species, and can be transmitted in many different ways, including person-to-person contact, contact with geophilic organisms in the soil during outdoor activities (especially in children), spread via animal vectors, that is intimate association with pet animals, even indirectly through inanimate objects carrying the fungi and poor personal hygiene may contribute to the spread of these infections among children (Enemuor and Amedu, 2009**)**.

Fungal infections are primarily localized to the hair, nails, and outer layers of the skin, reflecting the affinity of the fungi for keratin and the inability of the fungi to invade deeper tissues or organs in immunocompetent skin (MacLeod *et al.,* 2009). Infections can cause a number of signs and symptoms, including extensive inflammation, itching, and cracking of the skin. Furthermore, circular shape of the skin lesions often

has a red periphery surrounding a clear healed area and infected nails appear creaked and yellowed from infection. Bald areas on the scalp may appear at the site of infection (Gupta and Summerbell, 2000). These infections are referred to as dermatomycoses.

Dermatophytes are the most common cause of fungal infections worldwide, affecting approximately 20% of the population (Marques *et al.,* 2000). Many epidemiological studies have investigated the prevalence of etiologic agents of superficial mycoses in different parts of the world (Ayadi *et al.,* 1993; Venugopal and Venugopal, 1993; Ellabib and Khalifa, 2001; Anosike *et al.,* 2005). Host susceptibility may be enhanced by moisture, warmth, specific skin chemistry, composition of sebum and perspiration, age, heavy exposure and genetic predisposition (Brooks *et al.,* 2004). Dermatomycoses is highly contagious and represents a significant public health problem in Nigeria, particularly Lagos State an urban city with higher incidence of dermatophytosis due to hot humid climates and crowded living condition. (Adetosoye, 1977; Higgins *et al.,* 2000; Omar, 2000; Fathi and Al-Samaria, 2001; Anosike *et al.,* 2005). A number of studies on the prevalence and etiological aspects of dermatomycoses have been carried out in different parts of Nigeria (Soyinka, 1978; Egere and Gugnani, 1980; Ajao and Akintunde, 1985; Ogbonna *et al.,* 1985; Obasi and Clayton, 1989; Ive, 1996; Anosike *et al.,* 2005; Nweze and Okafor, 2005; Ozumba and Nlemadim, 2005; Mbata and Nwajagu, 2007).

A reasonable number of antifungal agents exist currently on the pharmaceutical market, together with some derivatives of these drugs that have become less toxic, with enhanced potencies and pharmacokinetics (Martinez-Rossi *et al.,* 2008). These

antifungal drugs interfere with the normal life cycle of fungi by inhibiting normal functioning of one or several vital cellular entities. The effects of these agents are reflected in altered patterns of growth, differentiation, transformation, and viability of the fungus. Sub cellular entities that may be injured reversibly or irreversibly are the Cell Wall, Plasma Membrane, Nucleus, Mitochondria, Microtubules, Ribosomes, Intracellular Membranes, and Peroxisomes. Damage to any of these organelles affects cellular functioning in general (Martinez-Rossi *et al.,* 2008). The detailed knowledge, acquired mainly in the last decade, of the structure, composition, and biochemistry of the fungal cell has contributed to the understanding of the mechanism of action of many antifungal drugs. With few exceptions, the primary sites of action of the antifungal agents have been established.

There is an expanding arsenal of therapeutic agents with selective toxicity for fungi. These drugs vary in potency and range of activity. The main compounds that are used widely today in antifungal therapy are the polyenes, antibiotics, allylamines and thiocarbomates, pyrimidine, echinocandins and the azole derivatives (Martinez-Rossi *et al.,* 2008). The usefulness of these substances vary considerably. Their potency, spectrum of activity and limitations are closely related to their modes of action. Another primary determinant of their usefulness is the mode of administration; some of the drugs must be given topically and others systemically. Barriers to systemic use are inadequate absorption from the gastrointestinal tract, metabolic inactivation, toxicity, and the intrinsic complexity of intravenous treatment (Borgers, 1980).

1.2 STATEMENT OF PROBLEM

Dermatophytes being human pathogenic fungi infect almost every human alive at some point over the course of his or her lifetime. Over USD $ 500,000,000 per year is spent worldwide for drugs targeted against dermatomycoses (Kane *et al.,* 1997). Current treatments are able to control infection to some certain level, but there is a major problem of recurrence and it is unclear whether this is due to occurrence of resistance in the clinical isolates that leads to failure in the treatment, or wrong diagnosis and identification of the dermatophytes at the species level and beyond, based on the use of conventional methods (microscopic identification of hyphae from lesion materials and *in-vitro* culture) at various mycology laboratories.

Although many antifungal drugs have been developed during the last two decades, they are confined to a relatively few chemical classes. In addition, the occurrence of resistance in clinical isolates leads to failure in the treatment of dermatomycoses. Thus, the effective control of dermatophytes will necessarily involve the development of a new generation of potent broad spectrum antifungal agent with selective action against new targets in the fungal cells, without irreversible side effects in the host. An antifungal resistance mechanism in dermatophytes is very possible, since the development of resistant strains to any new drug is inevitable.

Clinically it is important to identify the species causing infections in order to enable appropriate treatment. However, conventional methods do not give precise identification of the pathogen due to lack of stable characteristics such as colony morphology or microscopic appearance. These characteristics are not easily noticeable when subcultured, or might be artifacts due to growth conditions or

contaminating bacteria (Shin *et al.,* 2003). Thus, hampering the identification of strain-related differences, which could be useful in answering certain epidemiological questions such as sources of infection or strains belonging to a specific geographical area. This would also help in identifying the types of infection caused, in addition to determining the cause of relapse either being a failure in treatment or infection by a new strain (Baeza and Giannini, 2004).

The current diagnosis of dermatomycoses in almost all mycology laboratories in Lagos state, Nigeria is based upon conventional methods, that is, microscopic identification of hyphae directly from lesion materials followed by *in-vitro* culture. Although rapid and economical, but this microscopical examination is not species-specific. Therefore its role in dermatophyte diagnosis is limited to giving an initial, quick screen for hyphae and other mycotic elements, and further follow up with *in-vitro* culture is invariably required irrespective of the microscopic observations, as effective treatment and prevention of dermatophytosis depend on knowledge of the specific dermatophyte involved.

Recently, molecular approaches for species identification have been used successfully to identify dermatophytes precisely and rapidly (Liu *et al.,* 2002; Baeza and Giannini 2004). Several methods for identifying dermatophytes using molecular techniques have been reported to date, some of which include steps such as DNA extraction, polymerase chain reaction and restriction fragment length polymorphism targeting (PCR-RFLP) and DNA sequencing (Jackson *et al.,* 1999; Shin *et al.,* 2003). Even though complex, have been found to be more specific (Sahgal and Magan, 2008). The identification of species or strain of dermatophytes is an important part of laboratory

investigations of dermatophytic infections because dermatophytes are among the most frequently observed organisms in biomedicine, yet there has never been stability in the taxonomy, identification and naming of the approximately forty pathogenic species involved (Gräser *et al.*, 2006).

1.3 THE AIM AND OBJECTIVES OF STUDY

In view of the above-mentioned statement of problem, it therefore becomes necessary to accurately isolate, identify and characterize these pathogenic dermatophyte fungi using molecular techniques with the aim to provide more data on their resistance and susceptibility to available antifungal drugs in the Nigerian market. This will also help in designing new effective drugs with potent broad spectrum antifungal action.

The objectives are to:

1. Collect, isolate and identify (conventional laboratory methods) pathogenic fungi that cause skin diseases from patients at different tertiary health institutions in Lagos state, Nigeria.
2. Confirm the identity of the pathogenic fungi using molecular techniques and document the DNA sequence data generated from the isolated dermatophytes species/strains.
3. To investigate the growth patterns of the dermatophytes on different growth media.
4. Determine the sensitivities of the pathogenic fungi to different orthodox antifungal drugs, antiseptic soaps, detergents and extracted oils from three plant seeds.

1.4. SIGNIFICANCE OF THE STUDY

Based on the recent reports by several authors on the major problems of recurrence of dermatomycoses which is not clear whether this is due to resistance in these fungal isolates that leads to failure in the treatment, or wrong diagnosis and identification of the dermatophytes at the species level and beyond. This study is therefore designed to provide accurate identification to the available taxa of this group of organisms in Lagos State, Nigeria; investigate the actual cause of the recurrence of dermatomycoses in patients and determine the best medium for the cultivation of these groups of organisms.

1.5 RESEARCH QUESTIONS

1. Are there different strains of dermatophytes and related fungi in Lagos state, Nigeria?
2. Will there be a difference in identification of these fungi using modern molecular techniques as compared with conventional methods?
3. What medium is best to culture this group of fungi?
4. Is this group of human pathogenic fungi actually resistant to some antifungal drugs in Nigeria markets?
5. What are the reactions of the isolated dermatophytes to some antiseptic soaps and detergents in Nigeria'markets?
6. What are the reactions of the isolated dermatophytes strains to the various antifungal drugs available in the Nigeriam market?

7. Can medicinal plants (extracted oils from plant seeds) be of help in curing the infectious caused by these groups of organisms?

1.6 OPERATIONAL DEFINITION OF TERMS

Anthroconidium: is a type of vegetative fungal propagule typically produced by segmentation of pre-existing fungal hyphae.

Anthropophilic: These are fungi usually associated with humans only; transmission from man to man is by close contact or through contaminated objects.

Dermatomycoses: Fungal disease of the skin, (cutaneous mycoses).

Dermatophyte: A fungus belonging to the genera *Epidermophyton*, *Microsporum*, or *Trichophyton* with the ability to utilize keratin to infect hair, nail and skin.

Dermatophytosis: (also called a Tinea) is a dermatophytic infection and normally classified according to location on the body.

Ectothrix: Natural hair invasion by a dermatophyte characterized by arthroconidia on the outside of the hair shaft.

Endothrix: Natural hair invasion by a dermatophyte characterized by the development of arthroconidia within the hair shaft only.

Geophilic: Literarily "earth loving." Species that occur naturally in soil, presumably as a saprobe, but is capable of infecting both humans and animals.

Mycelium (pl. mycelia): The mass of hyphae making up the thallus of a fungus.

Mycology: The study of fungi.

Mycoses: Diseases of warm blooded animals caused by fungi.

Pleomorphic: Having more than one form.

Resistance: the capacity of a species or strain of microorganism to survive exposure to a toxic agent (as a drug) formerly effective against it.

Spore: a reproductive propagule formed by either meiosis or mitosis. However, if by asexual means, cleavage of cytoplasm is usually involved.

Susceptibility: the quality or state of being susceptible; lack of ability to resist some extraneous agent (as a pathogen or drug).

Zoophilic: Literarily animal loving. The group of fungi (dermatophytes) that infect animals. Diseases can be transmitted from animals to humans.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 MYCOLOGY

Fungi are living organisms that are more closely related to plants than animals. Most of them are composed of living filaments that grow in a mass, and their size and shape are determined by the environment. Like plants they have rigid cell walls, but they differ from plants in their lack of chlorophyll, the green matter that enables plants to manufacture their own food from water and carbon dioxide in the presence of sunlight. As a result, all fungi are heterotrophs and derive their food from organic materials like green plants, animals and even man. They therefore live as saprophytes if their source of food is dead organic material or as parasites if they are able to feed on living organisms (Uma, 2009).

Fungi secrete extracellular enzymes to help degrade the various organic materials before absorbing them into their system for growth, reproduction and survival. Fungi can reproduce sexually, asexually and parasexually (Vashishta and Sinha, 2005). Fungi are studied under mycology; this is a discipline that deals with the nature, characteristic, taxonomy, phylogeny, physiology and reproduction of fungi (Bhattachanga *et al.,* 2011). What is usually termed pure mycology concerns the detailed structure, cytology and mode of development of fungi. Field mycologist are interested in the fungi which are to be found in fields and woods, both the larger forms, known as mushrooms and toadstools, which grow on the ground or as parasites of forest trees, and the microscopic forms found on plant debris or as parasites of plants. The taxonomist studies structure with a view to classifying fungi, so as to show relationships and facilitate identifications by others. Industrial mycology

includes both the harmful activities of fungi in rotting or spoiling industrial raw materials and manufactured goods, and the uses of fungi in industrial fermentations. Medical mycology deals with the fungi which cause diseases in man. There are several other aspects of mycology, like agricultural mycology and veterinary mycology to mention a few (Onions *et al*., 1981).

Human pathogenic fungi are of five categories which are distinguishable, based on the degree of tissue penetration and mode of entry. They include: superficial, subcutaneous, cutaneous (dermatophytes), systemic and opportunistic mycoses (Uma, 2009). Superficial mycoses are fungal cosmetic infections of the skin and hair shaft. No living tissue is invaded and there are no cellular responses from the host. Example of organism involved in this type of mycoses include; *Tinea versicolor* (McClellan *et al.,* 1999). Subcutaneous mycoses are infections caused by fungi which are saprophytic inhabitants of soil and decaying vegetation. They enter the subcutaneous tissue by means of an open wound that comes in contact with the infected soil carrying the fungus spores (McClellan *et al.*, 1999).

Cutaneous mycoses are infections caused by three genera of fungi known as **dermatophytes**, which include *Epidermophyton*, *Microsporium* and *Trichophyton.* The infection could be transmitted through person to person contacts. The fungus also occurs in domestic animals, from whom it can be transmitted to humans. An example is *Microsporium canis* which resides on the body of cats. Systemic mycoses are infection of susceptible patients. These fungi are dimorphic. They can exhibit parasitic yeast like phase, but most are acquired by inhalation of spores from soil, in which the mould phase resides. An example is *Blastomyces dematitidis,* this fungus is

budding yeast in humans and a mould in culture (McClellan *et al.,* 1999). Opportunistic mycoses are infections that are normally harmless and are pathogenic only in immune compromised hosts. Example is Aspergillosis (Barchiesi *et al.,* 1995).

2.2 DERMATOPHYTES

Historically, medical mycology, specifically relating to human disease, began with the discovery of the fungal etiology of "favus" and centered around three European physicians in the mid-19th century: Robert Remak, Johann L. Schonlein, and David Gruby (Weitzman and Summerbell, 1995). Although dermatophytes undoubtedly existed in prehistoric times and have plagued lower animals and man for millions of years, the infections were long endured before their true nature was realized. The first recorded reference to a dermatophyte infection is attributed to Aulus Cornelius Celsus, the Roman encyclopedist, who in his 'De Re Medicina' written around 30 A.D., described a suppurative infection of the scalp that came to be known as the kerion of Celsus (Rosenthal, 1961). In his era and down through the Middle Ages the various dermatophytoses were described as *Tineas.* The term *Tinea* was first used for the clothes moth and in fact, the generic name of the various species of keratin destroying moths is *Tinea* (Ajello, 1953). Since the holes made by moths in woolen garments are circular and dermatophyte lesions are ring like on smooth skin, our Anglo Saxon ancestors coined the word 'ringworm' for these infections at least as far back as the 16th century. This term described the form of the lesion and relates it to the Roman *tinea.*

Not until the 19th century was the mycotic etiology of these skin infections discovered. In 1837 Robert Remak, a Polish physician on the medical faculty of

Berlin University, noted hyphae in the crusts of the disease known as favus (Alkiewicz, 1967; Kisch, 1954). This was an epochal discovery since for the first time a microorganism was incriminated as being the cause of a human disease. In 1841 Remak's discovery was independently confirmed by David Gruby, a Hungarian physician practising in Paris, who carefully but succinctly went on to describe several different types of dermatophyte infections*: Tinea favosa*, ectothrix and endothrix trichophytosis and microsporiosis as cited by Ajello (1974).

The real founder of dermatomycology was David Gruby on the basis of his discoveries during 1841 to 1844, his communications to the French Academy of Science and his publications during this period (Weitzman and Summerbell, 1995). Independently, and unaware of the work of Remak and Schoenlein, he described the causative agent of favus, both clinically and in microscopic details of the crusts, and established the contagious nature of the disease. He also described ectothrix invasion of the beard and scalp, naming the etiologic agent of the latter *Microsporum* (referring to the small spores around the hair shaft) *audouinii*, and described endothrix hair invasion by *Herpes* (*Trichophyton*) *tonsurans*. In addition to his observations on dermatophytes, he also described the clinical and microscopic appearance of thrush in children. Raimond Sabouraud, one of the best known and most influential of the early medical mycologists, began his scientific studies of the dermatophytes around 1890, culminating in the publication of his classic volume, *Les Teignes*, in 1910 as cited by Weitzman and Summerbell (1995).

Sabouraud’s contributions included his studies on the taxonomy, morphology, and methods of culturing the dermatophytes and the therapy of the dermatomycoses. He

classified the dermatophytes into four genera, *Achorion*, *Epidermophyton*, *Microsporum*, and *Trichophyton*, primarily on the basis of the clinical aspects of the disease, combined with cultural and microscopic observations. The medium that he developed is in use today for culturing fungi (although the ingredients are modified) and is named in his honor, Sabouraud dextrose (glucose) agar (Odds, 1991). Sabouraud's treatment of *Tinea capitis* by a one-dose, single-point roentgenologic epilation achieved cures in 3 months as opposed to the then therapy of manual epilation and topical application of medications (Kwon-Chung and Bennet, 1992). In 1934, Chester Emmons modernized the taxonomic scheme of Sabouraud and others and established the current classification of the dermatophytes on the bases of spore morphology and accessory organs. He eliminated the genus *Achorion* and recognized only the three genera *Microsporum*, *Trichophyton*, and *Epidermophyton* on the basis of mycological principles. Gruby also discovered and named the genus *Microsporum* in 1843 and described *M. audouinii* on the basis of the appearance of the fungus in clinical materials. The genus *Microsporum* is one of the three genera of the Fungi Imperfecti (Deuteromycotina) in which the dermatophytes are classified today. *Trichophyton*, the second genus, was described in 1845 by Per Hendrik Malmsten, the Swedish investigator, *Epidermophyton*, the third and last one, was established by Raymond Sabouraud in 1910 as cited by Ajello (1974).

Dermatophytes have long been divided into anthropophilic, zoophilic, and geophilic species on the basis of their primary habitat associations (Ajello, 1962). Anthropophilic dermatophytes are primarily associated with humans and rarely infect other animals (Mayr, 1989). Zoophilic dermatophytes usually infect animals or are associated with animals but occasionally infect humans. Geophilic dermatophytes are

primarily associated with keratinous materials such as hair, feathers, hooves, and horns after these materials have been dissociated from living animals and are in the process of decomposition. These species may cause human and animal infection. Geophilic species are thought to have been ancestral to the pathogenic dermatophytes, preadapted to cutaneous pathogenesis by their ability to decompose keratin and their consequent close association with animals living in hair and feather-lined nests in contact with soil (Chmel, 1980).

The dermatophyte structure most commonly associated with contagion, especially in the poorly conidial anthropophilic dermatophytes, is the oblong to rounded, persistent ‘‘spore,’’ ‘‘arthroconidium,’’ or ‘‘chlamydospore’’ found within or attached to the exterior of infected hairs and within skin scales. These structures, particularly in certain species, may persist for years in the environment (Shimmura, 1985) and are highly heat resistant (Sinski *et al.*, 1980), particularly when embedded in hair or skin scales (Stockdale, 1953). In some anthropophilic species studied in detail, arthroconidia have a tendency to adhere *in vitro* to corneocytes derived from particular body sites (Aljabre *et al*., 1993). It is possible that they may dissociate from skin cells in the environment and come in contact with new potential hosts as disseminated arthroconidia. Their persistence as an environmental source of contagion may lead to recurrent outbreaks of dermatophytosis in individuals and in community (Mackenzie, 1961).

Rippon (1988) stated that the arthroconidia of *Trichophyton rubrum* do not survive as long as do those of other species, e.g., *Epidermaphyton floccosum*. The transition from potentially sexual to asexual life histories in the non-soil associated

dermatophytes appears to have led to adaptive radiation, at least in the anthropophilic dermatophytes (Tanaka *et al.*, 1992). By most estimates, approximately two-thirds of the recognized dermatophyte species primarily associated with mammalian pathogenesis are anthropophiles (Aljabre *et al*., 1993). Within the anthropophiles, polymorphous morphological variation is common, and numerous atypical and variant types are recognized (Young, 1972), probably indicating further genetic drifts. Allopatric speciation appears to have been common in anthropophilic dermatophytes but rare in zoophiles, and several anthropophilic species have well-defined areas of endemicity while others, such as *T. rubrum* and *T. tonsurans*, are now cosmopolitan but appear to have had a more restricted distribution in the past, having been transported widely as a result of human migration (the anthropophiles travel with their human hosts). Also, spatial and ecological sympatric isolation appears to have been a predisposer to speciation in the anthropophiles: human-associated dermatophytes, unlike zoophiles, often have marked affinities for particular body sites. Most recognized asexual anthropophilic dermatophyte species are distinctive in morphology, physiology, and body site preference (Kane *et al*., 1990).

Dermatophyte colonization is characteristically limited to the dead keratinized tissue of the stratum corneum and results in either a mild or intense inflammatory reaction. Although the cornified layers of the skin lack a specific immune system to recognize this infection and rid itself of it, nevertheless, both humoral and cell-mediated reactions and specific and nonspecific host defense mechanisms respond and eventually eliminate the fungus, preventing invasion into the deeper viable tissue. This array of defense mechanisms thought to be active against dermatophytes consists of α2-macroglobulin keratinase inhibitor (Yu *et al.*, 1972), unsaturated transferrin

(King, 1975), epidermal desquamation (Berk *et al*., 1976), and lymphocytes, macrophages, neutrophils, and mast cells (Calderon, 1989).

There are two major classes of dermatophyte antigens: glycopeptides and keratinases. The protein portion of the glycopeptides preferentially stimulates cell-mediated immunity (CMI), whereas the polysaccharide portion preferentially stimulates humoral immunity. Keratinases, produced by the dermatophytes to enable skin invasion, elicit delayed-type hypersensitivity (DTH) responses when injected intradermally into the skin of animals (Grappel and Blank, 1972). Keratinases are produced by all dermatophytes studied. According to Apodaca and Mckerrow (1989), *T. rubrum* has been extensively studied and shown to produce two strongly keratinolytic proteinases with molecular weights of 93,000 and 71,000, as well as keratinolytic, trypsin- or chymotrypsin-like general proteinase with a molecular weight of 27,000. These proteinases all have a pH optimum of approximately 8. In another study with *T. rubrum*, a chymotrypsin-like acidic proteinase with a pH optimum of 4.5 was detected (Tanaka *et al*., 1992). Activity of the enzyme increased during the first 2 weeks of growth but then dropped and was superceded by the activity of neutral proteinases (Apodaca and Mckerrow, 1989). Human skin has a weakly acidic pH, and it is noteworthy that proteinases with an optimal activity under acidic conditions are reported to be important virulence factors in *T. mentagrophytes* (MacLeod *et al.,* 2009). The production of elastase has been associated with inflammatory dermatomycoses (Rippon, 1967).

Dermatophytes infect skin, hair and nails. On the skin and scalp, the lesion are often roughly circular with a rapid border but may coalesce to form confluent area of dry, scaling skin, itching and scratching, which in severe cases may ulcerate (Enemuor and

Amedu, 2009). Dermatophytes differ from most other pathogenic fungi since they are not opportunitic and frequently infect healthy individuals. Sites of infection include hair, nail and the stratum corneum of the skin (Woodfolk and Platts-Mills, 1998). The successful initiation of infection is a process closely related to the capability of the infecting dermatophyte to overcome the host resistance mechanisms. Cutaneous barriers against dermatophytes adherence, germination of arthroconidia and hyphae penetration into the stratum corneum include the intact keratinized layers of the skin and mucosal surfaces, the presence of fungistatic fatty acids on the skin undecanoic acid, for instance and other effectors such as skin pH. The adherence capability has been attributed to the presence of glycoproteins containing mannans in the cell wall of these fungi, which is presumed to correlate with virulence (Ogawa *et al*., 1998). In addition, for dermatophytes to be successful in their installation in the host the arthroconidia must germinate very rapidly and the hyphae must penetrate the body surface; otherwise they will be eliminated by the continuous desquamation of the epithelium (Paniz-Mondolfi *et al.*, 2012). Once installed, the dermatophytes must scavenge nutrients for growth, a process based on the induction of structural proteins, permeases and enzymes of the cell wall, in addition to the secretion of a variety of proteins and hydrolytic enzymes such as nucleases, lipases, nonspecific proteases and keratinases, among others, which occur in response to a short supply of essential nutrients in the host (Giddey *et al.*, 2007).

2.3 ANTIFUNGAL AGENTS

Fungal infections may become an important cause of human death or at least a significant cause of reduced quality of human living standards. On this basis, it is necessary to have antifungals available for the efficient control of fungal infections.

The cell wall of fungi contains compounds that are not found elsewhere in nature. Some of these components may provide selective targets for antifungal drugs without target-associated toxicity in mammalian hosts. The fungal cell wall consists of a multilayer structure composed of glucan, chitin, mannan, and mannoprotein (Poulain *et al*., 1978; Sullivan *et al.,* 1983). The appropriate antifungal therapy and selection of drug should be based on criteria such as immune status of the host, site of infection, characteristics of the infection (the fungal species and its susceptibility to different antifungal drugs), and pharmacokinetic characteristics of the drug (e.g., absorption, elimination, and toxicity). Nearly all antifungal agents have the ability to kill fungal cells in test-tube susceptibility assays, although the range of effective concentrations is quite broad for certain drugs or, for a given drug and the species of fungi tested (Hector, 2005).

The antifungal drugs that are routinely used for treatment of dermatophytoses include the polyenes, azoles, and the newest class; the echinocandins (Martinez-Rossi *et al*., 2008). An additional drug that is less frequently used, primarily for life threatening yeast infections, is flucytosine (5-fluorocytosine). For cutaneous mycoses, particularly infections caused by dermatophytes, the azoles, allylamines, and griseofulvin are most commonly used. Antifungal agents have been the focus of pharmaceutical industries, since dermatophytosis are increasing worldwide. Although allylamines, azole and griseofulvin have been used in the treatment of dermatophytoses, there is a necessity for more effective management of these drugs and discovery of new broad-spectrum antifungals. One of the major challenges in developing antifungal drugs lies in the similarities shared between fungi and their hosts. To develop new drugs, it is necessary to put into consideration that an efficient antifungal should be able to act in

a wide range of fungi, having no or low toxicity to the host (Martinez-Rossi *et al*., 2008).

Innovation in dermatophyte treatment has involved marketing of wide-spectrum topical agents, use of topical agents with anti-inflammatory as well as antifungal actions, and use of a combination of existing oral antifungal agents, or oral/topical antifungal agents, in attempts to improve on monotherapy cure rates. A wide variety of topical antifungal agents are available, in cream, gel, lotion, antiseptic soaps and shampoo formulations. A majority of the topical agents are of the 'azole' antifungal class (Gupta and Cooper, 2008). Though, the use of topical antifungal agents alone are not recommended for dermatomycoses, as it may lead to suppression of physical signs of infection, with lack of symptoms being wrongly associated with clearance of infection, leading to treatment relapse (Gupta *et al.,* 2003).

2.3.1 Polyenes

The polyenes, represented by amphotericin B (Fig.1) and nystatin (Fig. 2), were the first truly effective antifungal drugs introduced for treatment of life-threatening mycoses (Wasan *et al.,* 1998). These fermentation by-products of the soil organism *Streptomyces* were first described in the 1950s, when nystatin was discovered (Ruge *et al.,* 2005). It was amphotericin B, however, that quickly became the "gold standard" by which all subsequent antifungals were compared because of its broad spectrum of activity against diverse fungi and the ability to cure otherwise recalcitrant life-threatening fungal infections (Wasan *et al.,* 1998).

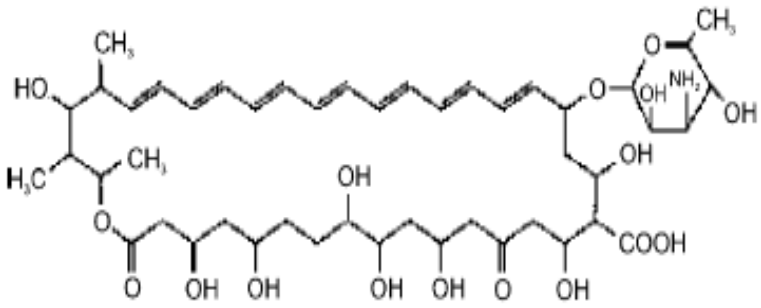


Fig.1 Chemical structure of Amphotericin B

Source: Ruge *et al.,* (2005).

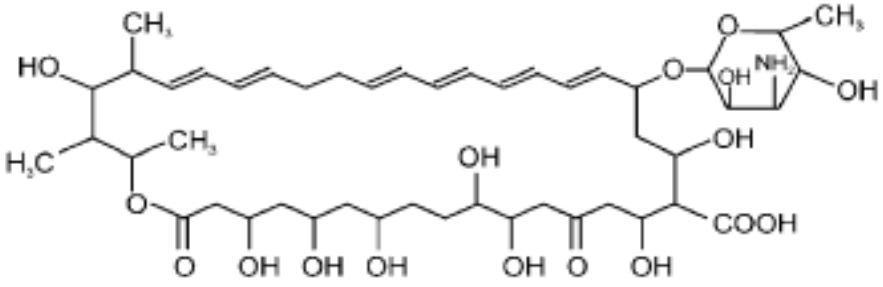


Fig.2. Chemical structure of Nystatin

Source: Ruge *et al.,* (2005).

2.3.1.1 Amphotericin B

Amphotericin B, chemically a macrocyclic polyketide (Fig.1), is an amphoteric compound that is poorly soluble in water. Because it is not absorbed by the oral route, the primary formulation (Fungizone), complexed with the bile salt desoxycholate, is used for intravenous or intrathecal administration. Solutions or topical formulations are also available for treatment of gastrointestinal, vaginal, and other superficial infections. Amphotericin B binds to sterols, and the primary fungal target is the ergosterol component of the fungal cell membrane. The compound self-aggregates within the ergosterol, literally creating a pore through which the cytoplasmic contents

of the fungal cell leak out, resulting in cell death (Matsuoka and Murata, 2002). Although the compound has a higher affinity for ergosterol, it also binds to mammalian cholesterol, with the resulting complexes contributing to the associated nephrotoxicity of the compound (Wasan *et al.*, 1998). The ensuing complications of renal toxicity and a long list of other toxicities are well documented. Thus, the benefits of this powerful antifungal have to be balanced against the inevitable side effects and its difficulty in administration. Amphotericin B has potent activity against a broad-spectrum of medically important yeasts and filamentous fungi, including the dimorphic pathogenic fungi (Nakai *et al.*, 2003). There are, however, select fungi that are inherently more resistant, including *Aspergillus terreus*, *Trichosporon beiglelii*, *Fusarium spp*., and possibly *Pseudoallerscheria boydii*, (Arikan *et al.*, 1999) as well as individual isolates of several species of *Candida (*Meyer, 1992; Terrell and Hughes, 1992*).* Acquired resistance has also been reported in several human clinical isolates of *Candida* species, (Drutz and Lehrer, 1978) and is likely a result of reductions in ergosterol content of membranes during prolonged or repeated usage in chronic infections (Dick *et al.*, 1980). Thus, awareness of this possibility should be kept in mind in cases of chronic treatment of yeast infections with amphotericin B. Although a controversial subject, there have been reports in the literature about antagonism between azoles and amphotericin B, particularly when therapy is initiated with an azole and then amphotericin B is subsequently added to or substituted for the azole.

2.3.2 Azoles

The azoles are a broad class of synthetic compounds that were co-discovered in the late 1960s by scientists of Janssen Pharmaceutica and Bayer AG to have activity

against medically important fungi. Although both companies were seeking compounds that could be administered by the oral route, Bayer's clotrimazole was only effective topically and Janssen's miconazole was marketed as topical and parenteral formulations. It was not until Janssen's discovery of ketoconazole (Nizoral) in 1979 that clinicians had, for the first time, the opportunity to use an oral medication for both serious and non-life threatening mycoses. These first azoles were all of the imidazole class, containing two nitrogens in the azole ring (Hector, 2005). In the 1980s, a new generation of more potent azoles were discovered by the creation of analogues containing three nitrogens in the azole ring; the triazoles. Janssen again led the way with itraconazole (Sporonox), a substantially more potent analog that also had a more favorable safety profile than ketoconazole. Pfizer introduced fluconazole (Diflucan), a drug that greatly expanded the market for antifungal by virtue of its good activity against economically important mycoses, its comparatively favorable safety profile, and a high price that helped push its world-wide sales figures to the billion dollars per year. The newest triazole used in treatment of life-threatening mycoses is vorizonazole, recently launched by Pfizer. A number of azoles for topical use are also available in a variety of formulations; most are imidazoles. The primary mechanism of action of all azoles is the blocking of ergosterol biosynthesis; a key component of the cytoplasmic membrane of fungi (VanDen Bossche *et al.,* 2003).

2.3.2.1 Clotrimazole and Miconazole

Although these two drugs are the oldest commercial members of the azole class, they still retain an important role in the treatment of superficial mycoses because of their good potency against yeasts and filamentous fungi, the availability of different formulations, and their comparatively low cost. As mentioned, these agents are

distinguished from the newer analogs in that they are imidazoles rather than triazoles. While the chemical distinction is not central to this review, it may partially explain a unique property that adds to their effectiveness as topical agents. Miconazole and clotrimazole, but not ketoconazole or itraconazole, have been demonstrated to have a direct, lytic effect on fungal membranes, in addition to the azole's ability to inhibit ergosterol synthesis (Brasseur *et al.,* 1983; Mathews *et al.,* 1998). This property was turned to advantage when methods were developed to successfully topically treat sinus infections because of *Aspergillus* spp. in dogs through the placement of catheters and one or more short-term infusions of a clotrimazole solution. The method has also been successfully utilized with enilconazole, another imidazole (Zonderland *et al.,* 2002).

2.3.2.2 Fluconazole

Fluconazole (Fig. 3) is a widely used antifungal agent that is unique among the azole class. Despite not being a potent antifungal, as judged by *in-vitro* susceptibility testing, it is nevertheless remarkably effective against a variety of mycoses. This is, in large part, because of its pharmacokinetic properties. Unlike virtually all other commercially available azoles, fluconazole is essentially water soluble, has very low protein binding in most animal species and in humans (10-12%), and therefore distributes throughout the body and into most body fluid (Como and Dismukes, 1994; Ripa *et al.,* 1993). These properties have been confirmed in cats, dogs and horses. Absorption of orally administered fluconazole is not influenced by food or gastric acidity. The major route of elimination of the drug is by renal clearance, with about 70% of the dose being excreted by this route. The drug is not generally metabolized; studies with radio labeled fluconazole in dogs showed that about 90% of the dose was

recovered as unchanged drug in urine and feces. Thus, fluconazole is commonly used for fungal infections of the urinary tract other than those caused by *Aspergillus* spp. or other filamentous fungi (Humphrey *et al*., 1985).

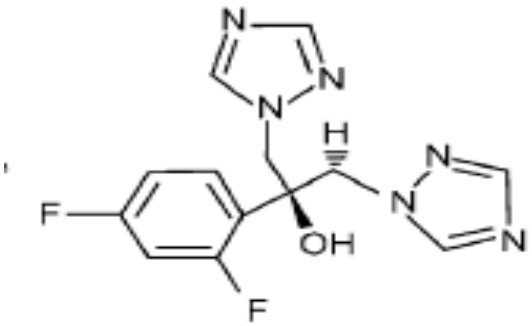


Fig. 3. Chemical structure of fluconazole

Source: Hof (2008).

2.3.2.3 Itraconazole

Itraconazole is a highly lipophilic compound (Fig. 4). Approximately 99% of the circulating drug is protein bound and its penetration into cerebrospinal fluid (CSF) and other aqueous body fluids is very limited. It has, however, a large volume of distribution, and there is considerable accumulation of the drug in lung, kidney, liver, bone, muscle, skin, and nails to concentrations higher than the circulating plasma levels; a characteristic that is key to its efficacy (VanDen Bossche *et al.,* 2003). Absorption of itraconazole delivered by oral capsule can be variable and unpredictable, and is influenced by food and gastric acidity (Legendre *et al.,* 1996). The relatively poor and variable oral bioavailability of the capsule form of itraconazole ultimately led to the introduction of oral solution and intravenous formulations. In humans, the oral solution has 30% better bioavailability than the capsules and achieves higher serum concentrations. The intravenous formulation can

be a valuable therapeutic option to amphotericin B in life-threatening infections. Itraconazole is extensively metabolized by the liver, but its primary metabolite retains good antifungal activity (VanDen Bossche *et al.*, 2003).

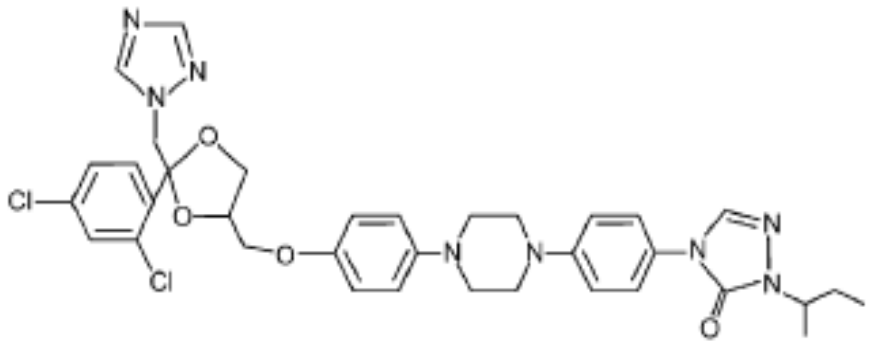


Fig. 4 Chemical structure of Itraconazole

Source: Hof (2008).

Itraconazole is generally well tolerated, with gastrointestinal upset and effects on the liver enzymes as the most common findings. Itraconazole is broadly active against most fungi of veterinary importance, including dermatophytes. In the latter category, it has been reported to be effective in cats in both experimental and natural infections because of *Microsporum canis*. Indeed, because the main delivery route for itraconazole to the skin is the sebum, it is possible to achieve high concentrations in the skin and, because of its protein binding, it is also retained in skin and nails for an extended period of time (Cauwenbergh *et al.*, 1988). Because of its retention at the target sites, it is therefore possible to use intermittent dosing with itraconazole in the treatment of superficial mycoses. In humans, it is usually the drug of choice against mild-to-moderate cases of histoplasmosis, blastomycosis and sporotrichosis, and is widely used for treatment of coccidioidomycosis. Its utility in canine blastomycosis is well-documented, while the data are more limited for veterinary coccidioidomycosis

and histoplasmosis (Hodges *et al.*, 1994). Lastly, a generic form of itraconazole was approved for human use by the Food and drugs Adminstration (FDA) in mid-2004 (Hector, 2005).

2.3.2.4 Ketoconazole

Ketoconazole is the oldest of the orally administered azoles on the market and remains the only member of the imidazole class that is currently used for oral treatment of systemic infections. Ketoconazole is active against yeasts, dimorphic fungi, and dermatophytes, but is less active against many other filamentous fungi (Sharp and Sullivan, 1989). However, its potency is limited compared to that of fluconazole and itraconazole, collectively (St-Germain *et al.*, 1995). Ketoconazole (Fig. 5) remains an extensively used compound, despite the fact that it is both less potent and more toxic than the newer triazole antifungals. Some of the reasons may include a long history of use and, thus, an appreciation for proper dosing schedules for a given animal species, as well as the comparatively lower cost (note that recently introduced generic forms of fluconazole and expectations of a generic form of itraconazole may reduce this advantage). However, the side-effects profile, particularly hepatotoxicity at high doses and a long list of drug-drug interactions are likely to limit its use as a systemic to that of an alternative drug for specific indications (e.g. treatment of *Pityriasis versicolor* in dogs) (Guillot *et al*., 2003). Interestingly, the drug-drug interaction with cyclosporine has led to the use of ketoconazole as a cyclosporinesparing agent (Robson, 2003).

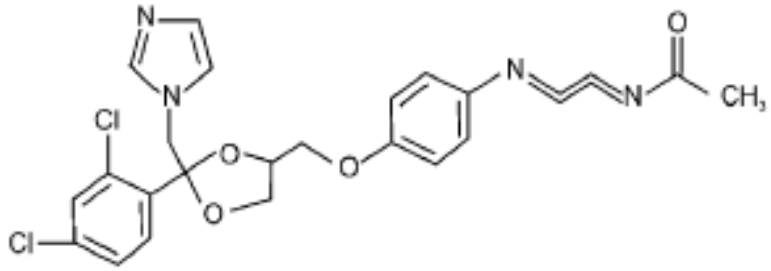


Fig. 5. Chemical structure of Ketoconazole

Source: Hof (2008).

2.3.2.5 Voriconazole

Voriconazole is a triazole (Fig. 6) that is structurally related to fluconazole. Despite its structural similarity to fluconazole, the drug is largely eliminated by metabolism rather than excretion, meaning that less active drug appears in the urine, compared with fluconazole. The pharmacokinetic behaviour and elimination of the drug appears to be quantitatively variable across animal species as well as for animals compared with humans, and so use of the human-application literature with this drug should be done with caution (Roffey *et al.,* 2003). Voriconazole has favourable *in vitro* activity against a variety of fungi, but is considered to be somewhat more active against filamentous and dimorphic fungi than yeasts (Espinel-Ingroff, 2003). Voriconazole activitiy against infections resulting from *Aspergillus* spp., however, that has generated the most interest for this drug in human use. In a landmark study of invasive aspergillosis, investigators determined that therapy with voriconazole led to better responses, improved survival, and fewer severe side effects compared to therapy with amphotericin B (Herbrecht *et al.,* 2002). Despite the importance of evaluating newer, more potent agents to ascertain the advantages that they confer, their use in veterinary medicine should be balanced against an almost complete lack

of published information on veterinary clinical use, the documented side effects in humans (Sheehan *et al.,* 1999).

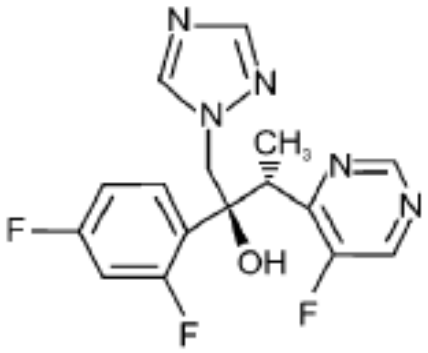


Fig. 6. Chemical structure of Voriconazole

Source: Hof (2008).

2.3.3 Pyrimidine

Flucytosine (5-fluorocytosine, or 5-FC) is a fluorinated pyrimidine. The mode of action of flucytosine (5-FC) (Fig. 7) is very unique within the antifungal agents. Uptake of 5-FC into fungal cells is mediated by the cytosine permease. 5-FC is subsequently deaminated to 5-fluorouracil (5-FU), converted to the nucleoside triphosphate and incorporated into RNA where it causes miscoding (Ruge *et al.,* 2005). In addition, 5-FU is converted to deoxynucleoside, which inhibits thymidylate synthase and thereby DNA biosynthesis (Georgopapadakou and Walsh, 1996). Although seldom used as monotherapy, because this orally administered, water-soluble drug penetrates into aqueous body fluids and is also excreted by the kidneys, it is sometimes used as an adjunct to amphotericin B for the treatment of serious yeast infections, especially cryptococcal meningitis (Dismukes *et al.,* 1987). Given its short plasma half-life, the emergence of resistance on prolonged therapy, and its known side effects profile, the clinical utility of this drug is limited (Malik *et al.,* 1996).

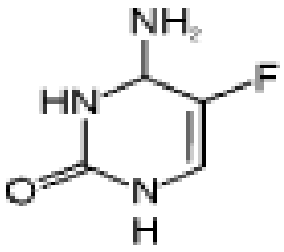


Fig7. Chemical structure of Flucytosine

Source: Ruge *et al.,* (2005).

2.3.4 Echinocandins

First described in the 1970s, these unique compounds target the fungal cell wall by selective inhibition of beta-1,3-glucan synthesis (Kurtz *et al.,* 1996). While the general mechanism of action is understood, the precise molecular interaction with the target has not been definitively established. Nevertheless, in fungi that have a high component of beta-1,3-glucan in the cell wall, exposure to this compound leads to catastrophic failure of the cell wall, not unlike what beta-lactams do to most bacteria. This class of compounds, although somewhat narrow in spectrum of activity, is highly lethal to most species of *Candida* and is also finding clinical use against infections because of *Aspergillus* spp. Although caspofungin is the only echinocandin currently marketed, two additional analogs, micafungin and anidulafungin, are in advanced stages of development and will likely become commercially available.

2.3.4.1 Caspofungin

Caspofungin, like all members of the echinocandin class, is a cyclic polypeptide with a long-chain lipid "tail."(Fig.8). Because of its structure, it is not orally absorbed and is therefore only available as a parenteral formulation. The echinocandins act at the actively growing tips and branching points of hyphae, leading to distorted and slower

growth forms that hypothetically are more susceptible to the animal's host defences (Espinel-Ingroff, 2003). However, they do not act on the intact portions of the hyphae and are not, therefore, fungicidal compounds against filamentous fungi. Nevertheless, the compound is being used for human infections because of aspergillosis, although often in conjunction with other antifungals (Deresinski and Stevens, 2003). Indeed, its initial approval by the Food and Drug Administration **(**FDA) was for salvage therapy of aspergillosis in humans; it was subsequently approved for use against candidiasis. With some exceptions, caspofungin is not considered active against other commonly encountered filamentous fungi (Diekema *et al.*, 2003).

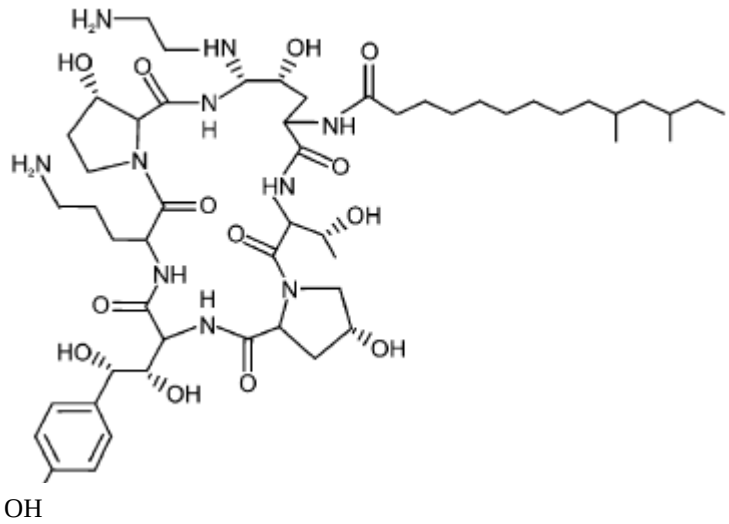


Fig. 8. Chemical structure of Caspofungin

Source: Ruge *et al.*, (2005).

2.3.5 Allylamines

The allylamines, discovered in the mid-1980s, are a class of synthetic compounds that also interfere with ergosterol biosynthesis in the fungal membrane, although at an earlier step in the enzymatic pathway compared to azoles. The major compound of the class, introduced to the market in 1995, is terbinafine (Lamisil); naftifine is another

analog that has been used. They are particularly active against dermatophytes (Hector, 2005).

2.3.5.1 Terbinafine

Terbinafine is one of the main treatment options for dermatophyte infections. Compared to the older agent, griseofulvin, it is more effective, as well as being significantly less toxic. Moreover, the required duration of therapy is also shorter with terbinafine (Mancianti *et al.,* 1999). After oral administration, the drug is rapidly distributed to the skin, nails, and hair, both through the sebum and by diffusion through the dermis, which contributes to its efficacy against dermatophytes. However, there is a report in experimentally induced *Microsporum canis* infections in cats that concluded that prolonged and/or higher doses were required to achieve clinical resolution (Kotnik, 2002). A finding of note is a conclusion by the author that the superior effects achieved in the higher dose was because of a high accumulation of the drug in the hair; evidence that pharmacodynamic principles are also relevant at sites exterior to the body. Terbinafine (Fig. 9) is available in both oral and topical formulations, but is not licensed for veterinary use. Topically, 1% cream and solution formulations are used. Terbinafine appears to be well tolerated. Reported adverse reactions in humans are, in general, transient and mild and mostly involve the gastrointestinal system and the skin (Hall *et al.,* 1997).

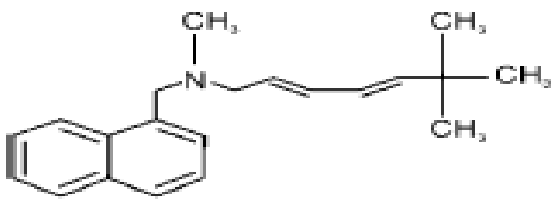


Fig. 9. Chemical structure of Terbinafine

Source: Ruge *et al.,* (2005).

2.3.6 Griseofulvin

Griseofulvin (Fig. 10) was isolated from *Penicillium griseofulvum*, has been shown to possess fungistatic action, in particular against dermatophytes. Reports have conflicted concerning the effects of griseofulvin on different subcellular sites. The activity of the drug against chitinous fungi has been attributed to inhibition of the synthesis of hyphal cell wall material (Gull and Trinci, 1973). Other investigators have suggested that this drug affects nucleic acid synthesis and mitosis. Susceptibility of fungi to griseofulvin appears to be related to the binding of the drug to RNA (Jessup *et al.,* 2000). It has been clearly established that griseofulvin is active only against growing cells, and this observation may indicate the primary involvement of microtubular changes in the mechanism of action of the drug. Indeed, recent work has shown that griseofulvin interferes not only with microtubules of the mitotic spindle, acting as a spindle poison, but also with cytoplasmic microtubules. In view of the presumed role of microtubules in the transport of secretory material through the cytoplasm toward the periphery of the cell (Gull and Trinci, 1973), the destruction of the microtubules may lead to impaired processing of newly synthesized cell wall constituents at the growing tips of hyphae. Griseofulvin's history dates back even further than the polyenes, with its first isolation as a fungal metabolite in 1939 and commercial introduction in 1956. Griseofulvin acts by inhibition of microtubule function, thereby disrupting the mitotic spindle necessary for fungal cell division during mitosis, as well as other cytoplasmic functions (Gull and Trinci, 1973). The highly lipophilic compound is well absorbed by the oral route, particularly when given with a heavy, fatty meal, and reaches the skin and hair in therapeutic concentrations. Griseofulvin is active only against dermatophytes, and is less potent,

in vitro, compared with terbinafine, itraconazole, and voriconazole (Jessup *et al.,* 2000).

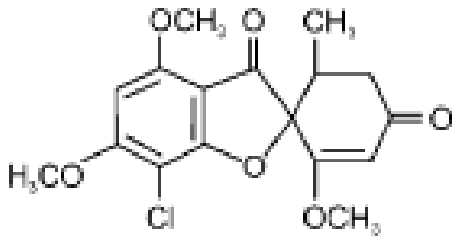


Fig. 10. Chemical structure of Griseofulvin

Source: Ruge *et al.,* (2005).

2.4 ANTIFUNGAL DRUG RESISTANCE

The term "resistance" is used to describe a relative in- sensitivity of a microbe to an antimicrobial drug as tested *in vitro* and compared with other isolates of the same species (Rex, 1997**)**. Primary resistance occurs in organisms never exposed in that host to the drug of interest. In contrast, secondary resistance, also defined as acquired resistance, arises only after exposure of the organism to the drug. Intrinsic resistance is defined as resistance of all or almost all isolates of one species to a certain drug (Rex *et al.,* 1997**).** The appropriate antifungal therapy and selection of drug should be based on criteria such as immune status of the host, site of infection, characteristics of the infection (the fungal species and its susceptibility to different antifungal drugs), and pharmacokinetic characteristics of the drug (e.g., absorption, elimination, and toxicity).

2.4.1 Polyenes

Amphotericin B is active against a wide variety of fungi, including yeasts and molds such as *Candida* spieces and *Aspergillus* species, Cryptococcus neoformans,

Zygomycetes, dimorphic fungi, and some dematiaceous fungi. The target structure is plasma membrane ergosterol, where amphotericin B forms a channel. Through this channel, the fungal cell leaks potassium ions, resulting in a disruption of the proton gradient. It is hypothesized that a ring of 8-10 polyene molecules form aqueous pores within the membrane bilayer structure. In addition, amphotericin B causes oxidative damage to plasma membranes. In higher concentrations, polyenes also inhibit chitin synthase, a cell wall synthetic enzyme localized in the membrane. The interaction between amphotericin B and human cell membranes containing cholesterol results in toxic side effects of the drug. The most important side effect is an impairment of renal function due to decreased filtration in the glomeruli (VanDen Bossche *et al.,* 1994).

Mechanism of resistance. The cause of resistance to polyenes is a significant alteration of the lipid composition in the plasma membrane. This circumstance leads to a lower affinity of amphotericin B to the plasma membrane, probably the result of a lack of the binding site (Hector, 2005). However, for a highly resistant strain of *Aspergillus terreus*, it was shown that the major sterol in its plasma membrane was ergosterol, suggesting that resistance to amphotericin B was not related to a modified sterol composition (Dannaoui *et al.,* 2000). Another cause for amphotericin B resistance may be an altered content of 1,3 glucans in the fungal cell wall. These components, which increase the stability of the cell wall, influence the access of large molecules such as amphotericin B to the plasma membrane (Gale, 1986).

2.4.2 Azoles

Mechanism of resistance; Different potential molecular mechanisms of azole resistance are possible. Some of these mechanisms are well known from antibacterial resistance. Modified target enzymes have been well characterized as a mechanism of

resistance to penicillin in *Streptococcus pneumoniae* and *Neisseria gonorrhoeae* (Dowson *et al.,* 1994), efflux pumps have been implicated in resistance to chloramphenicol and tetracycline (Cohen *et al.,* 1993), and resistance to aminoglycosides has been associated with alterations in the plasma membrane. Sometimes development of resistance to an azole drug leads to cross-resistance to other azoles, and sometimes the resistance is azole specific (Stevens and Stevens, 1996); this will depend on the specificity of the resistance mechanism (i.e., the affinity of the target enzyme or the pump for a particular molecular structure). Rustad *et al*. 2002 reported a correlation of azole resistance in *Candida albicans* with homozygosity at the mating-type-like (MTL) locus, suggesting that key resistance genes may be regulated by this locus or in the vicinity of this locus. Azoles act as ergosterol synthesis inhibitors by binding to the lanosterol demethylase. The gene encoding this protein is ERG11, also known as ERG16, and the enzyme has been referred to as CYP51A1 in *C. albicans*. Alterations of this enzyme caused by defined point mutations in ERG11 have been described to be associated with azole resistance. White (1997) described the presence of the amino acid substitution of lysine for arginine at position 467 (R467K). The substitution is located near the heme cofactor and thus causes structural and/or functional enzyme alterations. A substitution at amino acid 464 (G464S) in azole resistant *C. albicans* isolates has been described (Loffler *et al.*, 1997).

Cohen *et al.* (1993) confirmed that this mutation, resulting in changes in the heme-binding domain, causes fluconazole resistance through substantially reducing the inhibitory effect of fluconazole and described its association with perturbation of the heme environment. The sterol composition of the plasma membrane can also affect

the influx of drug into the cell. Alterations in the plasma membrane composition affect membrane fluidity and asymmetry, leading to a decreased uptake of drug (Parks and Casey, 1996). It was demonstrated that altered phospholipid and fatty acid composition of the plasma membrane of *C. albicans* may lead to resistance to miconazole (Mago and Khuller, 1989). Hitchcock and Whittle (1993) noted a larger lipid content and a lower polar-lipid-to-neutral- lipid ratio in an azole-resistant *C. albicans* isolate compared with wild-type strains, potentially causing a decreased permeability of the membrane and thus a reduced penetration of azoles. In a fluconazole-resistant *C. albicans* isolate, a decreased amount of ergosterol and a lower phosphatidylcholine: phos- phatidylethanolamine ratio in the plasma membrane, which might be responsible for an altered uptake and thus for a reduced intracellular accumulation of fluconazole, was demonstrated (Loffler *et al.*, 2000). In an itraconazole-resistant *C. krusei* isolate, it was indicated that reduced accumulation of drug accounts for resistance rather than drug efflux or modifications in the ergosterol (Venkateswarlu *et al.*, 1996).

2.4.3 Pyrimidine

Mechanism of resistance; there are two mechanisms of resistance proposed. First is a mutational decrease of activity of the cytosine permease or deaminase, leading to a decreased uptake or conversion of the drug. This mechanism is responsible for primary and intrinsic resistance (Vanden Bossche *et al.,* 1994). Second is a loss of activity of uracil phosphoribosyltransferase, an enzyme responsible for conversion of 5-fluorouracil to 5-fluorouridylic acid (Polak and Scholer, 1975; Diasio *et al.*, 1978). Whelan and Kerridge (1984) described decreased activity of the uridine monophosphate pyrophosphorylase associated with resistance to 5-FC in C. albicans.

In 29 clinical isolates of *C. albicans* with resistance to 5-FC, no cytosine permease deficiency mutants were found. In contrast, for *C. parapsilosis*, a 5-FC-resistant strain was described that evolved while the patient received therapy and had <7% of the cytosine deaminase activity compared with the parent susceptible isolate (Hoeprich *et al.*, 1974).

2.4.4 Echinocandin

Three antifungal compounds (caspofungin, FK-463, and VER-002) belonging to the class of echinocandins are currently available for clinical use or are under development (Hector, 1993). All compounds inhibit the fungus-specific biosynthesis of 1,3 3- D-glucan (Denning, 1997). They have potent in vitro activity against *Candida* spp. and some activity against *Aspergillus*, dimorphic molds, and *Pneumocystis carinii*. As expected, isolates resistant to antifungal triazoles did not show cross-resistance to the 3 compounds (Vazquez *et al.,* 1997). Treatment of fungi with O-glucan synthase inhibitors causes non-competitive inhibition of 1,3 O-glucan synthase with secondary effects on other constituents, such as an increase in the chitin content of the cell wall and a reduction in the ergosterol content of the fungal cell membrane (Pfaller *et al.*, 1989).

Mechanism of resistance; Kurtz and Douglas (1997) presumed that the glucan synthase complex is encoded by 2 genes and regulated by a third and mutations in one of the genes, FKS1, cause resistance to echinocandins by alteration of 0-glucan synthase. Echinocandins do not penetrate into the cytoplasm of the fungal cell. Thus, resistance mechanisms described for azoles, such as increased activity of efflux pumps or altered sterol composition of the plasma membrane, seem to be irrelevant.

2.5 MEDICINAL PLANTS

African plants constitute a rich pool of natural products. Research in the field of indigenous plants is a significant aspect to develop a safer antifungal agent since huge number of these pathogenic microorganisms that are developing resistance against the indigenous drugs day by days (Doughari and Obidah, 2008). Scientific investigations of medical plants have been initiated in many countries because of their contributions to health care. The primary benefits of using plant-derived medicines are that they are relatively safer than synthetic alternatives, offering profound therapeutic benefits and more affordable treatment. Besides, the rapid evolution of numerous defense mechanisms against antimicrobial agents and resistance to old and newly produced drugs underlies the urgency with which alternative chemotherapeutic agents with possible novel mechanisms of action to the microbes need to be developed which informed the need for the use of the listed medicinal plants in this study; *Azadirachta indica, Treculia africana and Detarium senegalense.*

2.5.1 ***Treculia africana*** Decne (African bread fruit, "Afon") belongs to the family Moraceae. There are three species in the genus *Treculia*, *Treculia africana* (decne), *T. obvoidea* and *T. acuminata* (Keay *et al*., 1964). The first two species are used for furniture making and as fuel wood, but the third one is a slender forest shrub used for construction work. *Treculia africana* is generally found along streams and water courses. Its distributions extend from latitude 150N of Angola and down to Saotome Island. It is widely distributed in the following town of Nigeria mostly in Oyo, Ogun, Anambra, Cross River, Delta and Imo States (Okunomo and Egho, 2010). *Treculia africana* has a bole usually fluted at the base without buttress. The bark is smooth, light grey to green in colour. It has a copious milky latex when the tree is cut that comes out of it. It grows up to 37 m in length and 3 m girth. Branches and young

foliage are glabrous and softy hairy. Leaves are 4 – 8 inch wide. Fruit is greenish, spongy in texture and it is 15 cm long, more in diameter containing numerous individual fruit-like orange pips. The seeds are round, sometimes elongated about 1.3 m long (Keay *et al.*, 1989).

Treculia africana is commonly roasted, cooked, mashed and consumed either directly as snack food or as flour for use in soup thickening and cake (Fasasi *et al*., 2004). The *seeds* are rich in amino acids, minerals and fatty acid (Lawal and Bassir, 1987). The crude extract from different parts of the plant has been used in the folk medicine in the treatment of various ailments. It is used either singly or in combination with other herbs in the traditional herbal preparation by different communities to treat various diseases. Decoction from different parts of the plant is used as an anti-inflammatory agent and in the treatment of whooping cough. The crushed leaves juice is applied on the tongue as a treatment for thrush in children; the latex is applied as an antibacterial agent in eardrops and as chewing stick. The sap of the male tree is applied locally on cotton wool to carious tooth for its removal. The root, immature leaves and bark are part of the concoction used locally for treating cough, constipation, edema and rheumatism (Aderibigbe *et al*., 2010). The pulps of *Treculia africana* have been shown to be useful in the treatment of Ascaris and Guinea Worm (Ogunleye and Parakoyi, 1992). Proximate chemical composition of the fruit and seed showed that it contains high level of carbohydrate and protein but is relatively low in fat, ash and fibre (Osabor *et al.,* 2009). The water soluble and ethylacetate fractions have been shown to reduce the fasting blood sugar levels (Oyelola *et al*., 2007). Three compounds were isolated from *Treculia africana* and they are identified as

Phyllocoumarin; Catechin and 6,9 – dihydroxy-megastigmane -3 –one with antibacterial and antifungal properties (Ogbonnia *et al*., 2008).

2.5.2 *Azadirachta indica*

Azadirachta indica A. Juss (Neem, "Dogoyaro") is a member of the Meliaceae (Mahogany) family. It has been referred to in the past by the botanical names, *Melia azadarach* (chinaberry). The tree is also known as neem, margosa, limba, minba, nimba, kohomba, and India lilac (Almas *et al*., 1999). Neem is a small to medium sized tree with a short, straight bole. The stem branches at 2 – 5m forming a broad, dense, round or oval crown. Total height is 15-25cm, occasionally reaching up to 30 cm; with a stem diameter ranging from 30 to 90cm. Neem is characterized by a long, penetrating lateral root system, which can extend up to 15m, with a relatively short tap root. Neem has moderately thick, fissured, grey outer bark, with a reddish brown inner bark. It is evergreen or deciduous depending on the climate. Leafless periods are usually brief, occurring during extended drought. Leaves are at the tip of branches. The tree produces many small, sweet-scented, white or cream colored, bisexual flowers. The fruit is a smooth, ellipsoidal drupe, 1.2-2.0 long, containing usually one seed. The fruit is initially green and turns yellow as it ripens in about 12 weeks after full bloom. Neem trees are prolific fruit producers, starting as early as 3-5 years, and becoming fully productive at 10-12 years (Dash, 2004). Neem adapts to a broad range of climate and soil conditions. It is normally found at elevations between sea level and 700m. Neem can grow at altitudes up to 1500m, as long as temperatures remain moderate, as it does not withstand cold or frost. Neem tolerates extremely high temperature, but its normal range is about 9.5^0C – 37^0C. It is also highly drought tolerant, and once established, it can survive 7-8 month dry seasons. It requires as

little as 150mm rainfall per year in areas where the root system can access ground water within 9-12m of the surface, however, it performs best in zones receiving 450-1200mm per year (Dash, 2004).

The principal active compound in the leaves of *Azadirachta indica* is azadirachtin, which repels pests, acts as an anti-feedant, and disrupts insects' growth and reproduction. Several bio active compounds are found in the leaves and other tissues; however, the neem seed kernel's are the main source of azadirachtin. Neem seed contains the most concentrated and accessible amounts of other potentially useful compounds as well. Neem seed and leaf extracts are effective against both chloroquine resistant and sensitive strain malarial parasites. Neem based pesticides have already been approved for various applications and are being produced commercially in several countries (Randhawa and Parmar, 2007). Various parts of the neem tree have been used in traditional medicine. In India neem oil and thebark and leaf extracts have been therapeutically used as folic medicine to control leprosy, helminthiasis, respiratory disorders, and constipation and also as a general health promoter (Akinlandeswani *et al.,* 2003). Neem oil finds use to control various skin infections. Bark, leaf, root, flower and fruit together cure blood morbidity, biliary infections, itching, skin ulcers and so on (Almas, 1999).

2.5.3 *Detarium senegalense*

Detarium senegalense J. Gmelin ("Ogbogbo" in Yoruba language) is a leguminous plant belonging to the subdivision Caesalpinoideae (Balogun and Fetuga, 1986) and is considered to be synonymous with *Detarium microcarpum* (FAO, 1988). The wood of *Detarium senegalense* is said to be not easy to work, but is used in Africa for

canoes, boat-ribs, planks, house-posts, furniture, fences, firewood, tool-handles, etc. In Sierra Leone, the pulped-up bark is eaten as remedy fur tuberculosis (Irvine, 1961). In Ivory Coast, though the existence of two varieties is recognized, their uses are alike, powdered for skin conditions, leprosy, inflamed buboes and wounds as a haemostatic and healing agent. In Liberia a bark-decoction is given to women at childbirth to eject the placenta should it become retained (Irvine, 1961). Relief is said to be immediate. The "Igbos" of Nigeria use the plant as a post-partum haemostatic medication (Iwu, 1993).Ash from the burnt bark is used in Sierra Leone to make soap (Dalziel, 1995). The seed is globular, slightly flattened, 5-8 cm in diameter. It is deep purplish-brown in colour, more or less oily and edible. In Nupe, the seed is pounded into a cake for cattle-feed, and has been taken in Northen Nigeria as a famine-food (Dalziel and Hutchinson, 1958). In Senegal, the seed is burnt as a mosquito repellant.

2.6 MOLECULAR CHARACTERIZATION

The use of molecular diagnostic assays in the clinical setting has been on the increase over the past decade. Both target and signal amplification allow for the rapid and early diagnosis of various dermatologic diseases with high sensitivity and specificity not commonly seen with other conventional techniques (Mandhaniya *et al.,* 2012). For many years, a conventional laboratory methods based on the detection of phenotypic characteristics such as microscopy and *in-vitro* culture, have played an essential role in identification. However, these procedures generally suffer from the drawbacks of being slow or non-specific since the tools necessary to reliably distinguish strains belonging to a singular species of microorganism are lacking.

The DNA base composition studies of Davison *et al.* (1980) began a new era of molecular biological investigations of dermatophytes. Though dermatophyte species proved to be relatively closely related to one another, early molecular tests such as restriction fragment typing (Mochizuki *et al.*, 1996) were able to distinguish common species. Full entry into the modern era of biosystematics came with the first molecular phylogeny of the dermatophytes and their Arthrodermataceous relatives by Graser *et al*. (1999), with additional molecular phylogenetic analyses rapidly following (Graser *et al*., 2000). All these phylogenies tended to be strongly influenced by the close relationships among dermatophyte species that evolved on hosts that are themselves relatively recently evolved, particularly humans as well as animals such as cats, cattle and horses that have undergone rapid selection and habitat change in the course of domestication. The revelation of this close relationship coincided with a significant collapse in the number of recognized species. The use of molecular techniques which are expensive and require an experienced staff can be useful when the identification of a strain is not possible with conventional methods. Molecular techniques like PCR in conjunction with restriction fragment length polymorphisms (RFLP) to target fungal mitochondrial DNA and sequencing of the Internal Transcribed Spacer (ITS) regions will provide a practical solution to the difficulties encountered in the identification of characteristic in conventional procedure and a technological advance in the laboratory diagnosis of dermatomycoses.

This present study was built on the fact that Lagos State which has often been referred to as the dirtiest, most disorganized, and the most unsafe mega-city in Nigeria. Lagos is seen as an intolerable place, which offers minimum resources for a healthy, safe, and productive life. The problems in the city are similar to all the other mega-cities;

traffic jams make transportation inefficient, waste management is malfunctioning leaving tons of waste on the streets, water resources are overused or polluted and inadequate housing, as well as slums, are becoming reality for an increasing number of inhabitants (Kuvaja, 2007). It is the smallest state in geographical terms in the Nigerian Federation; it occupies an area of 357,700 hectares, 22% of which consists of lagoons and creeks. At the center of the riverine State is Lagos Metropolis which at present occupies about 132,350 hectares of the land area (Kuvaja, 2001). The provision of medical facilities has not increased as fast as the demands of a growing population. Though private medicare has emerged to complement Government efforts, a yawning gap still exists between the citizens' needs and available facilities. Despite the twenty-five general hospitals in Lagos State, there are few mycological laboratories in which they lack experts. The technicians and biologist upon the problems encountered on identification still use conventional methods for the diagnosis of dermatomycoses.

CHAPTER THREE

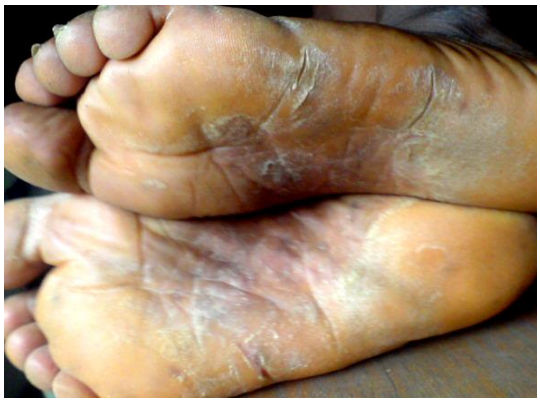
3.0 MATERIALS AND METHODS

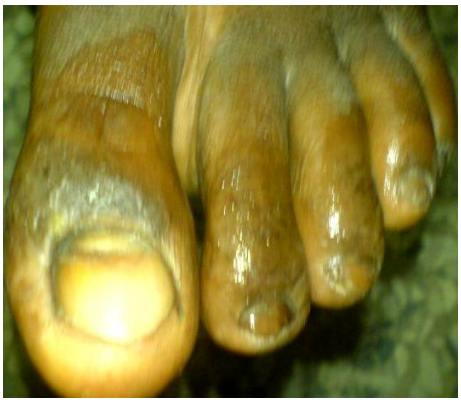
3.1 SAMPLE COLLECTION

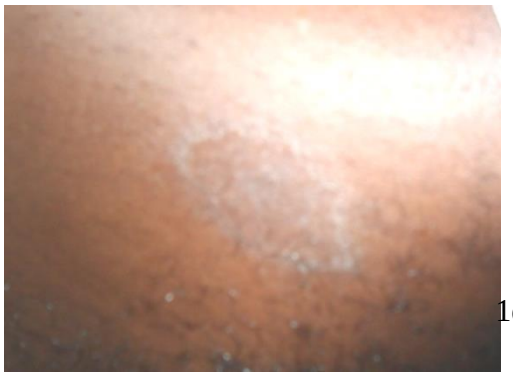
Samples were collected weekly from two of the three tertiary health institutions in Lagos state; Lagos University Teaching Hospital (LUTH) Idi-Araba, and Central Public Health Laboratory (CPHL) Yaba, for seventy-eight consecutive weeks (August, 2009 – January, 2011). This was made possible with the permission from Lagos State Health Service Commission (HSC) (Appendix A).

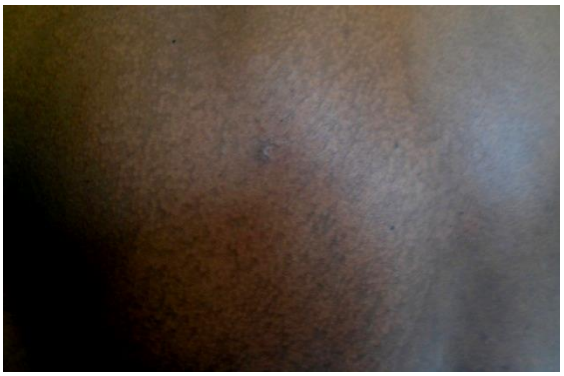
3.2 SPECIMEN COLLECTION

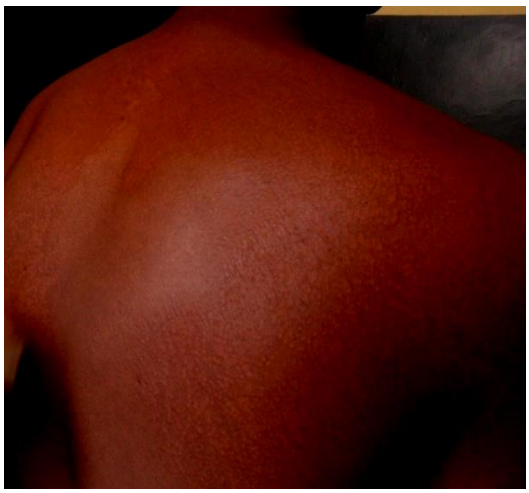
Scrapings were collected from infected areas which correspond to the active zone of the lesion from the different infected area on patient body (Plate 1a-f) from two different tertiary health institutions in Lagos state. The specimens were harvested in a sufficient amount and taken from the edge of the infected area; this was done by an experienced staff with scalpel blades. To improve the efficiency of mycological examination, patients were advised not to apply any local or systemic topical antifungal treatment on the infected area prior to samples collection. Also, the infected area was cleaned with methylated spirit before scrapping was done inorder to remove contaminants such as bacteria and some opportunistic fungi that may be present at the surface of the lesion area (Elewski, 1998). Specimens were collected in sterile envelop. Envelopes with collected specimen were then labelled with some necessary information concerning the patient. Informations such as name, gender, age and infection site were collected and recorded (Appendix B).

1a 

1b 

1c 

1d 

1e 

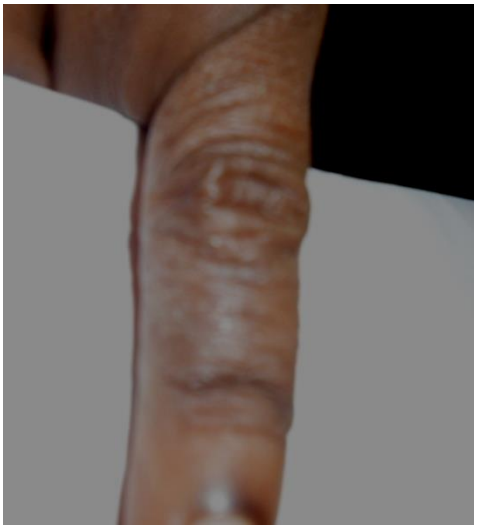
1f 

Plate 1a –f: Different infected sites on the human body (1a. infected feet, 1b. infected toes, 1c.infected abdomen, 1d. infected chest, 1e. infected back, 1f. infected finger).

3.2.1 Direct Examination

Direct examination is essential, as it allows one to be sure of fungal presence in the infection (the clinician to start treatment), pending culture. Here, a little part of the scrapping was placed on a glass slide, and one to two drops of 10–20% potassium hydroxide (KOH) with dimethyl sulfoxide (DMSO) which is the most commonly used was added, then viewed under the microscope for fungal hypae (Lilly *et al.*, 2006).

3.3 ISOLATION AND IDENTIFICATION OF THE PATHOGENIC FUNGI

3.3.1 Medium Preparation

Sabouraud dextrose agar containing antibiotics like chloramphenicol, gentamicin and cycloheximide were used as primary isolation medium (Appendix C). Sabouraud media were used for the isolation, cultivation, and maintenance of saprophytic and pathogenic fungi; it supplies peptone as the protein source and dextrose as the carbohydrate source for nourishment. As for antibiotics, they inhibit the growth of many bacterial species and opportunistic moulds that could hamper the recovery of dermatophytes since skin and hair are susceptible to contain many bacteria or conidia of saprophytic fungi. Sixty-five grammes (65g) of commercially produced Sabouraud dextrose agar was measured and dissolved in one litre of sterile distilled water and boiled to dissolve the medium completely. The agar was sterilised by autoclaving at 121^0C for 15 minutes and allowed cool to 50°C, before pouring the melted medium into a sterile Petri plates (15-cm diameter Petri plates at a depth of 4.0 mm), and cool at room temperature to solidify. For maximum shelf life, the medium was stored in refrigerator prior usage.

3.3.2 Isolation of Fungi

Prior to inoculation, the poured plates were allowed to cool down to room temperature. The scrapped specimens, was then lightly embedded on the agar. After seeding the prepared agar plates with the scrapped materials, cultures were incubated at 30^0C (Robert and Pihet, 2008). Culture plates were examined at least twice a week since some morphological traits can appear transiently, even though some can take up to twelve to fifteen days before showing any morphological characteristics.

3.3.3 Identification of Fungi

Once growth occurred, in order to accurately identify these isolated human pathogenic fungi, each specific type of colony morphology by gross appearance (topography, texture, and pigmentation) is noted. A little portion of the growth colony was teased with an inoculation needle and mount in a drop of Lactophenol Cotton Blue on a clean microscope slide. Covered with a cover slip, this was squash with the butt of the inoculation needle and the excess fluid then blot off. The preparation was examined under a light microscope with an attached camera (Motic McCamera [2000] 2.0 megapixel digital coloured camera) connected to a computer, for the microscopic photography of the Fungi. This was to observe the precise arrangement of the conidiophore and the way in which their spores are produced. The identities of these fungi were certified using their cultural, morphological as well as comparing them with confirmed representatives of different species in relevant texts such as Alexopolous *et al*. (2007) and Ellis *et al*. (2007).

3.4 DNA EXTRACTION

Extraction of fungal DNA was done using 4 different protocols namely Cetyltrimethyl ammonium bromide (CTAB) protocol, Modified CTAB protocol, Zymo kit by Zymo Research Corp (Hatfield-Pretoria 0028, South Africa) and GNOME kit by Qbiogene Corp.

3.4.1 DNA extraction using CTAB protocol

Ten millilitres (10 ml) of sterilized distilled water was added to a freshly pure culture of each isolates to be extracted, a suspension was made by gently probing the colony with the tip of a sterilised Pasteur pipette. The suspension was transferred into an eppendorf tube, and was spin for 30 seconds. The supernatant was decanted and 400 µl of CTAB buffer and 75 µl of 10% SDS were added to the pellet. This was then incubated in water bath at the temperature of 65^{o}C for fifteen minutes. Five hundred microlitres (500 µl) of chloroform was added and this was mixed by rocking on an orbital shaker at 100 rpm for 5 minutes, which was then spinned at 10,000 rpm for 10 minutes. The supernatant was collected into fresh eppendorf tube and 500 µl of iso-propanol was added to the supernatant. This was kept at -20 ^{o}C for one hour after which it was spin at 14,000 rpm for ten minutes. The supernatant was decanted gently and the pellet (DNA) was washed with 70% ethanol then, the pellet was air dried for thirty minutes. The dried pellet was then re-suspended in 200 µl of sterile distilled water.

3.4.2 DNA extraction using modified CTAB protocol

Ten millilitre (10 ml) of isolation buffer (10X CTAB) containing 80 µl of meta-mercaptoethanol in 50 ml blue cap tubes was pre-heated in 65 ^{o}C water bath. One

gramme (1g) of freshly scrapped pure culture plates of each fungal isolates was added to the preheated isolation buffer in each tube. The mixture was then incubated at 65^0 C for 15 minutes. Ten millitre (10 ml) of SEVAG (24:1 chloroform: Isoamyl achohol), then mixed gently but thoroughly. The cap of the tubes containing the mixture was then open to release gas, which was re-tightened then rocked the tubes using an orbital shaker (100 rpm) for 60 minutes. After rocking, the tubes were spinned at 4000 rpm at 25^0 C for 20 minutes. The mixture gave a clear and colourless aqueous solution at the top (containing the DNA), the clear colourless aqueous was removed into another set of tubes with the aid of a plastic transfer pipette. Two-third volume of iso-propanol was added to the aqueous solution. The mixture was then mixed gently and stored in freezer (-20^0 C) for 24 hours. This was to allow the DNA in it to precipitate. The mixture was later spinned in centrifuge at 3000 rpm for 5 minutes to collect the precipitate, and the liquid in the tube was discarded. Then 3 ml of 70% ethanol was added to dislodge the pellet in order to facilitate 'washing'. It was spinned again at 3000 rpm for 5 minutes, the liquid was discarded and the alcohol was allowed to evaporate by leaving the tubes open and on its side. The DNA was resuspended in 1.5 ml of water and stored at -20^0 C prior shipments for PCR and Sequencing.

3.4.3 DNA extraction using GNOME kit

A fresh pure culture of each fungal isolates was scraped into a microcentrifuge tube and suspended in 200 µl extraction buffer (2% Triton X-100, 1% SDS, 100 mM NaCl, 10 mM Tris-HCl, pH 8.0,1 mM Na_2EDTA). The suspension was centrifuged for 5 min at 13,500 rpm. The supernatant was transferred to a new microcentrifuge tube, and the nucleic acids were precipitated by adding 1 ml absolute ethanol. Suspensions were mixed and centrifuged for 2 min (13,500 rpm). The pellet was resuspended in

400 µl Tris-EDTA, pH 8.0, 3 µl RNAse (10 mg/ml) and incubated for 5 min at 37° C. Then, 10 µl ammonium acetate (4 M) and 1 ml absolute ethanol were added and gently mixed. This mixture was centrifuged for 3 min at 13,500 rpm and the supernatant discarded. The DNA pellet was dried in airflow for 15 min and finally resuspended in 40 µl sterile distilled water.

3.4.4 DNA extraction using ZYMO kit

Ten grammes of freshly prepared pure fungal culture plate of each isolate was scrapped and suspended in 50 µl of water to the tubes of a ZR BashingBead™ Lysis Rack. Four hundred microlitres (400 µl) of Lysis Solution was added to each tube. Tubes cap were tight to prevent leakage. The ZR BashingBead™ Lysis Rack was Centrifuge at 5,500 rpm for 5 minutes. Two hundred and fifty microlitres (250 µl) of the supernatant was transferred to each well of a Deep-Well Block. Seven hundred and fifty microlitres (750 µl) of Fungal DNA Binding Buffer was added to the supernatant in the Deep-Well Block from Step 3. Completely covered with cover foil then mixed thoroughly by vortexing block for 2 minutes. The Deep-Well Block was centrifuge at 5,000 rpm for 5 minutes. After removing cover foil and 500 µl of each of the supernatants from Step 5 was transferred to the wells of a Silicon-A™ Plate on a collection plate. The assembly was centrifuge at 5,000 rmp for 5 minutes. The flow was discard through from the collection plate and repeat Step 5. Two hundred microlitres (200 µl) of DNA Pre-Wash Buffer was added to the wells of the Silicon-A™ Plate on the emptied Collection Plate and centrifuge the assembly at 5,000 rpm for 5 minutes. Five hundred microlitres 500 µl Fungal DNA Wash Buffer was added to the wells of the Silicon-A™ Plate on the collection plate and centrifuge the assembly at 5,000 rpm for 5 minutes. The Silicon-A™ Plate was transferred to an

Elution Plate and 100 µl DNA Elution Buffer was added directly to the matrices in the plate. The assembly was centrifuged at 5,000rpm for 5 minutes. After which eluted, ultra-pure DNA is now ready for PCR and Sequencing.

3.5 DNA VERIFICATION ON 1% AGAROSE GEL ELECTROPHORESIS

One percent (1%) of agarose gel was prepared by mixing 1.5 g agarose with150 ml 1X TBE Buffer. The mixture was boiled in a microwave until all agarose has dissolved (usually, two minutes). This was cooled down under running cold tap water; after which, 6 µl of Ethidium Bromide (this was done in the fumehood) was added and swirled to mix. The gel was then poured into the tray and allowed to stand for at least thirty minutes before removing the combs. The gel was then placed in an electrophoresis tank. Five microliters of loading dye was spotted for 10 µl of each DNA sample on a parafilm paper. Using a pipette, 15 µl of each sample was loaded into each "well" of the prepared 1% agarose gel; also one-half microgram of 1kb ladder (Lambda DNA Hindlll) was loaded into the "marker" well. It was run for 45 minutes at 110 Milli Amps.

3.6 POLYMERASE CHAIN REACTION (PCR) AND DNA SEQUENCE

The extracted DNA samples were contracted out to Macrogen Corp. Great Seneca Hwy. Rockville, MD, USA for the PCR and DNA sequence analysis. The Polymerase Chain Reaction-Restriction Fragment Length Polymorphism (PCR-RFLP) is used to amplify the internal transcribed spacer (ITS) region of the rDNA. The PCR was performed using the universal primer pair ITS1-ITS4 (Jackson *et al.*, 1999). The thermal cycler was programmed for 4 min at 94 °C followed by 35 cycles of 1 min at 94 °C, 2 min at 58 °C and 1.5 min at 72 °C. The nucleotide sequences were

determined automatically using the cycle sequencing protocol by the BigDye Terminator v3.1 Cycle Sequencing Kit in the ABI PRISM 377-DNA Sequencer.

The sequence data generated from the DNA extraction, PCR and sequences were sent to Prof Michael Arabatzis, a researcher and mycologist at University of Athens, School of Medicine, Greece for further identification and BLAST in order to confirm their identities. Basic Local Alignment Search Tool (BLAST) is a program used to infer functional and evolutionary relationships between sequences as well as to help identify members of gene families. This is achieved by comparing nucleotide sequences data that was produced from the fungal DNA extracted to sequences database at the Genbank and determine the statistical significance of matches.

3.7 ANTIFUNGAL ACTIVITY TEST USING DIFFERENT CLASSES OF ANTIFUNGAL DRUGS, EXTRACTED OIL FROM SOME NIGERIA PLANTS, SOME ANTISEPTIC SOAPS AND DETERGENTS.

3.7.1 Medium preparation

Sabouraud's dextrose agar containing antibiotics like chloramphenicol, gentamicin and cycloheximide was used as the medium for the sensitivity test. The medium was prepared according to the one described in section 3.3.1 above.

3.7.2 Antifungal agents' preparation

Eight different antifungal drugs, listed below were used: (1) *"Flucamed"* (fluconazole by Drugfield Phar. Ltd), (2) "*Diflucan*" (fluconazole by Pfizer), (3) "*Nystatin*" (Polyene by Mekophar), (4) "*Itranox*" (itraconazole by Hanmi Pharm. Co.Ltd), (5) "*Sporanox*" (itraconazole by Janseen-cilag), (6) "*Griseofulvin"* (Hovid Bhd), (7) "*Nizoral*" (ketoconazole by Janseen-cilag) and (8) "*ketoconazole"* (Janseen-cilag) (Plates 2-9).

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Plate 2: A pack of Itranox antifungal tablet.
Plate 3: A pack of Diflucan antifungal tablet.
Plate 4: A pack of Nystatin antifungal tablet.
Plate 5: A pack of Grisofulvin antifungal tablet.

6 

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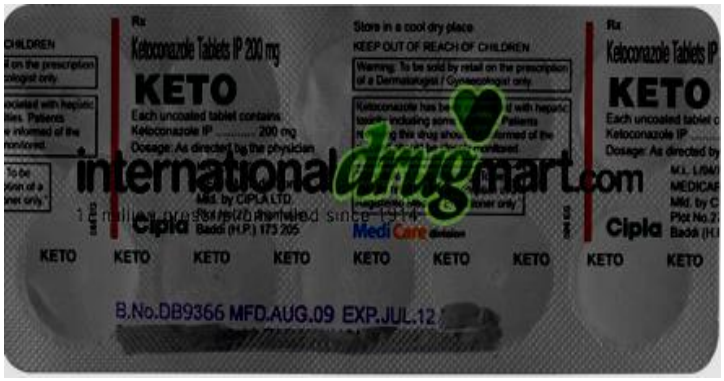
9 

Plate 6: A pack of Flucamed antifungal tablet.
Plate 7: A pack of Sporanox antifungal tablet.
Plate 8: A pack of Nizoral antifungal tablet.
Plate 9: A sachet of Ketoconazole antifungal tablet.

The above listed antifungal agents were dissolved in sterile distilled water to serve as stock solutions. Based on the manufacturers' prescrition, different grammes of the drugs were dissolved in 10 ml of sterile distilled water. 100 mg of itraconazole in 10 ml of water, 200mg per 10 ml of water for the ketoconazole, 500mg per 10 ml of water for the griseofulvin, 50mg per 10ml of water for the fluconazole and 500000IU per 10ml of water for the nystatin. All stock solutions were stored in the refrigerator until use.

3.7.3 Extraction of vegetable oil from three plant seeds

The seeds of *Azadirachta indica, Treculia africana and Detarium senegalense,* used in this study (Plates 10-12) had some foreign materials and dirt which was separated by hand picking. These seeds were sun dried in the open for five days, and were further dried in the oven at 40°C for 7hrs to a constant weight in order to reduce its moisture content, which was initially at about 5 to 7%. Mortar and pestle were used to crush the seeds into smaller particles in order to weaken or rupture the cell walls to release oil for extraction. One hundred and fifty grammes of each of the seeds was packed into a thimble and was inserted into the soxhlet extractor which was in turn inserted into the quick fit round bottom flask containing the solvent to be used that is, petroleum ether. The soxhlet cover was placed on top of the soxhlet. The condenser with inlet and outlet was placed unto the soxhlet cover, while the inlet of the condenser was connected unto a running tap, its run-off (waste water) passed through the outlet. The whole set-up was placed on a regulated heating mantle (usually 40^0 C to 60^0 C). When the solvent started boiling, the vapour rises through the vertical tube into the condenser at the top. The liquid condensate drips into the filter paper thimble in the centre, which contains the solid sample to be extracted. The extract seeps

10

11

12

Plate 10: *Azadirachta indica* seeds
Plate 11: *Detarium senegalense* seeds
Plate 12: *Treculia africana* seeds

through the pores of the thimble and fills the siphon tube, where it flows back down into the round bottom flask. This was allowed to continue for 30 minutes. It was then removed from the tube, dried in the oven, cooled in the desiccators and weighed again to determine the amount of oil extracted (Akpan *et al.*, 2006).

3.7.4 Inocula preparation

Stock inoculum suspensions of the isolated fungi were prepared from young growing culture medium. Ten millilitres of sterilized distilled water was added to colonies of fresh young growing culture of each fungus, and the suspensions were made by gently probing the colony with the tip of a sterilised Pasteur pipette. The resulting mixture of conidia and hyphal fragments was drawn and transferred to sterile tubes and kept in the refrigerator prior use.

3.7.5 Sensitivity test against antifungal agent and extracted oil

The disc diffusion agar method by Irobi and Daramola (1994) was used for the antifungal testing. Five millilitres (5ml) of the stock inoculum suspensions of each fungus was spread over the entire agar surface using a sterile inoculating loop on the previously prepared agar plates (SDA). This is to ensure an even distribution of the inoculum and these were dried at room temperature for 15 minutes before applying four of the antifungal agents' discs (the discs were prepared by the perforation of Whatman's filter paper into disks of 5 mm in diameter. The paper discs were then wrapped in an aluminium foil and sterilised in an autoclave at 121^0 C, for 15 minutes to kill any chance containminant before soaking in previously prepared antifungal agent solution and extracted oils twenty-four hours prior inoculation). The discs were left to warm to room temperature before use. The four disks were placed evenly (not

closer than 24 mm from center to center) on the surface of the agar plate with the aid of a sterile forceps. The inoculated plates were then incubated at 30° C aerobically. Each plate was labelled with necessary information (like agent name, fungus name and time of inoculation). Culture plates were examined twice a week, zone of inhibition were read and measured using a transparent ruler according to the methods of Booths, (1971).

3.7.6 Sensitivity test against some antiseptic soaps and detergents.

The following antiseptic soaps and detergents with the brand names were used, (1) "*Tetmosol*" antiseptic soap, (2) "*Dettol"* antiseptic soap, (3) *"Delta*" antiseptic soap, (4) "*Life bouy*" antiseptic soap, (5) *"Omo"* (multiactive) detergent, (6) "*Ariel*" (Pro-zim), (7) "*Klin"* and (8) "*Zip"* (Plate 13 and 14) with the chemical constituent as stated in Table 1 .Five grammes (5 g) of each detergent and bar soap mentioned above were measured and dissolved in fifteen milliliters of sterilized distilled water in clean petri dishes (i.e. ratio 1:3). Enough sterilized perforated paper discs were then soaked in these prepared solution for twenty four hours prior the test. Four discs from each solution were then placed evenly (not closer than 24 mm from center to center) with the aid of a sterile forceps on the surface of the agar plate which has been evenly seeded with a fresh stock inoculum suspensions of each fungus isolates tested against. The inoculated plates were then incubated at 30°C aerobically. Each plate was labelled with necessary information (like agent name, fungus nam e and time of inoculation). Culture plates were examined at least twice a week, zone of inhibition were read and measured using a transparent ruler according to the methods of Booths, *et al.* (1971).

13a 

13b 

13c 

13d 

Plate 13 a-d: A sachet of **"***Omo*" (multiactive) detergent, b. A sachet of "*Klin"* detergent,c. A sachet of "*Zip"* detergent, d. A sachet of "*Ariel* "(Pro-zim).

14a 

14b 

14c 

14d 

Plate 14a-d: a. A bar of "*Tetmosol"* antiseptic soap, b. A bar of "*Delta*" antiseptic soap, c. A bar of *"Lifebuoy*" antiseptic soap, d. A bar of "*Dettol*" antiseptic soap.

Table 1: Chemical constituent of the antiseptic soaps and detergents used.

S/N	AGENT	CHEMICAL CONSTITUENT
1	Lifebuoy	1%w/w Trichlorocarbanilide, Sodium Tallowate, Aqua, Titanium dioxide, *Helianthus annuus* seed oil, Sodium Lauyl Sulfate and *Curcuma aromatica* root oil.
2	Dettol	0.6%w/w Trichlorocarbanilide, water, Glycerine, soap base, Menthol and Colour fragrance.
3	Delta	0.5%w/w Trichlorocarbanilide, water and soap base
4	Tetmosol	0.5%w/w Trichlorocarbanilide, Sodium Tallowate, Monosulfram and Citronella
5	Klin	Triclosan, Linear Alkyl Benzene Sulfonate, Sodium Tripolyphoshate, Natrium Carbonate and Natrium Sulphate
6	Zip	Triclosan, and Antiredeposition agent
7	Arial	Triclosan, Linear Alkyl Benzene Sulfonate, Sodium Tripolyphoshate, Natrium Carbonate, Natrium Sulphate and Sodium Carbonate
8	Omo	Triclosan, Linear Alkyl Benzene Sulfonate, Sodium Sulphate, Sodium Tripolyphoshate and Sodium Carbonate

3.8 GROWTH STUDIES USING DIFFERENT TYPES OF MEDIA

3.8.1 Preparation of different types of media used.

Growth studies of all isolated fungi were carried out using the modified methods of Khattab (2006), to determine the best media for their cultivation. The mentioned grammes of each medium (Appendix C) were dissolved in 1 litre distilled water and boiled to dissolve the medium completely. The mixture was then sterilized by autoclaving at 121^0 C for 15 minutes. Each medium was poured in triplicates into Petri dishes and a cross was drawn with a permanent marker at the bottom of each Petri dish to indicate the centre as the origin. After which fresh pure culture of each fungus that was used for the study was transferred by cutting with a cork borer of 6 mm in diameter and placed at the centre of each marked petri dishes. The plates were then incubated at 30^0 C. The plates were observed daily at twenty four hours interval. The growth diameter were measured and recorded.

3.9 PHYTOCHEMICAL SCREENING

The extracted oils used in this study were subjected to preliminary phytochemical screening using stardard test to show the different types of chemical constituents present.

3.9.1 Test for alkaloids

Five millitres (5 ml) of each oil extract was measured and stirred with 5 ml of 1% aqueous hydrochloric acid on steam bath; 1ml of the filtrate was treated with a few drops of mayer's reagent and a second 1 ml portion was treated similarly with freshly prepared Dragendoff's reagent and wagner's reagent. Turbidity or precipitation with

any of these reagents was taken as evidence for the presence of alkaloid in the extract being screened (Evans, 1989; Harborne, 1998; Ogundipe and Oladipo, 2001).

3.9.2 Test for tannins

Five ml of each oil extract was measured and stirred with 10 ml of distilled water, filtered and 5% Ferric Chloride reagent was added to the filtrate. A blue, green, or blue-green precipitate was taken as evidence for the presence of tannins (Evans, 1989; Houghton and Raman, 1998).

3.9.3 Test for flavonoids

The methods of Harborne (1973) and Sofowora (1993) was adopted to determine the presence of flavonoids in the plant extracts by measuring 5 ml of dilute ammonia solution into the filterate of the extract followed by addition of sulphuric acid. A yellow colouration indicates the presence of flavonoids.

3.9.4 Test for saponins

The ability of saponins to produce frothing in aqueous solution and to haemolyse red blood cells was used as screening test for the sample. For frothing tests the method described by Farnsworth (1966) was used. Five (5) ml of each oil extract was measured and shaken with water in a test tube and filtered. Frothing, which persisted on warming, is a preliminary evidence for presence of saponins. In order to remove ‘‘false-positive’’ result, the blood haemolysis test was performed on the filtrate of the frothed extract, 2 ml of normal saline solution was added to two test tubes. To one, of these, 2 ml of distilled water was added and 2 ml of extract to other. The concentration of sodium chloride in each tube is isotonic with blood serum. Five

drops were added to each test tube and inverted gently to mix the content. Complete haemolysis of red blood cells in the test tube with the oil extract was taken as further evidence of presence of saponin.

3.9.5 Test for cardiac glycosides

To test for the presence of cardiac glycosides**,** 0.5 g of the oil extracts was dissolved in pyridine and a few drops of 2% sodium nitroprusside together with a few drops of 20% Sodium hydroxide were added (Sofowora, 1993).

3.9.6 Test for anthraquinones glycoside

For the presence of anthraquinones glycoside, 0.1 g of the oil extracts was dissolved in 10 ml of hot water and this was steamed for five minutes, then the filtrates were extracted with chloroform. The chloroform layer was taken off; this layer was washed with 5 ml water and was shaken with 5 ml of dilute ammonia solution (Van- Buren and Robinson, 1981).

3.9.7 Test for reducing compounds

The methods of Harborne (1973); Van- Buren and Robinson (1981); Obadoni and Ochuko (2001) were used to determine the presence of reducing compounds in these oils used as a control means against these pathogenic fungi. 0.5 g of the oil extract was boiled with 1% aqueous hydrochloride acid to test for the presence of reducing compounds/sugars.

3.10 QUANTITATIVE DETERMINATION OF THE BIOACTIVE COMPOUNDS

3.10.1 Alkaloid determination using Harborne **(**1973) method: Five (5) g of the sample was weighed into a 250 ml beaker and 200 ml of 10% acetic acid in ethanol was added and covered and allowed to stand for 4 h. This was filtered and the extract was concentrated on a water bath to one-quarter of the original volume. Concentrated ammonium hydroxide was added in drops to the extract until the precipitation was complete. The whole solution was allowed to settle and the precipitated was collected and washed with dilute ammonium hydroxide and then filtered. The residue is the alkaloid, which was dried and weighed.

3.10.2 Tannin determination by Van-Burden and Robinson (1981) method: Five hundred (500) mg of the sample was weighed into a 50 ml plastic bottle. 50 ml of distilled water was added and shaken for 1 h in a mechanical shaker. This was filtered into a 50 ml volumetric flask and made up to the mark. Then 5 ml of the filtered was pipetted out into a test tube and mixed with 2 ml of 0.1 M $FeCl_3$ in 0.I N HCl and 0.008 M potassium ferrocyanide. The absorbance was measured at 120 nm within 10 minutes.

3.10.3 Cardiac glycosides determination: The method used was that of Obadoni and Ochuko (2001). The samples were ground and 20 g of each were put into a conical flask and 100 cm^3 of 20% aqueous ethanol were added. The samples were heated over a hot water bath for 4 h with continuous stirring at about 55° C. The mixture was filtered and the residue re-extracted with another 200 ml 20% ethanol. The combined extracts were reduced to 40 ml over water bath at about 90° C. The concentrate was transferred into a 250 ml separatory funnel and 20 ml of diethyl ether was added and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded. The purification process was repeated. 60 ml of n-butanol was

added. The combined n-butanol extracts were washed twice with 10 ml of 5% aqueous sodium chloride. The remaining solution was heated in a waterbath. After evaporation the samples were dried in the oven to a constant weight; the saponin content was calculated as percentage.

3.10.4 Reducing compounds determination by the method of Boham and Kocipai-Abyazan (1974): Ten (10) g of the plant sample was extracted repeatedly with 100 ml of 80% aqueous methanol at room temperature. The whole solution was filtered through whatman filter paper No 42 (125 mm). The filtrate was later transferred into a crucible and evaporated into dryness over a water bath and weighed to a constant weight.

3.11 STATISTICAL ANALYSIS

The data obtained from the growth studies and sensitivity test against the eight antifungal drugs, extracted oils, antiseptic soaps and detergents were analysed using analysis of variance (ANOVA) and Duncan‘s multiple range test (DMRT).

CHAPTER FOUR

4.0 RESULTS

4.1 COLLECTION OF SAMPLES

The weekly collection for seventy-eight consecutive weeks produced two hundred and forty- nine isolate in all. These were collected from different parts of the body, with the following percentage: 3.6% of the population from the face; 15.6% from the nail; 4.4% from the head and 21.0% from the leg and foot part. 6.0% and 49.4% from the private part and the body part (i.e. the chest, back, laps and buttocks) respectively (Fig.11). Of the two hundred and forty-nine scrapes, one hundred and fifteen (46.19%) were collected from male while the remaining one hundred and thirty-four (53.81%) were from females (Fig. 12). There are also variations with age, for the age group zero months to age twelve, 14.45% were generated (i.e. thirty-six scrapes); 3.2% (i.e. eight scrapes) for the age thirteen to eighteen while the adult age group produced 82.32% (i.e. two hundred and five scrapes) (Fig. 13).

4.2 ISOLATION AND IDENTIFICATION OF THE PATHOGENIC FUNGI

Out of the two hundred and forty nine isolates Appendix (B), only seven isolates did not yield any fungal growth with no respect to the body part they are isolated from. The remaining two hundred and forty- two produced twenty five morphological different species of fungi, of which twelve were suspected to belong to the dermatophytic group based on the morphologically comparsion with that of a stardard mycology text books Alexopolous *et al*. (2007) and Ellis *et al*. (2007). These fungi were later confirmed by the molecular techniques to give the following species of dermatophytes; the genera *Microsporum*, *Trichophyton* and *Epidermaphyton* (Fig 14). These included: *Epidermophyton floccosum, Microsporum*

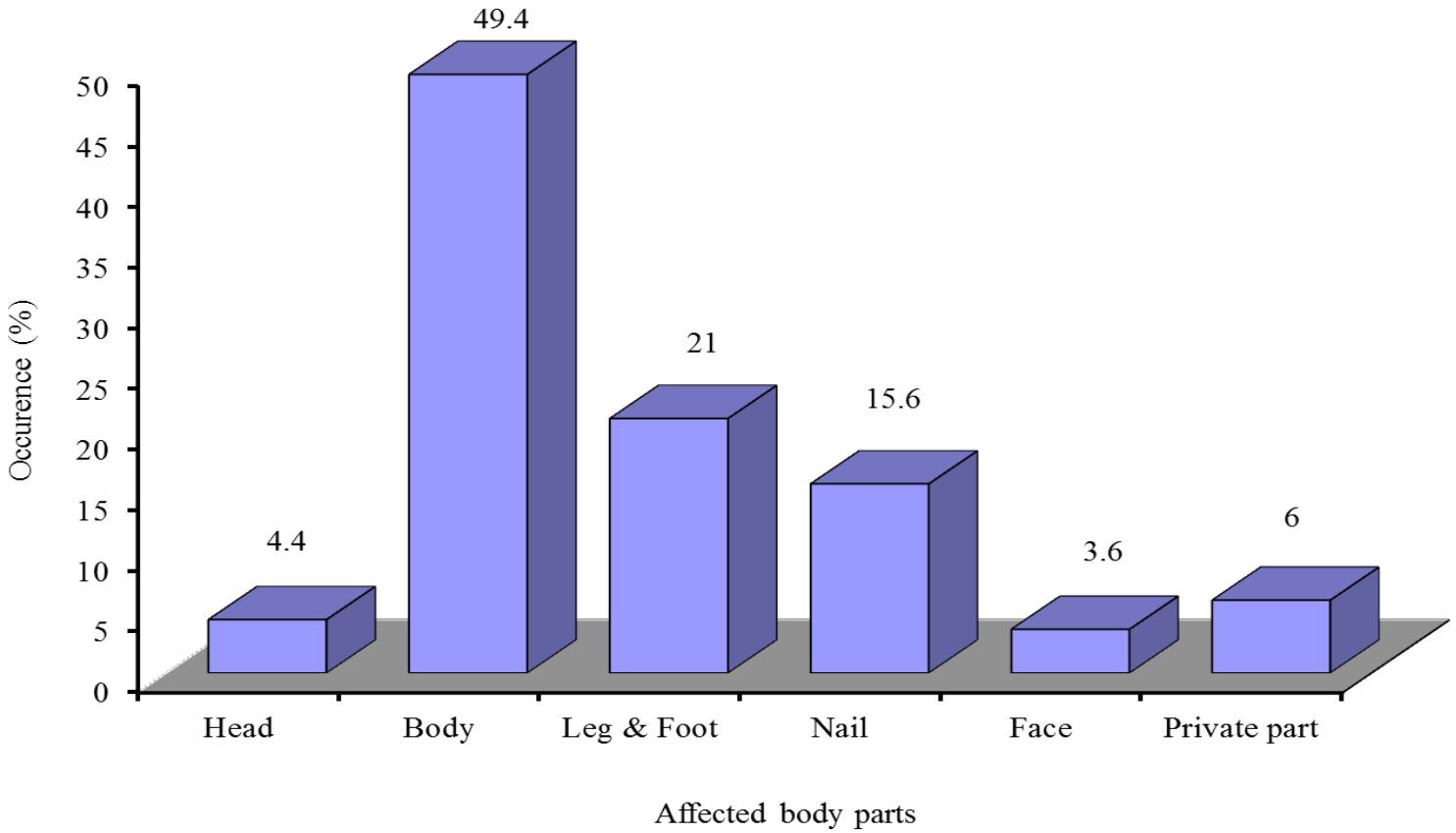


Fig. 11: Percentage occurrence of dermatomycoses from all infected body parts

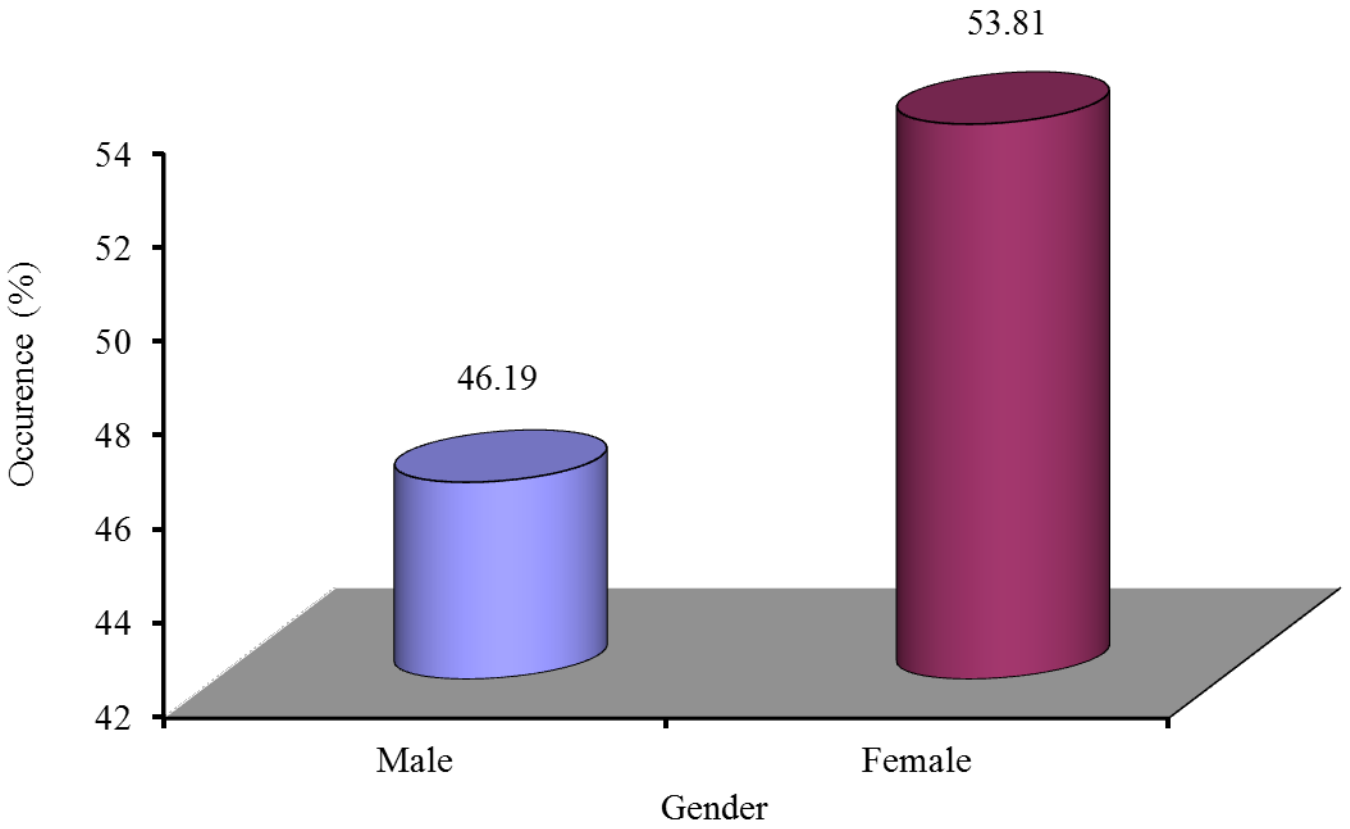


Fig. 12: Percentage occurrence of dermatomycoses in male and female.

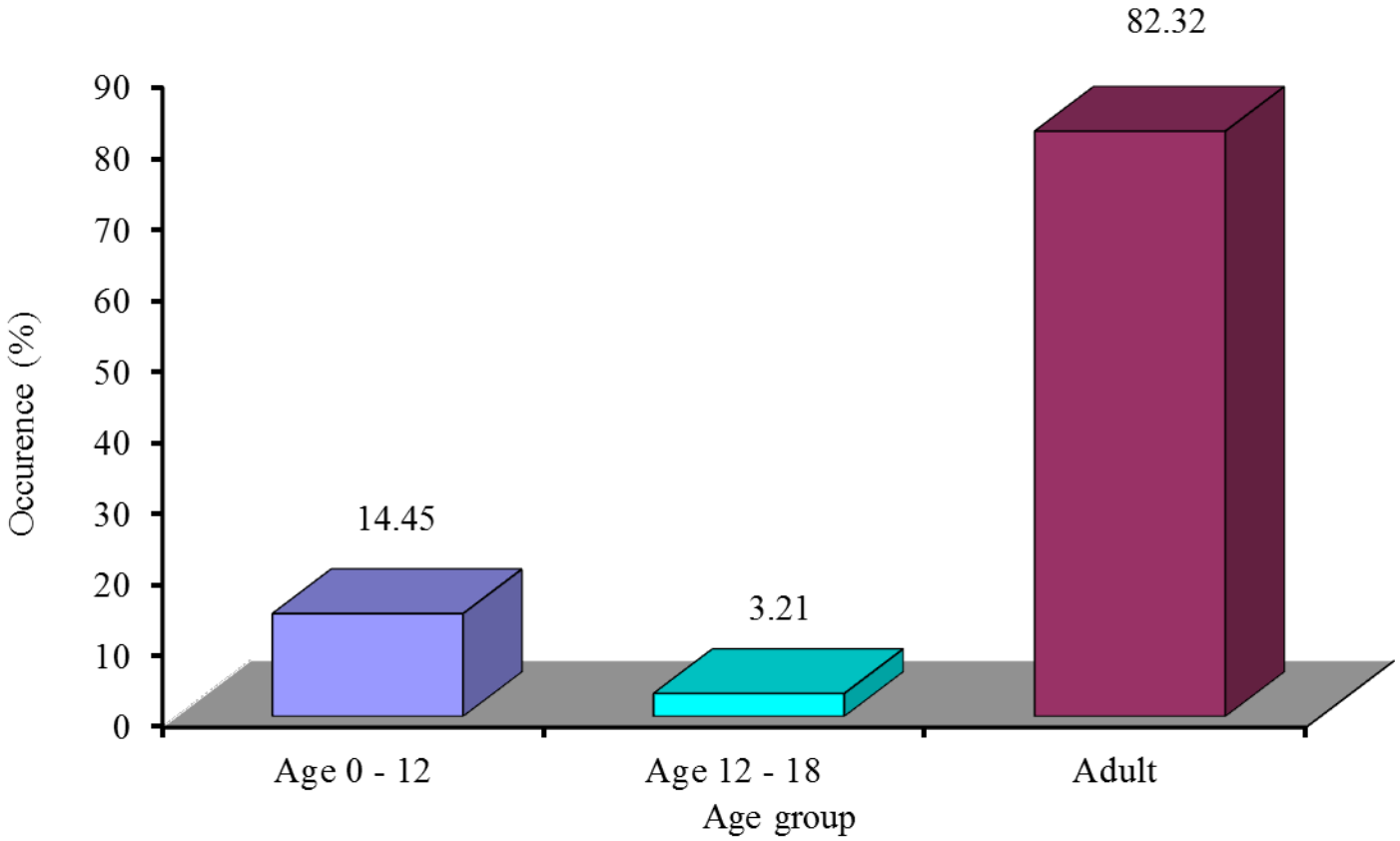


Fig. 13: Percentage occurrence of dermatomycoses among different age groups.

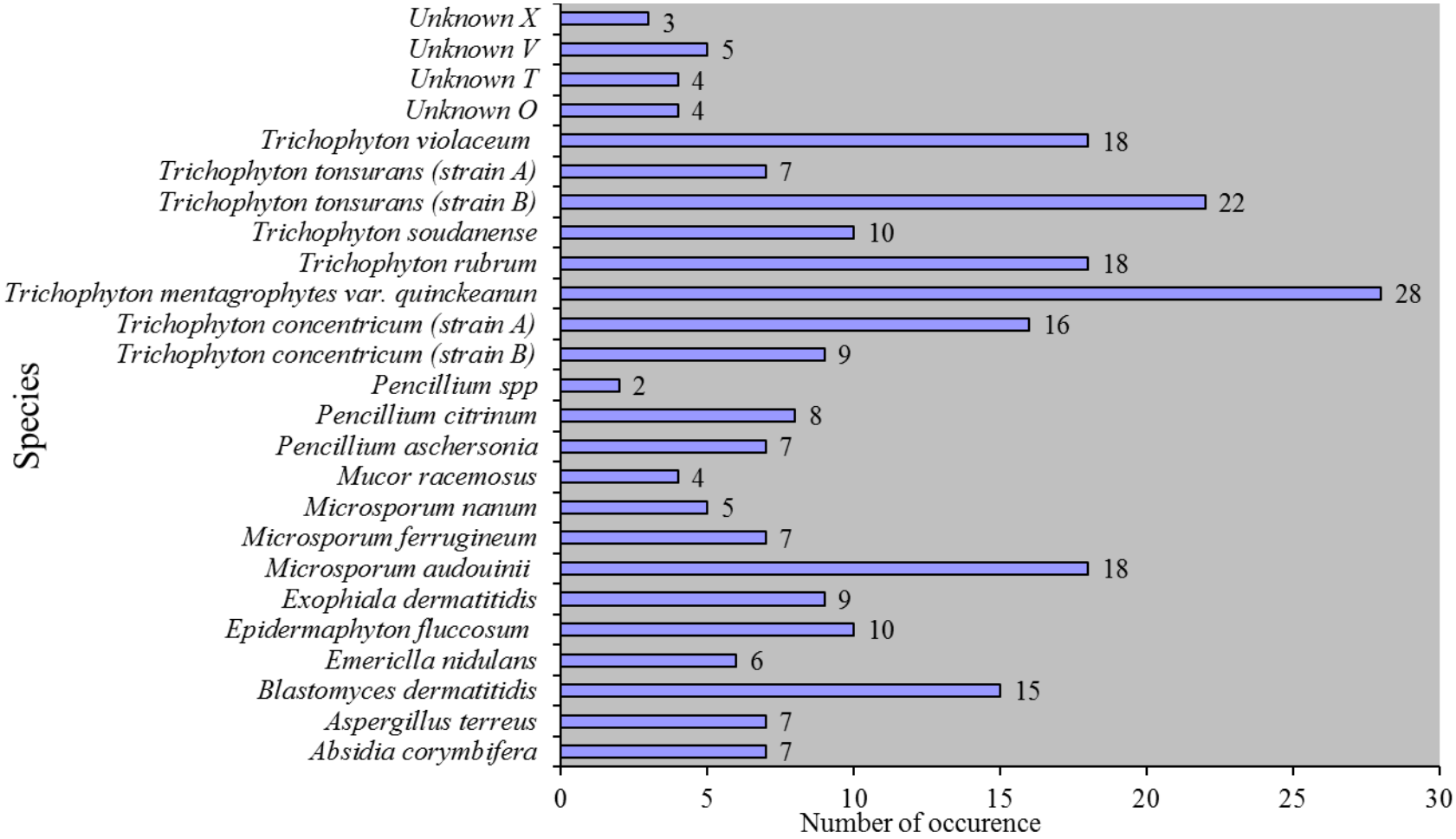


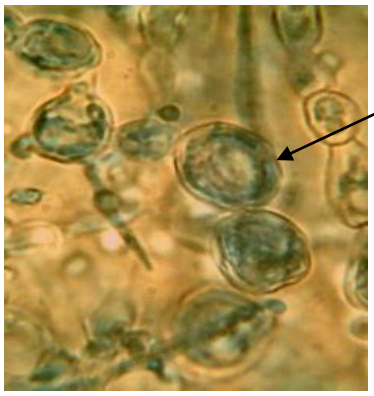
Fig. 14: Frequency of occurrence of all isolated fungi.

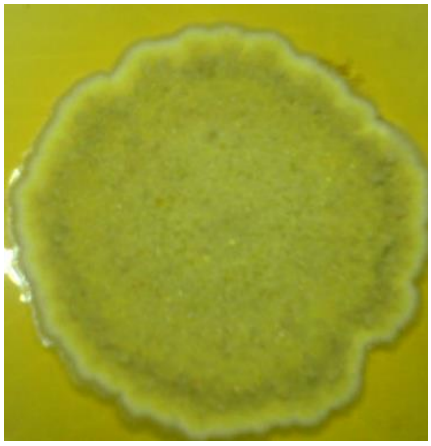
audouinii, Microsporum ferrugineum, Microsporum nanum, Trichophyton concentricum strain A, *Trichophyton concentricum* strain B, *Trichophyton mentagrophytes var. quinckeanun, Trichophyton rubrum, Trichophyton soudanense, Trichophyton tonsurans* strain A, *Trichophyton tonsurans* strain B and *Trichophyton violaceum* (Fig. 14). On the basis of their primary habitat associations, the isolated above mentioned dermatophytes are of three groups. These are the anthropophilic, zoophilic, and geophilic (Table 2). Two species of these isolated fungi belong to the systemic mycoses group otherwise known as the "deep mycoses" and these are *Blastomyces dermatitidis* and *Exophiala dermatitidis* The other seven species of the isolated fungi belong to the opportunistic mycoses group, and these included: *Absidia corymbifera, Aspergillus terreus, Emericlla nidulans, Mucor racemosus, Pencillium aschersonia, Pencillium citrinum, and Pencillium* species, while the last four isolated species remain unidentified. Plate 17 – 39 showed the photographs and the microphotograhs of the each isolated fungi species/strain cultures. Table 3 contains the morphology and microscopic descriptive characters of each isolated fungi. It was noted that some strain of isolated fungi are peculiar to a particular body site. For example, *Trichophyton rubrum, Trichophyton violaceum* and *Epidermophyton* f*locossum* were isolated from the nail more than any other dermatophytes species. *Trichophyton soudanense was* isolated more from private part and head while *Microsporum audouinii* was more of the body especially that of the school children age.

Table 2: The three groups of isolated dermatophytes based on their primary habitat

S/N	Anthropophilic	Zoophilic	Geophilic
1	*Epidermophyton floccosum*	*Trichophyton mentagrophytes var. quinckeanum*	*Microsporum nanum*
2	*Microsporum audouinii*		
3	*Microsporum ferrugineum*		
4	*Trichophyton rubrum*		
5	*Trichophyton soudanense*		
6	*Trichophyton violaceum*		
7	*Trichophyton concentrium(*strainA*)*		
8	*Trichophyton concentrium (*strain B*)*		
9	*Trichophyton tonsurans (*strain A)		
10	*Trichophyton tonsurans (*strain B)		

15a 

15b 

16a 

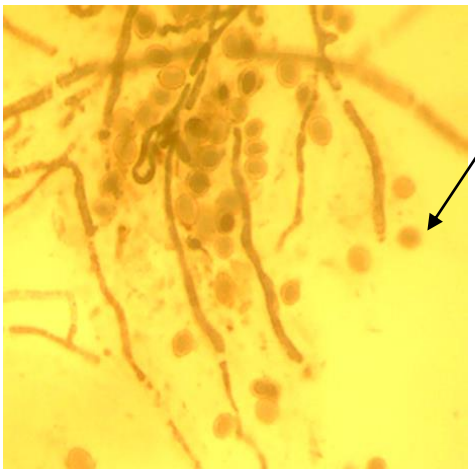
16b 

Microphotograph and culture photograph of isolated fungi on Sabouraud dextrose agar

Plate 15a: Culture of *Trichophyton concentricum* (strain A)
Plate 15b: Photomicrograph of *Trichophyton concentricum* (strain A*)*
(i) Chlamydospore (Mag. x400)

Plate 16a: Culture of *Trichophyton tonsurans* (strain A)
Plate 16b: Photomicrograph of *Trichophyton tonsurans* (strain A)
(Mag. x400)

17a 

17b 

18a 

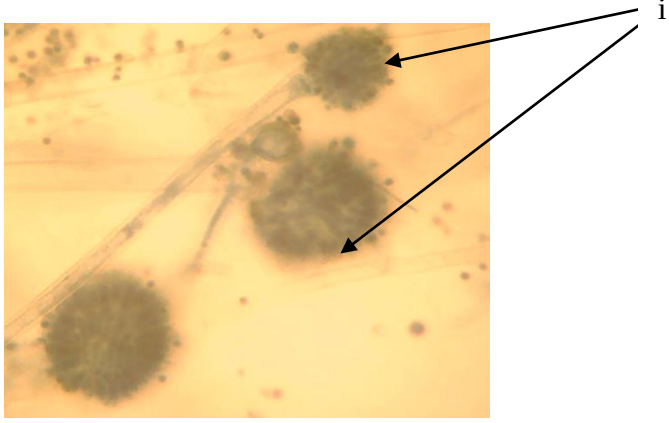
18b 

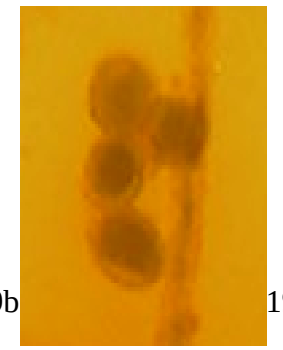
Plate 17a: Culture of *Trichophyton mentagrophytes var. quinckeanun*

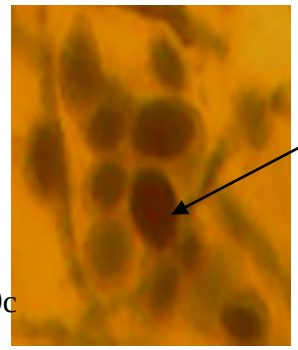
Plate 17b: Photomicrograph of *Trichophyton mentagrophytes var. quinckeanun* (i) microconidium (Mag. x400)

Plate 18a: Culture of *Aspergillus terreus*

Plate 18b: Photomicrograph of *Aspergillus terreus* (i) conidial heads (Mag. x400)

19a 

19b 

19c 

20a 

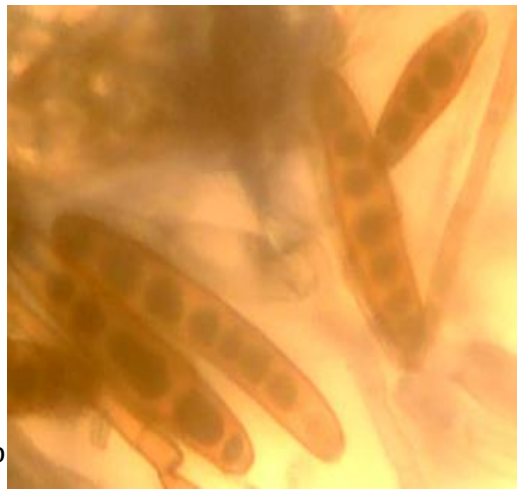
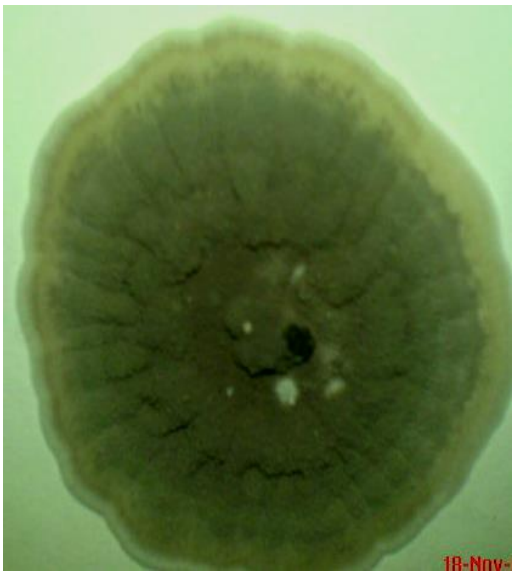
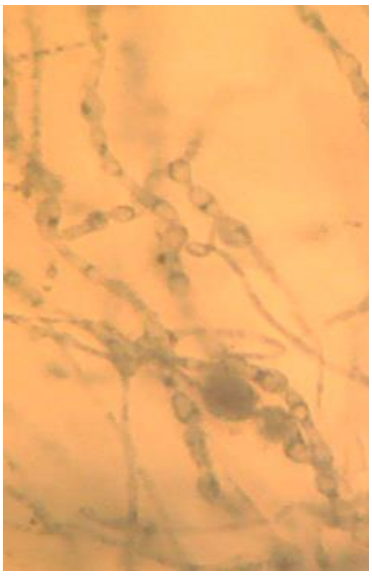
20b 

Plate 19a: Culture of *Blastomyces dermatitidis*
Plate 19b: Photomicrograph of *Blastomyces dermatitidis*
Plate 19c: Photomicrograph of *Blastomyces dermatitidis* (i) unipolar Budding yeast like cell (Mag. x400)

Plate 20a: Culture of *Trichophyton tonsurans* (strain B)
Plate 20b: Photomicrograph of *Trichophyton tonsurans* (strain B) (Mag. x400)

21a 

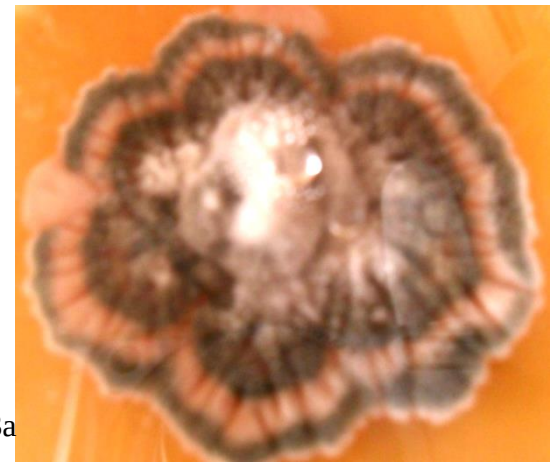
21b 

22a 

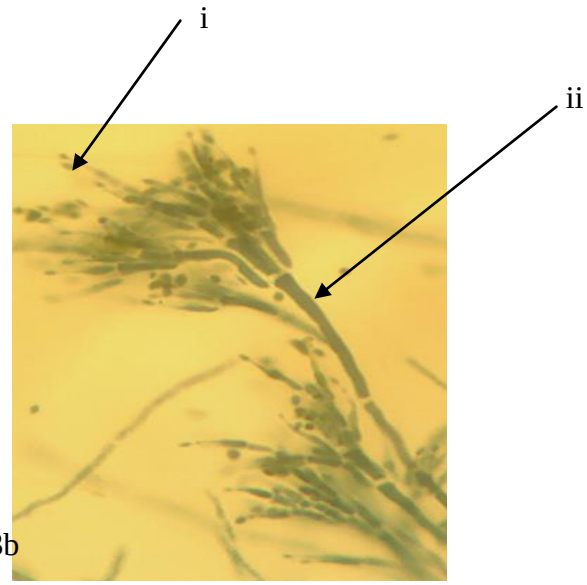
22b 

Plate 21a: Culture of *Exophiala dermatitidis*
Plate 21b: Photomicrograph of *Exophiala dermatitidis* (Mag. x400)

Plate 22a: Culture of *Microsporum audouinii*
Plate 22b: Photomicrograph of *Microsporum audouinii (i) intercalary chlamydospore* (Mag. x400)



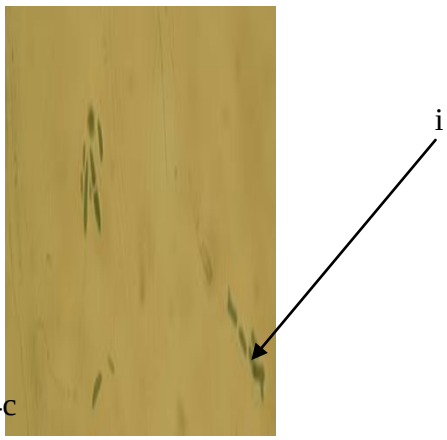
23a



23b

24a

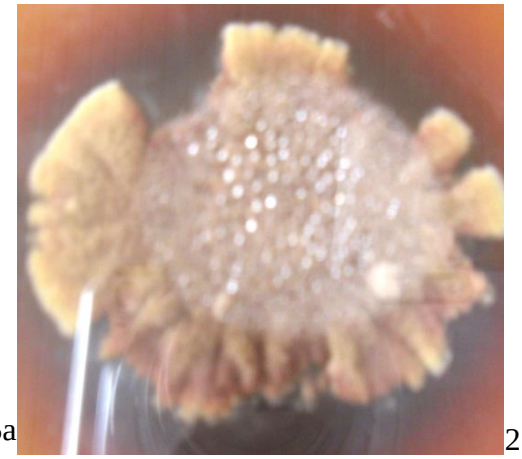
24b



24c

Plate 23a: Culture of *Penicillium citrinum*
Plate 23b: Photomicrograph of *Penicillium citrinum* (i) Condia (ii) Condiophore (Mag. x400)

Plate 24a: Culture of *Trichophyton rubrum*
Plate 24b: Photomicrograph of *Trichophyton rubrum*
Plate 24c: Photomicrograph of *Trichophyton rubrum* (i) microconidia (Mag. x400)

25a  25b 

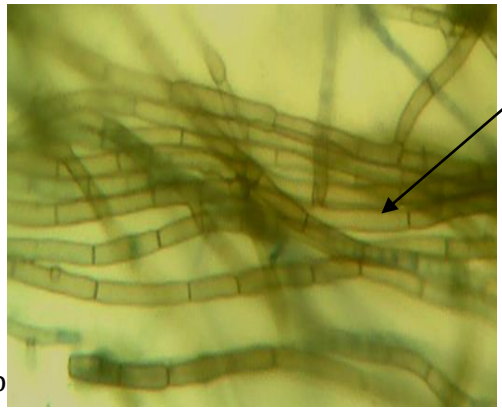
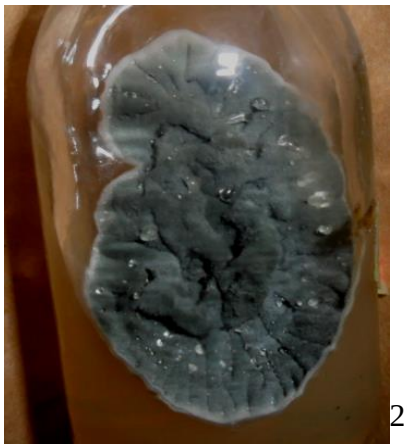
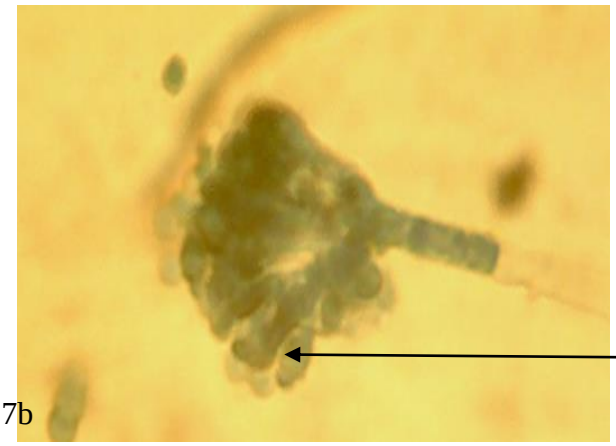
26a  26b 

Plate 25a: Culture of *Trichophyton soudanense*
Plate 25b: Photomicrograph of *Trichophyton soudanense* (Mag. x400)

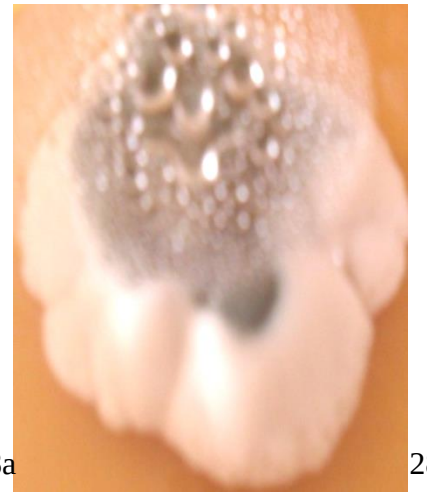
Plate 26a: Culture of *Microsporum ferrugineum*
Plate 26b: Photomicrograph of *Microsporum ferrugineum* (i) "bamboo Hyphae" (Mag. x400)



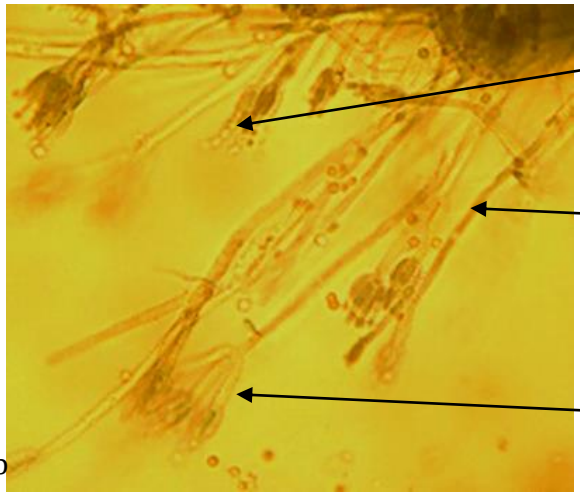
27a



27b



28a



28b

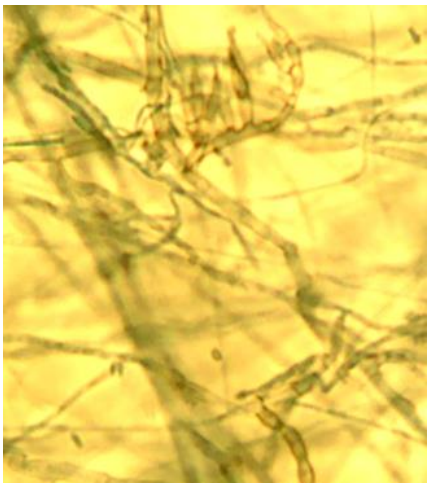
Plate 27a: Culture of *Penicillium aschersonia*

Plate 27b: Photomicrograph of *Penicillium aschersonia (i)* conidia (Mag. x400)

Plate 28a: Culture of *Penicillium* species

Plate 28b: Photomicrograph of *Penicillium* species (i) conidia in unbranched chain (ii) specialized hypha (iii) phialide (Mag. x400)

29a 

29b 

30a 

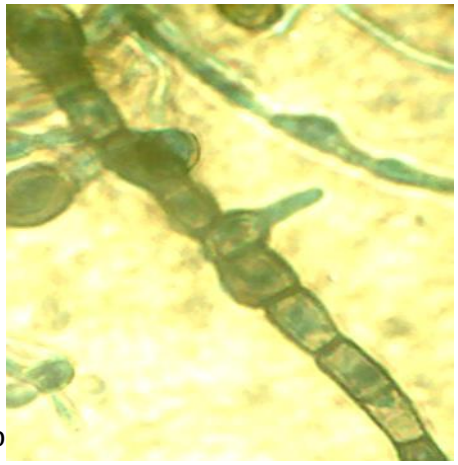
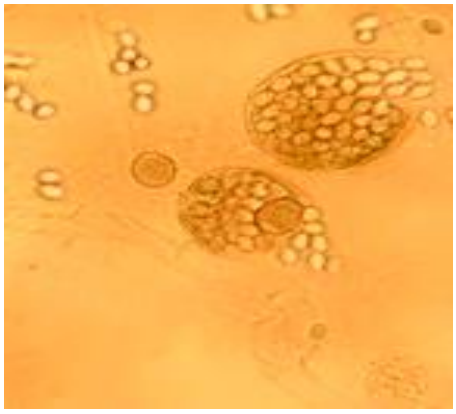
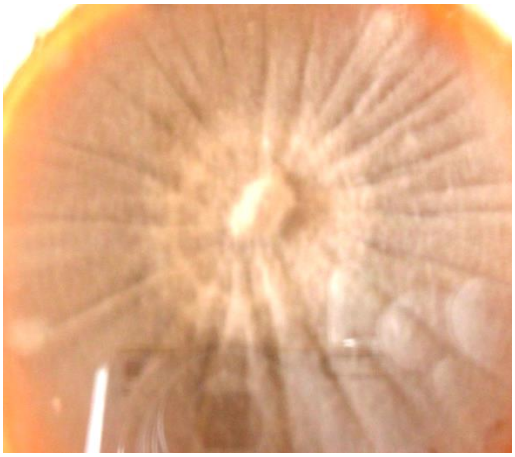
30b 

Plate 29a: Culture of *Unidentified* O
Plate 29b: Photomicrograph of *Unidentified* O(Mag. x400)

Plate 30a: Culture of *Epidermaphyton fluccosum*
Plate 30b: Photomicrograph of *Epidermaphyton fluccosum* (Mag. x400)

31a 

31b 

32a 

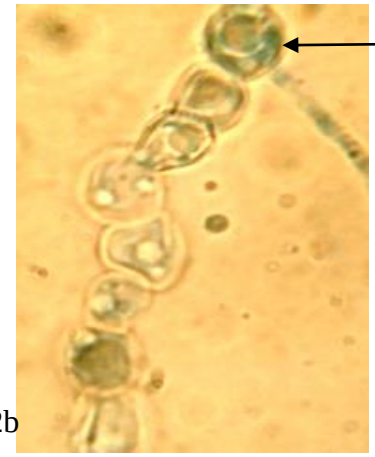
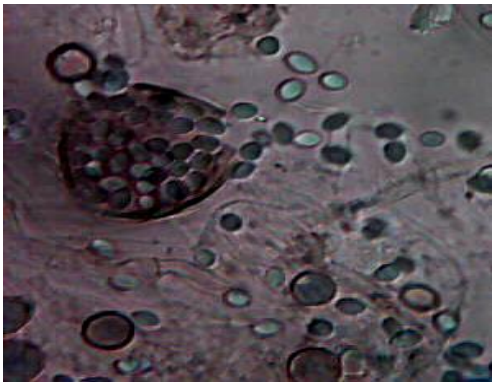
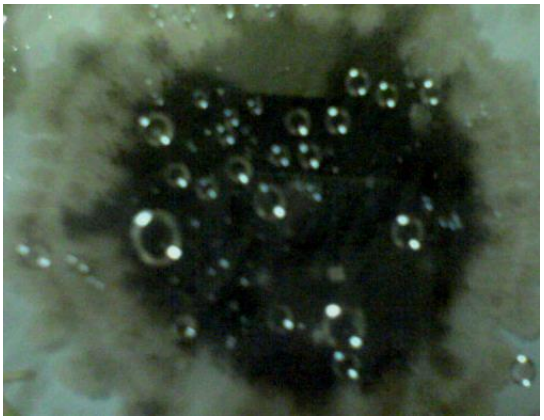
32b 

Plate 31a: Culture of *Mucor racemosus*
Plate 31b: Photomicrograph of *Mucor racemosus* (Mag. x400)

Plate 32a: Culture of *Trichophyton concentricum* (strain B)
Plate 32b: Photomicrograph of *Trichophyton concentricum* (strain B)
(i) ballon-shaped chlamydospore (Mag. x400)

33a 

33b 

34a 

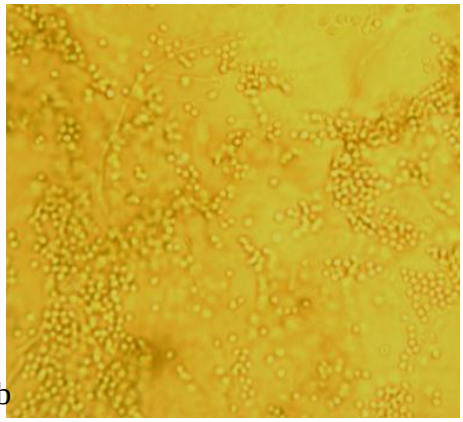
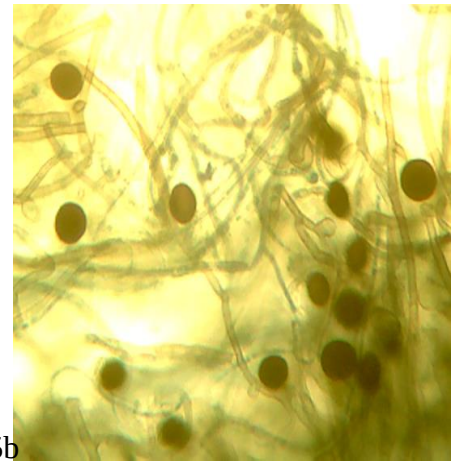
34b 

Plate 33a: Culture of *Absidia corymbifera*
Plate 33b: Photomicrograph of *Absidia corymbifera* (Mag. x400)

Plate 34a: Culture of *Unidentified T*
Plate 34b: Photomicrograph of *Unidentified T* (Mag. x400)

35a  35b 

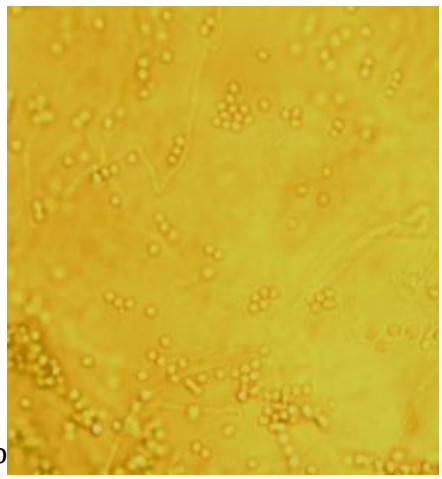
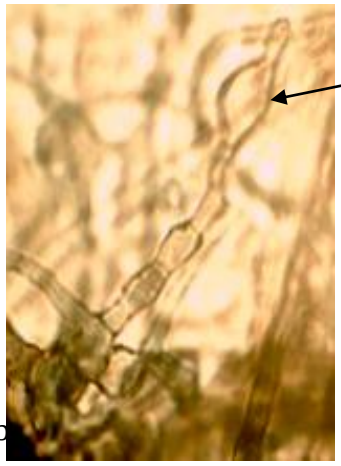
36a  36b 

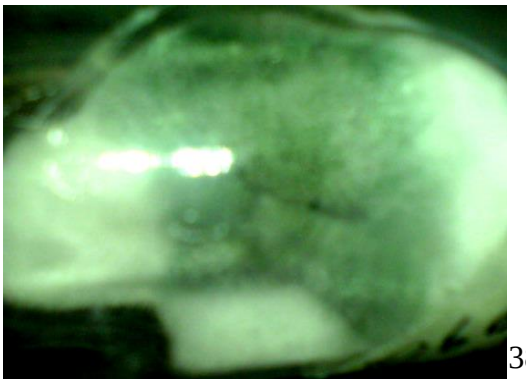
Plate 35a: Culture of *Microsporum nanum*
Plate 35b: Photomicrograph of *Microsporum nanum* (Mag. x400)

Plate 36a: Culture of *Unidentified V*
Plate 36b: Photomicrograph of *Unidentified V* (Mag. x400)

37a 



37b


38a

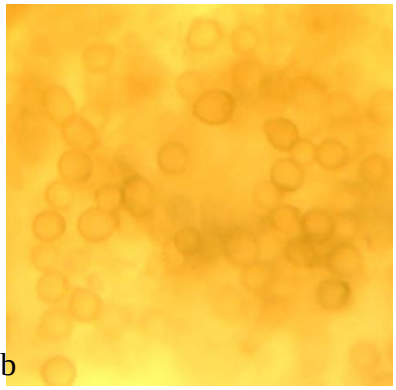
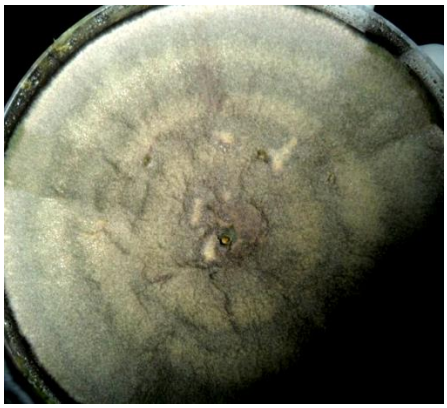

38b

Plate 37a: Culture of *Trichophyton violaceum*
Plate 37b: Photomicrograph of *Trichophyton violaceum*
(i) Chlamydospore (Mag. x400)

Plate 38a: Culture of Unidentified X
Plate 38b: Photomicrograph of Unidentified X (Mag. x400)

39a 

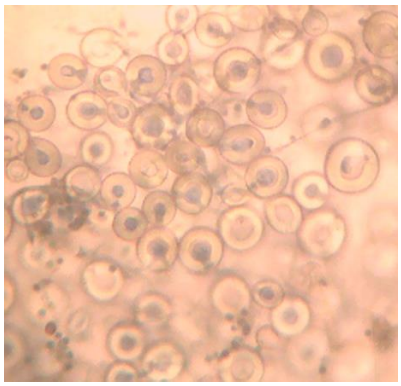
39b 

Plate 39a: Culture of *Emericlla nidulans*
Plate 39b: Photomicrograph of *Emericlla nidulans* (Mag. x400)

Table 3: FUNGI MORPHOLOGY AND MICROSCOPIC DESCRIPTION

FUNGI	CHARACTERISTICS
Albsidia corymbifera	It is a fast growing fungus, floccose, white at first becoming pale grey with age. Sporangia are small and are typically pyriform in shape with a characteristic conical-shaped columella and pronounced apophysis, often with a short projection at the top.
Aspergillus terreus	This fungus colony is typically suede-like and cinnamon-buff to sand brown in colouration. Conidial heads are short columnar and biseriate, with short, brownish and smooth-walled Stipes.
Blastomyces dermatitidis	This fungus grows slowly as glabrous, tan, nonsporulating colony with unipolar budding yeast like cells in culture.
Emericella nidulans	The colony is typically dirty green in colour. Conidial heads are short columnar and biseriate. Stipes are usually short, brownish and smooth-walled.

Epidermophyton floccosum	This is a slow growing fungus with a khaki-coloured colony. Culture shows characteristic smooth, thin-walled macro conidia which are often produced in clusters growing directly from the hyphae.
Exophiala dermatitidis	Colony here is slow growing, suede-like, olivaceous grey with the development of aerial mycelium with age, while the culture flask-shaped to cylindrical annellides are produced with the development of mycelium.
Microsporum audouinii	The colony here has a dense suede-like to downy surface, suggestive of mouse fur in texture with no reverse pigment. Its culture produces an occasional thick-walled terminal or intercalary chlamydospores.
Microsporum ferrugineum	The fungus is slow growing, forming a waxy, glabrous colony. Colouration changes from cream to yellow. There is presence of irregular branching hyphae with prominent cross walls often referred

	to as “bamboo hyphae”.
Microsporum nanum	Cultures produce numerous small ovoid to pyriform macroconidia with one to three cells, with relatively thin, finely echinulate (rough) walls, and broad truncate bases.
Mucor racemosus	Colonies are very fast growing, cottony to fluffy, white to yellow colouration. The culture consists of spherical, non-apophysate sporangia with pronounced columellae and conspicuous collarette at the base of the columella following sporangiospore dispersal.
Penicillium aschersonia	Colony is fast growing, in shades of green and white. Cultures consisting of chains of single-celled conidia are produced in basipetal succession from a specialised conidiogenous cell called a phialide.
Penicillium citrinum	Colony is fast growing, in shades of green and white. Cultures consisting of chains of single-celled conidia are produced in basipetal succession from a specialised

	conidiogenous cell called a phialide. The phialides are produced singly in groups or from branched metulae, giving a brush-like appearance.
Penicillium species	Colony here is elevated and fast growing, in shades of green and white. Cultures consisting of chains of single-celled conidia are produced in basipetal succession from a specialised conidiogenous cell called a phialide.
Trichophyton rubrum	This fungus possesses a flat to slightly raised colony with a white to cream colouration. Culture has numerous clavate to pyriform microconidia.
Trichophyton soudanense	This is a slow-growing fungus with a flat to folded, suede-like surface and broad fringe of submerged growth colony. Both surface mycelium and reverse pigment are characteristically deep apricot-orange in colour. Culture often produces hyphae showing reflexive or right-angle branching.

Trichophyton violaceum	This is a very slow growing organism with a glabrous, heaped and folded colony. Hyphae are relatively broad, tortuous, much branched and distorted.
Trichophyton concentrium (strain A)	This is a slow growing fungus, raised and folded colony, often deeply folded into the agar which may produce splitting of the medium into a few portions. This fungus is orange-brownish in colour. The culture consists of clavate to pyriform microconidia.
Trichophyton. concentricum (strain B)	This fungus is a slow growing organism, with a raised and folded colony, glabrous becoming suede-like, mostly white to cream-coloured. The isolate produces clavate to pyriform microconidia.
Trichophyton mentagrophytes var. quinckeanum	The colony here is slightly raised and folded, white to cream and suede-like in texture. In the culture, numerous microconidia are borne laterally along the sides of the hyphae.

Trichophyton tonsurans (strainA)	This fungus is powdery, flat with a raised centre and radial grooves. It shown pale-buff to yellow colouration. There is presence of numerous characteristic microconidia which vary in size and shape from long clavate to broad pyriform.
Trichophyton tonsurans (strain B)	The fungus possesses a yellow-brown coloured colony with irregular hyphae and numerous septa. Also, there is the presence of numerous characteristic microconidia which vary in size and shape from long clavate to broad pyriform.
Unidentified O	The fungus possesses a relatively slow growing colony, flat surface and folded, in shades of black and white colouration.
Unidentified T	Colony here is a relatively fast growing one, with a flat surface, with shades of black and white colouration.
Unidentified V	Colony here is an elevated, folded and fast growing, It is in shades of white, black

	and yellow colouration. Culture consists of chains of double or multiple celled conidia scattered all over the culture.
Unidentified X	Colony here is elevated and fast growing, in shades of green and white.

4.3 MOLECULAR CHARACTERIZATION

The extracted DNA from each isolated fungi which were size seperated on 1% agarose gel was then viewed under the ultraviolet trans-illuminator for DNA quality and yield assessments and the photograph of the gel was taken as shown in the electrophorogram below (Plate 40 – 43). Plate 40 showed the **e**lectrophorogram of DNA extracted with the CTAB protocol, Plate 41 showed the **e**lectrophorogram of DNA extracted with the modified CTAB protocol, Plate 42 showed the **e**lectrophorogram of DNA extracted with the GNOME protocol, while the **e**lectrophorogram of DNA extraction done with Zymo protocol was showed in plate 43.

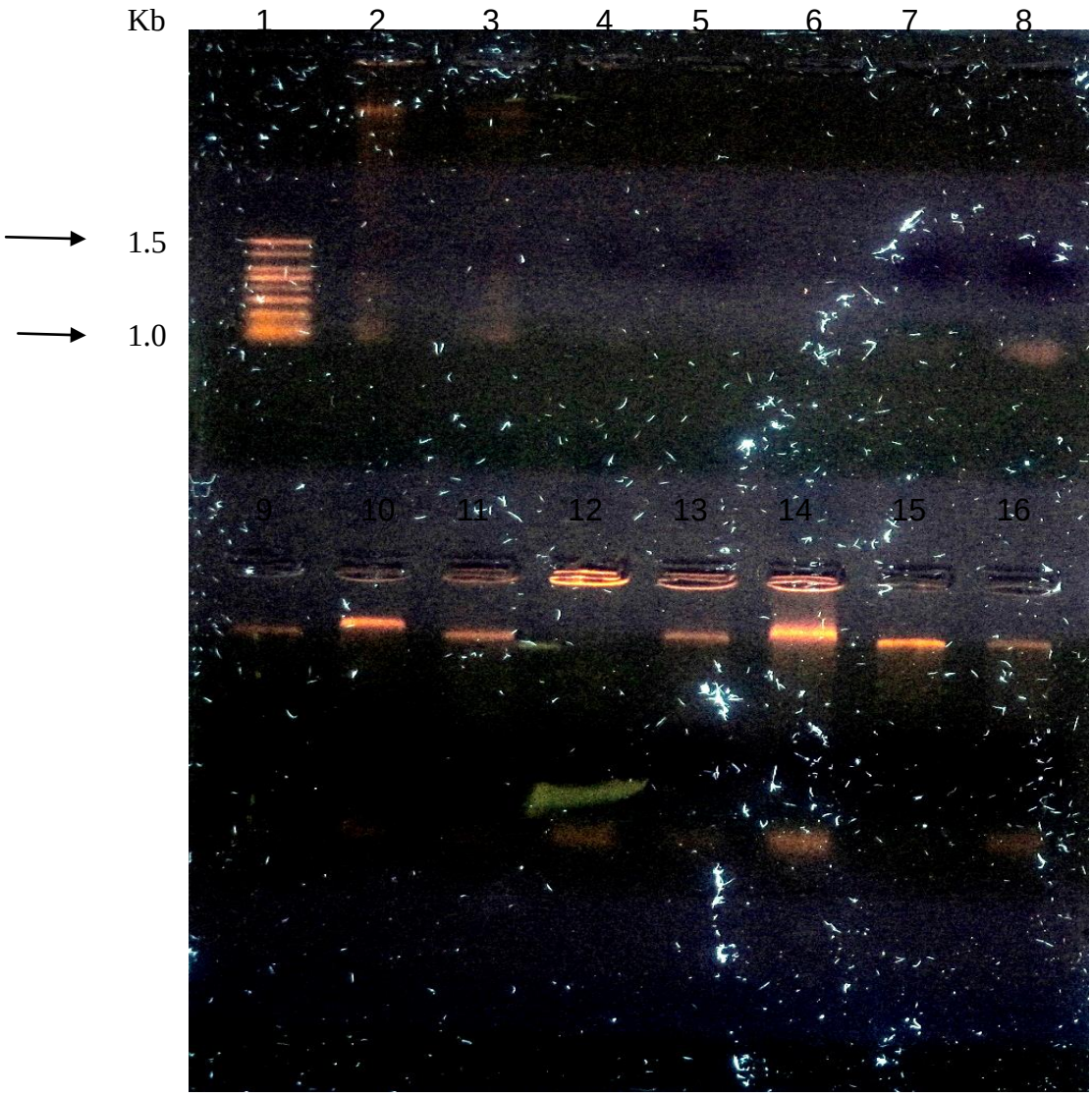


Plate 40: **Electrophorogram of extracted DNA samples using CTAB protocol.** The first lane contained the marker, sample number C123b, C117, L015, L030, B9, L033, C017. The second lane contained sample number C051b, C069b, C053b, C012, L002, C08, C077 and L032.

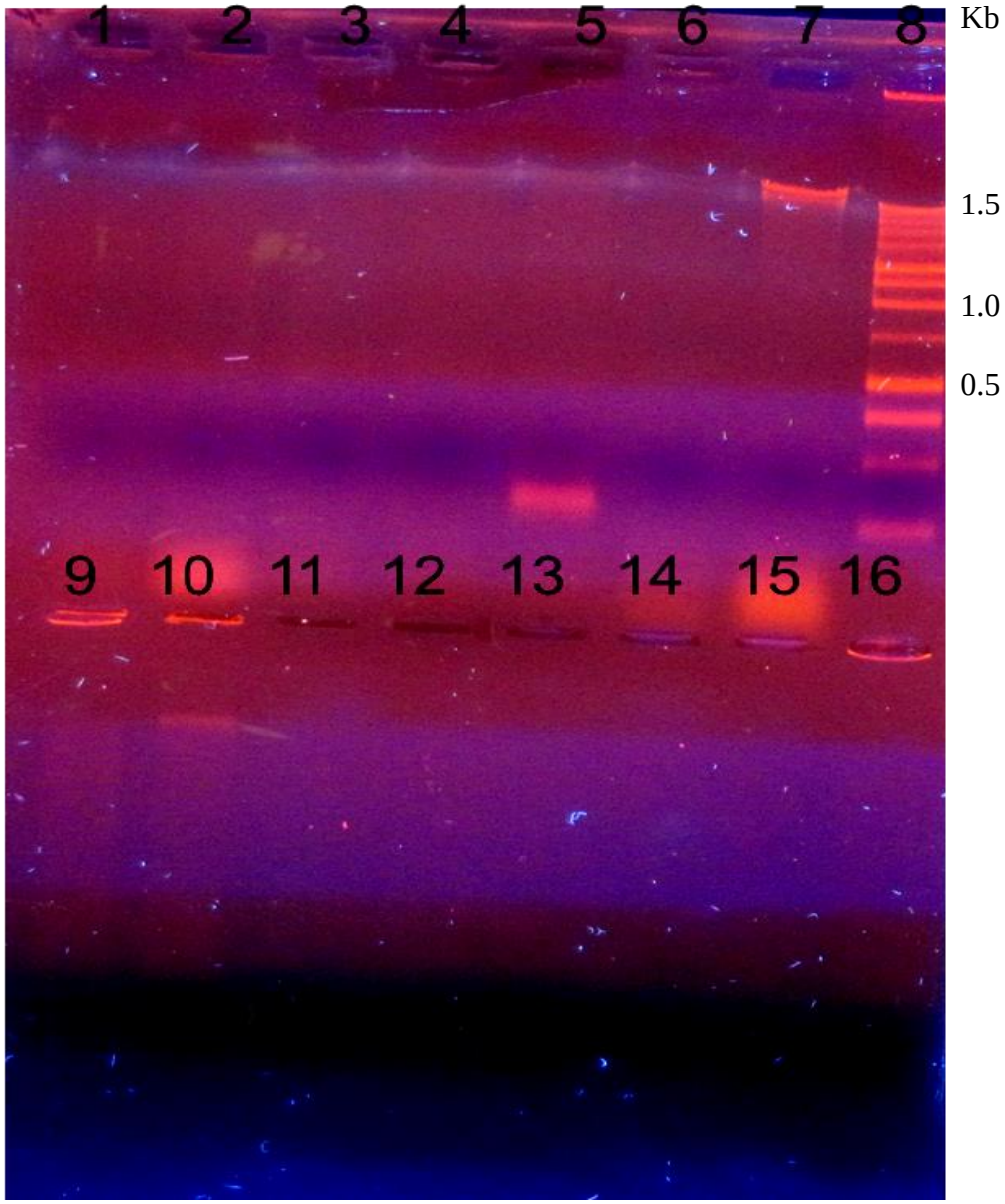


Plate 41: **Electrophorogram of extracted DNA samples using modified CTAB protocol.** The first lane contained sample number C038, C042, C052, C063, C065, L042, C122 and the marker. The second lane contained sample number A7b, L035, L012, C013, L068, C137, C140 and C016.

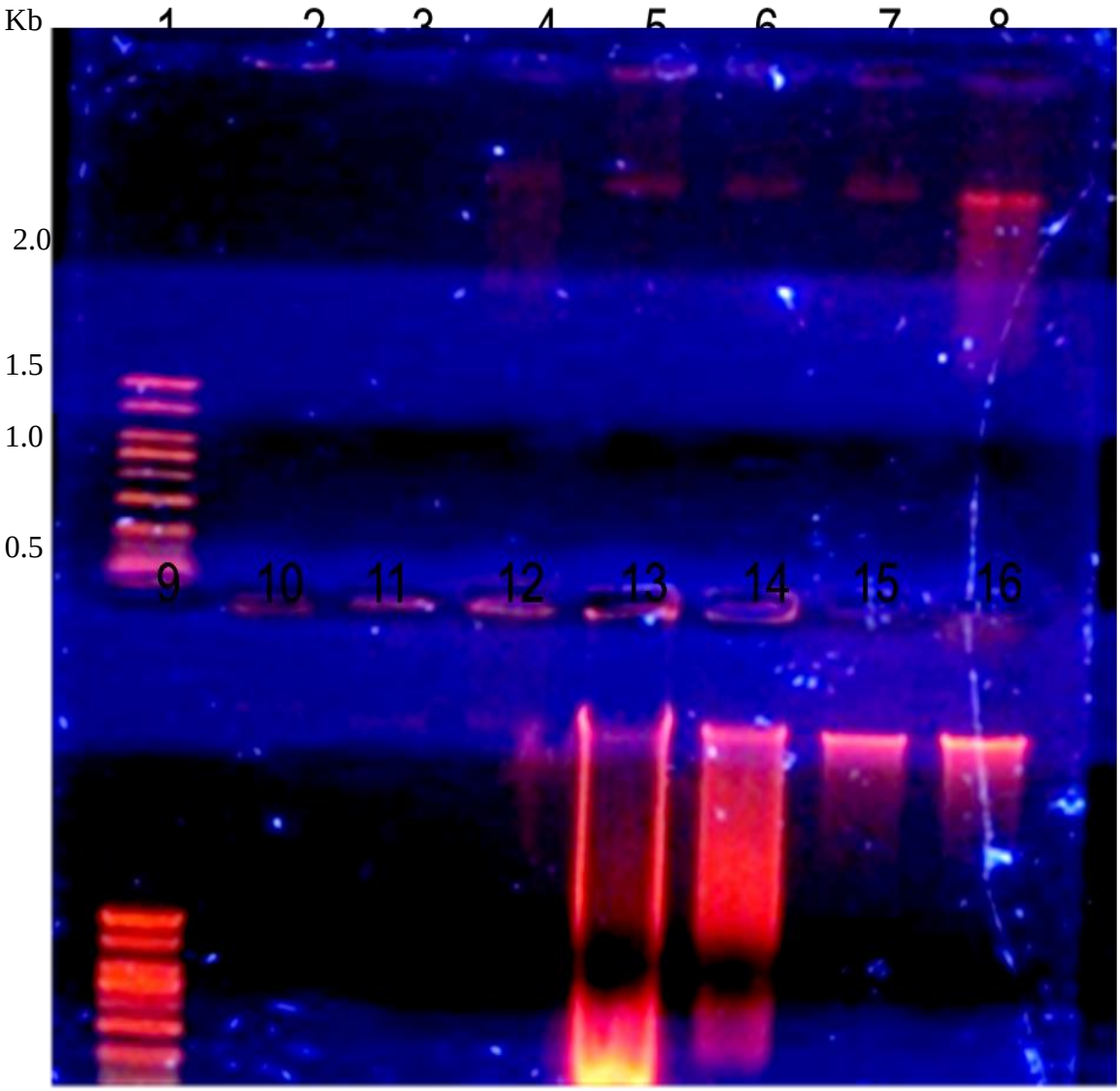


Plate 42: **Electrophorogram of extracted DNA samples using GNOME kit.** The first lane contained the marker, sample number C070, C088, C08, L002, L019a, L019b, C140. The second lane contained the marker, sample number C075, C138, C112, L025b, C99b, C077b and L066b.

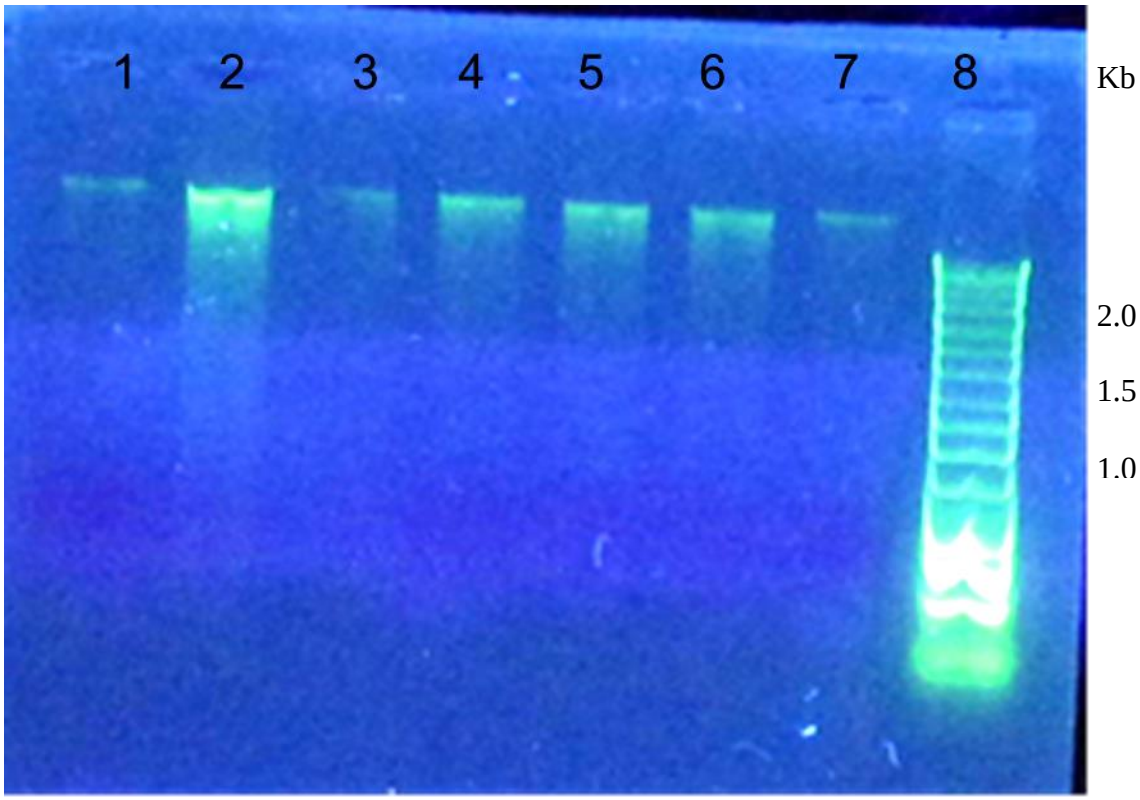


Plate 43: **Electrophorogram of extracted DNA samples using ZYMO kit.** The lane labelled 1,2,3,4,5,6,7,8 contained sample number L012a, C081a, L016a, B9a, C101a, C129a,C003a and the Marker respectively.

Below is the generated sequence data from the isolated fungi and the percentage of their significance of matches with the sequences database at the Genbank.

110719-03_C07_TOA-ITS1

AACATGCGTGAGATTGTAAGTCTAGCTTTCCCCCTGTCCATCACTTCCCCT
CTATTTGCTTGCACCATTAGTATCACTGGCTCTCACCAGGCATGGGAAGGC
TGTGGCACAGCTGCACCGTGGGCTCCCGACTATGTGGAACTCAGATGTGC
CTGCGCTGGTATCCAAACGGAATGGACTACCGGCTGCGCGGGCCCAAGGA
CATTACGTGGGTGGAGATCTCTGGCTGCTCATTCCTCATTGTTAAGACCAC
GGCGGGCGGCATCAGCATTGAGGAGGCCGTATCCATGATCACTAAAATTG
TCCTGGATGTCCGACCCACGGACAAAGTTTGGGACATGTGCGCTGCGCCT
GGTTCAAAGACAGCCTAGCTCAATGAGGCTCTGCACGCTGCTCCCGACGA
ACACCAGATTCTTCCCGGCTCTGCT
This sequences data was 98% significant matched with *Trichopyhton concentrum*

110719-09_O05_TOB-ITS4

AACATGCGTGAGATTGTAAGTCTAGCTTTCCCCCTGTCCATCACTTCCCCT
CTATTTGTCAACATGGAGTAACGTGATCTTAATAAACTTATAACACAGCTT
TGAACAACGGAACTCTTGGTTCTAATCCATCAAAAACGTAGCATAGCGCC
ATATGTAATGCGAGGTGCTATTCTGTGAATCTTCCTGTCTTTCAACGAAAC
TCGCGCTCAAAGGTATTCCTTTGTACACGTTGGTTTATTAATTATCAACAC
CCCTGCCACATAATTTTGACGGAAATGGAAATGAGNGTTTCGGATGAGAT
ACCAATTCTATACCTGACTTTCGGCTCAGTTAACGTTCACCTAGGATCTCC
TTAGGACCCAAACATTCCTCA
This sequences data was 96% significant matched with *Trichophyton tonsurans*

110719-03_A07_TOC -ITS1

AACATGCGTGAGATTGTAAGTCTAGCTTTCCCCCTGTCCATCACTTCCCCT
CTATTTGGATAATGTTGCCCACTTAAGCGTCCGCTTCACCAGCAGACGATT
TGATCCTTCCTCATACTTTATGGTATCATGTTTGCGGAGTTCTCGCCCCGCT
ATCGGACAAAGAGCACGGTCCCTGCTTTTGGGCCATATGCTGGCCGACGG
GAAGTGCCTATAATGCTCCACCTACACCGCATGGACGACCACGTTGATTTT
CTTTGGCAAATTCCGAAATTCGAACACCGCTATAAGACATCTGTGATGTG
GTAGTAACCTGTTTAAATCACTACATTGGTTGCTGCATTTGCCCATACCGC
TCATGTGTGGGATAGTGCACCGGGGCTCTGGGGCATTATGGGGGCTTCCC
TGCCGTACCAAGTAAGAC
This sequences data was 99% significant matched with *Trichophyton mentagrophytes var. quinckeanum*

110719-09_I05_TOD-ITS4

GGACGAGGGCCCAACACACAAGCCGGGCTTGAGGGCAGCAATGACGCTC
GGACAGGCATGCCCCCCGGAATACCAGGGGGCGCAATGTGCGTTCAAAG
ACTCGATGATTCACTGAATTCTGCAATTCACATTAGTTATCGCATTTCGCT
GCGTTCTTCATCGATGCCGGAACCAAGAGATCCATTGTTGAAAGTTTTAAC
TGATTGCAAAGAATCACACTCAGACTGCAAGCTTTCAGAACAGGGTTCAT
GTTGGGGTCTCCGGCGGGCACGGGCCCGGGGGGCGAGTCGCCCCCCGGCGG
CCAGCAACGCTGGCGGGCCCGCCGAAGCAACAAGGTACAATAGTCACGG
GTGGGAGGTTGGGCCATAAAGACCCGCACTCGGTAATGATCCTTCCGCA
This sequences data was 99% significant matched with *Aspergillus terreus*

110719-03_K05_TOE-ITS1

TGGCCACTTGAACACAACTTGTTGGACCACGCTTACATGCATGTGGGTTCA
TTTTTCGAACCGACGCCGCCTTCTAACTCCCAAATTGTGTGAGGGATATCG
CGTCATTATCCCCTTATTGGCCGGCCAACGCCTCTTCTGTTCGCCCGGAGG
TCTAACATCTTACAGGCCCATTGCTGACCACTTAGCCAATTGATGGGCCCC
CCCAAAGGTGAAAGGAAACTGTAAGACAACAGACATGGTTAATTTCCCCG
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GAATGCGCGGTAGCACCGGGACATTCTTCCGATTCGGCTGATTGTCGCAC
AATTTCGGTCCGCCTTACCTATCCTAATTATCACCCCGTCCGCATTCTGGG
TGCCGTTAAAACCCNAACAAGACCGTGGCTGGTTACGCGAATGTGGACAT
AGTCAGCCATGAGGTGTTGAGCCCCGTCTCGTACA
This sequences data was 99% significant matched with *Blastomyces dermatitidis*

110719-03_I05_TOF-ITS4

AGTCTGAGACCTATAACGAAATTAGTTAAAACTTTCAACAACGGATCTCT
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CAGCGGCGGCACCGCGTCCGGTCCTCGAGCGTATGGGGCTTCGTCACCCG
CTCTAGTAGGCCCGGCCGGCGCCAGCCGACCCCCAACCTTTAATTATCTCA
GGTTGACCTCGGATCAGGTAGGGATACCCGCTGAACTTAAGCATATCAAT
AAACGGAGGAA
This sequences data was 95% significant matched with *Trichophyton tonsurans*

110719-03_E07_TOG-ITS4

CACTGAACTTCATGCCTGAGAGTGATGCAGTCTGAGCCTGAATACAAATC
AGTCAAAACTTTCAACAATGGATCTCTTGGTTCCGGCATCGATGAAGAAC
GCAGCGAACTGCGATAAGTAATGTGAATTGCAGAATTCAGTGAATCATCG
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CGAGCGTCATTGCTGCCCTCAAGCCCGGCTTGTGTGTTGGGTCGTCGTCCC
CCCCGGGGGACGGGCCCGAAAGGCAGCGGCGGCACCGTGTCCGGTCCTC
GAGCGTATGGGGCTTTGTCACCCGCTCGATTAGGGCCGGCCGGGCGCCAG
CCGGCGTCTCCAACCTTATTTTTCTCAGGTTGACCTCGGATCAGGTAGGGA
TACCCGCTGAACTTAAGCATATCAATAAGCGGGGGAAGTTGCCCTCTTGG
GATATCGGTAGGGGTGCACCGANTGCCGGTTCGATCAGCACACGAAAGTC
This sequences data was 97% significant matched with *Exophiala dermatitidis*

110719-09_A07_TOH-ITS

CGGCTGGCGCCCGGCCGGCCCTAATCGAGCGGGTGACAAAGCCCCATACG
CTCGAGGACCGGACACGGTGCCGCCGCTGCCTTTCGGGCCCGTCCCCCGG
GGGGGACGACGACCCAACACACAAGCCGGGCTTGAGGGCAGCAATGACG
CTCGGACAGGCATGCCCCCCGGAATGCCAGGGGGCGCAATGTGCGTTCAA
AGACTCGATGATTCACTGAATTCTGCAATTCACATTACTTATCGCAGTTCG
CTGCGTTCTTCATCGATGCCGGAACCAAGAGATCCATTGTTGAAAGTTTTG
ACTGATTTGTATTCAGGCTCAGACTGCATCACTCTCAGGCATGAAGTTCAT
GGTCCCCCGGCGGCTCGCCCCTAGGGGGCTCCCCACCGAAGCAACAGTGT
TAGGTAGTCACGGGTGGGAGGTTGGGCGCCCGGAGGCAGCCCGCACTCG
GTAATGATCCTTCCGCAGGTCCCTACGG
This sequences data was 98% significant matched with *Microsporum audouinii*

110719-03_G07_TOI-ITS1

ACCACTGAACTTCATGCCTGAGAGTGATGCAGTCTGAGCCTGAATACAAA
TCAGTCAAAACTTTCAACAATGGATCTCTTGGTTCCGGCATCGATGAAGA
ACGCAGCGAACTGCGATAAGTAATGTGAATTGCAGAATTCAGTGAATCAT
CGAGTCTTTGAACGCACATTGCGCCCCCTGGCATTCCGGGGGGCATGCCT
GTCCGAGCGTCATTGCTGCCCTCAAGCCCGGCTTGTGTGTTGGGTCGTCGT
CCCCCCCGGGGGACGGGCCCGAAAGGCAGCGGCGGCACCGTGTCCGGTC
CTCGAGCGTATGGGGCTTTGTCACCCGCTCGATTAGGGCCGGCCGGGCGC
CAGCCGGCGTCTCCAACCT
This sequences data was 97% significant matched with *Penicillium citrinum*

110719-09_C07_TOJ-ITS4

AACATGCGTGAGATTGTAAGTCTAGCTTTCCCCCTGTCCATCACTTCCCCT
CTATTTGGATTCACTGAATTCTGCAATTCACATTACTTATCGCAGTTCGCT
GCGTTCTTCATCGATGCCGGAACCAAGAGATCCATTGTTGAAAGTTTTGAC
TGATTTGTATTCAGGCTCAGACTGCATCACTCTCAGGCATGAAGTTCAGTG
GTCCCCCGGCGGCTCGCCCCTAGGGGGCTCCCCGCCGAAGCAACAGTGTT
AGGTAGTCACGGGTGGGAGGTTGGGCGCCCGGAGGCAGCCCGCACTCGG
TAATGATCCTTCCGCAGGTTCACCTACGGAATCATTACCGAGTGCGGGCT
GCCTCCTGGCGCCCAACCTCCACCNGGACTACTNAATNTGTTTGCTTCNGN
GGGAGCCCCCTAGGAGCGAGCCGCCCGGGACCACTGAACTTCACCTGAAG
TGATGCCTGAGCCTGAATACATCCAAAACTTTCACAATGGATCTTNGTTCG
CCATAAAAAGCACAGCGATATAATGTGAATTGCAAAATCAAATCATCGAG
TCTTGAACGCAATTG

This sequences data was 98% significant matched with *Trichophyton rubrum*

110719-03_I07_TOK-ITS1

AACATGCGTGAGATTGTAAGTCTAGCTTTCCCCCTGTCCATCACTTCCCCT
CTATTTGCTTTGTAATCAGTTAAAACTTTCAACAATGGATCTCTTGGTTCC
GGCATCGATGAAGAACGCAGCGAAATGCGATAACTAATGTGAATTGCAG
AATTCAGTGAATCATCGAGTCTTTGAACGCACATTGCGCCCCCTGGTATTC
CGGGGGGCATGCCTGTCCGAGCGTCATTACTGCCCTCAAGCCCGGCTTGT
ATTGGGTCCTCGTCCCCCCGGGGACGGGCCCGAAAGGCAGCGGCGGCACC
GCGTCCGGTCCTCGAGCGTATGGGGCTTTGTCACCCGCTCTGTAGGCCCGG
CCGGCGCCAGCCCACGCAGATCATCCTTTTTTTCAGGTTGACCTCGGATCA
GGTAGGGATACCCGCTGAACTTAAGCATATCAATAAGCGGAGGA

This sequences data was 99% significant matched with *Trichophyton soudanense*

110719-03_K07_TOL-ITS1

GCACTCCCGAGCGTGATTGCTCGGTTTCTTTTACTCTACAATATGGCCGTA
TAATGCACCACTTGGTGGAATTGGCTAGAATCTAGACCTTATACATGTTGG
TAACCCGGATGCCGACTTCTTCCTCCAGATGGTTACAATGATTAGGTACCG
AATTTTGGATGTAGGCCGGCACGCCTATGCCTTGCCTCGTTCGTGAAACAT
CTTGAATCCGCATTGCCGCGCACTAAGGAGGGCTGGGGGCCCCCCCTAAG
ATGAAAGGAAACTATACCATCTGCCAGAGGATACTAACCCCTAAGGAACC
CCATAGGTTGCCGCGCTCGTGGTTTCCCTCTGGCTGCGAGAGTGGACGGC
CGCTCCGGGGCATTATTGGGGGTTCGGTTCATAACCACAATTCACTCCTTA
GGCTTAGCCTGCTGGCGGCCCCGAAGCATTGTAGGNCTTGTTGCATTCCTG
GGCAATCGGATCATGCAGAATGCGGGCACTATCAGCATTAGTGTTTCTGA
TTTG

This sequences data was 98% significant matched with *Microporum ferrugineum*

110719-03_M07_TOM-ITS1

CTCCGTCAGGCTTTCTTCTCCAGTTTTTGACAGGGATTTCTTGTCGTTATAC
CTATTTCCCTACAATTACCTCCTGCCTTCGATTGCTGGTGTAACATCCAAC
AGGCTCATGCCCACCTCCTTTCCCAACTTTTGCGCCACTCGAATGGTCAAT
GGAAACTGTAAGCNATCAGACTCAGTAAACATTCCACTAAAATATCTTCA
AATCAGGCCACTCTTGAACNCCCAGGTCTTAGTTTGGTAAAGTNTCCTGCC
TCCCGGCATCTTCATTTACGGTTCAGTGTCATTCCCTTCGGACCATTAGTC
CCATTAAAGGGGTGGACTATTCATTCAAAGGGTGAAAAACCTCTATCATG
GCGATGGCTCACTACGTGAACCATCAGTATAATTAACTTTTTTGGGNTCCA
GGTGCCGTAAAGTTCAAAT

This sequences data was 99% significant matched with *Penicillium aschersonia*

110719-09_G07_TON-ITS4

CAGTTCACCTGCGAAAATTATTTTTTGTGCTGACTGCCGGCTTTTTCAGGG
AGCGGGGAATGAAACCAAGAATCTCCAGGACAGGACGCGGTGCCTCCGC
TGCCTTTCGGGCCGCCCCGCCTGGGATAGCCCGGGGCCCATCCCTCAAGA
CGTCATTGAGGGCTTGTAACATGAGCTTAATAAACATATAAAACAACTCT
AAACGGAAGAATTCTGGTTAAATCCTCATAAAAAACGTANCTCAGCGCCC
TATGTTTTGCGAGGGGCATATCCTGTTNTCATCGATTCTTTCACCAAAACT
TGCGTTGAGGTATTCCATTGAATACNCCTGTTTGAAATCAAAAACGCCCC
AGCCATATTTTCTTGAGGTGAATGGGAATGAGAGTTTCGGCGTTATGGCTG
GGTCTTTAAATTTCTTTCGGCTGAGTTATTATTCACCTTGGCCCAGTTGGA
AACCCGATGAACTAAGCTCTAGTGAAAAACAAAAGGTGCCTCCCAAAGA
AAAGATCCTA

This sequences data was 99% significant matched with *Pencillium* species

110719-03_O07_TOO-ITS1.

ACTGTTGCTTCGGCGGGGAACCCCCTCGGGGGCGAGCCGCCGGGGACTAC
TGAACTTCATGCCTGAGAGTGATGCAGTCTGAGTCTGAATATAAAATCAG
TCAAAACTTTCAACAATGGATCTCTTGGTTCCGGCATCGATGAAGAACGC
AGCGAACTGCGATAAGTAATGTGAATTGCAGAATTCAGTGAATCATCGAG
TCTTTGAACGCACATTGCGCCCCCTGGCATTCCGGGGGGCATGCCTGTCCG
AGCGTCATTGCTGCCCATCAAGCCCGGCTTGTGTGTTGGGTCGTCGTCCCC
CCCGGGGGACGGGCCCGAAAGGCAGCGGCGGCACCGTGTCCGGTCCTCG
AGCGTATGGGGCTTTGTCACCCGCTCGACTAGGGCCGGCCGGGCGCCAGC
CGACGTCTCCAACCATTTTTCTTCAGGTTGACCTCGGATCAGGTAGGGATA
CCCGCTGAACTTAAGCATATCAATAAGCGGAGGAAACTCGNNGGCGGTG
GCTCACGGCGGGCCAGCCCCCTCCTCAAATCAGAACAAATCAGAGGTTGC
GTCGGGTACA

This sequences data was significant matched with none *(*Unknown O)

110719-09_K07_TOP-ITS4

GGCCCTAGTCGAGCGGGTGACAAAGCCCCATACGCTCGAGGACCGGACA
CGGTGCCGCCGCTGCCTTTCGGGCCCGTCCCCCGGGGGGGACGACGACCC
AACACACAAGCCGGGCTTGATGGGCAGCAATGACGCTCGGACAGGCATG
CCCCCCGGAATGCCAGGGGGCGCAATGTGCGTTCAAAGACTCGATGATTC
ACTGAATTCTGCAATTCACATTACTTATCGCAGTTCGCTGCGTTCTTCATC
GATGCCGGAACCAAGAGATCCATTGTTGAAAGTTTTGACTGATTTTATATT
CAGACTCAGACTGCATCACTCTCAGGCATGAAGTTCAGTAGTCCCCGGCG
GCTCGCCCCCGAGGGGGT

This sequences data was 99% significant matched with *Epidermaphyton fluccosum*

110719-03_A09_TOQ-ITS1

ACTTCATGCCTGAGAGTGATGCAGGCTGGCTCTGAATCTAAAGCAAAACT
TTCTTCCTGAATCTTTTGCTTCCCGCCTCCATTAATCGCACCAAACTGAAG
AAGGTGCCGTCGCGAATGTACCGGACNGATCCTGGACAACCCAAACCACG
GGTAGATCCCCGTTNCGACATTGGTTCATCTGGTTGATGAATCATGGGCAT
TGGGGTCCGCGGACTGCCCCTGGCAGAACAAAAAGTCAGCATGTGGATGG
GTTTCTATCATTGAGAAATAATGGGTTACATGCCATGCCACTGTATGCTCG
TCTGCGACAGATAGGGCCTTGGCTTCTGTGAGCATCCCATGGGGCCCCCCT
TGGATCAAGGGACTGGCCTGATTTTACATCACTGAGTGGGAAGCAATAAT
CAGGACTGTCANAGTATTCGCTATGTCTATCTGATTCNNTGTGCAGGGGAT
GGTGAGCATTGATGCCCGCTATGTGCCTCACAGTCGCGCCCTTCATTACCC
ATCTCAACCCGCAACCTCTCCTGAG

This sequences data was 98% significant matched with *Mucor racemosus*

110719-03_C09_TOR-ITS1

AACATGCGTGAGATTGTAAGTCTAGCTTTCCCCCTGTCCATCACTTCCCCT
CTATTTGTGGTAGATTGCGACGTGCGGCGAAAACATTAAGTAATCAACGG
GGGGGTATATTTATGATGAAAAACCAACAAAGNTTGATTGCAAGACCCCA
TCCTCCTTGGCTCAACTAAATAATTGGCATCCTGGAGCATTAGGCGGGGTT
CCGNTTCAATGCACACTCTTCATCATATTTNGAATCTCTTAAAACGGGAGA
GCAAATTGTTACCAGTAATGNAATATTGGAATAAGAGATGACTCACGAAC
ATCTCGCCGGATATCGTTAACCTTGATTCCCTGAGAGTGATAAAGATACG
GACCNTGCATGCCTCTGCCCAACCCCCACTCCNCCATGACCATCATCTTCT
CCAATTTGTCTGTGCGTGACTAGCCCTCGCCCAAGCACAGCTTCACGCTCG
TTCGACTAGGAGCCCTTCACAACCTCGTCTTGATCCTGATGATAAGCGCAA
CTAAAGC

This sequences data was 95% significant matched with *Trichophyton concentricum*

110719-09_O07_TOS-ITS4

TAACCATGTGGATGGGCAACGAACATGTTTACCGGCCTGATAATTTTGCTT
AAGCCTGGTATCCTATTAAAACTCTACCTTCAGATCTCAACAAAATAATTG
TAACGGGGTGTTAATAAACTTGTAAAACAACTTTTAACTGGGATCTCTTGG
TTCTCGGTTGNTGAAAAACGTCGATGATGCGATAACTATTGTGAATTGCAT
ATTTATCGCATCATCCTGTCTTCTTCATCNACTCGCGAACCAAGGGATTCC
TTGGTACACGCTGGTTTCTAAATCTTAAACACCCCTGTTACATTTATTTTAC
GGAAATGGAAATGGTAANTTCGGATAGGAGNTGAATTCTTTACCTGACTT
TCGGCTGAGTTAACATTCTCTTAGCCTATCCCTATGGC

This sequences data was 96% significant matched with *Absidia corymbifera*

110719-09_K05_TOU-ITS4

ATGCGTCGGCGGGCGCCGGCCGGGCCTACGGAGCGGAAGACGAAGCCCC
ATACGCTCGAGGACCGGACGCGGTGCCGCCGCTGCCTTTCGGGCCCGTCC
CCCGGGAGCCGGGGGACGAGGGCCCAACACACAAGCCGGGCTTGAGGGC
AGCAATGACGCTCGGACAGGCATGCCCCCCGGAATACCAGGGGGCGCAA
TGTGCGTTCAAAGACTCGATGATTCACTGAATTCTGCAATTCACATTAGTT
ATCGCATTTCGCTGCGTTCTTCATCGATGCCGGAACCAAGAGATCCATTGT
TGAAAGTTTTAACTGATTGCAAAGAATCACACTCAGACTGCAAGCTTTCA
GAACAGGGTTCATGTTGGGGTCTCCGGCGGGCACGGGCCCGGGGGCGAGT
CGCCCCCCGGCGGCCAGCAA

This sequences data was 98% significant matched with *Microsporum nanum*

110719-09_I05_TOW-ITS4

AACATGCGTGAGATTGTAAGTCTAGCTTTCCCCCTGTCCATCACTTCCCCT
CTATTTGCCCCCGGGAGCCGGGGGACGAGGGCCCAACACACAAGCCGGG
CTTGAGGGCAGCAATGACGCTCGGACAGGCATGCCCCCCGGAATACCAGG
GGGCGCAATGTGCGTTCAAAGACTCGATGATTCACTGAATTCTGCAATTC
ACATTAGTTATCGCATTTCGCTGCGTTCTTCATCGATGCCGGAACCAAGAG
ATCCATTGTTGAAAGTTTTAACTGATTGCAAAGAATCACACTCAGACTGC
AAGCTTTCAGAACAGGGTTCATGTTGGGGTCTCCGGCGGGCACGGGCCCG
GGGGCGAGTCGCCCCCCGGCGGCCAGCAACGCTGGCGGGCCCGCCGAAG
CAACAAGGTACAATAGTCACGGGTGGGAGGTTGGGCCATAA

This sequences data was 99% significant matched with *Trichophyton violaceum*

110719-03_E05_TOY-ITS1

ACGGATCTCTTGGTTCCGGCATCGATGAAGAACGCAGCGAAATGCGATAA
CTAATGTGAATTGCAGAATTCAGTGAATCATCGAGTCTTTGAACGCACATT
GCGCCCTCTGGTATTCCGGAGGGCATGCCTGTCCGAGCGTCATTGCTGCCC
TCAAGCCCGGCTTGTGTGTTGGGCCCCGTCCCCCCCGCCGGGGGGACGGG
CCCGAAAGGCAGCGGCGGCACCGCGTCCGGTCCTCGAGCGTATGGGGCTT
CGTCACCCGCTCTAGTAGGCCCGGCCGGCGCCAGCCGACCCCCAACCTTT
AATTATCTCAGGTTGACCTCGGATCAGGTAGGGATACCCGCTGAACTTAA
GCATATCAATAAACGGAGGAACATTCNNGTGCCGTATCCTTGCTGGCCCC
CACTCTACTGGAGGAGCGAACAAAGNACAAAGTTAG

This sequences data was 99% significant matched with *Emericella nidulans*

The above result of the blasted DNA sequence database of the isolated dermatophytes revealed that no sequence data was 100% homologous with those in the Genbank judging from their percentage of significance of matches with the sequences database at the Genbank. This simply implies that the above listed isolates are different strains of the matched fungi species at the Genbank (table 4 below show the checklist of all isolated fungi from the study).

Table 4: CHECKLIST OF ALL ISOLATED FUNGI

S/N	DERMATOPHYTES	SYSTEMIC MYCOSES FUNGI	OPPORTUNISTIC MYCOSES FUNGI	UNIDENTIFIED FUNGI SPECIES
1	*Epidermophyton floccosum*	*Blastomyces dermatitidis*	*Absidia corymbifera*	Unidentified species O
2	*Microsporum audouinii*	*Exophiala dermatitidis*	*Aspergillus terreus*	Unidentified species T
3	*Microsporum ferrugineum*		*Emericella nidulans*	Unidentified species V
4	*Microsporum nanum*		*Mucor racemosus*	Unidentified species X
5	*Trichophyton concentricum* strain A		*Pencillium aschersonia*	
6	*Trichophyton concentricum* strain B		*Pencillium citrinum*	
7	*Trichophyton mentagrophytes var. quinckeanun*		*Pencillium* species	
8	*Trichophyton rubrum*			
9	*Trichophyton soudanense*			
10	*Trichophyton tonsurans* strain A			
11	*Trichophyton tonsurans* strain B			
12	*Trichophyton violaceum*			

4.4 SENSITIVITY TEST AGAINST DIFFERENT ANTIFUNGAL DRUGS AND THE EXTRACTED OIL.

All isolates of dermatophytes and opportunistic fungal tested produced detectable growth at time points ranging from 2 to 15 days. The range of mean inhibition zone for the disk test of diffusion test of eight antifungal agents and three extracted oil are summarized in Table 5. When all the strains were considered together, flucamed (Fluconazole) was apparently the least effective, while Nystatin (Polyene) possessed the highest antifungal activity against all isolated fungi.

"Flucamed" belongs to the fluconazole "family"of the class azole, it was able to inhibit the growth of *Trichophyton mentagrophytes var. quinckeanum* (Table 6) with zone of inhibition 12.50±0.62 mm, *Blastomyces dermatitidis* with 19.50±1.59 mm zone of inhibition, *Trichophyton tonsurans* strain B with 13.00±1.00 mm mean zone of inhibition, *Microsporum audouinii* with 12.75±1.50 mm zone of inhibition (Table 7), *Trichophyton soudanense* with 13.75±1.14mm zone of inhibition, *Microsporum ferrugineum* with 12.37±0.70 mm zone of inhibition (Table 8), 13.12±0.78 mm zone of inhibition against *Trichophyton concentrium* strain B, 20.25±0.95 mm zone of inhibition against *Absidia corymbifera* (Table 10) and 14.00±0.62 mm zone of inhibition Unidentified V (Table 11).

"Diflucan", another member of the fluconazole in the azole class, also inhibit a number of isolated dermatophytes and other human pathogenic fungi. This include; *Trichophyton concentricum* strain A and B with zone of inhibition of 21.75±0.95 mm and 15.75±1.03 mm respectively (Tables 6&10). *Trichophyton tonsurans* strain A and B with 13.50±0.8 mm and 18.50±1.54mm zone of inhibition respectively (Tables

Table 5: Summarized table on sensitivity activities of the isolated fungi against eight antifungal agents and three extracted seed oils.

Fungi	Flucamed	Diflucan	Itranox	Ketoconazole	Griseofulvin	Nizoral	Sporanox	Nystatin	*Azadirachta indica*'oil	*Treculia africana*'oil	*Detarium senegalense*'oil	Distilled water
A.terreus	-	+	+	-	+	-	-	+	-	-	-	-
Absidia corymbifera	+	+	-	-	+	+	-	+	-	-	+	-
Blastomyces dermatitidis	+	+	-	-	-	-	+	+	-	-	-	-
E.floccosum	-	-	+	+	-	+	+	+	-	-	-	-
Emericella nidulans	-	-	-	-	+	-	+	+	+	-	+	-
Exophilia dermatitidis	-	-	+	-	-	-	+	+	-	-	-	-
M. nanum	-	-	-	-	+	-	-	+	-	-	-	-
M.audouinii	+	+	-	+	+	+	-	-	+	-	+	-
M.ferrugineum	+	+	-	+	+	+	-	+	-	-	+	-
Mucor racemosus	-	+	+	-	-	-	+	+	+	-	-	-
P.aschersonia	-	-	+	+	+	-	+	+	-	-	+	-
P.citrinum	-	+	+	+	-	+	+	+	+	-	-	-
*P.*species	-	-	+	+	-	+	-	+	-	-	-	-
T.concentricum A	-	+	-	-	+	+	+	+	+	-	+	-
T.concentricum B	+	+	-	+	+	+	+	+	-	-	+	-
T.ment.var.quinck	+	+	-	-	+	-	-	+	-	-	-	-
T.rubrum	-	-	+	-	+	+	+	+	-	-	-	-
T.soudanense	+	+	-	-	+	-	+	+	-	-	+	-
T.tonsurans. A	-	+	+	+	+	+	+	-	-	-	-	-
T.tonsurans. B	+	+	-	+	+	+	+	+	-	-	-	-
T.violaceum	-	-	+	+	+	-	+	+	-	-	-	-
Unknown O	-	-	-	+	-	+	-	+	-	-	+	-
Unknown T	-	-	-	-	+	-	-	+	-	-	-	-
Unknown V	+	-	+	+	-	-	+	-	-	-	-	-
Unknown X	-	-	+	-	+	+	+	+	-	-	+	-

–,resistance, +,susceptibility.

6 & 7). *Trichophyton mentagrophytes var. quinckeanum* with zone of inhibition of 19.75±0.70 mm, 12.00±0.65 mm zone of inhibition against *Aspergillus terreus* (Table 6). *Blastomyces dermatitidis* and *Microsporum audouinii* with the zone of inhibition of 13.75±1.16 mm and 14.75±0.99 mm respectively (Table 7), 18.80±2.06 mm mean of inhibition against *Penicillium citrium* and *Trichophyton soudanense* with 16.00± 0.92mm. *Microsporum ferrugineum* and *Mucor racemosus* with zone of inhibition 13.50± 1.05 mm, 14.62±1.17 mm respectively, and *Absidia corymbifera* with zone of inhibition 18.50± 1.63 mm (Tables 8 & 10).

"Itranox" belongs to the itraconazole group which is another member of the class azole. This antifungal drug also demonstrates activities against a quite number of isolated dermatophytes and other human pathogenic fungi. It inhibits *Trichophyton tonsurans* strain A and *Trichophyton rubrum* with mean of 12.12±0.51 mm, 13.50±0.90 mm respectively (Tables 6 & 8), 14.87± 1.12 mm against *Exophiala dermatitidis* and *Penicillium citrium* with 17.75±1.97 mm mean (Tables 7 & 8). Also with *Aspergillus terreus* and *Penicillium aschersonia* (Tables 6a & d), it showed zone of inhibition of 12.00±0.53 mm, 18.00±1.41 mm mean respectively. With *Epidermophyton floccosum*, 17.75±0.88 mm mean; 17.75±0.88 mm with *Penicillium* species and *Absidia corymbifera* with 14.12±0.85 mm mean. While it showed 17.50±0.90 mm and 16.50±0.90 mm of inhibition zone against *Trichophyton violaceum* and Unidentified X (Tables 9, 10 & 11).

"Ketoconazole", the oldest of the class azole also showed inhibition zone against quite a number of the isolated fungi. This include; *Trichophyton tonsurans* strain A and B with 18.87±2.08 mm and 17.00±1.55 mm respectively (Tables 6 & 7),

Table 6: Sensitivity test on eight antifungal agents, three extracted oils against *Trichophyton concentricum* strain A, *T. tonsurans* strain A, *T. mentagrophyton var. quinckeanum* and *Aspergillus terreus*.

Antifungal agents & Extracted seed oils	*Mean inhibition zone diameter ± **SE (mm)			
	Trichophyton concentricum strain A	*T. tonsurans strain A*	*T. mentagrophyton var. quinckeanum*	*Aspergillus terreus*
Flucamed	*0.00±0.00^{A}	0.00±0.00^{A}	12.50±0.62^{B}	0.00±0.00^{A}
Diflucan	21.75±0.95^{E}	13.50±0.80^{B}	19.75±0.70^{D}	12.00±0.65^{B}
Itranox	10.00±0.00^{A}	12.12±0.51^{A}	0.00±0.00^{A}	12.00±0.53^{B}
Ketoconazole	10.00±0.00^{A}	18.87±2.08^{B}	0.00±0.00^{A}	0.00±0.00^{A}
Griseofulvin	13.12±0.69^{CD}	27.50±1.11^{D}	15.37±1.34^{C}	12.50±0.73^{B}
Nizoral	13.62±0.84^{D}	25.12±1.18^{CD}	0.00±0.00^{A}	0.00±0.00^{A}
Sporanox	11.25±0.31^{AB}	23.25±2.47^{C}	0.00±0.00^{A}	0.00±0.00^{A}
Nystatin	11.37±0.59^{AB}	0.00±0.00^{A}	11.50±0.42^{A}	19.50±0.98^{C}
Azadirachta indica'oil	10.37±0.26^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Trecula africana'oil	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Detarium senegalense'oil	12.12±0.47^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Sterile distilled Water	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}

*Means (n = 3) with the same superscript letter in a column are not significantly different (p>0.05), while means with different superscript letter are significantly the same (p<0.05) when subjected to Duncan multiple range test, DMRT (ANOVA posthoc).
**SE = Standard error ***n = number of observations

12.50±0.70 mm *Trichophyton concentrium* strain B, *Microsporum audouinii* with 13.62±0.92 mm (Tables 7 & 10), *Penicillium citrinum* with 25.00±1.88 mm, *Penicillium aschersonia* with 14.12±0.91 mm, *Penicillium* species with 13.62±0.84mm and 13.62±0.84 mm mean of inhibition zone against *Epidermaphyton fluccosum* (Tables 8 & 9).

"Nystatin", a member of the class Polyene demonstrated the highest activities against the isolated fungi compared to other antifungal agent used in this study. It showed zone of inhibition against twenty-two species strains of the twenty-five isolated fungi in this study except in *Trichophyton tonsurans* strain B, *Microsporum audouinii* and Unidentified V.

"Griseofulvin" is a member of class Grisovid, also showed inhibition zone against quite a number of the isolated fungi and this include; *Trichophyton concentricum* strain A and B with zone of inhibition of 13.12±0.69 mm and 11.12±0.22 mm respectively (Tables 6 & 10), *Trichophyton tonsurans* strain A with 27.50±1.11 mm, *Trichophyton mentagrophytes var. quinckeanum* with 15.37±1.34 mm, *Aspergillus terreus* with 12.50±0.73 mm, *Trichophyton rubrum* with 14.00±0.92mm, 14.37±0.80 mm zone of inhibition against *Trichophyton soudanense*, *Microsporum ferrugineum* with zone of inhibition of 13.37±0.77 mm (Tables 6 & 8), *Absidia corymbifera* with 15.37±0.73 mm, 12.25±0.41 mm zone of inhibition of inhibition against *Trichophyton violaceum*, *Microsporum nanum* with mean of 14.25±0.70 mm (Tables 10 & 11), *Penicillium aschersonia* with 13.00±0.65mm, *Emericella nidulans* with 15.75±1.03 mm (Tables 9 & 12), Unidentified T and Unidentified X with 14.25±0.70 mm and 12.87±0.44 mm zone of inhibition respectively (Tables 10&11).

Table 7: Sensitivity test on eight antifungal agents, three extracted oils against *Blastomyces dermatitidis, Trichophyton tonsurans* strain B, *Exophiala dermatitidis and Microsporum audouinii*.

Antifungal agents & Extracted seed oils	*Mean inhibition zone diameter ± **SE (mm)			
	Blastomyces dermatitidis	*Trichophyton tonsurans* strain B	*Exophiala dermatitidis*	*Microsporum audouinii*
Flucamed	*19.50±1.59^{A}	13.00±1.00^{ABC}	0.00±0.00^{A}	12.75±1.50^{ABC}
Diflucan	13.75±1.16^{B}	18.50±1.54^{E}	0.00±0.00^{A}	14.75±0.99^{C}
Itranox	0.00±0.00^{A}	0.00±0.00^{A}	14.87±1.12^{B}	0.00±0.00^{A}
Ketoconazole	0.00±0.00^{A}	17.00±1.55^{DE}	0.00±0.00^{A}	13.62±0.92^{BC}
Griseofulvin	0.00±0.00^{A}	14.25±0.95^{BCD}	0.00±0.00^{A}	12.25±0.59^{ABC}
Nizoral	0.00±0.00^{A}	18.75±2.47^{E}	0.00±0.00^{A}	19.62±2.25^{D}
Sporanox	11.37±0.46^{A}	14.62±0.98^{CD}	18.75±2.16^{C}	0.00±0.00^{A}
Nystatin	11.50±0.18^{A}	11.12±0.29^{AB}	18.25±1.43^{C}	0.00±0.00^{A}
Azadirachta indica'oil	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	10.87±0.29^{A}
Trecula africana'oil	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Detarium senegalense'oil	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	11.62±0.46^{A}
Sterile distilled Water	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}

*Means (n = 3) with the same superscript letter in a column are not significantly different (p>0.05), while means with different superscript letter are significantly the same (p<0.05) when subjected to Duncan multiple range test, DMRT (ANOVA posthoc).
**SE = Standard error ***n = number of observations

"Nizoral" is another member of the ketoconazole compound in the class azole, which inhibited the growth of some isolated fungi in this study. It inhibits the growth of *Trichophyton concentricum* strain A and B with the zone of inhibition of 13.62±0.84 mm and 19.75±2.05 mm respectively (Tables 6 & 10), 25.12±1.18 mm and 18.75±2.47 mm zone of inhibition against *Trichophyton tonsurans* strain A and B (Tables 6 & 7). 19.62±2.25 mm zone of inhibition against *Microsporum audouinii*, it inhibit *Penicillium citrinum* 23.00±1.77 mm zone of inhibition (Tables 7 & 8). Against *Trichophyton rubrum* and *Microsporum ferrugineum*, it produced inhibition zone of 13.50±0.96 mm and 13.00±0.96 mm respectively (Table 8), while it is effective against *Penicillium* species with mean of 14.50±0.73 mm, Unidentified O with mean of 15.50±1.18 mm, *Epidermatophyton Fluccosum* with zone of inhibition of 13.75±0.86 mm (Table 9), *Absidia corymbifera* with mean of 16.00±1.13 mm and Unidentified X with 15.75±0.95 mm zone of inhibition (Tables 10 & 11).

"Sporanox" belongs to the the itraconazole group of the class azole, and this also showed inhibition zone against quite number of fungi in this study. This include; *Trichophyton concentricum* strain A and B, with 11.25±0.31 mm and 16.00±2.05 mm zone of inhibition respectively, 23.25±2.47 mm and 14.62±0.98 mm zone of inhibition against *Trichophyton tonsurans* strain A and B, respectively (Tables 7&10). *Blastomyces dermatitidis* with zone of inhibition 11.37±0.46 mm, *Exophiala dermatitidis* 18.75±2.16 mm (Table 7), *Penicillium citrinum* with zone of inhibition *22.25*±1.97 mm, *Trichophyton rubrum 19.87*±2.07 mm, *Trichophyton soudanense* with zone of inhibition of 15.37±1.05 mm (Table 8), *Penicillium aschersonia* with zone of inhibition 19.50±1.18 mm (Table 9),

Table 8: Sensitivity test on eight antifungal agents, three extracted oils against *Penicillium citrium, Penicillium citrium, T. soudanense, Microsporum ferrugineum*

Antifungal agents & Extracted seed oils	*Mean inhibition zone diameter ± **SE (mm)			
	Penicillium citrium	*Trichophyton rubrun*	*Trichophyton soudanense*	*Microsporum ferrugineum*
Flucamed	*0.00±0.00^{A}	0.00±0.00^{A}	13.75±1.14^{BC}	12.37±0.70^{B}
Diflucan	18.50±2.06^{B}	0.00±0.00^{A}	16.00±0.92^{D}	13.50±1.05^{B}
Itranox	17.75±1.97^{B}	13.50±0.90^{B}	0.00±0.00^{A}	0.00±0.00^{A}
Ketoconazole	25.00±1.88^{C}	0.00±0.00^{A}	0.00±0.00^{A}	12.00±0.62^{B}
Griseofulvin	10.00±0.00^{A}	14.00±0.92^{B}	14.37±0.80^{CD}	13.37±0.77^{B}
Nizoral	23.00±1.77^{C}	13.50±0.96^{B}	0.00±0.00^{A}	13.00±0.96^{B}
Sporanox	22.25±1.97^{C}	19.87±2.05^{C}	15.37±1.05^{CD}	0.00±0.00^{A}
Nystatin	30.50±0.92^{D}	12.50±2.07^{AB}	12.50±0.46^{B}	13.12±0.76^{B}
Azadirachta indica'oil	10.25±0.70^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Trecula africana'oil	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Detarium senegalense'oil	0.00±0.00^{A}	0.00±0.00^{A}	12.25±0.55^{B}	13.12±0.61^{B}
Sterile distilled Water	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}

*Means (n = 3) with the same superscript letter in a column are not significantly different (p>0.05), while means with different superscript letter are significantly the same (p<0.05) when subjected to Duncan multiple range test, DMRT (ANOVA posthoc).
**SE = Standard error ***n = number of observations

Table 9: Sensitivity test on eight antifungal agents, three extracted oils against *Penicillium aschersonia, P.* species, Unknown O and *Epidermophyton fluccosum*

Antifungal agents & Extracted seed oils	*Mean inhibition zone diameter ± **SE (mm)			
	Penicillium aschersonia	*Penicillium* species	Unidentified O	*Epidermophyton fluccosum*
Flucamed	*0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Diflucan	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Itranox	18.00±1.41^{C}	17.75±0.88^{C}	0.00±0.00^{A}	17.75±0.88^{C}
Ketoconazole	14.12±0.91^{B}	13.62±0.84^{B}	13.25±0.64^{B}	13.62±0.84^{B}
Griseofulvin	13.00±0.65^{B}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Nizoral	0.00±0.00^{A}	14.50±0.73^{B}	15.50±1.18^{C}	13.75±0.86^{B}
Sporanox	19.50±1.18^{C}	0.00±0.00^{A}	0.00±0.00^{A}	21.00±1.92^{D}
Nystatin	19.50±1.45^{C}	19.75±1.03^{D}	17.00±1.13^{C}	27.50±1.63^{E}
Azadirachta indica'oil	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Trecula africana'oil	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Detarium senegalense'oil	12.25±0.94^{AB}	0.00±0.00^{A}	13.00±0.75^{B}	0.00±0.00^{A}
Sterile distilled Water	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}

*Means (n = 3) with the same superscript letter in a column are not significantly different (p>0.05), while means with different superscript letter are significantly the same (p<0.05) when subjected to Duncan multiple range test, DMRT (ANOVA posthoc).
**SE = Standard error ***n = number of observations

Mucor racemosus with 11.75±0.45, 13.25±1.08 mm zone of inhibition against Unidentified V, *Trichophyton violaceum with* 20.75±1,19 mm zone of inhibition, Unidentified X and *Emericella nidulans* with zone of inhibition of 13.87±1.28 mm and 12.87±0.63 mm respectively (Tables 11 & 12).

The three extracted oil were weighed after the extraction and the following weight were recorded. As for *Azadirachta indica*, 27.7 g of oil was gotten, while 18.2 g and 12.5 g were gotten from *Treculia africana* and *Detarium senegalense'* seed respectively. Of the three extracted oil (*Azadirachta indica*, *Treculia africana* and *Detarium senegalense*) used in this study, only two (*Azadirachta indica and Detarium senegalense*) were able to inhibited some strain of isolated fungi. As for the *Azadirachta indica,* this extracted oil was able to inhibit only five of the twenty five isolated fungi. The isolated fungi include; *Trichophyton concentricum* strain A with 10.37±0.26 mm mean of inhibition, *Microsporum audouinii* with 10.87±0.29 mm mean (Tables 6 & 7), *Penicillium citrinum* with mean of 10.25±0.25 mm mean of inhibition, *Mucor racemosus* and *Emericella nidulans* with means of 11.37±0.46 mm and 11.87±0.44 mm mean of inhibition respectively (Tables 8, 10 & 12).

Detarium senegalense oil was more effective than *Azadirachta indica* oil*;* it was able to inhibit ten isolated fungi. These included; *Trichophyton concentricum* strain A and B with zone of inhibition 12.12±0.47 mm and 13.00±0.65 mm respectively (Tables 6 & 10), *Exophiala dermatitidis* with zone of inhibition of 11.00±0.26mm, *Microsporum audouinii* with 11.62±0.46 mm zone of inhibition (Table 7), *Trichophyton soudanense* with 12.25±0.55 mm zone of inhibition, *Microsporum ferrugineum with* 13.12±0.61 mm zone of inhibition (Table 8), *Penicillium*

Table 10: Sensitivity test on three antifungal agents, three extracted oil against *Mucor racemosus, Trichophyton concentricum* strain B, *Absidia corymbifera and* Unidentified T.

Antifungal agents & Extracted seed oils	*Mean inhibition zone diameter ± **SE (mm)			
	Mucor racemosus	*Trichophyton concentricum* strain B	*Absidia corymbifera*	Unknown T
Flucamed	*0.00±0.00^{A}	13.12±0.78^{B}	20.25±0.95^{C}	0.00±0.00^{A}
Diflucan	14.62±1.17^{C}	15.75±1.03^{C}	18.50±1.63^{C}	0.00±0.00^{A}
Itranox	14.12±0.85^{C}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Ketoconazole	0.00±0.00^{A}	12.50±0.70^{AB}	0.00±0.00^{A}	0.00±0.00^{A}
Griseofulvin	0.00±0.00^{A}	11.12±0.22^{AB}	15.37±0.73^{B}	14.25±0.70^{B}
Nizoral	0.00±0.00^{A}	19.75±2.05^{D}	16.00±1.13^{B}	0.00±0.00^{A}
Sporanox	11.75±1.28^{B}	16.00±1.19^{C}	0.00±0.00^{A}	0.00±0.00^{A}
Nystatin	11.75±1.58^{B}	12.750.83^{AB}	11.12±0.47^{A}	16.50±1.18^{C}
Azadirachta indica'oil	11.37±1.30^{AB}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Trecula africana'oil	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Detarium senegalense'oil	0.00±0.00^{A}	13.00±0.65^{B}	11.37±0.32^{A}	0.00±0.00^{A}
Sterile distilled Water	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}

*Means (n = 3) with the same superscript letter in a column are not significantly different (p>0.05), while means with different superscript letter are significantly the same (p<0.05) when subjected to Duncan multiple range test, DMRT (ANOVA posthoc).
**SE = Standard error ***n = number of observations

Table 11: Sensitivity test on eight antifungal agents, three extracted oil against *Microsporum nanum,* Unidentified V, *Trichophyton violaceum* and Unidentified X

Antifungal agents & Extracted seed oils	*Mean inhibition zone diameter ± **SE (mm)			
	Microsporum nanum	Unidentified V	*Trichophyton violaceum*	Unidentified X
Flucamed	*0.00±0.00^{A}	14.00±0.62^{C}	0.00±0.00^{A}	0.00±0.00^{A}
Diflucan	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Itranox	0.00±0.00^{A}	11.50±0.46^{B}	17.50±0.90^{C}	16.50±0.90^{CD}
Ketoconazole	0.00±0.00^{A}	11.87±0.51^{B}	0.00±0.00^{A}	0.00±0.00^{A}
Griseofulvin	14.25±0.70^{B}	0.00±0.00^{A}	12.25±0.41^{B}	12.87±0.44^{B}
Nizoral	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	15.75±0.95^{C}
Sporanox	0.00±0.00^{A}	13.25±1.08^{C}	20.75±1.19^{D}	13.87±1.28^{B}
Nystatin	16.50±1.18^{C}	0.00±0.00^{A}	11.87±0.44^{B}	18.00±1.19^{D}
Azadirachta indica'oil	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Trecula africana'oil	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Detarium senegalense'oil	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	12.37±0.53^{B}
Sterile distilled Water	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}

*Means (n = 3) with the same superscript letter in a column are not significantly different (p>0.05), while means with different superscript letter are significantly the same (p<0.05) when subjected to Duncan multiple range test, DMRT (ANOVA posthoc).
**SE = Standard error ***n = number of observations

Table 12: Sensitivity test on eight antifungal agents, three extracted oils against *Emericella nidulans*

Antifungal agents & Extracted seed oils	*Mean inhibition zone diameter ± **SE (mm) *Emericella nidulans*
Flucamed	*0.00±0.00^{A}
Diflucan	0.00±0.00^{A}
Itranox	0.00±0.00^{A}
Ketoconazole	0.00±0.00^{A}
Griseofulvin	15.75±1.03^{C}
Nizoral	0.00±0.00^{A}
Sporanox	12.87±0.63^{B}
Nystatin	21.75±2.25^{D}
Azadirachta indica'oil	11.87±0.44^{A}
Trecula africana'oil	0.00±0.00^{A}
Detarium senegalense'oil	12.62±0.56^{B}
Sterile distilled Water	0.00±0.00^{A}

*Means (n = 3) with the same superscript letter in a column are not significantly different (p>0.05), while means with different superscript letter are significantly the same (p<0.05) when subjected to Duncan multiple range test, DMRT (ANOVA posthoc).
**SE = Standard error ***n = number of observations

aschersonia with zone of inhibition of 12.25±0.94 mm, Unidentified O with zone of inhibition of 13.00±0.75 mm and 12.37±0.53 mm with Unidentified X (Tables 9 & 11), *Absidia corymbifera* and *Emericella nidulans* with 11.37±0.32 mm and 12.62±0.56 mm of inhibition zone respectively (Tables 10 & 12).

4.5 SENSITIVITY TEST AGAINST SOME ANTISEPTIC SOAPS AND DETERGENTS.

The antiseptic soaps and detergents used in this sensitivity test were able to produce zone of inhibition against the fifteen isolated fungi tested, except the antiseptic soap "*Lifebuoy*". This antiseptic soap was able to inhibit the growth of only five of the fifteen isolated fungi used. This include; *Trichophyton rubrum* with 10.50±0.50 mm zone of inhibition, *Trichophyton soudanense* with 11.25±1.25 mm zone of inhibition, *Microsporum ferrugineum* with 10.75±0.75 mm zone of inhibition, *Penicillium aschersonia* with 10.75±0.36 mm zone of inhibition and *Penicillium* species with 11.12±0.35 mm zone of inhibition Tables (14-17).

Table 13: Summarized table on sensitivity activities of the isolated fungi against four antiseptic soaps and four detergents.

Fungi	**Dettol**	**Delta**	**Lifebuoy**	**Tetmosol**	**Klin**	**Arial**	**Omo**	**Zip**	**Distilled water**
A.terreus	+	+	-	+	+	+	+	+	-
Blastomyces dermatitidis	+	+	-	+	+	+	+	+	-
E.fluccosum	+	+	-	+	+	+	+	+	-
M.audouinii	+	+	-	+	+	+	+	+	-
M.ferrugineum	+	+	+	+	+	+	+	+	-
P.aschersonia	+	+	+	+	+	+	+	+	-
P.citrinum	+	+	+	+	+	+	+	+	-
*P.*species	+	+	+	+	+	+	+	+	-
T. rubrum	+	+	+	+	+	+	+	+	-
T.concentricum A	+	+	-	+	+	+	+	+	-
T.ment.var.quinckeanum	+	+	-	+	+	+	+	+	-
T.soudanense	+	+	+	+	+	+	+	+	-
T.tonsurans. A	+	+	-	+	+	+	+	+	-
T.violaceum	+	+	-	+	+	+	+	+	-
Unknown O	+	+	-	+	+	+	+	+	-

–, resistance, +, susceptibility.

Table 14: Sensitivity test on four antiseptic soaps and four detergents against *Trichophyton concentricum* strain A, *T. tonsurans* strain A, *T. mentagrophytes var.quinckeanum* and *Aspergillus terreus*.

Antiseptic soaps & Detergents	*Mean inhibition zone diameter ± **SE (mm)			
	Trichophyton concentricum strain A	*T. tonsurans strain A*	*T. mentagrophytes var.quinckeanum*	*Aspergillus terreus*
Dettol	*15.12± 0.22^{B}	14.37±0.46^{B}	15.12±0.22^{B}	15.25±0.36^{B}
Delta	20.00± 0.53^{DE}	18.75± 0.75^{DE}	20.00±0.53^{EF}	20.00±0.53^{D}
Lifebuoy	0.00± 0.00^{A}	0.00+0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}
Tetmosol	18.25± 0.55^{CD}	18.25±0.55^{CD}	17.75±0.79^{CD}	18.25±0.55^{CD}
Klin	20.50± 0.90^{E}	20.25± 0.88^{E}	20.50±0.90^{F}	18.50±0.82^{CD}
Arial	18.50± 1.29^{CD}	17.75± 1.33^{CD}	18.50±1.29^{DE}	18.50±1.29^{CD}
Omo	11.62± 0.46^{A}	11.12±0.35^{A}	11.62±0.46^{A}	11.62±0.46^{A}
Zip	17.12± 0.35^{C}	16.37± 0.53^{C}	15.87±0.54^{BC}	17.12±0.35^{C}
Distilled water	0.00±0.00^{A}	0.00±0.00^{A}	0.00±00^{A}	0.00±0.00^{A}

*Means (n = 3) with the same superscript letter in a column are not significantly different (p>0.05), while means with different superscript letter are significantly the same (p<0.05) when subjected to Duncan multiple range test, DMRT (ANOVA posthoc).
**SE = Standard error ***n = number of observations

Table 15: Sensitivity test on four antiseptic soaps and four detergents against *Blastomyces dermatitidis, Trichophyton. violaceum, Epidermatophyton fluccosum* and *Microsporum audouinii*.

Antiseptic soaps & Detergents	*Mean inhibition zone diameter ± **SE (mm)			
	Blastomyces dermatitidis	*T. violaceum*	*Epidermatophyton fluccosum*	*Microsporum audouinii*
Dettol	*15.12±0.22^{B}	15.12±0.22^{B}	15.62±0.41^{B}	15.12±0.22^{B}
Delta	20.00±0.53^{DE}	20.00±0.53^{DE}	17.50±0.62^{BC}	20.00±0.53^{DE}
Lifebuoy	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00A
Tetmosol	18.25±0.55^{CD}	18.25±0.55^{CD}	17.75±0.77^{C}	18.25±0.55^{CD}
Klin	20.50±0.90^{E}	20.50±0.90^{E}	20.50±0.90^{D}	20.50±0.90^{E}
Arial	18.50±1.29^{CD}	18.50±1.29^{CD}	18.50±1.29^{C}	18.50±1.29^{CD}
Omo	11.62±0.46^{A}	10.87±0.35^{A}	12.00±0.53^{A}	11.37±0.32^{A}
Zip	17.12±0.35^{C}	17.25±0.36^{C}	17.12±0.35^{BC}	17.12±0.35^{C}
Distilled water	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}

*Means (n = 3) with the same superscript letter in a column are not significantly different (p>0.05), while means with different superscript letter are significantly the same (p<0.05) when subjected to Duncan multiple range test, DMRT (ANOVA posthoc).
**SE = Standard error ***n = number of observations

Table 16: Sensitivity test on four antiseptic soaps and four detergents against *Penicillium citrinum, Trichophyton rubrum, T. soudanense and Microsporum ferrugineum*

Antiseptic soaps & Detergents	*Mean inhibition zone diameter ± **SE (mm)			
	Penicillium citrinum	*T .rubrum*	*T. soudanense*	*Microsporum ferrugineum*
Dettol	*15.12±0.41^{B}	13.75±0.55^{B}	14.75±0.52^{BC}	14.25±0.79^{BC}
Delta	17.50±0.62^{DE}	15.50±0.90^{CD}	14.25±0.79^{BC}	15.50±0.90^{CD}
Lifebuoy	0.00±0.00^{A}	10.50±0.50^{A}	11.25±1.25^{A}	10.75±0.75^{A}
Tetmosol	18.62±0.56^{CD}	17.37±1.05^{D}	18.50±0.98^{DE}	18.75±0.92^{EF}
Klin	21.00±1.00^{E}	20.50±0.90^{E}	20.50±0.90^{E}	20.50±0.90^{F}
Arial	18.50±1.29^{CD}	17.00±1.25^{D}	17.00±1.25^{CD}	17.00±1.25^{DE}
Omo	14.62±0.88^{A}	11.62±0.96^{AB}	12.37±0.92^{AB}	12.37±0.92^{AB}
Zip	15.87±0.54^{C}	16.25±0.95^{CD}	16.12±0.93^{CD}	16.25±0.95^{CDE}
Distilled water	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}

*Means (n = 3) with the same superscript letter in a column are not significantly different (p>0.05), while means with different superscript letter are significantly the same (p<0.05) when subjected to Duncan multiple range test, DMRT (ANOVA posthoc).
**SE = Standard error ***n = number of observations

Table 17: Sensitivity test on four antiseptic soaps and four detergents against *Penicillium aschersonia, Unknown O* and *Penicillium species*.

Antiseptic soaps & Detergents	*Mean inhibition zone diameter ± **SE (mm)		
	Penicillium aschersonia	*Unknown O*	*Penicillium species*
Dettol	*15.25±0.31^{C}	15.00±0.75^{B}	16.62±0.56^{C}
Delta	19.75±0.59^{DE}	17.00±0.84^{C}	20.00±0.53^{DE}
Lifebuoy	10.75±0.36^{AB}	0.00±0.00^{A}	11.12±0.35^{AB}
Tetmosol	18.25±0.55^{D}	17.75±0.59^{C}	18.25±0.55^{CD}
Klin	20.50±0.90^{E}	20.50±0.90^{D}	20.50±0.90^{E}
Arial	18.50±1.29^{DE}	18.50±1.29^{C}	18.50±1.29^{CD}
Omo	12.12±0.29^{B}	11.62±0.46^{A}	12.75±0.36^{B}
Zip	19.00±0.75^{DE}	17.12±0.35^{C}	17.12±0.35^{C}
Distilled water	0.00±0.00^{A}	0.00±0.00^{A}	0.00±0.00^{A}

*Means (n = 3) with the same superscript letter in a column are not significantly different (p>0.05), while means with different superscript letter are significantly the same (p<0.05) when subjected to Duncan multiple range test, DMRT (ANOVA posthoc).
**SE = Standard error ***n = number of observations

The antiseptic soap "*Dettol*"is effective against all the fifteen isolated fungi used. It inhibits the growth of *Trichophyton concentricum* strain A with 15.12±0.22 mm zone of inhibition; *Trichophyton tonsurans* strain A with 14.37±0.46 mm zone of inhibition; *Trichophyton mentagrophytes var. quinckeanum* with 15.12±0.22 mm zone of inhibition; *Aspergillus terreus* with 15.25±0.36mm zone of inhibition; *Blastomyces dermatitidis* with 15.12±0.22 mm zone of inhibition; *Trichophyton violaceum* with 15.12±0.22 mm zone of inhibition; *Epidermatophyton fluccosum* with 15.62±0.41 mm zone of inhibition; *Microsporum audouinii* with 15.12±0.22mm zone of inhibition; *Penicillium citrinum* with 15.12±0.41 mm zone of inhibition; *Trichophyton rubrum* with 13.75±0.55 zone of inhibition; *Trichophyton soudanense* with 14.75±0.52 zone of inhibition; *Microsporum ferrugineum* with 14.25±0.79 zone of inhibition; *Penicillium aschersonia* with 15.25±0.31mm zone of inhibition; *Penicillium* species with 16.62±0.56 mm zone of inhibition and Unknown O with 15.00±0.75 zone of inhibition.

"*Delta*" is another antiseptic soap used against the fifteen isolated human pathogenic fungi. This antiseptic soap proved to be effective against all the fifteen isolated fungi with the highest zone of inhibition compared to other antiseptic soaps used. It inhibits the growth of *Trichophyton concentricum* strain A with 20.00±0.53mm zone of inhibition; *Trichophyton tonsurans* strain A with 18.75±0.75mm zone of inhibition; *Trichophyton mentagrophytes var. quinckeanum* with 20.00±0.53mm zone of inhibition; *Aspergillus terreus* with 20.00±0.53mm zone of inhibition; *Blastomyces dermatitidis* with 20.00±0.53mm zone of inhibition; *Trichophyton violaceum* with 20.00±0.53mm zone of inhibition; *Epidermatophyton fluccosum* with 17.50±0.62mm zone of inhibition; *Microsporum audouinii* with 20.00±0.53mm zone of inhibition;

Penicillium citrinum with 17.50±0.62mm zone of inhibition; *Trichophyton rubrum* with 15.50±0.90 zone of inhibition; *Trichophyton soudanense* with 14.25±0.79 zone of inhibition; *Microsporum ferrugineum* with 15.50±0.90 zone of inhibition; *Penicillium aschersonia* with 19.75±0.59mm zone of inhibition; *Penicillium* species with 20.00±0.53mm zone of inhibition and Unknown O with 17.00±0.84mm zone of inhibition.

The antiseptic soap "*Tetmosol*" is another effective agent used; this antiseptic soap also inhibits the growth of all the fifteen isolated fungi tested against it. It inhibits the growth of *Trichophyton concentricum* strain A with 18.25±0.55mm zone of inhibition; *Trichophyton tonsurans* strain A with 18.25±0.55mm zone of inhibition; *Trichophyton mentagrophytes var. quinckeanum* with 17.75±0.79mm zone of inhibition; *Aspergillus terreus* with 18.25±0.55mm zone of inhibition; *Blastomyces dermatitidis* with 18.25±0.55mm zone of inhibition; *Trichophyton violaceum* with 18.25±0.55mm zone of inhibition; *Epidermatophyton fluccosum* with 17.75±0.77mm zone of inhibition; *Microsporum audouinii* with 18.25±0.55mm zone of inhibition; *Penicillium citrinum* with 18.62±0.56mm zone of inhibition; *Trichophyton rubrum* with 17.37±1.05 zone of inhibition; *Trichophyton soudanense* with 18.50±0.98 zone of inhibition; *Microsporum ferrugineum* with 18.75±0.92 zone of inhibition; *Penicillium aschersonia* with 18.25±0.55mm zone of inhibition; *Penicillium* species with 18.25±0.55mm zone of inhibition and Unknown O with 17.75±0.59mm zone of inhibition.

Of all the detergents used in this part of the study, detergent "*Klin*" proved to be the most effective compared to other detergent used. While detergent "*Omo* multiactive

"was the least effective detergent against all tested the isolated fungi strains. Detergent "*Klin*" inhibits the growth of *Trichophyton concentricum* strain A with 20.50±0.90mm zone of inhibition; *Trichophyton tonsurans* strain A with 20.25±0.88mm zone of inhibition; *Trichophyton mentagrophytes var. quinckeanum* with 20.50±0.90mm zone of inhibition; *Aspergillus terreus* with 18.50±0.82mm zone of inhibition; *Blastomyces dermatitidis* with 20.50±0.90mm zone of inhibition; *Trichophyton violaceum* with 20.50±0.90mm zone of inhibition; *Epidermatophyton fluccosum* with 20.50±0.90mm zone of inhibition; *Microsporum audouinii* with 20.50±0.90mm zone of inhibition; *Penicillium citrinum* with 21.00±1.00mm^{D} zone of inhibition; *Trichophyton rubrum* with 20.50±0.90mm zone of inhibition; *Trichophyton soudanense* with 20.50±0.90mm zone of inhibition; *Microsporum ferrugineum* with 20.50±0.90mm zone of inhibition; *Penicillium aschersonia* with 20.50±0.90mm zone of inhibition; *Penicillium* species with 20.50±0.90mm zone of inhibition and Unknown O with 20.50±0.90mm zone of inhibition.

Detergent "*Omo*" though the least effective in its group still inhibited the growth of all isolated fungi tested against it. It inhibits the growth of *Trichophyton concentricum* strain A with 11.62±0.46mm zone of inhibition; *Trichophyton tonsurans* strain A with 11.12±0.35mm zone of inhibition; *Trichophyton mentagrophytes var. quinckeanum* with 11.62±0.46mm zone of inhibition; *Aspergillus terreus* with 11.62±0.46mm zone of inhibition; *Blastomyces dermatitidis* with 11.62±0.46mm zone of inhibition; *Trichophyton violaceum* with 10.87±0.35mm zone of inhibition; *Epidermatophyton fluccosum* with 12.00±0.53mm zone of inhibition; *Microsporum audouinii* with 11.37±0.32mm zone of inhibition; *Penicillium citrinum* with 14.62±0.88.mm zone of inhibition; *Trichophyton rubrum* with 11.62±0.96mm zone of inhibition;

Trichophyton soudanense with 12.37±0.92mm zone of inhibition; *Microsporum ferrugineum* with 12.37±0.92mm zone of inhibition; *Penicillium aschersonia* with 12.12±0.29mm[B] zone of inhibition; *Penicillium* species with 12.75±0.36mm zone of inhibition and Unknown O with 11.62±0.46mm zone of inhibition.

"*Ariel*" is another effective detergent used against the fifteen isolated human pathogenic fungi. It inhibits the growth of *Trichophyton concentricum* strain A with 18.50±1.29mm zone of inhibition; *Trichophyton tonsurans* strain A with 17.75±1.33mm zone of inhibition; *Trichophyton mentagrophytes var. quinckeanum* with 18.50±1.29mm zone of inhibition; *Aspergillus terreus* with 18.50±1.29mm zone of inhibition; *Blastomyces dermatitidis* with 18.50±1.29mm zone of inhibition; *Trichophyton violaceum* with 18.50±1.29mm zone of inhibition; *Epidermatophyton fluccosum* with 18.50±1.29mm zone of inhibition; *Microsporum audouinii* with 18.50±1.29mm zone of inhibition; *Penicillium citrinum* with 18.50±1.29mm zone of inhibition; *Trichophyton rubrum* with 17.00±1.25 zone of inhibition; *Trichophyton soudanense* with 17.00±1.25 zone of inhibition; *Microsporum ferrugineum* with 17.00±1.25 zone of inhibition; *Penicillium aschersonia* with 18.50±1.29mm zone of inhibition; *Penicillium* species with 18.50±1.29mm zone of inhibition and Unknown O with 18.50±1.29mm zone of inhibition.

"*Zip*", the forth detergent used is another effective detergent used against the fifteen isolated human pathogenic fungi. It inhibits the growth of *Trichophyton concentricum* strain A with 17.12±0.35mm zone of inhibition; *Trichophyton tonsurans* strain A with 16.37±0.53mm zone of inhibition; *Trichophyton mentagrophytes var. quinckeanum* with 15.87±0.54mm zone of inhibition; *Aspergillus terreus* with 17.12±0.35mm zone

of inhibition; *Blastomyces dermatitidis* with 17.12±0.35mm zone of inhibition; *Trichophyton violaceum* with 17.25±0.36mm zone of inhibition; *Epidermatophyton fluccosum* with 17.12±0.35mm zone of inhibition; *Microsporum audouinii* with 17.12±0.35mm zone of inhibition; *Penicillium citrinum* with 15.87±0.54mm zone of inhibition; *Trichophyton rubrum* with 16.25±0.95 zone of inhibition; *Trichophyton soudanense* with 16.12±0.93 zone of inhibition; *Microsporum ferrugineum* with 16.25±0.95 zone of inhibition; *Penicillium aschersonia* with 19.00±0.75mm zone of inhibition; *Penicillium* species with 17.25±0.36mm zone of inhibition and Unknown O with 17.25±0.36mm zone of inhibition.

4.5.2 Significant differences in zone of inhibition of Isolated Fungi tested with the antiseptic soaps and detergents.

Analysis of variances revealed that there are significant differences ($P<0.05$) in the zone of inhibition exhibted by all the antiseptic soaps and detergents tested against all the isolated fungi (Appendix E). Further analysis using Duncan multiple range test (DMRT), showed that there is no significantly differences ($P>0.05$) in the zone of inhibition when *Trichophyton concentricum* strain A was tested against antiseptic soap "*Lifebuoy*", detergent "*Omo*" and sterile distilled water, but the zone of inhibition by the antiseptic soap "*Dettol"* was significantly different from that of "*Lifebuoy"* and "*Omo*" and every other antiseptic soaps and detergents used. While the zone of inhibition by detergent "*Klin*" was not significantly different from that of "*Delta*" but significant different from all other used against this fungus. DMRT also revealed that that there is no significant different ($P>0.05$) in the zone of inhibition when *Trichophyton tonsurans* strain A was tested against the antiseptic soap "*Lifebuoy*", sterile distilled water and detergent "*Omo*", but the zone of inhibition by

the antiseptic soap "*Dettol*" was significantly different ((P<0.05)) from every other antiseptic soaps and detergents used.

With the fungus *Trichophyton mentagrophytes var.quinckeanum,* DMRT showed that there is no significant differences (P>0.05) in the zone of inhibition by detergent "Klin" and that of the the antiseptic soap "Delta" but the zone of inhibition by detergent "Klin" was significantly different (P<0.05) from all other detergents and antiseptic soaps. DMRT revealed that there is no significantly differences (P>0.05) in the zone of inhibition in *Aspergillus terreus* by the agents; the antiseptic soap "Tetmosol" and "Delta", and detergents "Klin" and "Ariel" while the zone of inihibition by antiseptic soap was significantly different (P<0.05) from all other antiseptic soaps and detergent inhibition. In *Blastomyces dermatitidi*s, DMRT showed that there is no significantly differences in the zone of inhibition by detergent "Zip", "Ariel" and that of the the antiseptic soap "Tetmosl" but the zone of inhibition by antiseptic saop "Dettol" was significant different from all other detergents and antiseptic soaps.

In the fungus *Trichophyton violaceum,* DMRT showed that there is no significantly differences (P>0.05) in the zone of inhibition by antiseptic soap "Tetmosol", "Delta", and detergent "Ariel", but the zone of inhibition by the antiseptic soap "Dettol" was significantly different (P<0.05) from every other antiseptic soaps and detergents used. In *Epidermatophyton fluccosum,* DMRT revealed that detergent "Klin" was significantly different (P<0.05) from every other antiseptic soaps and detergents against the fungus. While in fungus *Microsporum audouinii,* DMRT showed that there is no significantly differences (P>0.05) in the zone of inhibition by detergent

"Klin" and the antiseptic soap "Delta" but detergent "Klin" was significantly different ($P<0.05$) from every other antiseptic soaps and detergents used.

DMRT also revealed there is no significantly differences ($P>0.05$) in the zone of inhibition by antiseptic soap "Dettol", detergent "Omo" and "Zip" in fungus *Penicillium citrium.*

4.6 GROWTH STUDIES ON DIFFERENT MEDIA.

Based on the growth rate, the isolated fungi in this study are divided into three groups. These are (i) the slow growing group which are the dermatophytes, (ii) the fast growing group, which are the opportunitics fungi that are also present at the infected sites, and (iii) the third group are moderately slow in growing and this group contained the systemic mycoses organisms, that usually affect the tissues beneath the skin but do have its mycelia on the skin surface.

The dermatophytes isolated in this study include; *Trichophyton concentricum* strain A and B, *Trichophyton tonsurans* strain A and B, *Trichophyton mentagrophytes var.quinckeanum, Trichophyton rubrum, Trichophyton soudanense, Trichophyton violaceum, Epidermaphyton fluccosum, Microsporum audouinii, Microsporum nanum* and *Microsporum ferrugineum.* This group had their optimal growth on 1% peptone agar and Sabouraud dextrose agar. *Trichophyton concentricum* strain A and B had the highest growth rate on 1% peptone agar followed by growth on Sabouraud dextrose agar (SDA) (Figs. 15 and 20). *Trichophyton tonsurans* strain A and B also had the highest growth rate on 1% peptone agar followed by growth on Sabroud dextrose agar (Figs. 15 and 16). *Trichophyton mentagrophytes var.quinckeanum, Trichophyton rubrum* and *Trichophyton violaceum* had their highest growth rate on both 1%

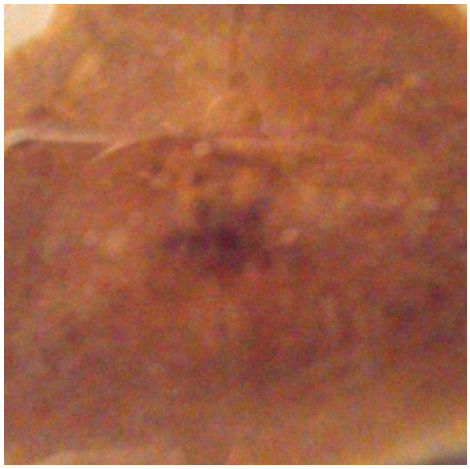
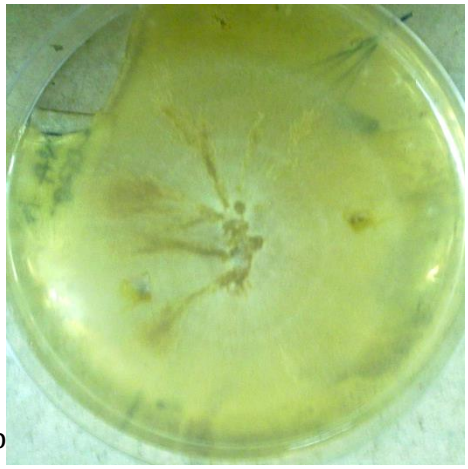
peptone agar and Sabouraud dextrose agar (Figs. 16, 17 and 18). *Trichophyton soudanense* and *Epidermaphyton fluccosum* had their highest growth rate on 1% peptone agar followed by growth on Sabouraud dextrose agar (Figs. 17 and 18).

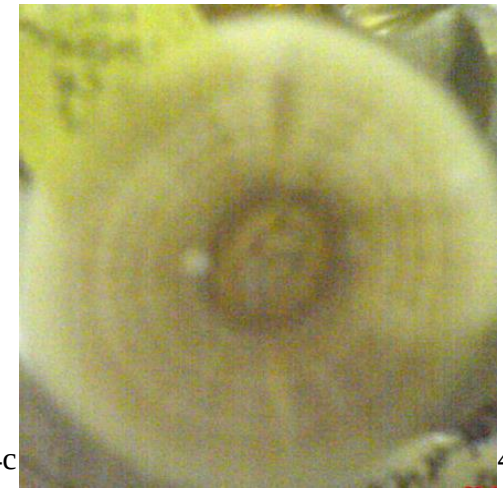
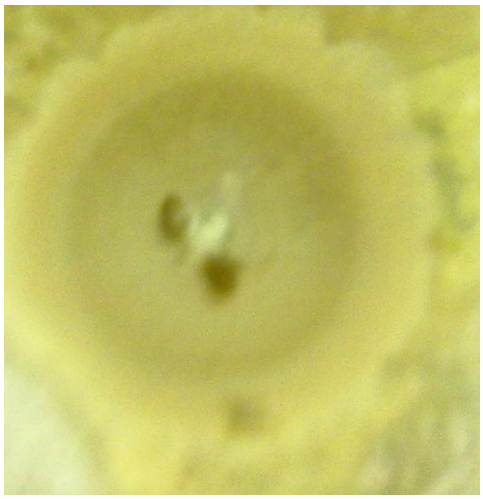
Unlike the *Trichophyton* species and *Epidermaphyton fluccosum, Microsporum audouinii* had its best growth rate on Sabouraud dextrose agar followed by growth on Potato dextrose agar while *Microsporum ferrugineum* had its highest growth rate on Sabroud dextrose agar followed by same growth rate on Potato dextrose agar, Nutrient agar and Malt extract agar (Fig. 19). *Microsporum nanum* had the same growth rate on the following media*;* Sabouraud dextrose agar, Potato dextrose agar, Nutrient agar and Malt extract agar with the least growth rate on Yeast extract agar (Fig. 20). The second group which is the fast growing organism had the best growth rate on different media. *Absidia corymbifera* had its best growth rate recorded on Potato dextrose agar, followed by the growth rate on Malt extract agar while *Aspergillus terreus* had the highest growth rate recorded on Sabouraud dextrose agar followed by growth on Malt extract agar (Fig. 21). *Emericella nidulans*, *Penicillium aschersonia, Penicillium citrinum* and *Penicillium* species recorded the best growth rate on Malt extract agar (Figs. 22, 23 and 24).

Mucor racemosus had its highest growth rate on Potato dextrose agar while recorded the same growth rate on both Malt extract agar and Yeast extract agar with the least growth rate on 1% peptone agar (Fig. 22). The four unknown species isolated in this study belong to the fast growing group, and while unknown species O, T and X had recorded their best growth on Malt extract agar (Figs. 26 and 27), unknown species V recorded its best growth rate on Potato dextrose agar (Fig. 27). The third isolated

group of organism in this study is moderately slow growing. These are *Blastomyces dermatitidis* and *Exophiala dermatitidis.* These two organisms grew best on Yeast extract agar with the least growth on Sabouraud dextrose agar (Fig. 25).

Another remarkable observation from the growth studied was that of the variation of colour and texture of the colony same organism on different media. This was observed with two isolates. *Epidermaphyton flucossum* showed different morphological character on Sabouraud dextrose agar and 1% Peptone agar (Plates 44a-b) and same morphological characteristic differences was observed in *Trichophyton tonsurans* strain B on Sabround dextrose agar and 1% Peptone agar (Plates 44c-d).

44a  44b 

44c  44d 

Culture photograph of two fungi isolates exhibiting pleomorphism

Plate 44a: *Epidermaphyton flucossum* on Sabouraud dextrose agar

Plate 44b: *Epidermaphyton flucossum* on 1% Peptone agar

Plate 44c: *Trichophyton tonsurans* strain B on Sabouraud dextrose agar medium

Plate 44d: *Trichophyton tonsurans* strain B on 1% Peptone agar

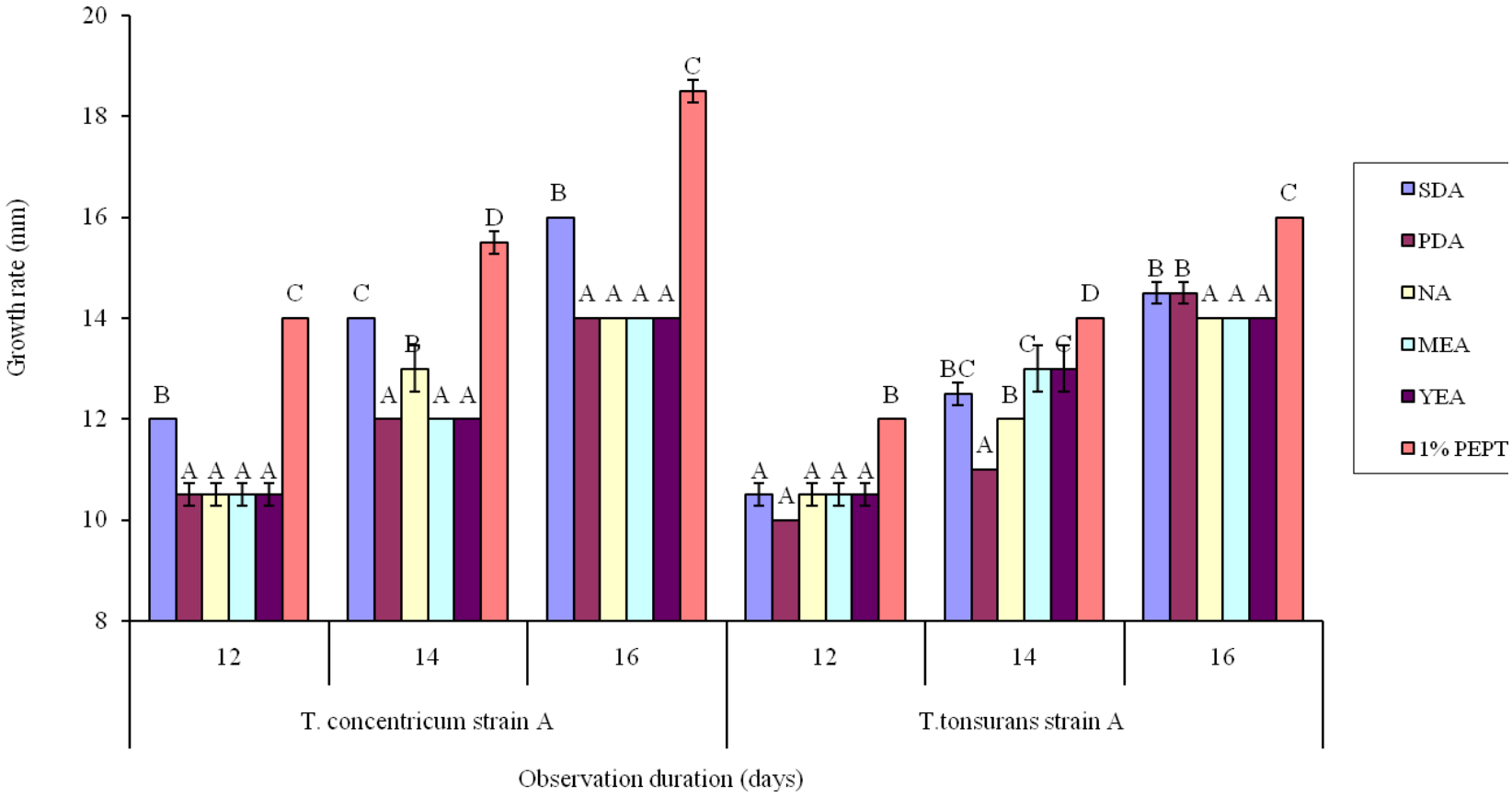


Fig 15: Growth rate of *Trichophyton concentricum* strain A and *T. tonsurans* strain A on six different media. Means ± SE bars with the same superscript letter in each growth period are not significantly different (p>0.05; DMRT)

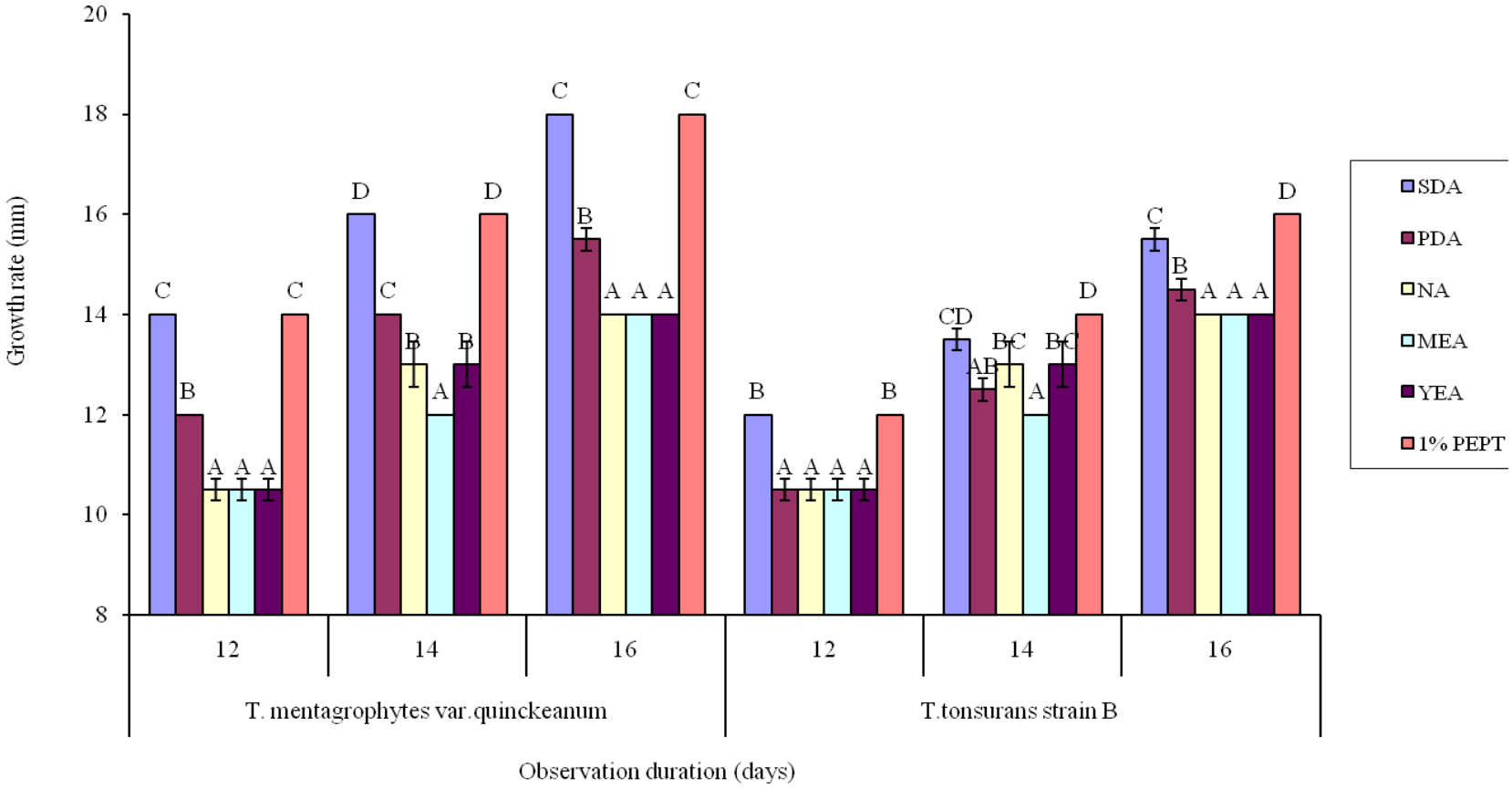


Fig 16: **Growth rate of *T.mentagrophytes var.quinckeanum* and *T.tonsurans* strain B on six different media. Means ± SE bars with the same superscript letter in each growth period are not significantly different (p>0.05; DMRT).**

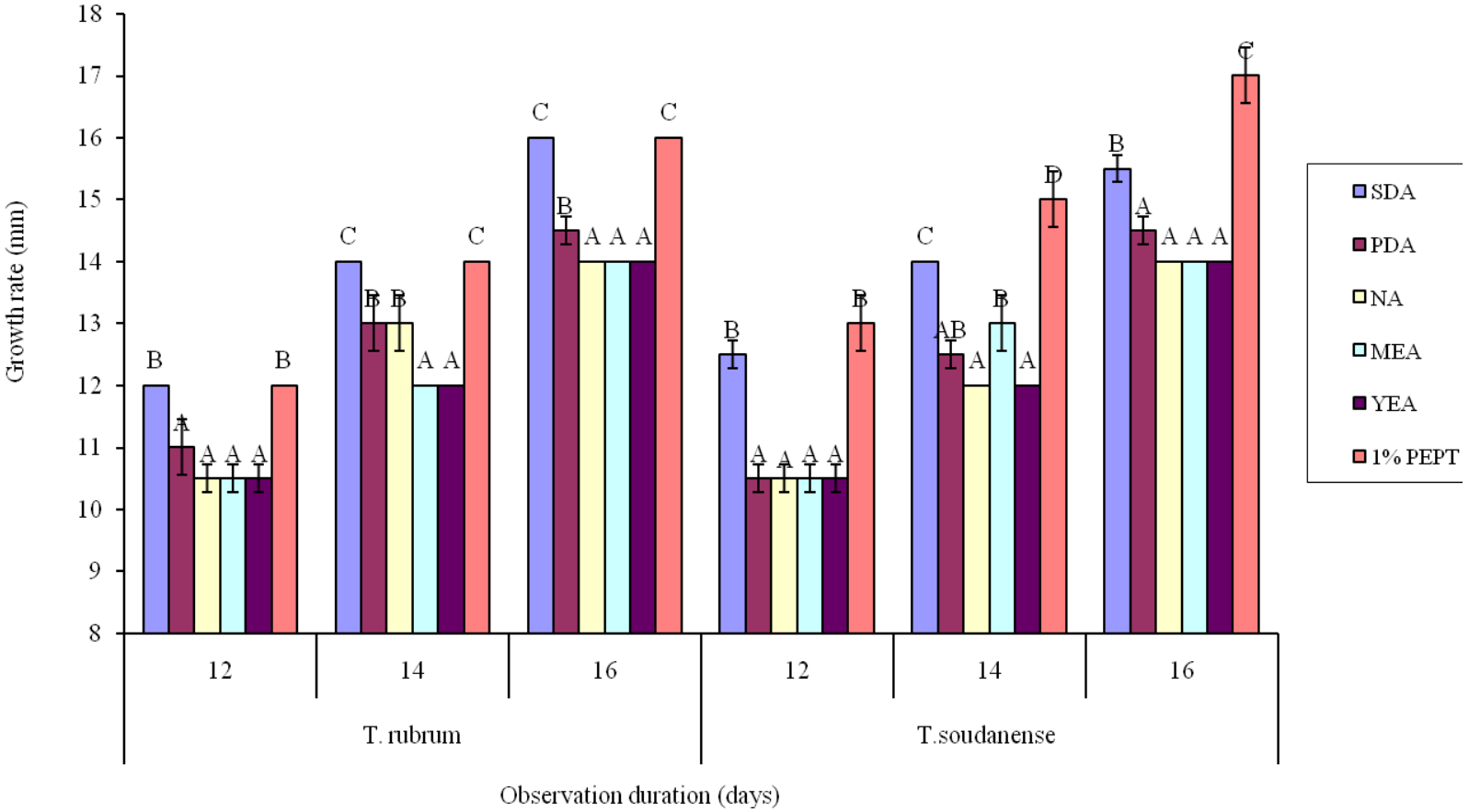


Fig 17: Growth rate of *T. ru*brum and *T. soudanense* on six different media. Means ± SE bars with the same superscript letter in each growth period are not significantly different (p>0.05; DMRT).

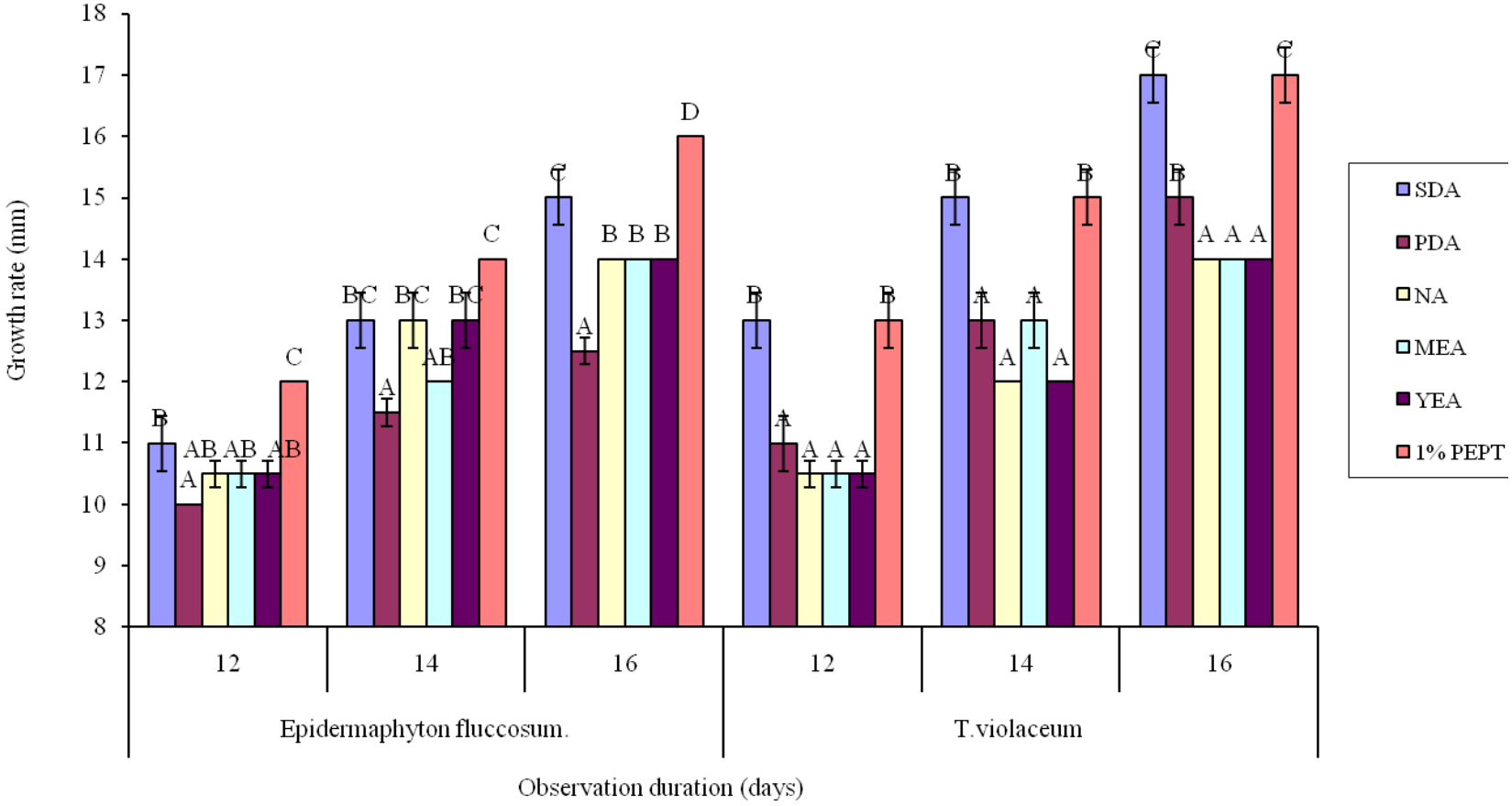


Fig 18: Growth rate of *Epidermaphyton floccosum* and *T.violaceum* on six different media. Means ± SE bars with the same superscript letter in each growth period are not significantly different (p>0.05; DMRT).

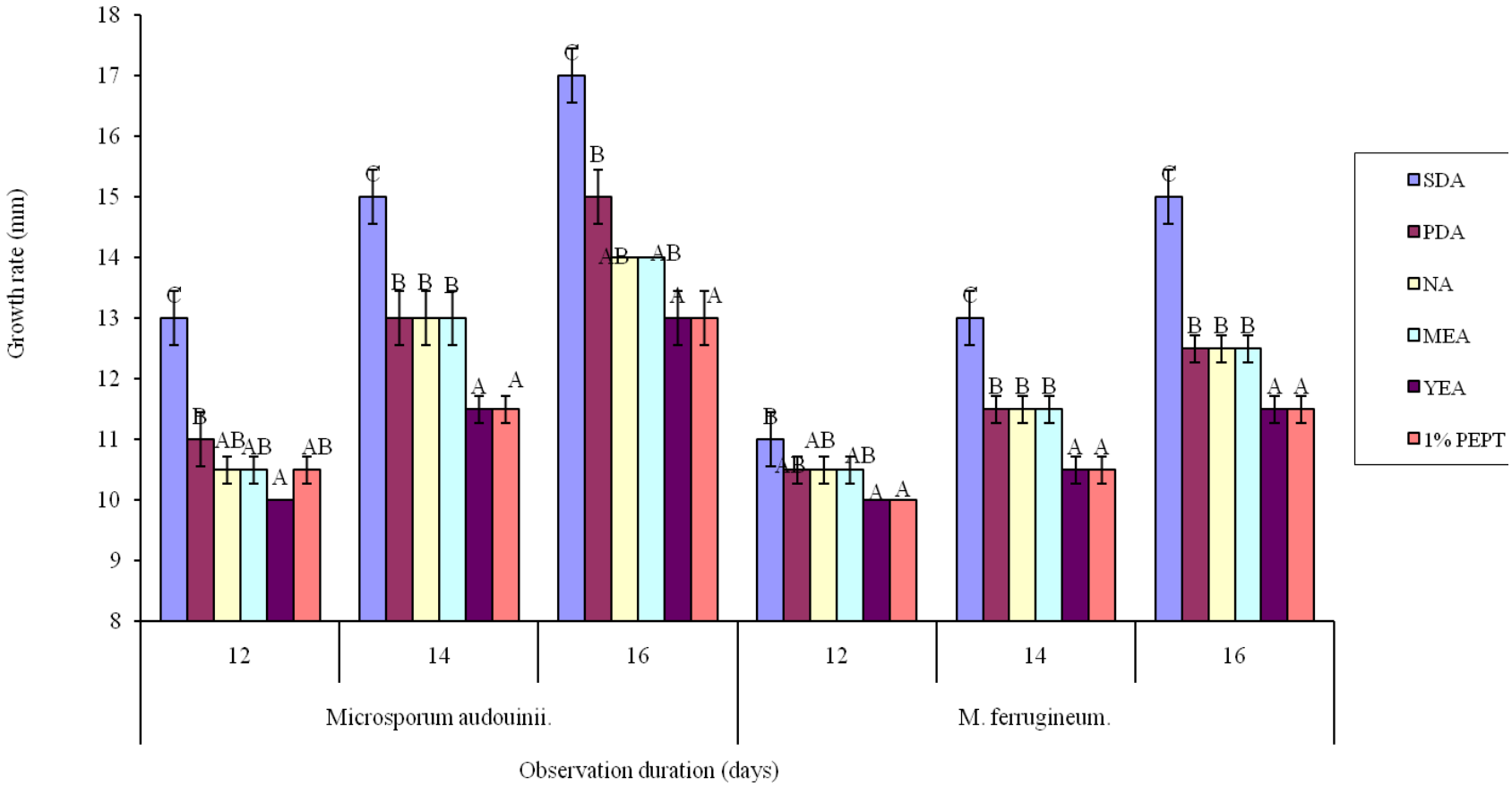


Fig 19: Growth rate of *Microsporum audouinii* and *M. ferruginum* on six different media. . Means ± SE bars with the same superscript letter in each growth period are not significantly different (p>0.05; DMRT).

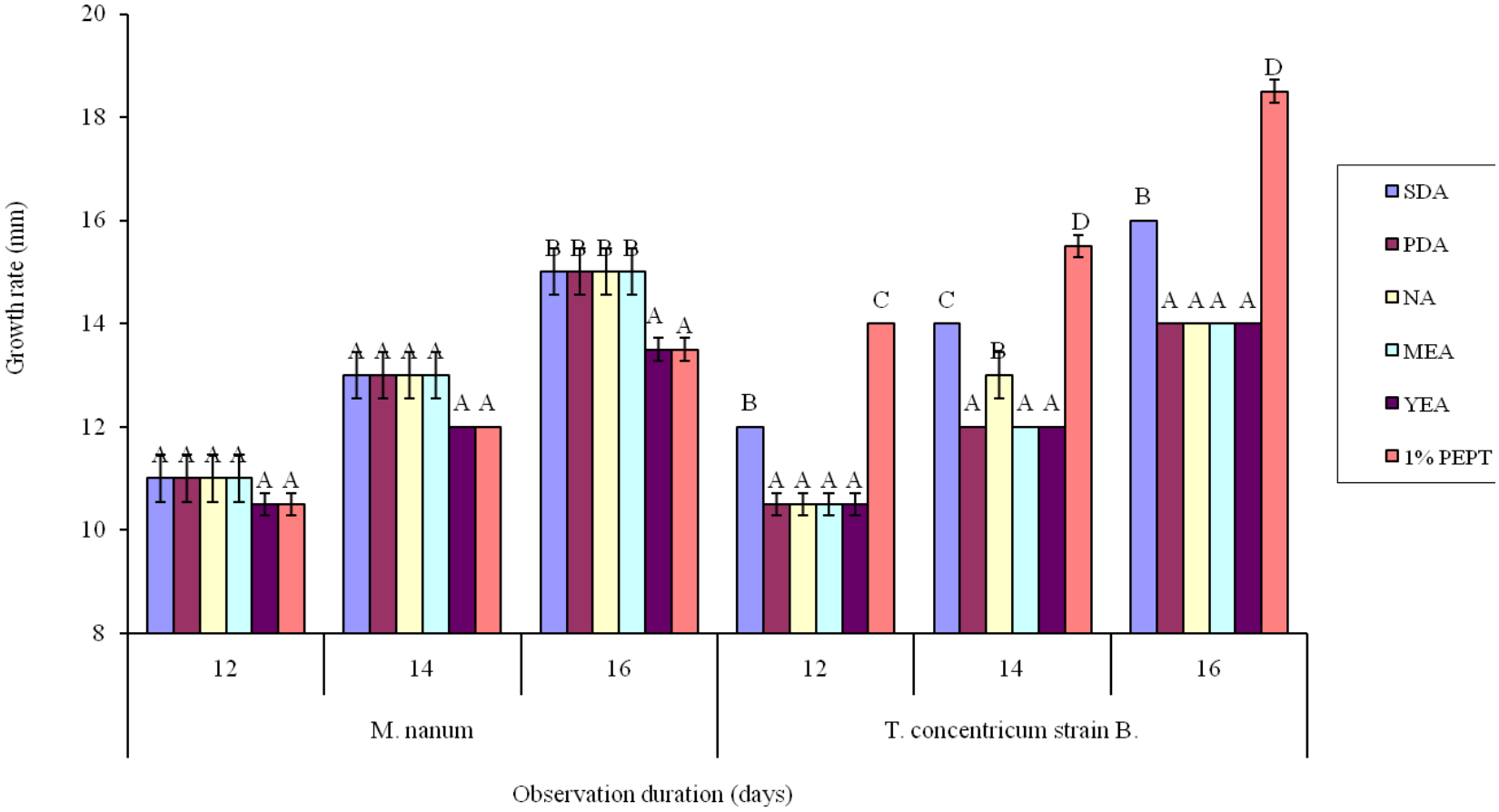


Fig 20: Growth rate of *M. nanum* and *T.concentricum* strain B on six different media. . Means ± SE bars with the same superscript letter in each growth period are not significantly different (p>0.05; DMRT).

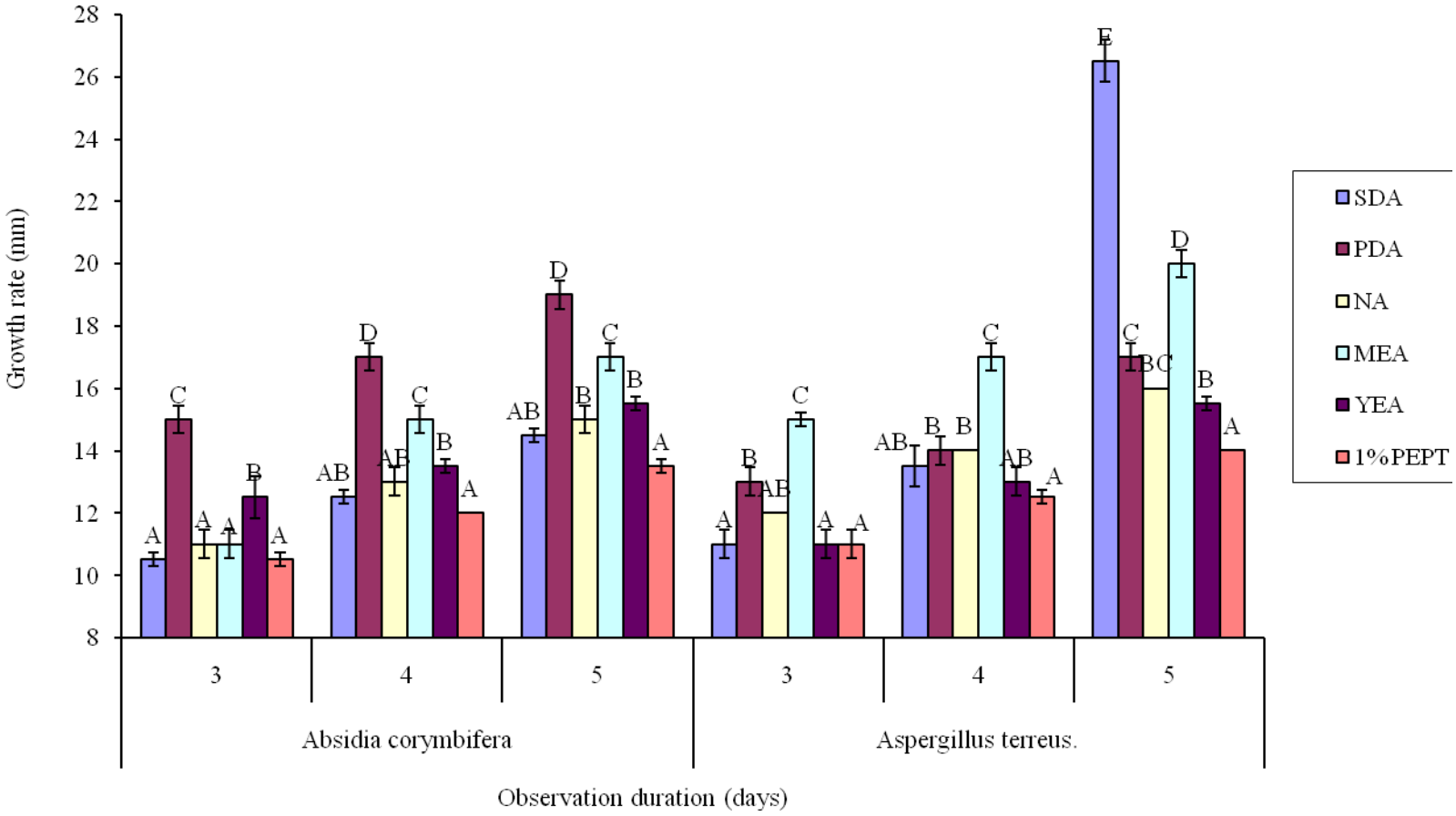


Fig 21: Growth rate of *Absidia corymbifera* and *Aspergillus terreus* on six different media. . Means ± SE bars with the same superscript letter in each growth period are not significantly different (p>0.05; DMRT).

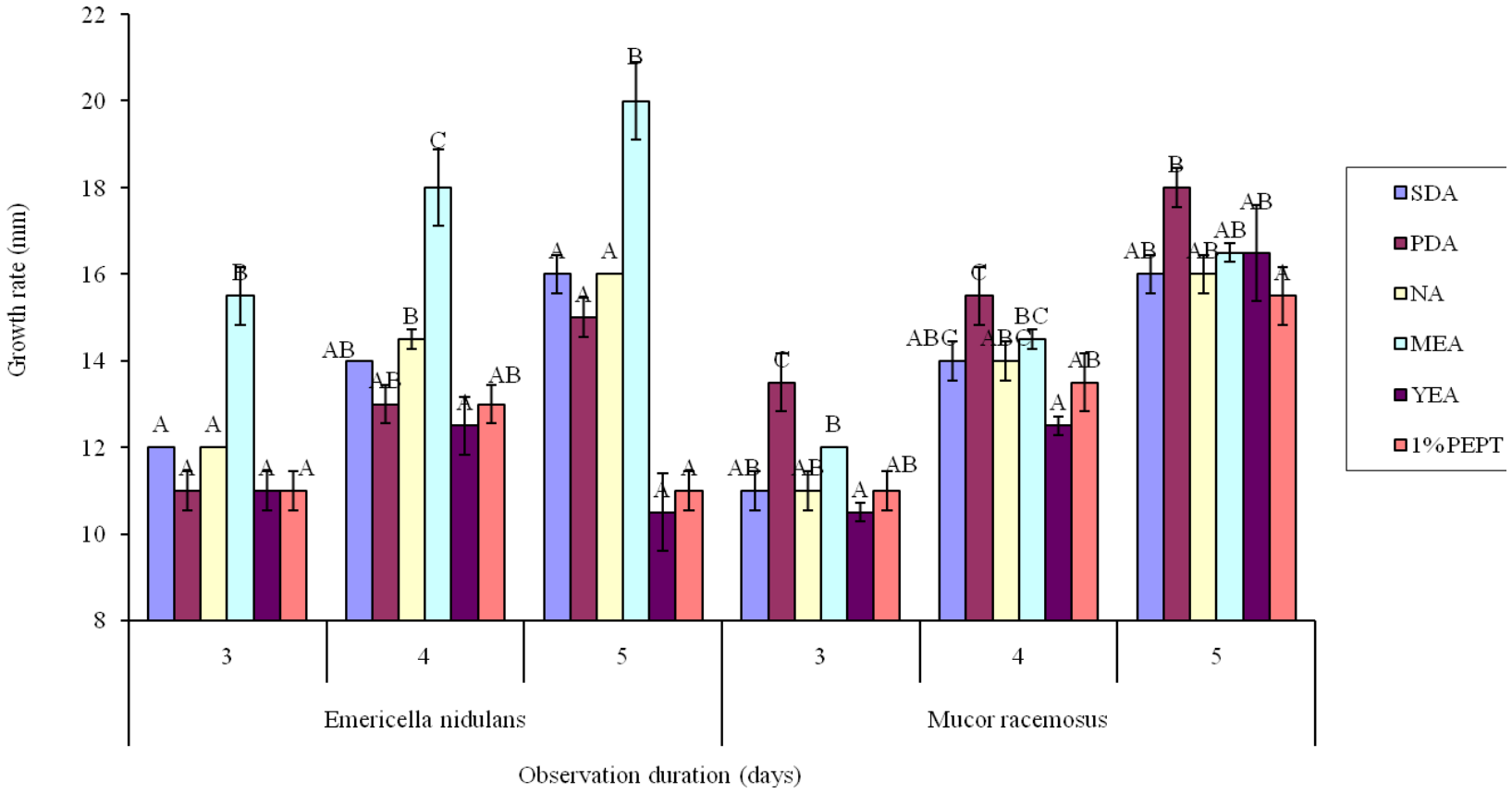


Fig 22: Growth rate of *Emericella nidulans* and *Mucor racemosus* on six different media. Means ± SE bars with the same superscript letter in each growth period are not significantly different (p>0.05; DMRT).

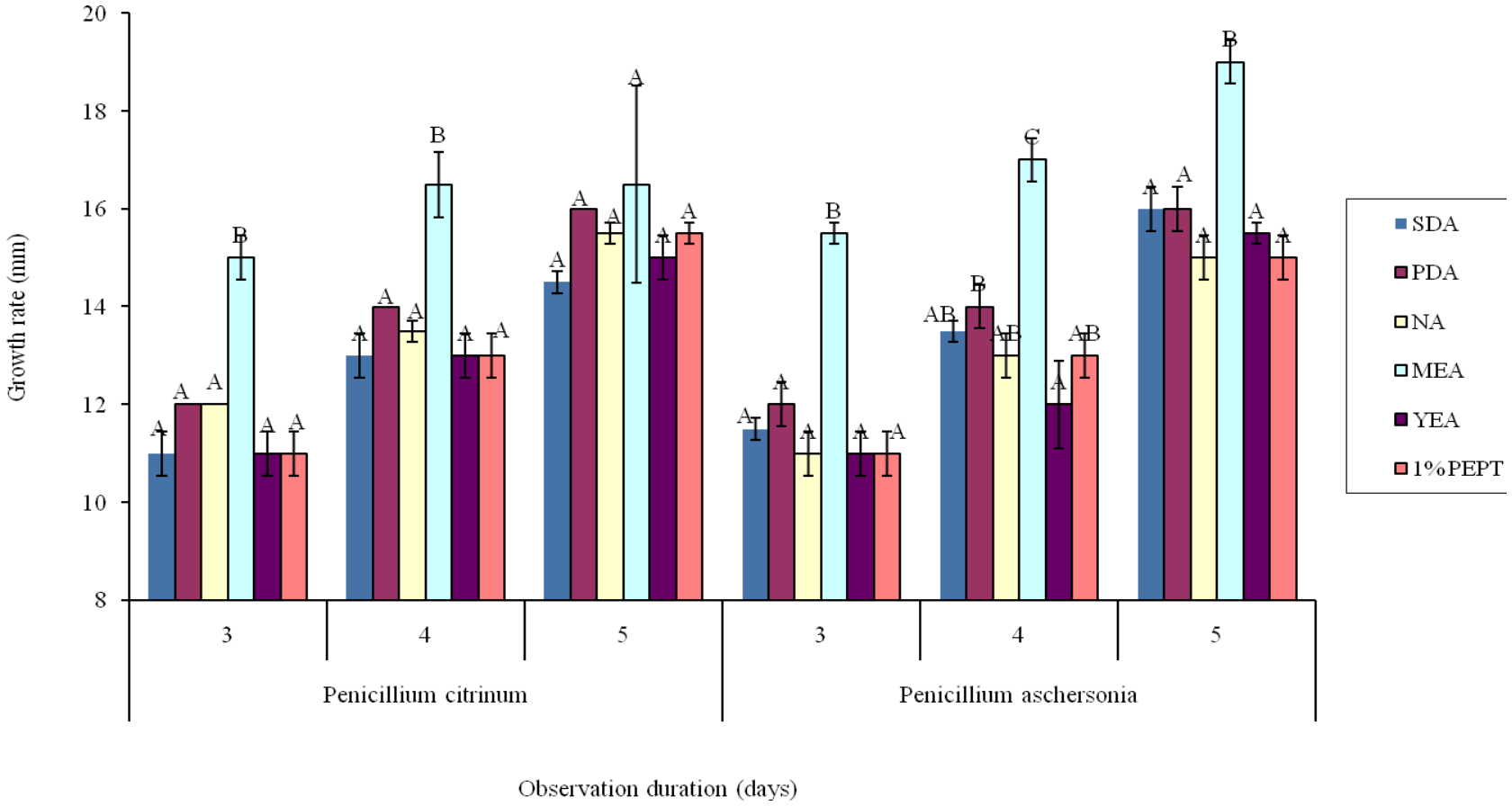


Fig 23: Growth rate of *Penicillium citrinum* and *P. aschersonia* on six different media. Means ± SE bars with the same superscript letter in each growth period are not significantly different (p>0.05; DMRT).

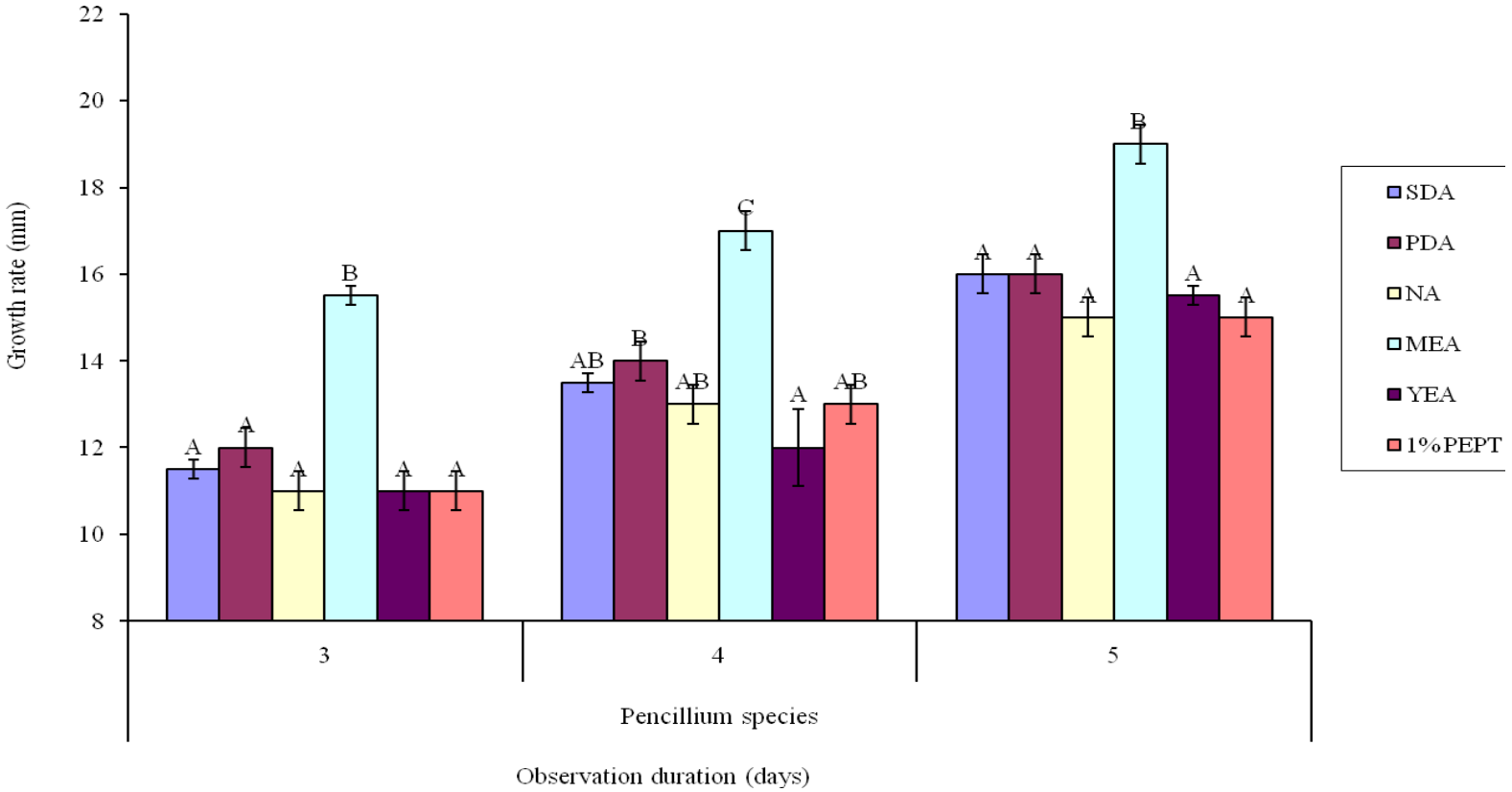


Fig 24: Growth rate of *Penicillium* sp on six different media. Means ± SE bars with the same superscript letter in each growth period are not significantly different (p>0.05; DMRT).

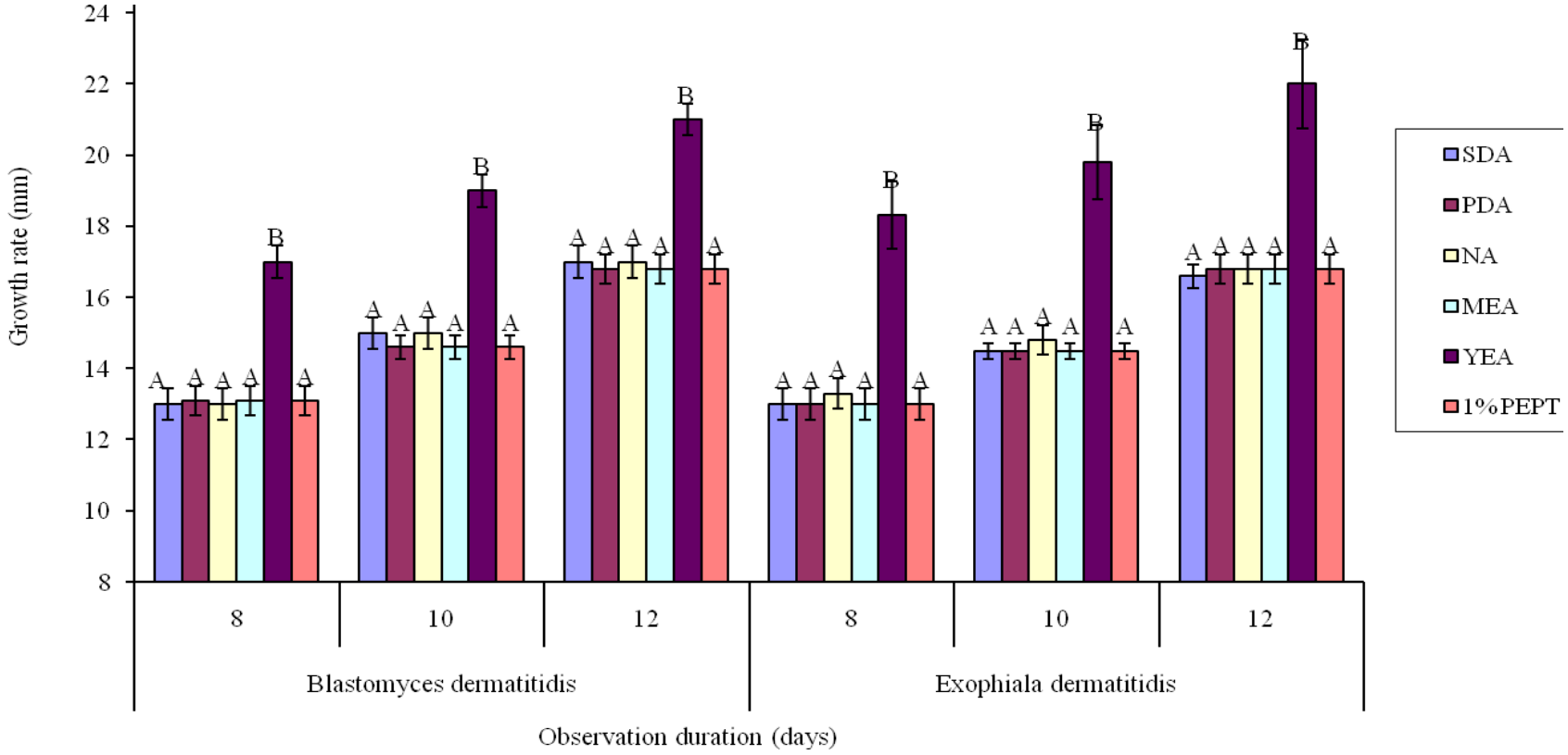


Fig 25: Growth rate of *Blastomyces dermatitidis* and *Exophiala dermatitidis* on six different media. Means ± SE bars with the same superscript letter in each growth period are not significantly different (p>0.05; DMRT).

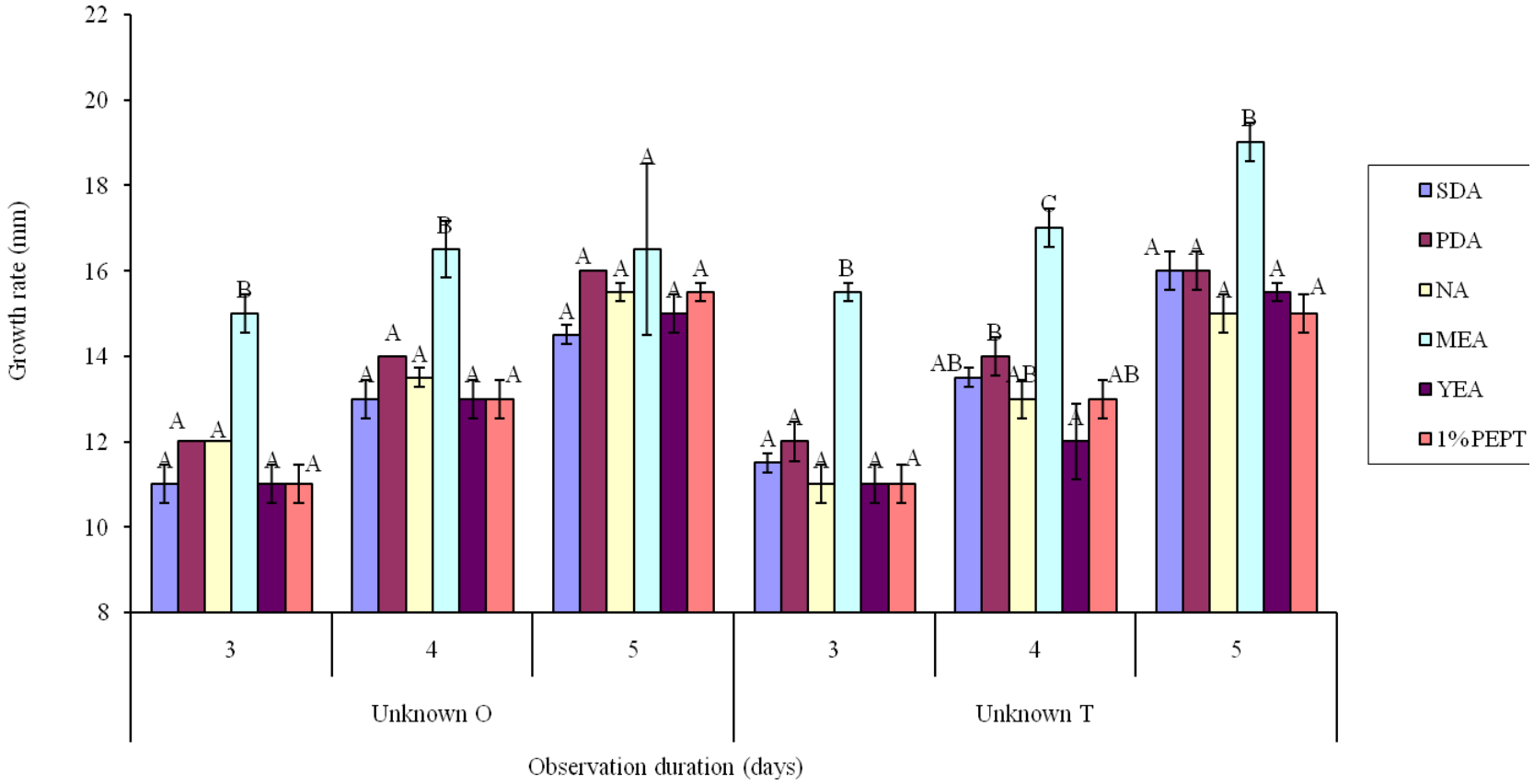


Fig 26: Growth rate of Unknown O and Unknown T on six different media. Means ± SE bars with the same superscript letter in each growth period are not significantly different (p>0.05; DMRT).

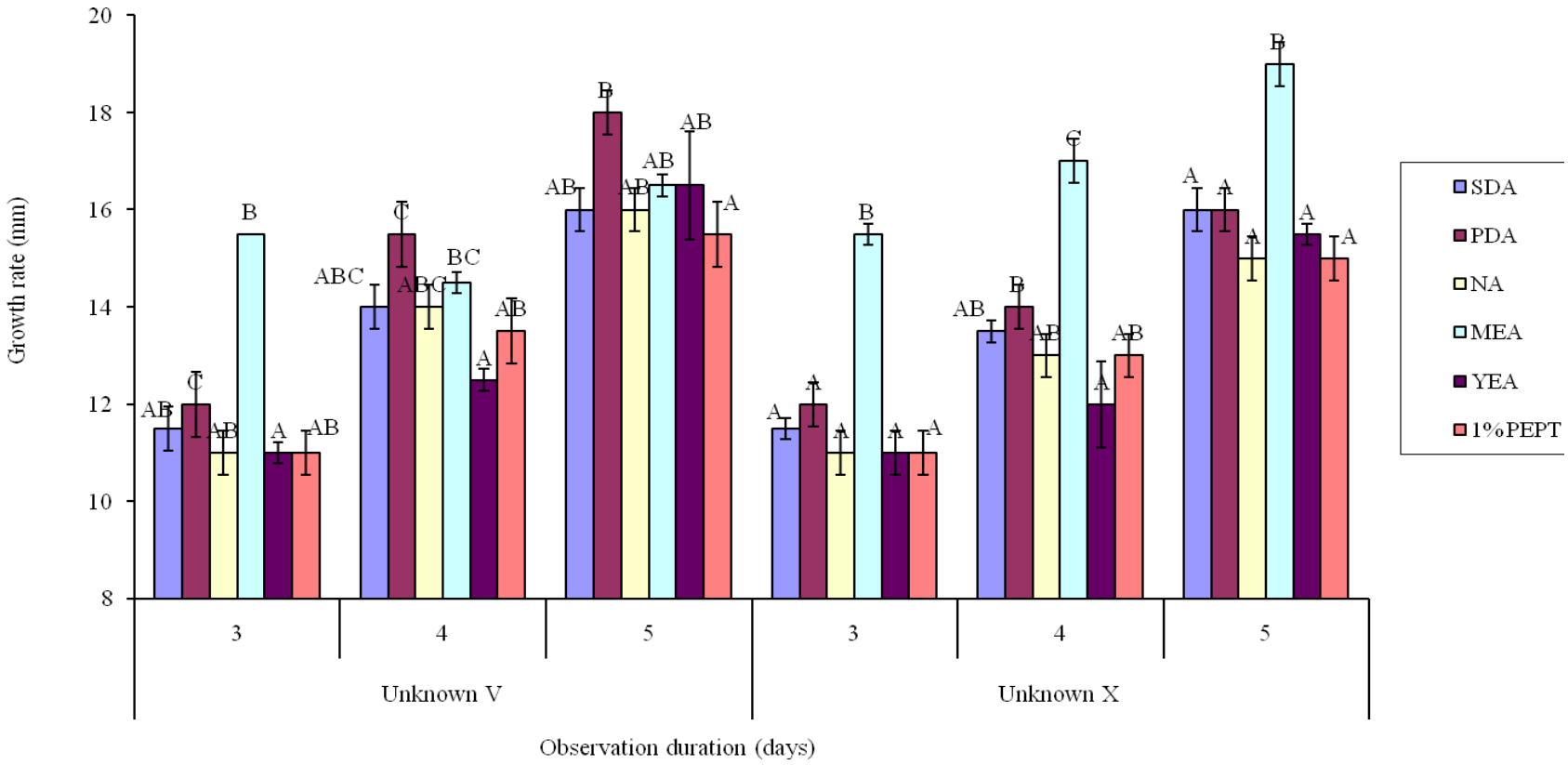


Fig 27: Growth rate of Unknown V and Unknown X on six different media. Means ± SE bars with the same superscript letter in each growth period are not significantly different (p>0.05; DMRT).

4.7 PHYTOCHEMICAL SCREENING

Based on the phytochemical screening carried out on the three extracted seed' oils used in this study, the three oils were found to contain alkaloid, tannins, cardiacglycosides and reducing compounds. The test on the quantitative estimation of the percentage of the active compounds present in the three extracted oils revealed that *Detarium senegalense* oil contains the highest percentage of alkaloid, tannins, reducing compounds and cardiacglycosides, followed by the *Azadirachta indica* seed*'* oil which also contains alkaloid, cardiacglycosides and reducing compounds with tannins in a higher percentage. *Treculia africana* seed oil was found to contain the least percentage of alkaloid, tannins, cardiacglycosides and reducing compounds (Table 18).

Table 18: Phytochemical screening and percentage of the active compounds present in the oil extracts of *Azadirachta indica, Treculia africana, Detarium senegalense'* seeds.

Oil extracts	Alkaloid	Flavonoids	Tannins	Cardicglycosides	Anthraquinone	Phlobatannins	Saponins	Reducing compounds
Azadirachta indica	0.51	-	0.90	0.51	-	-	-	0.31
Treculia Africana	0.30	-	0.54	0.50	-	-	-	0.29
Detarium senegalense	1.83	-	1.07	0.91	-	-	-	0.54

CHARPTER FIVE

5.0 DISCUSSION

The survey result in this present study revealed that dermatomycoses can occur irrespective of age, gender, occupation, custom and tradition of people. Although, several works have been published on the prevalence of dermatomycoses in different parts of Nigeria, most are reports among children of school age (Adetosoye, 1977; Ogbonna *et al.*, 1985; Gugnani and Njoku-Obi, 1986; Ekanem and Gugnani, 1987; Ayanwale and Alabi, 1988; Nweze, 2001; Anosike *et al*., 2005; Kao, 2005; Nweze and Okafor, 2005; Onayemi *et al*., 2005; Mbata and Nwajagu, 2007; Enemuor and Amedu, 2009 and Chukwu *et al.*, 2011). However, some researchers (Gugnani *et al*., 1975; Soyinka, 1978; Alabi, 1980; Gugnani, 1982 and Yahya, 2007) did not limit their sampling to the age group of school children only. The ratio of school children below the age of twelve years to adults encountered in this present study was approximately 1: 6. This may be due to the fact that most parents do not take dermatomycoses in their kids/wards serious to the extent of seeking medical help. They tend to resort to the use of some medicinal plants or sometimes go for self medication. Recently, Adekunle and Ikumapayi (2006) had reported the use of the aqueous extracts of *Funtumia elastica* (bark) and *Mallotus oppositifolius* (leaf) by the Nigerian populace to treat skin disease in children. Adekunle *et al*. (2011) also reported the use of *Acalypha wilkesiana* leaves by Nigerian naives to cure skin diseases (skin rashes in children) caused by *Candida albicans, Trichophyton mentagrophyte and Microsporium audonii.* They provided some scientific justification for the utilization of extracts from these plants, by reporting that the antifungal activities of the extracts might be due to the presence of phytochemical

compounds in the plants. They concluded that this is probably the rationale behind the use of these plants by the local populace.

The prevalence of dermatomycoses is higher in female patients than in the male patients encountered. According to Kane *et al*. (1997), they reported that reduction in triacylglycerides in sebum (an oily substance secreted by the sebaceous glands in mammalian skin which helps to make the skin and hair waterproof and to protect them from drying out) may predispose postmenopausal women to the development of dermatomycosis more frequently than other adults. Yahya (2007) and Atraide *et al.* (2011) also observed a higher prevalence of dermatomycoses in female patients in their studies. All these agreed with the findings in this study. Also, the results from the prevalence study of dermatomycoses in this study showed the skin lesion (49.4%) to have the highest percent of prevalence compared to other dermatomycoses encountered in this study. This finding was supported by the findings of Alabi (1980) and Yahya (2007).

It was also observed in this study that some dermatophytes are peculiar to certain parts of the human body for their manifestation. For example, *Trichophyton rubrum* was more frequently isolated from the nails and foot than any other part of the human body, *Trichophyton soudanense* was isolated more from hairy region like the head and "private part" while *Trichophyton violaceum* was found to be associated with the head, feet and the nail regions. All these agree with the findings of Zurita and Hay (1967); Weitzman and Summerbell (1995); Gupta and Summerbell (2000) and Ellis *et al*. (2007). In their works, they reported that the anthroconidia or chlamydopspores of dermatophytes species have a tendency to adhere *in vitro* to corneocytes derived from

a particular body part. Also, *Microsporum audouinii* an anthropophilic fungus was found to be peculiar to the human skin, most especially on the skin of children below the age of twelve. Kane *et al.* 1997 and Ellis *et al.* 2007 reported that this is possibly due to an increase in fungistatic action of triglycerides in the sebum that is produced following puberty.

According to Fredricks *et al.* (2005) no single DNA extraction method amongst those currently available is optimal for all analyzed fungi. Zhang *et al.* (1996) and Muller *et al.* (1998) reported that the major challenge for isolation of DNA of good quality and quantity from fungi lies in breaking the rigid cell walls, due to fungal nucleases and high polysaccharide contents of the cell wall. Melo *et al.* (2006) agreed with Zhang *et al.* (1996) and Muller *et al.* (1998) by concluding that DNA isolation from some fungal species is difficult because they have cell walls or capsules that are relatively unsusceptible to lysis. The result from this present study agrees with all the above statements as it revealed that no single DNA extraction protocol can isolate fungal DNA from the three groups of fungi encountered. Of the four DNA extraction protocols employed in the study, only the Zymo DNA protocol was able to extract DNA from the dermatophytic group.

Ten species and two different strains of dermatophytes were isolated in this study. These include; *Microsporum audouinii, Microsporum ferrugineum, Microsporum nanum, Trichophyton concentricum strain* A and B*, Trichophyton mentagrophytes var. quinckeanum, Trichophyton rubrum, Trichophyton soudanense, Trichophyton tonsurans* strain A and B and *Trichophyton violaceum.* The identification of two different strains of *Trichophyton concentricum and Trichophyton tonsurans* was made

possible by the application of the molecular techniques which provide the accurate identification. Based on the result from DNA sequences data, which revealed that no single organism sequence data was 100% homologous with those in the Genbank judging from their percentage of significance of matches with the sequences database at the Genbank. This implies that the isolated dermatophytes are different strains. This is probably the first work to report the identification of dermatophytes using molecular methods and to report this quantity of dermatophytes (species/strains) in Lagos State, Nigeria. According to Adekunle (2011), the major problem of curing dermatomycoses caused by this group of fungi is correct identification (fungal taxonomy) in order to determine the appropriate drug to employ. Kac (2000), had earlier reported that the accurate identification and characterization of dermatophytes through conventional method is sometimes difficult based on their overlapping characteristics and pleomorphism.

Woodgyer (2004) also supported Elias-Costa *et al.* (2000) by reporting that the use of molecular methods to explore the existing taxonomy of dermatophytes is an exciting prospect for the accurate identification of this group of organisms. Makimura *et al.* (1999) reported that because members of the dermatophytes are taxonomically and phylogenetically closely related, methods such as G+ C content, DNA hybridization studies, restriction fragment length polymorphism (RFLP) of mitochondrial DNA, and the base sequence of the 18S, 28S rRNA, or rRNA gene (rDNA), lack the resolving power to distinguish between some species. As a result of this, Kac (2000) and Balajee *et al.* (2007) were able to use sequence analysis of the highly variable internal transcribed spacer (ITS) 1 and 2 of the ribosomal DNA (rDNA) to explore the identification and phylogeny of some members of this group of fungi. Graser *et al.*

(2000), reported that the advantage of using the Internal Transcribed Spacer (ITS) region sequencing is due to the fact that it is the only existing marker for complete database identification. Iwen (2003) also supported that ITS sequencing of other pathogenic fungi (non-dermatophytes) involved in dermatomycoses can also be identified through this means. The findings in this present study agrees with all these findings as the different isolated strains of *Trichophyton concentricum* and *Trichophyton tonsurans,* even though they are phenotypically different, yet they were genetically identical judging from their percentage of significance of matches with the sequences database at the Genbank.

According to Graser *et al*. (2006), molecular methods come into play to determine the degree of distinction among closely related species. An example of phenotypic identity that was long known to be illusory was that of *Trichophyton mentagrophytes* in the traditional concept of Emmons (1934). This was used for many years as an 'aggregate' species name for the anamorph of two teleomorphic species, *Arthroderma benhamiae* and *A. vanbreuseghemii.* Takashio (1973) reported that these species were recognized as separate in terms of the biological species concept and were later shown to be strongly phylogenetically separate in molecular studies focusing on the ribosomal Internal Transcribed Spacer (ITS) region. Another example was found in the case of the taxa *Trichophyton mentagrophytes* According to Graser *et al*. (1999) and Probst *et al.* (2003), prior to the use of molecular biological investigations of dermatophytes by Davison *et al.* (1980), this 'aggregate' species was formerly treated together as *Trichophyton mentagrophytes* sensu Emmons. Out of this complex five distinct taxa have been segregated. These are *T. mentagrophytes* var. *erinacei*; *T.*

mentagrophytes var. *interdigitale*; *T. mentagrophytes* var. *mentagrophytes; T. mentagrophytes* var. *nodular*; *T. mentagrophytes* var. *quinckeanum.*

Gupta *et al.* (2001) reported in their work that segregation is necessary, due to the fact that treatment regimens may differ for different strains of dermatophytes. This statement was confirmed in this study when *T. concentricum* strain A was resistance to Flucamed and Ketoconazole, *T. concentricum* strain B was susceptible to the two drugs. The same observation was made with the strain A and B of *T. tonsurans,* while strain B showed susceptiblility towards Nystatin and Flucamed, strain A was resistance towards the two drugs. This is in contrast with the findings of Nweze *et al.* (2006), who reported that dermatophytes species (*T. tonsurans, T. soudanense, T mentagrophytes*; *T. violaceum*; *T. rubrum; M. audouinii* and *Epidermatophton flocossum*) present in Nigeria were susceptibile to Ketoconazole used in their work. Sugar and Liu (1996), Espinel-Ingoff *et al*. (2001), Fernandez- Torres *et al*. (2001), Espinel-Ingoff (2003) and Ellis *et al.* 2007, reported the resistance in some dermatophytes strains against some antifungal agents and their findings agree with the results in this study. According to Hayes and Wolf, (1990) resistance can be classified as either intrinstic or acquired. Intrinstic resistance allows all normal members of a species to tolerate a particular drug. In this case, a specific characteristic responsible for resistance is inherent to the species and has arisen through the process of evolution. Acquired resistance is a term used when a resistant strain emerges from a population that was previously drug-sensitive.

Resistance is inevitable (intrinsic or acquired) even at low frequency of gene mutation. If the selective pressure exerted by the constant use of antifungal drugs

continues, resistant strains will eventually become predominant in Lagos state, Nigeria. Unless the mutation that renders dermatophytes resistant also reduces their adaptability, the mutant strains will persist even in the absence of selective pressure of the drug. Gupta *et al.* (2001) reported that expansion of information on *in-vitro* susceptibility testing of dermatophytes will provide data that will help in developing new potent broad spectrum antifungal drug. In addition, Fernandez-Torres *et al.* (2003) reported that the various cases of imported dermatophytes infection across different countries and the variability in the properties of similar strains obtained across different geographical locations of the world make exchange of information on susceptibility data across different countries necessary. The data from the *in-vitro* sensitivity tests of eight antifungal drugs on the various species/strains isolated in this study will also be of help to choose appropriate antifungal agents for successful treatment.

According to Adekunle and Ikumapayi (2006), research on bioactive substances from plant sources has great scope and could lead to the development of antifungal agents that can combat fungal resistance. This present work showed that the oil extracted from *Azadirachta indica* and *Detarium senegalense* seeds had the potential of controlling these human pathogenic fungi. The choice of oil used in this study is as a result of the fact that oil serves as one of the bases for the production of most topical antifungal body cream. Therefore, if natural oil with antifungal potency could be discovered, then it will be used as an additive in the production of new effective topical antifungal creams. Adekunle and Ozonweke (2001) had previously reported the use of oil from the seed of *Glycine max* in the production of body and hair creams to reduce the fungal growth. From the report, they suggested that the oil extract might

be host specific due to variations in the zone of inhibition. The antifungal activity of the *Detarium senegalense* has not been reported in literature. The findings of the present study indicate that *D. Senegalense* seed oil has potential as an antifungal agent against fungi that are pathogenic to humans. This extracted oil act as a broad-spectrum agent that inhibites not only dermatophytes, but also non-dermatophytic fungi (*Albsidia corymbifera*, *Emericella nidulans* and *Penicillium aschersonia*) and some azole-resistant species. The findings on the antifungal activities of *D. senegalense* seed' oil used in this study was justified by the reports of the following authors. Keay *et al.* (1989) and Abreu and Relva, (2002) who reported that the stem barks, seeds, leaves and roots extracts of *D. senegalense* are widely used in herbal medicine in Nigeria. They are prepared as infusions or decoctions to treat veneral diseases, urogenital infections, hemorrhoids, rheumatism, stomach ache, intestinal worms and diarrhea. According to Okwu and Uchegbu, (2009), the isolation, characterization and structural elucidation of the anthocyanidine alkaloid; 2 – methoxyamine 3,4,5,7 – tetrahydroxyanthocyanidines from *D. senegalense* stem bark provide the reason for its antimicrobial activities. Wang *et al.* (1996) also showed that *D. senegalense* seed contains a large amount of water soluble, non-starch polysaccharide, (zyloglucan). This suggests that it has considerable commercial potential in food, drugs and pharmaceutical industries. Sowemimo *et al.* (2011) have reported on the chemical composition; proximate analysis and mineral content of the seed of *D. senegalense* which they believed probably contribute to the medicinal value and help in treatment of skin diseases.

The oil extracts of *Azadirachta indica* (neem) seeds was also effective in inhibiting the growth of a few number of dermatophytes (*Microsporum audouinii* and

Trichophyton concentricum strain A) and three other non-dermatophytic fungi. A lot of work has been done on the medicinal use of various parts of *Azadirachta indica*. Adesida and Taylor (1967) and National Research Council (1992) reported that *Azadirachta indica* leaf when rub on the body, protect it from fungal attack. Gahukar (1995) also reported that natural products derived from neem can inhibit and retard growth of bacteria and fungi. Biswas *et al.,* (2002) support this by stating in their report that every part of *Azadirachta indica* can be used as traditional medicine for household remedy against various human ailments. Dua *et al*. (2009) showed that neem seed oil formulation is effective in controlling mosquito larvae in different breeding sites under natural field conditions, and that these toxic effects might be due to the presence of aflatoxin and other toxic compounds in the oil. Koga *et al*. (1987), reported that mechanistic investigations indicate that *Azadirachta indica* (neem) seed oil uncouples mitochondrial oxidative phosphorylation, thus inhibiting the respiratory chain. They also report that it decreases intramitochondrial levels of acetyl CoA and acid-soluble CoA esters and reduces the mitochondrial ATP content. According to Adekunle and Uma (2005), *Azadirachta indica* leaf extract have fungistatic properties towards a number of opportunistic fungi. National Research Council (1992) reported oil extract from neem seed to be effective against certain human fungi, including *Trichophyton*, *Epidermophyton*, *Microsporum*, *Trichosporon*, *Geotricum* and *Candida*. The finding agree with the result from this present study as extracted seed oil of *Azadirachta indica* also possesses the ability to inhibit some human pathogenic fungi growth. Though, inhibiting just two species of dermatophytes might be as a result of the fact that the tested dermatophytes are of different strains.

Onadapo and Owonubi (1993) reported that tannins has antimicrobial activities and that at low concentration tannins can inhibit the growth of microorganisms and act as an antifungal agent at higher concentration by coagulating the protoplasm of the micro-organism. Reports by Barnabas and Nagarajan (1988) and Burapedjo and Bunchoo (1995) also revealed tannins to possess the ability to inhibit cell wall formation in fungi which could lead to the death of the organism even at low concentration. The result of the phytochemical test carried out in this study showed that the three extracted seed oils tested against the fungal isolates contained the same number of active ingredients (alkaloid, tannins, cardicglycosides and reducing compounds) but in different percentage. Therefore, the effectiveness (rate of growth inhibition) of the oils may probably be due to the quantity of the phytochemical compounds present in the oil extracts. However, it is important to point out that these natural oil extracts need to be further purified through antifungal activity guided fractionation to isolate and identify the compounds responsible for the biological activity.

The result from the sensitivity test with antiseptic soaps and detergents in the present study revealed that these two agents (antiseptic soaps and detergents) can help in eliminating some of the fungal inocula, through proper hygienic that is by bathing with antiseptic soaps and washing our clothes with detergents thereby reducing infection incident. Due to the fact that three of four types of antiseptic soaps were able to inhibit the growth of most fungal isolated tested against. This finding agrees with the reports of McMurray *et al*. (1998), Rook and Stanford (1998), McDonnell and Russsel, (1999) and Meade and Callahan (2000), in which they stated that 'Trichlorocarbanilide' a common ingredient in most antiseptic soap have

antimicrobial properties, which has the ability to kill or inhibit the growth of microbes like bacteria, virus and fungi. They further stated that the percentage of this chemical constituent also determine their effectiveness. '*Lifebuoy*' one of the anitseptic soaps used in this study was the least effective in its action and it was found to contain 1% 'Trichlorocarbanilide'. The three other antiseptic soaps used contain 5% 'Trichlorocarbanilide', which probably might be the reason behind their effectiveness. All the four detergents used were able to inhibit all the tested fungal isolates in this present study. In the studies by Stojanovic *et al.* (2001) and Stojanovic *et al*. (2011), they reported that 'Triclosan' which is the main ingredient in the production of most detergents have influence on the metabolic activities of fungi and disrupt fungi activities by lysis chemical composition of fungal walls. This is probably the reason for their abilities to inhibit the growth of all tested fungal isolates here.

Growth studies of the twenty five isolated fungi on six different media revealed that one percent (1%) peptone agar is the best medium for the cultivation of *Trichophyton* and *Epidermaphyton species.* This is due to the fact that the eight isolates of the genus *Trichophyton* and *Epidermaphyton* obtained their optimal growth on One percent (1%) peptone agar, compare to the other five media used in this study. The findings of Rebell and Tapliin (1970), de-Hoog *et al*. (1995) and Kane *et al.* (1997) in their various reports also agree with this fact. They reported that One percent (1%) peptone agar is a rich source of carbon and nitrogen content for the fungal growth. From the result, Sabouraud dextrose agar (SDA) was shown to be the best medium for the cultivation of *Microsporum* species and this agrees with the reports of Ajello (1977), Rippon (1988), and Kane *et al.* (1997). *Blastomyces dermatitidis* and *Exophiala*

dermatitidis had their optimal growth on Yeast extract agar (YEA), this also agrees with the reports of de-Hoog *et al*. (2006) and Dixon and Polak-Wyss (1991). According to Raper and Thom (1949) and Pitt (1979), Malt extract agar (MEA) is recommended for the cultivation of *Penicillium* species and this was found to agree with the finding in this present study as *Penicillium* species obtained the optimal growth on Malt extract agar (MEA).

From this growth study, it was also observed that some dermatophytes have the ability to change into different forms. This brings the subject of "Pleomorphism". Pleomorphism simply implies the ability of a species to change into various distinct forms in the life cycle. This phenomenon was experienced with the species *Epidermaphyton flucossum* and *Trichophyton tonsurans* in this study and this report agrees with the findings of Bistis, (1959) and Ellis *et al.* (2007). Bistis, (1959) in his work disagreed with the report of Chin and Knight (1957) on the fact that pleomorphism is brought about by changes in the environmental factors. Bistin, (1959) concluded that pleomorphism was as a result of nutrient constituent of the medium on which the dematophytes were grown.

SUMMARY OF FINDINGS

- This study revealed that there is significantly higher prevalence of dermatomycoses in female (53.81%) than in male (46.19%), also that body infection is the most frequent (49.4%) while face infection (3.6%) has the least frequency in the studied area.

- Twenty-five species of human pathogenic fungi were isolated, in which twelve different species/strains belong to the group dermatophytes (*Epidermophyton*, *Microsporum* and *Trichophyton*), two species belong to the systemic mycoses group, seven isolated species are of the opportunistic mycoses group and four are yet to be identified.

- This study reveals that no single DNA extraction protocol can isolate all groups of fungal DNA. The CTAB protocol, Modified CTAB protocol and the GNOME kit used in this work were only able to extract non-dermatophytes DNA. While only the Zymo DNA kit was able to isolate dermatophytes DNA.

- The resistance to the various antifungal drugs used in this study reveals the emerging threats of new strains of dermatophytes fungi in Lagos State, Nigeria.

- The *in-vitro* activities of various antiseptic soaps and detergents used against some isolated fungi in this study also indicated that these agents can help in eliminating some of these fungal inocula, thereby reducing infection incident.

- The crude phytochemical compounds (alkaloid, tannins, cardiac glycosides and reducing compounds) present in the extracted seed oils indicated that they possess antifungal properties.

- This study also shows that the extracted oil from some plant seeds possesses the active ingredients that could be employed in their purified form to cure dermatomycoses.

- For the cultivation of dermatophytes, the best medium to employ varies. While One percent (1%) peptone agar proved to be the best medium for the cultivation *Trichophyton* and *Epidermaphyton* species, *Microsporum* species attained their optimum growth on Sabouraud dextrose agar (SDA).

CONCLUSION

From the findings of this study, the taxa of dermatophytes present in Lagos State, Nigeria were isolated, identified, characterized and documented. This was made possible by employing the use of both the conventional laboratory methods and molecular (DNA sequencing of available strains of fungi) methods. The combination of the two methods brought to light the issue of the emergence of new strains of dermatophytes in Lagos state, Nigeria. In addition, the findings on DNA extraction methods which is the first step in all molecular studies showed that one DNA extraction method might not be able to extract all fungal DNA. The findings from the sensitivity test on the antifungal drugs revealed that since there is emergence of new species/strains of dermatophytes (*Epidermophyton, Microsporum and Trichophyton*), then resistance either intrinsic or acquired will continue to occur. Therefore, there will always be need for new effective broad spectrum antifungal drugs which will only be possible through exchange of documented details on *in-vitro* sensitivity test from different geographical locations. The natural oil extracted from *Detarium senegalense* seeds in this study has proved to be effectively useful in controlling dermatomycoses in man. Also, the study on the growth rate shows that for the cultivation of dermatophytes, the best media are One percent (1%) peptone agar and Sabouraud dextrose agar. The pleomorphism in some species/strains of isolated dermatophytes in Lagos state, Nigeria was also highlighted in this present study as a precaution to prevent wrong diagnoses of dermatomycoses in most of our mycological laboratories. Conclusively, it is therefore, important to employ the use of molecular methods which are accurate and reliable at identifying 'taxa' of human pathogens in order to prescribe appropriate antifungal therapy to treat dermatomycoses in our society.

CONTRIBUTIONS TO KNOWLEDGE

1. This study was able to isolate, identify, characterize and document DNA sequences of twelve species/strains of dermatophytes, and nine species/strains fungi that cause other types of mycoses present in Lagos state, Nigeria (this is a major contribution to mycology).

2. The pleomorphism in some species/strains of isolated dermatophytes in Lagos state, Nigeria was confirmed in this study. This will help in preventing wrong diagnoses of dermatophytes in most of our mycological laboratories.

3. The antifungal activities of the various antifungal drugs, antiseptic soaps and detergents in the Nigerian markets on the isolated dermatophytes were documented.

4. This study showed that natural oil extracted from *Detarium senegalense* seeds effectively inhibited growth of dermatophytes *in-vitro.*

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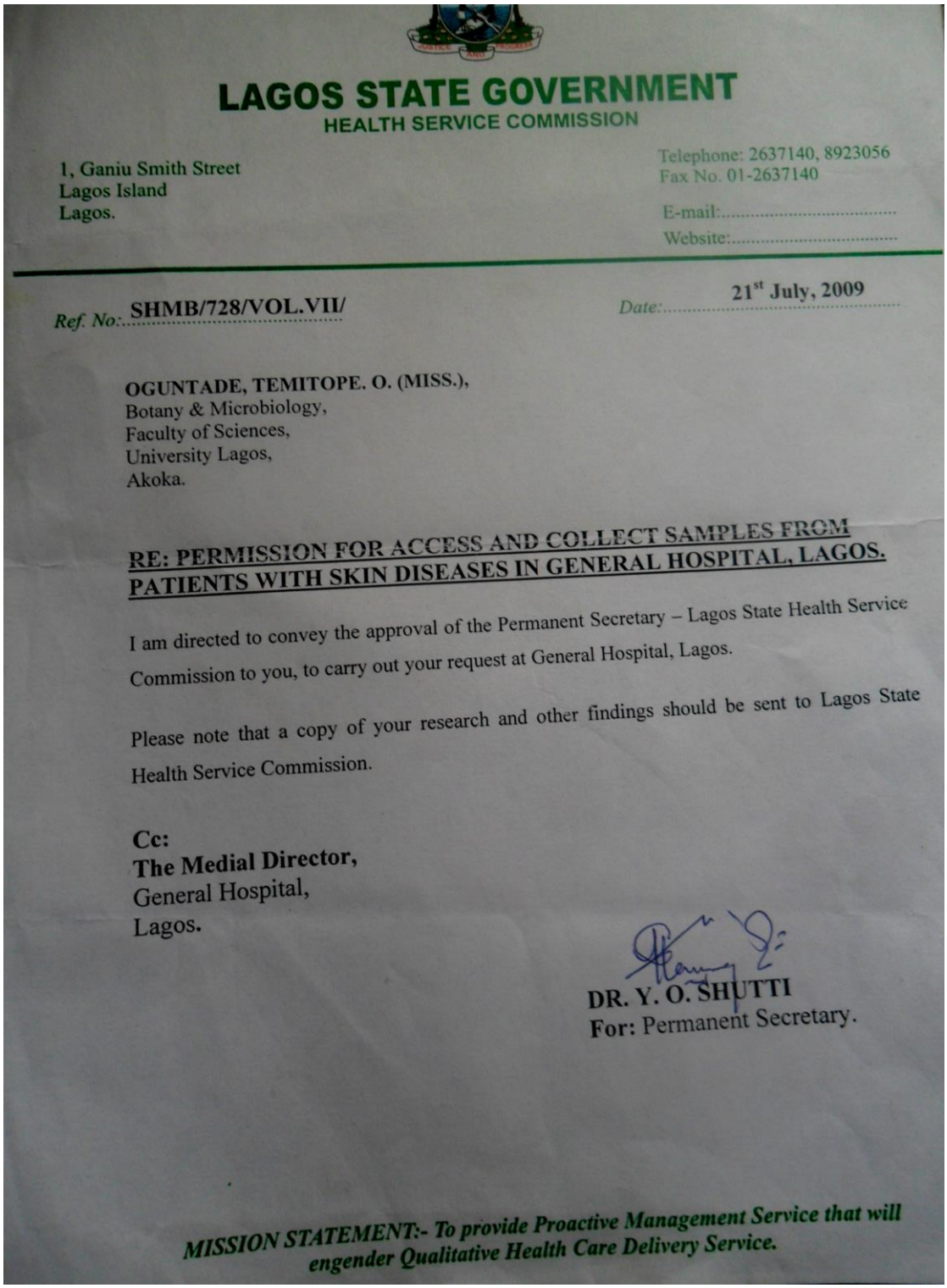
Appendix A: Permission letter from Lagos state Health Service Commission.



LAGOS STATE GOVERNMENT
HEALTH SERVICE COMMISSION

1, Ganiu Smith Street
Lagos Island
Lagos.

Telephone: 2637140, 8923056
Fax No. 01-2637140
E-mail:.....
Website:.....

Ref. No: SHMB/728/VOL.VII/ *Date:* 21st July, 2009

OGUNTADE, TEMITOPE. O. (MISS.),
Botany & Microbiology,
Faculty of Sciences,
University Lagos,
Akoka.

RE: PERMISSION FOR ACCESS AND COLLECT SAMPLES FROM PATIENTS WITH SKIN DISEASES IN GENERAL HOSPITAL, LAGOS.

I am directed to convey the approval of the Permanent Secretary – Lagos State Health Service Commission to you, to carry out your request at General Hospital, Lagos.

Please note that a copy of your research and other findings should be sent to Lagos State Health Service Commission.

Cc:
The Medial Director,
General Hospital,
Lagos.

DR. Y. O. SHUTTI
For: Permanent Secretary.

MISSION STATEMENT:- To provide Proactive Management Service that will engender Qualitative Health Care Delivery Service.

Appendix B: SAMPLES COLLECTION BETWEEN THE PERIODS OF AUGUST 2009 – JANUARY 2011

Sample	Place of Collection	Date	Body Part	Gender	Age	Varieties
C89	**CPHL**	**Aug. 2009**	Foot	Male	Adult	*T. rubrum*
C90	" " "	" " "	Neck	Female	Adult	*T. tonsurans (strain A)*
C91			Foot	Male	Adult	*E. fluccosum (strain)*
C92			Private Part	Male	Adult	*M. ferrugineum*
C93			Head	Female	11years	*T. soudanense*
C94			Foot	Female	Adult	*E. fluccosum (strain)*
C95			Belle	Male	Adult	*T. concentricum (strainA)*
L025	**LUTH**		Palm	Female	Adult	*M. nanum;* Unknown X
L024	" "		Nails	Male	Adult	*T. rubrum*
L003			Leg	Female	Adult	*E. dermatitidis; T.soudanense*
L004			Head & Leg	Male	Adult	*M. nanum*
C96	**CPHL**		Bottom		Adult	*T. concentricum (strainA)*
C97			Nail		Adult	*T. rubrum*
C98			Face		Adult	*B. dermatitidis*
C99			Leg		Adult	*E. dermatitidis; P. citrinum*
C100			Head		Adult	*T. soudanense*
C101	CPHL	Sept. 2009	Bottom	Female	Adult	*M. ferrugineum*
C102			Finger Nail	Female	Adult	*T. rubrum*
C103			Foot	Female	Adult	*E. fluccosum (strain)*
C104			Head &Back	Female	8years	*T. violaceum (strain)*
C105			Back	Male	9years	*T. mentagrophytes var.quinckeanun*
C106			Foot	Male	8years	*T. mentagrophytes var.quinckeanun*
C107			Hand	Male	6years	*T. concentricum (strainA)*
C108			Foot	Female	Adult	*T. rubrum*
C109			Foot	Female	Adult	*E. dermatitidis*
L012	LUTH	Sept. 2009	Finger nail	Female	Adult	*T. rubrum*
L013			Leg	Female	13years	*M. nanum*
L014			Neck	Male	Adult	*B. dermatitidis*
L015			Leg	Female	Adult	*T. tonsurans (strain A)*
L025			Buttom	Male	Adult	*T. violaceum (strain); A. Terreus*
L016	LUTH	Oct. 2009	Skin	Male	Adult	*T. mentagrophytes var.quinckeanun*
L019			Skin	Male	Adult	*T. tonsurans (strainA)*
L021			Nail	Male	Adult	*E. fluccosum (strain)*
L017			Skin	Female	Adult	*T. mentagrophytes var.quinckeanun; P. citrinum*
L018			Skin	Male	Adult	*T. tonsurans (*

						strain A)
L020			Head	Female	13years	*T. soudanense*
C112	CPHL	Oct. 2009	Foot	Female	Adult	*P. citrinum*
C113			Foot	Female	Adult	*T. violaceum (strain)*
C114			Finger	Female	15years	*T. rubrum*
C115			Bottom	Male	7years	*T. concentricum (strainA)*
C116			Foot	Female	Adult	*T. rubrum*
C110			Face	Male	Adult	*M. nanum*
C111			Finger	Female	10years	*T. tonsurans (strain B)*
L028	LUTH	Nov. 2009	Nail	Male	Adult	*T. rubrum*
L029			Skin	Female	Adult	*T. concentricum (strainA); M. racemosus*
A1			Body	Male	Adult	*T. mentagrophytes var.quinckeanun;*
A2			Body	Male	Adult	*T. tonsurans (strainA)*
A3			Body	Female	Adult	*T. tonsurans (strainA)*
A4			Body	Female	Adult	*T. mentagrophytes var.quinckeanun*
A5			Body	Male	Adult	*T. tonsurans (strain B)*
A6			Leg	Male	Adult	*T. tonsurans (strain A)*
A7			Nail	Male	Adult	*T. rubrum; A. Terreus*
C117	CPHL	Nov. 2009	Hand palm	Female	Adult	*A. corymbifera*
C118			Fingernail	Male	Adult	*E. fluccosum (strain)*
C119			Head	Male	2years	*B. dermatitidis; P. aschersonia*
C120			Foot	Female	16years	*A. terreus*
B1	LUTH	Dec. 2009	Back	Male	Adult	*E. fluccosum (strain)*
B2			Private part	Female	Adult	*T. soudanense*
B3			Body	Male	Adult	*T. mentagrophytes var. quinckeanun*
B4			Neck	Female	Adult	*T. concentricum (strainB)*
B5			Hand	Male	Adult	*T. tonsurans (strainA)*
B6			Fingers &toes nail	Female	Adult	*E. fluccosum (strain)*
B7			Skin	Female	Adult	*T. mentagrophytes var. quinckeanun*
B8			Bottom	Male	Adult	*T. soudanense*
B9			Skin & face	Female	Adult	*M. audouinii*
B10			Private part	Male	Adult	*E. fluccosum (strain)*
B11			Bottom & back and leg	Female	Adult	*T. violaceum (strain); Pencillium spp*
B12			Private part	Male	Adult	*B. dermatitidis*
B13			Body	Female	Adult	*T. tonsurans (strain A);* Unknown V
B14			Private part bottom	Male	Adult	*M. ferrugineum*
C121	CPHL	Dec. 2009	Foot	Male	Adult	*A. corymbifera*
C122			Foot	Female	Adult	*B. dermatitidis*
C123			Breast	Female	Adult	*T. tonsurans (strain A);* Unknown O
C124			Finger nail	Male	Adult	*T. violaceum (strain)*

C125			Back	Female	8years	*T. tonsurans (strain A)*
C126			Palm	Male	8years	*E. fluccosum (strain); P. citrinum*
C127			Palm	Female	Adult	*T. tonsurans (strain B); P. aschersonia*
C128			Foot	Female	Adult	*T. violaceum (strain)*
C129			Back	Male	Adult	*M. audouinii*
C130	CPHL	Jan. 2010	Face	Male	Adult	*T. concentricum)strainB);* Unknown V
C131			Hand	Male	Adult	*T. concentricum (strainB)*
C132			Belle	Female	Adult	*T. tonsurans (strainA)*
C133			Hand	Female	Adult	*E. fluccosum (strain);* Unknown T
C134			Lap	Male	5years	*T. violaceum (strain)*
C135			Palm	Male	5years	*T. violaceum (strain)*
C136			Foot	Female	5years	*E. dermatitidis*
C137			foot	Female	8years	*A. corymbifera*
C138			Leg	Female	Adult	*E. dermatitidis*
L005	LUTH	Jan. 2010	Skin	Male	Adult	*T. tonsurans (strainA)*
L006			Skin	Female	Adult	*M. audouinii; P. aschersonia*
L012			Hand	Male	Adult	*M. audouinii*
L013			Skin	Male	Adult	*T. mentagrophytes var. quinckeanun*
L014		Feb. 2010	Skin	Male	Adult	*B. dermatitidis*
L015			Skin	Male	Adult	*T. concentricum (strainA)*
L019			Skin	Male	Adult	*A. corymbifera; P. aschersonia*
L021			Nail	Female	Adult	*T. rubrum*
L023		March. 2010	Skin	Female	Adult	*E. dermatitidis*
L026			Skin	Female	Adult	*T. mentagrophytes var. quinckeanun*
L028			Nail	Female	Adult	*T. violaceum (strain)*
L029			Skin	Female	Adult	*T. mentagrophytes var. quinckeanun*
L030		April	Foot	Female	Adult	*T. soudanense*
L031			Skin	Female	Adult	*T. mentagrophytes var.quinckeanun; P. citrinum*
L033			Skin	Male	2years	*M. audouinii*
L035			Skin	Female	Adult	Unknown T; *P. citrinum*
L036			Private part	Female	Adult	*M. audouinii*
L037`			Skin	Female	Adult	*E. dermatitidis; P. citrinum*
C139	CPHL	Feb. 2010	Leg	Male	Adult	No isolate
C140			Arm	Male	Adult	*B. dermatitidis*
C141			Hand	Male	Adult	No isolate
C142			Belle	Female	Adult	*T. concentricum (strainA)*
C001			Hand	Male	Adult	Unknown X; *E. nidulans*
C002			Foot	Male	Adult	*T. violaceum (strain)*
C003			Head	Male	Adult	*M. audouinii*
C004			Finger nail	Female	Adult	*T. violaceum (differentstrain*
C005			Finger nail	Female	Adult	*T. rubrum*
C006	CPHL	March 2010	Belle	Female	Adult	*M. audouinii;* Unknown O
C007			Laps	Female	Adult	*T. mentagrophytes var. quinckeanun*
C008			Face	Female	Adult	*T. tonsurans (strain B)*
C009			Foot	Male	Adult	*M. nanum*

C010		Foot	Female	Adult	*E. fluccosum (strain)*
C011		Body	Male	Adult	*B. dermatitidis*
C012	April 2010	Hand palm	Female	Adult	*B. dermatitidis*
C013		Private part	Male	Adult	*T. soudanense*
C014		Foot	Female	Adult	*E. nidulans*
C015		Foot	Male	11years	*E. nidulans*
C016		Head	Male	Adult	*E. dermatitidis*
C021		Face	Female	Adult	No isolate
C022		Hand palm	Male	Adult	*E dermatitidis*
C023	May, 2010	Finger nail	Female	Adult	*T. rubrum*
C024		Finger nail	Female	Adult	*T. violaceum (strain)*
C025		Belle	Female	Adult	*T. concentricum (strain A); P citrinum*
C026		Leg	Male	Adult	*T. tonsurans (strain A)*
C027		Foot	Female	Adult	*E. nidulans*
C028		Finger	Male	Adult	*E. fluccosum (strain)*
C029		Chest	Female	Adult	*T. concentricum (strain A)*
C030		Hand	Male	Adult	*M. nanum*
C032		Hand	Male	Adult	*M. audouinii*
C033	June, 2010	Shoulder	Male	Adult	No isolate
C034		Back	Female	Adult	*T. mentagrophytes var.quinckeanun; A. corymbifera*
C035		Back	Female	Adult	*T. mentagrophytes var.quinckeanun;* Unknown X
C036		Head	Male	Adult	*T. mentagrophytes var.quinckeanun*
C037	July, 2010	Hand palm	Male	Adult	*B. dermatitidis*
C038		Laps	Female	12years	*M. ferrugineum*
C039		Head	Male	6years	*M. audouinii*
C040		Chest	Female	8years	No isolate
C041		Head	Male	11years	*M. audouinii*
C042		Hand	Male	Adult	*M. audouinii*
C043		Finger	Female	4years	*A. corymbifera*
C044	August, 2010	Hand	Female	Adult	*T.mentagrophytes var.quinckeanun*
C045		Hand	Male	10years	*M. audouinii*
C046		Hand	Female	18mnths	*M. audouinii*
C047		Finger nail	Female	Adult	*T. violaceum (strain)*
C048		Laps	Female	Adult	*T. rubrum*
C049		Leg	Female	Adult	*T. violaceum (strain)*
C050		Hand palm	Female	Adult	No isolate
C051		Foot	Female	Adult	*T. violaceum (strain); P.citrinum*
C052	September, 2010	Nail	Female	Adult	*T. violaceum (strain)*
C053		Nail	Female	Adult	*T. rubrum; P.aschersonia*
C054		Leg	Male	Adult	*T.concentricum (strain)*
C055		Hand palm	Female	12years	No isolate
C056		Belle	Female	Adult	*T.mentagrophytes var.quinckeanun;* Unknown V
C057		Shoulder back	Female	Adult	*T.mentagrophytes var.quinckeanun*
C058		Foot	Male	Adult	*A. terreus*
C059		Face	Female	Adult	*T. concentricum (strain)*
C060		Foot	female	Adult	*E. dermatitidis*
C061		Leg	Male	3years	*T. rubrum*
C062		Finger nail	Female	Female	*T.violaceum (strain)*
C063		Breast	Female	Adult	*M. ferrugineum;* Unknown V
C064		Foot	Female	Adult	Unknown T

C065			Back	Male	Adult	*T.mentagrophytes var.quinckeanun*
C066	October, 2010		Leg	Female	Adult	*T. tonsurans (strain B)*
C067			Hand palm	Female	6years	*T. rubrum*
C068			Head	Male	5years	*T. soudanense*
C069			Leg	Male	Adult	*T. tonsurans (strain B); E. nidulans*
C070			Finger	Female	Adult	*M. racemosus*
C071			Face	Male	Adult	*B. dermatitidis*
C072			Foot	Male	15years	*T. tonsurans (strain B)*
C073			Hand palm	Male	Adult	*B. dermatitidis*
C074	November, 2010		Hand	Male	16years	*M. audouinii*
C075			Hand	Female	Adult	*B. dermatitidis*
C076			Back of ear lope	Male	12years	*T. concentricum (strain); Absidia corymbifera*
C077			Hand	Male	6years	*M. racemosus; A.corymbifera*
C078			Face	Female	Adult	*T. tonsurans (strainA)*
C079			Leg	Female	8years	*T. soudanense*
C080	December, 2010		Hand	Female	Adult	*E. fluccosum (strain)*
C081			Foot	Female	Adult	*M. ferrugineum*
C082			Leg	Female	Adult	*T.concentricum (strainB)*
C083			Finger	Female	Adult	*E. fluccosum (different strain); Pencillium spp*
C084			arm	Female	Adult	*T.concentricum (strainB)*
C085			Head	Male	9years	*T. soudanense*
C086			Foot	Male	17years	*T. violaceum (strain)*
C087			Foot	Male	Adult	*T.concentricum (strainA)*
C088			Back	Male	Adult	*T.mentagrophytes var.quinckeanun; A. Corymbifera*
C089			Leg	Male	Adult	*T. tonsurans (strain A)*
L002	LUTH	March, 2010	Skin	Male	9years	*B. dermatitidis*
L017			Nail	Female	Adult	*T. rubrum*
L019			Skin	Female	Adult	*T.violaceum (strain);* Unknown X
L020			Nail	Female	Adult	*T.violaceum (strain); P.citrinum*
L021		June, 2010	Skin	Female	Adult	*T. tonsurans (strain A)*
L022			Nail	Male	7years	*E. dermatitidis*
L024			Skin	Female	Adult	*E. fluccosum (different strain);* Unknown T
L027			Skin	Male	Adult	*T.mentagrophytes var.quinckeanun*
L028		July, 2010	Nail	Female	Adult	*T. violaceum (different strain)*
L029			Skin	Female	Adult	*T.concentricum (strainA)*
L030			Skin	Male	9years	*T.concentricum (strainA)*
L032		August, 2010	Nail	Female	Adult	*E. dermatitidis*
L033			Skin	Female	Adult	*M. audouinii*
L034			Skin	Male	Adult	*M. audouinii*
L035			Skin	Female	Adult	*T.concentricum (strainA)*
L036			Nail	Female	Adult	*T. rubrum*
L040			Nail			*T. violaceum (strain)*
L041		September, 2010	Skin	Male	Adult	*M. audouinii*

L042			Skin	Female	Adult	*T.concentricum (strainA)*
L044			Skin	Male	Adult	*T.mentagrophytes var.quinckeanun*
L045			Skin	Male	Adult	Unknown V
L050			Skin	Male	6years	*M. audouinii*
L051		October, 2010	Skin	Female	Adult	*T. tonsurans (strain A)*
L059			Skin	Female	Adult	*T.mentagrophytes var.quinckeanun*
L066		November, 2010	Nail	Male	Adult	*T. violaceum (different strain); P. aschersonia*
L067			Skin	Male	Adult	*M. nanum; A. Terreus*
L068			Skin	Female	4month	*M. audouinii*
L069			Skin	Female	Adult	*T. concentricum (strainB)*
C01	**CPHL**	January, 2011	Back	Male	Adult	*M. audouinii*
C02			Hand	Female	14years	*T.mentagrophytes var.quinckeanun*
C03			Leg	Female	Adult	*A. terreus;* Unknown X
C04			Face	Female	Adult	*T.mentagrophytes var.quinckeanun*
C05			Back	Male	Adult	*T. concentricum (strainB);E.nidulans*
C06			Back	Male	Adult	*M. audouinii*
C07			Leg	Male	Adult	*M. ferrugineum; Pencillium spp*
C08			Leg	Male	Adult	*A. terreus*
C09			Finger	Female	Adult	*T. rubrum*
C010			Abdomen	Female	Adult	*M. audouinii*
C011			Private part	Female		*T.mentagrophytes var.quinckeanun*
C012			Foot	Male	Adult	*A. terreus*
C013			Foot	Female	Adult	*T. soudanense*
C014			Laps	Female	Adult	*T. tonsurans (strain B)*
C015			Hands	Male	Adult	*B. dermatitidis*
C016			Back	Male	Adult	*T.mentagrophytes var.quinckeanun;* Unknown V
C017			Hands	Female	Adult	Unknown O
C018			Foot	Female	Adult	*T.concentricum (strainB)*
L06	**LUTH**	January, 2011	Nail	Male	Adult	*T. rubrum*
L07			Skin	Male	Adult	*T.concentricum (strainB)*
L08			skin	Male	Adult	*T. violaceum (strain)*
L09			Skin	Male	Adult	*M. audouinii*
L010			Skin	Female	Adult	*T.mentagrophytes var.quinckeanun*
L011			Skin	Male	Adult	*T.mentagrophytes var.quinckeanun*
L012			Skin	Male	Adult	*T. tonsurans (strainA)*
L013			Skin	Female	Adult	*M. audouinii; P. aschersonia*

Appendix C: Media used in the study

Medium for the isolation	
Sabouraud Dextrose Agar	65.0 g
Cycloheximide	0.5 g
Chloramphenicol	50.0mg
Gentamicin	8.0mg
Distilled water	1000.0 ml
Sabouraud Dextrose Agar (Biolab):	
Pancreatic Digest of Casein .....5.0 g	
Peptic Digest of Animal Tissue 5.0	
Dextrose 40.0	
Agar 15.0	
65g/l	
Potato Dextrose Agar (LabM):	
Potato extract.....4.0	
Dextrose.....20.0	
Agar No.1.....15.0	
39g/l	
Malt Extract Agar (Biolab)	
Peptones.....5.0	
Malt extract 30.0	
Agar 15	
pH .....5,4 (approx.)	
50g/l	
Yeast extract agar (Merck)	
Yeast extract 5.0	
Glucose 10.0	
Agar-agar 20.0	
35g/l	
Nutrient agar (LabM)	
Peptone 5.0	
Beef extract 3.0	
Sodium Chloride 8.0	
Agar No.2 .....12.0	
28g/l	
One percent (1%) Peptone agar (Lab M)	
pH of 2% solution 7.2±0.2	
Total Nitrogen 12%±0.5	
Amino Nitrogen 5%±0.5	

Appendix D: One way analysis of variance (ANOVA) of the sensitivity activities of the isolated fungi against eight antifungal agents and three extracted seeds'oil

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Trichophyton concentricum strain A	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	21.7500	2.7124	.9590	19.4824	24.0176	18.00	26.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	5.00	8	13.1250	1.9594	.6928	11.4869	14.7631	10.00	16.00
	6.00	8	13.6250	2.3867	.8438	11.6297	15.6203	11.00	18.00
	7.00	8	11.2500	.8864	.3134	10.5089	11.9911	10.00	12.00
	8.00	8	11.3750	1.6850	.5957	9.9663	12.7837	10.00	15.00
	9.00	8	10.3750	.7440	.2631	9.7530	10.9970	10.00	12.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	12.1250	1.3562	.4795	10.9912	13.2588	10.00	14.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	11.9688	3.4655	.3537	11.2666	12.6709	10.00	26.00
Trichophyton tonsurans	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	13.5000	2.2678	.8018	11.6041	15.3959	10.00	16.00
	3.00	8	12.1250	1.4577	.5154	10.9063	13.3437	10.00	14.00
	4.00	8	18.8750	5.8904	2.0826	13.9505	23.7995	10.00	25.00
	5.00	8	27.5000	3.1623	1.1180	24.8563	30.1437	22.00	30.00
	6.00	8	25.1250	3.3568	1.1868	22.3187	27.9313	21.00	30.00
	7.00	8	23.2500	7.0051	2.4767	17.3936	29.1064	12.00	30.00
	8.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	15.0313	7.1118	.7258	13.5903	16.4722	10.00	30.00
T. mentagrophyton var. quinckeneanum	1.00	8	12.5000	1.7728	.6268	11.0179	13.9821	10.00	16.00
	2.00	8	19.7500	1.9821	.7008	18.0930	21.4070	16.00	22.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	5.00	8	15.3750	3.8149	1.3488	12.1857	18.5643	11.00	22.00
	6.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	7.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	8.00	8	11.5000	1.1952	.4226	10.5008	12.4992	10.00	14.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	11.5938	3.2072	.3273	10.9439	12.2436	10.00	22.00
Aspergillus terrus	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	12.0000	1.8516	.6547	10.4520	13.5480	10.00	15.00
	3.00	8	12.0000	1.5119	.5345	10.7361	13.2639	10.00	14.00
	4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	5.00	8	12.5000	2.0702	.7319	10.7693	14.2307	10.00	16.00
	6.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	7.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	8.00	8	19.5000	2.7775	.9820	17.1780	21.8220	16.00	24.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	11.3333	2.8825	.2942	10.7493	11.9174	10.00	24.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Trichophyton concentricum strain A	Between Groups	980.531	11	89.139	46.689	.000
	Within Groups	160.375	84	1.909		
	Total	1140.906	95			
Trichophyton tonsurans	Between Groups	4018.781	11	365.344	39.038	.000
	Within Groups	786.125	84	9.359		
	Total	4804.906	95			
T. mentagrophyton var. quinckeneanum	Between Groups	815.781	11	74.162	38.603	.000
	Within Groups	161.375	84	1.921		
	Total	977.156	95			
Aspergillus terrus	Between Groups	665.333	11	60.485	40.974	.000
	Within Groups	124.000	84	1.476		
	Total	789.333	95			

Post Hoc Tests

Homogeneous Subsets

Trichophyton concentricum strain A

Duncan[a]

		Subset for alpha = .05				
VAR00001	N	1	2	3	4	5
1.00	8	10.0000				
3.00	8	10.0000				
4.00	8	10.0000				
10.00	8	10.0000				
12.00	8	10.0000				
9.00	8	10.3750				
7.00	8	11.2500	11.2500			
8.00	8	11.3750	11.3750			
11.00	8		12.1250	12.1250		
5.00	8			13.1250	13.1250	
6.00	8				13.6250	
2.00	8					21.7500
Sig.		.093	.237	.151	.471	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Trichophyton tonsurans

Duncan[a]

		Subset for alpha = .05			
VAR00001	N	1	2	3	4
1.00	8	10.0000			
8.00	8	10.0000			
9.00	8	10.0000			
10.00	8	10.0000			
11.00	8	10.0000			
12.00	8	10.0000			
3.00	8	12.1250			
2.00	8	13.5000			
4.00	8		18.8750		
7.00	8			23.2500	
6.00	8			25.1250	25.1250
5.00	8				27.5000
Sig.		.052	1.000	.224	.124

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

T. mentagrophyton var. quinckeneanum

Duncan[a]

VAR00001	N	Subset for alpha = .05			
		1	2	3	4
3.00	8	10.0000			
4.00	8	10.0000			
6.00	8	10.0000			
7.00	8	10.0000			
9.00	8	10.0000			
10.00	8	10.0000			
11.00	8	10.0000			
12.00	8	10.0000			
8.00	8	11.5000	11.5000		
1.00	8		12.5000		
5.00	8			15.3750	
2.00	8				19.7500
Sig.		.070	.153	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Aspergillus terrus

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
1.00	8	10.0000		
4.00	8	10.0000		
6.00	8	10.0000		
7.00	8	10.0000		
9.00	8	10.0000		
10.00	8	10.0000		
11.00	8	10.0000		
12.00	8	10.0000		
2.00	8		12.0000	
3.00	8		12.0000	
5.00	8		12.5000	
8.00	8			19.5000
Sig.		1.000	.443	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Blastomyces dermatitides	1.00	8	19.5000	4.5040	1.5924	15.7346	23.2654	10.00	24.00
	2.00	8	13.7500	3.2842	1.1611	11.0044	16.4956	10.00	18.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	5.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	6.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	7.00	8	11.3750	1.3025	.4605	10.2861	12.4639	10.00	14.00
	8.00	8	11.5000	.5345	.1890	11.0531	11.9469	11.00	12.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	11.3438	3.1215	.3186	10.7113	11.9762	10.00	24.00
Trichophyton tonsuran strain B	1.00	8	13.0000	2.8284	1.0000	10.6354	15.3646	10.00	18.00
	2.00	8	18.5000	4.3753	1.5469	14.8422	22.1578	12.00	24.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	17.0000	4.4078	1.5584	13.3150	20.6850	10.00	22.00
	5.00	8	14.2500	2.7124	.9590	11.9824	16.5176	10.00	18.00
	6.00	8	18.7500	7.0051	2.4767	12.8936	24.6064	10.00	30.00
	7.00	8	14.6250	2.7742	.9808	12.3057	16.9443	11.00	20.00
	8.00	8	11.1250	.8345	.2950	10.4273	11.8227	10.00	12.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	13.1042	4.3949	.4486	12.2137	13.9947	10.00	30.00
Exophilla dermatitides	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	3.00	8	14.8750	3.1820	1.1250	12.2148	17.5352	11.00	20.00
	4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	5.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	6.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	7.00	8	18.7500	6.1354	2.1692	13.6207	23.8793	12.00	30.00
	8.00	8	18.2500	4.0620	1.4361	14.8541	21.6459	12.00	24.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	11.0000	.7559	.2673	10.3680	11.6320	10.00	12.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	11.9063	3.9205	.4001	11.1119	12.7006	10.00	30.00
Microsporum audouinii	1.00	8	12.7500	4.2678	1.5089	9.1820	16.3180	10.00	22.00
	2.00	8	14.7500	2.8158	.9955	12.3960	17.1040	10.00	18.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	13.6250	2.6152	.9246	11.4386	15.8114	10.00	18.00
	5.00	8	12.2500	1.6690	.5901	10.8546	13.6454	10.00	14.00
	6.00	8	19.6250	6.3682	2.2515	14.3011	24.9489	11.00	30.00
	7.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	8.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	9.00	8	10.8750	.8345	.2950	10.1773	11.5727	10.00	12.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	11.6250	1.3025	.4605	10.5361	12.7139	10.00	14.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	12.1250	3.6599	.3735	11.3834	12.8666	10.00	30.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Blastomyces dermatitides	Between Groups	694.281	11	63.116	22.914	.000
	Within Groups	231.375	84	2.754		
	Total	925.656	95			
Trichophyton tonsuran strain B	Between Groups	1055.208	11	95.928	10.334	.000
	Within Groups	779.750	84	9.283		
	Total	1834.958	95			
Exophilla dermatitides	Between Groups	1006.281	11	91.480	16.930	.000
	Within Groups	453.875	84	5.403		
	Total	1460.156	95			
Microsporum audouinii	Between Groups	721.500	11	65.591	9.999	.000
	Within Groups	551.000	84	6.560		
	Total	1272.500	95			

Homogeneous Subsets

Blastomyces dermatitides

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
3.00	8	10.0000		
4.00	8	10.0000		
5.00	8	10.0000		
6.00	8	10.0000		
9.00	8	10.0000		
10.00	8	10.0000		
11.00	8	10.0000		
12.00	8	10.0000		
7.00	8	11.3750		
8.00	8	11.5000		
2.00	8		13.7500	
1.00	8			19.5000
Sig.		.135	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Trichophyton tonsuran strain B

Duncan[a]

		Subset for alpha = .05				
VAR00001	N	1	2	3	4	5
3.00	8	10.0000				
9.00	8	10.0000				
10.00	8	10.0000				
11.00	8	10.0000				
12.00	8	10.0000				
8.00	8	11.1250	11.1250			
1.00	8	13.0000	13.0000	13.0000		
5.00	8		14.2500	14.2500	14.2500	
7.00	8			14.6250	14.6250	
4.00	8				17.0000	17.0000
2.00	8					18.5000
6.00	8					18.7500
Sig.		.093	.055	.320	.091	.284

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Exophilla dermatitides

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
1.00	8	10.0000		
2.00	8	10.0000		
4.00	8	10.0000		
5.00	8	10.0000		
6.00	8	10.0000		
9.00	8	10.0000		
10.00	8	10.0000		
12.00	8	10.0000		
11.00	8	11.0000		
3.00	8		14.8750	
8.00	8			18.2500
7.00	8			18.7500
Sig.		.477	1.000	.668

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Microsporum audouinii

Duncan[a]

VAR00001	N	Subset for alpha = .05			
		1	2	3	4
3.00	8	10.0000			
7.00	8	10.0000			
8.00	8	10.0000			
10.00	8	10.0000			
12.00	8	10.0000			
9.00	8	10.8750	10.8750		
11.00	8	11.6250	11.6250		
5.00	8	12.2500	12.2500	12.2500	
1.00	8	12.7500	12.7500	12.7500	
4.00	8		13.6250	13.6250	
2.00	8			14.7500	
6.00	8				19.6250
Sig.		.072	.057	.077	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Penicillium citrium	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	18.5000	5.8310	2.0616	13.6252	23.3748	12.00	30.00
	3.00	8	17.7500	5.5997	1.9798	13.0685	22.4315	12.00	30.00
	4.00	8	25.0000	5.3452	1.8898	20.5313	29.4687	20.00	30.00
	5.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	6.00	8	23.0000	5.0143	1.7728	18.8080	27.1920	20.00	32.00
	7.00	8	22.2500	5.5997	1.9798	17.5685	26.9315	14.00	30.00
	8.00	8	30.5000	.9258	.3273	29.7260	31.2740	30.00	32.00
	9.00	8	10.2500	.7071	.2500	9.6588	10.8412	10.00	12.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	16.4375	7.8523	.8014	14.8465	18.0285	10.00	32.00
Trichophyton	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	3.00	8	13.5000	2.5635	.9063	11.3569	15.6431	10.00	16.00
	4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	5.00	8	14.0000	2.6186	.9258	11.8108	16.1892	10.00	18.00
	6.00	8	13.5000	2.7255	.9636	11.2214	15.7786	10.00	18.00
	7.00	8	19.8750	5.8172	2.0567	15.0117	24.7383	12.00	30.00
	8.00	8	12.5000	2.0702	.7319	10.7693	14.2307	10.00	16.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	11.9479	3.5581	.3632	11.2270	12.6689	10.00	30.00
Trichophyton soudanense	1.00	8	13.7500	3.2404	1.1456	11.0410	16.4590	10.00	20.00
	2.00	8	16.0000	2.6186	.9258	13.8108	18.1892	12.00	20.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	5.00	8	14.3750	2.2638	.8004	12.4824	16.2676	11.00	18.00
	6.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	7.00	8	15.3750	2.9731	1.0511	12.8894	17.8606	11.00	20.00
	8.00	8	12.5000	1.3093	.4629	11.4054	13.5946	11.00	14.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	12.2500	1.5811	.5590	10.9281	13.5719	10.00	14.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	12.0208	2.7758	.2833	11.4584	12.5833	10.00	20.00
Microsporum ferugineum	1.00	8	12.3750	1.9955	.7055	10.7067	14.0433	10.00	15.00
	2.00	8	13.5000	2.9761	1.0522	11.0119	15.9881	10.00	18.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	12.0000	1.7728	.6268	10.5179	13.4821	10.00	14.00
	5.00	8	13.3750	2.1998	.7778	11.5359	15.2141	10.00	16.00
	6.00	8	13.0000	2.7255	.9636	10.7214	15.2786	10.00	18.00
	7.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	8.00	8	13.1250	2.1671	.7662	11.3132	14.9368	10.00	16.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	13.1250	1.7269	.6105	11.6813	14.5687	11.00	16.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	11.7083	2.2144	.2260	11.2597	12.1570	10.00	18.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Penicillium citrium	Between Groups	4795.125	11	435.920	34.463	.000
	Within Groups	1062.500	84	12.649		
	Total	5857.625	95			
Trichophyton	Between Groups	789.865	11	71.806	14.609	.000
	Within Groups	412.875	84	4.915		
	Total	1202.740	95			
Trichophyton soudanense	Between Groups	483.208	11	43.928	14.834	.000
	Within Groups	248.750	84	2.961		
	Total	731.958	95			
Microsporum ferugineum	Between Groups	214.333	11	19.485	6.508	.000
	Within Groups	251.500	84	2.994		
	Total	465.833	95			

Homogeneous Subsets

Penicillium citrium

Duncan[a]

		Subset for alpha = .05			
VAR00001	N	1	2	3	4
1.00	8	10.0000			
5.00	8	10.0000			
10.00	8	10.0000			
11.00	8	10.0000			
12.00	8	10.0000			
9.00	8	10.2500			
3.00	8		17.7500		
2.00	8		18.5000		
7.00	8			22.2500	
6.00	8			23.0000	
4.00	8			25.0000	
8.00	8				30.5000
Sig.		.907	.674	.148	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Trichophyton

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
1.00	8	10.0000		
2.00	8	10.0000		
4.00	8	10.0000		
9.00	8	10.0000		
10.00	8	10.0000		
11.00	8	10.0000		
12.00	8	10.0000		
8.00	8	12.5000	12.5000	
3.00	8		13.5000	
6.00	8		13.5000	
5.00	8		14.0000	
7.00	8			19.8750
Sig.		.056	.224	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Trichophyton soudanense

Duncan[a]

VAR00001	N	Subset for alpha = .05			
		1	2	3	4
3.00	8	10.0000			
4.00	8	10.0000			
6.00	8	10.0000			
9.00	8	10.0000			
10.00	8	10.0000			
12.00	8	10.0000			
11.00	8		12.2500		
8.00	8		12.5000		
1.00	8		13.7500	13.7500	
5.00	8			14.3750	14.3750
7.00	8			15.3750	15.3750
2.00	8				16.0000
Sig.		1.000	.103	.077	.077

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Microsporum ferugineum

Duncan[a]

VAR00001	N	Subset for alpha = .05	
		1	2
3.00	8	10.0000	
7.00	8	10.0000	
9.00	8	10.0000	
10.00	8	10.0000	
12.00	8	10.0000	
4.00	8		12.0000
1.00	8		12.3750
6.00	8		13.0000
8.00	8		13.1250
11.00	8		13.1250
5.00	8		13.3750
2.00	8		13.5000
Sig.		1.000	.140

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Penicillium aschersonia	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	3.00	8	18.0000	4.0000	1.4142	14.6559	21.3441	12.00	24.00
	4.00	8	14.1250	2.5877	.9149	11.9616	16.2884	11.00	18.00
	5.00	8	13.0000	1.8516	.6547	11.4520	14.5480	11.00	16.00
	6.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	7.00	8	19.5000	3.3381	1.1802	16.7093	22.2907	14.00	24.00
	8.00	8	19.5000	4.1057	1.4516	16.0675	22.9325	12.00	24.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	12.2500	2.6592	.9402	10.0268	14.4732	10.00	18.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	13.0313	4.2931	.4382	12.1614	13.9011	10.00	24.00
Penicillium species	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	3.00	8	17.7500	2.4928	.8814	15.6659	19.8341	14.00	22.00
	4.00	8	13.6250	2.3867	.8438	11.6297	15.6203	11.00	18.00
	5.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	6.00	8	14.5000	2.0702	.7319	12.7693	16.2307	12.00	18.00
	7.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	8.00	8	19.7500	2.9155	1.0308	17.3126	22.1874	16.00	24.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	12.1354	3.6176	.3692	11.4024	12.8684	10.00	24.00
Unknown O	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	13.2500	1.8323	.6478	11.7182	14.7818	10.00	16.00
	5.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	6.00	8	15.5000	3.3381	1.1802	12.7093	18.2907	12.00	22.00
	7.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	8.00	8	17.0000	3.2071	1.1339	14.3188	19.6812	12.00	22.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	13.0000	2.1381	.7559	11.2125	14.7875	10.00	16.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	11.5625	2.8314	.2890	10.9888	12.1362	10.00	22.00
Epidermophyton floccosum	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	3.00	8	17.7500	2.4928	.8814	15.6659	19.8341	14.00	22.00
	4.00	8	13.6250	2.3867	.8438	11.6297	15.6203	11.00	18.00
	5.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	6.00	8	13.7500	2.4349	.8609	11.7144	15.7856	11.00	18.00
	7.00	8	21.0000	5.4511	1.9272	16.4428	25.5572	14.00	30.00
	8.00	8	27.5000	4.6291	1.6366	23.6300	31.3700	20.00	30.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	13.6354	5.9207	.6043	12.4358	14.8351	10.00	30.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Penicillium aschersonia	Between Groups	1322.531	11	120.230	23.576	.000
	Within Groups	428.375	84	5.100		
	Total	1750.906	95			
Penicillium species	Between Groups	1070.365	11	97.306	47.281	.000
	Within Groups	172.875	84	2.058		
	Total	1243.240	95			
Unknown O	Between Groups	556.125	11	50.557	20.666	.000
	Within Groups	205.500	84	2.446		
	Total	761.625	95			
Epidermophyton floccosum	Between Groups	2847.365	11	258.851	45.029	.000
	Within Groups	482.875	84	5.749		
	Total	3330.240	95			

Homogeneous Subsets

Penicillium aschersonia

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
1.00	8	10.0000		
2.00	8	10.0000		
6.00	8	10.0000		
9.00	8	10.0000		
10.00	8	10.0000		
12.00	8	10.0000		
11.00	8	12.2500	12.2500	
5.00	8		13.0000	
4.00	8		14.1250	
3.00	8			18.0000
7.00	8			19.5000
8.00	8			19.5000
Sig.		.089	.120	.215

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Penicillium species

Duncan[a]

		Subset for alpha = .05			
VAR00001	N	1	2	3	4
1.00	8	10.0000			
2.00	8	10.0000			
5.00	8	10.0000			
7.00	8	10.0000			
9.00	8	10.0000			
10.00	8	10.0000			
11.00	8	10.0000			
12.00	8	10.0000			
4.00	8		13.6250		
6.00	8		14.5000		
3.00	8			17.7500	
8.00	8				19.7500
Sig.		1.000	.226	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Unknown O

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
1.00	8	10.0000		
2.00	8	10.0000		
3.00	8	10.0000		
5.00	8	10.0000		
7.00	8	10.0000		
9.00	8	10.0000		
10.00	8	10.0000		
12.00	8	10.0000		
11.00	8		13.0000	
4.00	8		13.2500	
6.00	8			15.5000
8.00	8			17.0000
Sig.		1.000	.750	.059

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Epidermophyton floccosum

Duncan[a]

VAR00001	N	Subset for alpha = .05				
		1	2	3	4	5
1.00	8	10.0000				
2.00	8	10.0000				
5.00	8	10.0000				
9.00	8	10.0000				
10.00	8	10.0000				
11.00	8	10.0000				
12.00	8	10.0000				
4.00	8		13.6250			
6.00	8		13.7500			
3.00	8			17.7500		
7.00	8				21.0000	
8.00	8					27.5000
Sig.		1.000	.917	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Mucor racemosus	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	14.6250	3.3354	1.1792	11.8365	17.4135	11.00	22.00
	3.00	8	14.1250	2.4165	.8543	12.1048	16.1452	11.00	18.00
	4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	5.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	6.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	7.00	8	11.7500	1.2817	.4532	10.6784	12.8216	10.00	14.00
	8.00	8	11.7500	1.5811	.5590	10.4281	13.0719	10.00	14.00
	9.00	8	11.3750	1.3025	.4605	10.2861	12.4639	10.00	14.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	11.1354	2.0704	.2113	10.7159	11.5549	10.00	22.00
Trichophyton concentricum strain B	1.00	8	13.1250	2.2321	.7892	11.2589	14.9911	10.00	16.00
	2.00	8	15.7500	2.9155	1.0308	13.3126	18.1874	12.00	20.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	12.5000	2.0000	.7071	10.8280	14.1720	10.00	16.00
	5.00	8	11.1250	.6409	.2266	10.5892	11.6608	10.00	12.00
	6.00	8	19.7500	5.8002	2.0507	14.9009	24.5991	12.00	28.00
	7.00	8	16.0000	3.3806	1.1952	13.1737	18.8263	12.00	22.00
	8.00	8	12.7500	2.3755	.8399	10.7641	14.7359	10.00	16.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	13.0000	1.8516	.6547	11.4520	14.5480	10.00	16.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	12.8333	3.7238	.3801	12.0788	13.5878	10.00	28.00
Absidia corymbifera	1.00	8	20.2500	2.7124	.9590	17.9824	22.5176	16.00	24.00
	2.00	8	18.5000	4.6291	1.6366	14.6300	22.3700	12.00	24.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	5.00	8	15.3750	2.0659	.7304	13.6479	17.1021	14.00	20.00
	6.00	8	16.0000	3.2071	1.1339	13.3188	18.6812	12.00	22.00
	7.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	8.00	8	11.1250	1.3562	.4795	9.9912	12.2588	10.00	14.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	11.3750	.9161	.3239	10.6091	12.1409	10.00	13.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	12.7188	4.0723	.4156	11.8936	13.5439	10.00	24.00
Unknown T	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	5.00	8	14.2500	1.9821	.7008	12.5930	15.9070	12.00	18.00
	6.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	7.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	8.00	8	16.5000	3.3381	1.1802	13.7093	19.2907	12.00	22.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	10.8958	2.3192	.2367	10.4259	11.3657	10.00	22.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Mucor racemosus	Between Groups	247.615	11	22.510	11.846	.000
	Within Groups	159.625	84	1.900		
	Total	407.240	95			
Trichophyton concentricum strain B	Between Groups	813.083	11	73.917	12.313	.000
	Within Groups	504.250	84	6.003		
	Total	1317.333	95			
Absidia corymbifera	Between Groups	1253.281	11	113.935	29.711	.000
	Within Groups	322.125	84	3.835		
	Total	1575.406	95			
Unknown T	Between Groups	405.458	11	36.860	29.348	.000
	Within Groups	105.500	84	1.256		
	Total	510.958	95			

Homogeneous Subsets

Mucor racemosus

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
1.00	8	10.0000		
4.00	8	10.0000		
5.00	8	10.0000		
6.00	8	10.0000		
10.00	8	10.0000		
11.00	8	10.0000		
12.00	8	10.0000		
9.00	8	11.3750	11.3750	
7.00	8		11.7500	
8.00	8		11.7500	
3.00	8			14.1250
2.00	8			14.6250
Sig.		.092	.612	.470

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Trichophyton concentricum strain B

Duncan[a]

		Subset for alpha = .05			
VAR00001	N	1	2	3	4
3.00	8	10.0000			
9.00	8	10.0000			
10.00	8	10.0000			
12.00	8	10.0000			
5.00	8	11.1250	11.1250		
4.00	8	12.5000	12.5000		
8.00	8	12.7500	12.7500		
11.00	8		13.0000		
1.00	8		13.1250		
2.00	8			15.7500	
7.00	8			16.0000	
6.00	8				19.7500
Sig.		.054	.151	.839	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Absidia corymbifera

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
3.00	8	10.0000		
4.00	8	10.0000		
7.00	8	10.0000		
9.00	8	10.0000		
10.00	8	10.0000		
12.00	8	10.0000		
8.00	8	11.1250		
11.00	8	11.3750		
5.00	8		15.3750	
6.00	8		16.0000	
2.00	8			18.5000
1.00	8			20.2500
Sig.		.239	.525	.077

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Unknown T

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
1.00	8	10.0000		
2.00	8	10.0000		
3.00	8	10.0000		
4.00	8	10.0000		
6.00	8	10.0000		
7.00	8	10.0000		
9.00	8	10.0000		
10.00	8	10.0000		
11.00	8	10.0000		
12.00	8	10.0000		
5.00	8		14.2500	
8.00	8			16.5000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Microsporum nanum	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	5.00	8	14.2500	1.9821	.7008	12.5930	15.9070	12.00	18.00
	6.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	7.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	8.00	8	16.5000	3.3381	1.1802	13.7093	19.2907	12.00	22.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	10.8958	2.3192	.2367	10.4259	11.3657	10.00	22.00
Unknown V	1.00	8	14.0000	1.7728	.6268	12.5179	15.4821	11.00	16.00
	2.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	3.00	8	11.5000	1.3093	.4629	10.4054	12.5946	10.00	14.00
	4.00	8	11.8750	1.4577	.5154	10.6563	13.0937	10.00	14.00
	5.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	6.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	7.00	8	13.2500	3.0589	1.0815	10.6927	15.8073	10.00	18.00
	8.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	10.8854	1.7703	.1807	10.5267	11.2441	10.00	18.00
Trichophyton violaceum strain	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	3.00	8	17.5000	2.5635	.9063	15.3569	19.6431	14.00	22.00
	4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	5.00	8	12.2500	1.1650	.4119	11.2761	13.2239	11.00	14.00
	6.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	7.00	8	20.7500	3.3700	1.1915	17.9326	23.5674	16.00	26.00
	8.00	8	11.8750	1.2464	.4407	10.8330	12.9170	10.00	14.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	11.8646	3.6350	.3710	11.1281	12.6011	10.00	26.00
Unknown X	1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	2.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	3.00	8	16.5000	2.5635	.9063	14.3569	18.6431	14.00	20.00
	4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	5.00	8	12.8750	1.2464	.4407	11.8330	13.9170	11.00	14.00
	6.00	8	15.7500	2.7124	.9590	13.4824	18.0176	12.00	20.00
	7.00	8	13.8750	3.6425	1.2878	10.8298	16.9202	10.00	22.00
	8.00	8	18.0000	3.3806	1.1952	15.1737	20.8263	12.00	22.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	11.00	8	12.3750	1.5059	.5324	11.1160	13.6340	10.00	14.00
	12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	96	12.4479	3.3525	.3422	11.7686	13.1272	10.00	22.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Microsporum nanum	Between Groups	405.458	11	36.860	29.348	.000
	Within Groups	105.500	84	1.256		
	Total	510.958	95			
Unknown V	Between Groups	183.365	11	16.670	12.243	.000
	Within Groups	114.375	84	1.362		
	Total	297.740	95			
Trichophyton violaceum strain	Between Groups	1109.365	11	100.851	58.074	.000
	Within Groups	145.875	84	1.737		
	Total	1255.240	95			
Unknown X	Between Groups	770.615	11	70.056	19.805	.000
	Within Groups	297.125	84	3.537		
	Total	1067.740	95			

Homogeneous Subsets

Microsporum nanum

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
1.00	8	10.0000		
2.00	8	10.0000		
3.00	8	10.0000		
4.00	8	10.0000		
6.00	8	10.0000		
7.00	8	10.0000		
9.00	8	10.0000		
10.00	8	10.0000		
11.00	8	10.0000		
12.00	8	10.0000		
5.00	8		14.2500	
8.00	8			16.5000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Unknown V

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
2.00	8	10.0000		
5.00	8	10.0000		
6.00	8	10.0000		
8.00	8	10.0000		
9.00	8	10.0000		
10.00	8	10.0000		
11.00	8	10.0000		
12.00	8	10.0000		
3.00	8		11.5000	
4.00	8		11.8750	
7.00	8			13.2500
1.00	8			14.0000
Sig.		1.000	.522	.202

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Trichophyton violaceum strain

Duncan[a]

VAR00001	N	Subset for alpha = .05			
		1	2	3	4
1.00	8	10.0000			
2.00	8	10.0000			
4.00	8	10.0000			
6.00	8	10.0000			
9.00	8	10.0000			
10.00	8	10.0000			
11.00	8	10.0000			
12.00	8	10.0000			
8.00	8		11.8750		
5.00	8		12.2500		
3.00	8			17.5000	
7.00	8				20.7500
Sig.		1.000	.571	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Unknown X

Duncan[a]

VAR00001	N	Subset for alpha = .05			
		1	2	3	4
1.00	8	10.0000			
2.00	8	10.0000			
4.00	8	10.0000			
9.00	8	10.0000			
10.00	8	10.0000			
12.00	8	10.0000			
11.00	8		12.3750		
5.00	8		12.8750		
7.00	8		13.8750		
6.00	8			15.7500	
3.00	8			16.5000	16.5000
8.00	8				18.0000
Sig.		1.000	.136	.427	.114

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Descriptives

Emerialla nidulans

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
1.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
2.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
4.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
5.00	8	15.7500	2.9155	1.0308	13.3126	18.1874	12.00	20.00
6.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
7.00	8	12.8750	1.8077	.6391	11.3637	14.3863	10.00	16.00
8.00	8	21.7500	6.3640	2.2500	16.4296	27.0704	14.00	30.00
9.00	8	11.8750	1.2464	.4407	10.8330	12.9170	10.00	14.00
10.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
11.00	8	12.6250	1.5980	.5650	11.2890	13.9610	10.00	14.00
12.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
Total	96	12.0729	3.9742	.4056	11.2677	12.8782	10.00	30.00

ANOVA

Emerialla nidulans

	Sum of Squares	df	Mean Square	F	Sig.
Betw een Groups	1105.865	11	100.533	21.400	.000
Within Groups	394.625	84	4.698		
Total	1500.490	95			

Post Hoc Tests

Homogeneous Subsets

Emerialla nidulans

Duncan[a]

		Subset for alpha = .05			
VAR00001	N	1	2	3	4
1.00	8	10.0000			
2.00	8	10.0000			
3.00	8	10.0000			
4.00	8	10.0000			
6.00	8	10.0000			
10.00	8	10.0000			
12.00	8	10.0000			
9.00	8	11.8750	11.8750		
11.00	8		12.6250		
7.00	8		12.8750		
5.00	8			15.7500	
8.00	8				21.7500
Sig.		.146	.390	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Appendix E: One way analysis of variance (ANOVA) of the sensitivity test on four antiseptic soaps and four detergents against some of the isolated fungi.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Trichophyton concentricum strain A	1.00	8	15.1250	.6409	.2266	14.5892	15.6608	14.00	16.00
	2.00	8	20.0000	1.5119	.5345	18.7361	21.2639	18.00	22.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	18.2500	1.5811	.5590	16.9281	19.5719	16.00	20.00
	5.00	8	20.5000	2.5635	.9063	18.3569	22.6431	16.00	24.00
	6.00	8	18.5000	3.6645	1.2956	15.4364	21.5636	12.00	24.00
	7.00	8	11.6250	1.3025	.4605	10.5361	12.7139	10.00	14.00
	8.00	8	17.1250	.9910	.3504	16.2965	17.9535	16.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	15.6806	4.3014	.5069	14.6698	16.6913	10.00	24.00
T. tonsurans strain A	1.00	8	14.3750	1.3025	.4605	13.2861	15.4639	12.00	16.00
	2.00	8	18.7500	2.1213	.7500	16.9765	20.5235	16.00	22.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	18.2500	1.5811	.5590	16.9281	19.5719	16.00	20.00
	5.00	8	20.2500	2.4928	.8814	18.1659	22.3341	16.00	24.00
	6.00	8	17.7500	3.7702	1.3330	14.5980	20.9020	12.00	24.00
	7.00	8	11.1250	.9910	.3504	10.2965	11.9535	10.00	12.00
	8.00	8	16.3750	1.5059	.5324	15.1160	17.6340	14.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	15.2083	4.1789	.4925	14.2263	16.1903	10.00	24.00
T. mentagr var. quinckeanum	1.00	8	15.1250	.6409	.2266	14.5892	15.6608	14.00	16.00
	2.00	8	20.0000	1.5119	.5345	18.7361	21.2639	18.00	22.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	17.7500	2.2520	.7962	15.8673	19.6327	16.00	22.00
	5.00	8	20.5000	2.5635	.9063	18.3569	22.6431	16.00	24.00
	6.00	8	18.5000	3.6645	1.2956	15.4364	21.5636	12.00	24.00
	7.00	8	11.6250	1.3025	.4605	10.5361	12.7139	10.00	14.00
	8.00	8	15.8750	1.5526	.5489	14.5770	17.1730	14.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	15.4861	4.2856	.5051	14.4791	16.4932	10.00	24.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Trichophyton concentricum strain A	Between Groups	1118.528	8	139.816	45.142	.000
	Within Groups	195.125	63	3.097		
	Total	1313.653	71			
T. tonsurans strain A	Between Groups	1013.250	8	126.656	35.209	.000
	Within Groups	226.625	63	3.597		
	Total	1239.875	71			
T. mentagr var. quinckeanum	Between Groups	1080.861	8	135.108	38.148	.000
	Within Groups	223.125	63	3.542		
	Total	1303.986	71			

Post Hoc Tests

Homogeneous Subsets

Trichophyton concentricum strain A

Duncan[a]

VAR00001	N	Subset for alpha = .05				
		1	2	3	4	5
3.00	8	10.0000				
9.00	8	10.0000				
7.00	8	11.6250				
1.00	8		15.1250			
8.00	8			17.1250		
4.00	8			18.2500	18.2500	
6.00	8			18.5000	18.5000	
2.00	8				20.0000	20.0000
5.00	8					20.5000
Sig.		.085	1.000	.145	.064	.572

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

T. tonsurans strain A

Duncan[a]

VAR00001	N	Subset for alpha = .05				
		1	2	3	4	5
3.00	8	10.0000				
9.00	8	10.0000				
7.00	8	11.1250				
1.00	8		14.3750			
8.00	8			16.3750		
6.00	8			17.7500	17.7500	
4.00	8			18.2500	18.2500	
2.00	8				18.7500	18.7500
5.00	8					20.2500
Sig.		.269	1.000	.065	.326	.119

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

T. mentagr var. quinckeanum

Duncan[a]

VAR00001	N	Subset for alpha = .05					
		1	2	3	4	5	6
3.00	8	10.0000					
9.00	8	10.0000					
7.00	8	11.6250					
1.00	8		15.1250				
8.00	8		15.8750	15.8750			
4.00	8			17.7500	17.7500		
6.00	8				18.5000	18.5000	
2.00	8					20.0000	20.0000
5.00	8						20.5000
Sig.		.107	.428	.051	.428	.116	.597

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Aspergillus terreus	1.00	8	15.2500	1.0351	.3660	14.3846	16.1154	14.00	17.00
	2.00	8	20.0000	1.5119	.5345	18.7361	21.2639	18.00	22.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	18.2500	1.5811	.5590	16.9281	19.5719	16.00	20.00
	5.00	8	18.5000	2.3299	.8238	16.5521	20.4479	16.00	22.00
	6.00	8	18.5000	3.6645	1.2956	15.4364	21.5636	12.00	24.00
	7.00	8	11.6250	1.3025	.4605	10.5361	12.7139	10.00	14.00
	8.00	8	17.1250	.9910	.3504	16.2965	17.9535	16.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	15.4722	4.0835	.4812	14.5126	16.4318	10.00	24.00
Blastomyces Dermatitidis	1.00	8	15.1250	.6409	.2266	14.5892	15.6608	14.00	16.00
	2.00	8	20.0000	1.5119	.5345	18.7361	21.2639	18.00	22.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	18.2500	1.5811	.5590	16.9281	19.5719	16.00	20.00
	5.00	8	20.5000	2.5635	.9063	18.3569	22.6431	16.00	24.00
	6.00	8	18.5000	3.6645	1.2956	15.4364	21.5636	12.00	24.00
	7.00	8	11.6250	1.3025	.4605	10.5361	12.7139	10.00	14.00
	8.00	8	17.1250	.9910	.3504	16.2965	17.9535	16.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	15.6806	4.3014	.5069	14.6698	16.6913	10.00	24.00
T. violaceum	1.00	8	15.1250	.6409	.2266	14.5892	15.6608	14.00	16.00
	2.00	8	20.0000	1.5119	.5345	18.7361	21.2639	18.00	22.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	18.2500	1.5811	.5590	16.9281	19.5719	16.00	20.00
	5.00	8	20.5000	2.5635	.9063	18.3569	22.6431	16.00	24.00
	6.00	8	18.5000	3.6645	1.2956	15.4364	21.5636	12.00	24.00
	7.00	8	10.8750	.9910	.3504	10.0465	11.7035	10.00	12.00
	8.00	8	17.2500	1.0351	.3660	16.3846	18.1154	16.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	15.6111	4.3849	.5168	14.5807	16.6415	10.00	24.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Aspergillus terreus	Between Groups	992.194	8	124.024	40.749	.000
	Within Groups	191.750	63	3.044		
	Total	1183.944	71			
Blastomyces Dermatitidis	Between Groups	1118.528	8	139.816	45.142	.000
	Within Groups	195.125	63	3.097		
	Total	1313.653	71			
T. violaceum	Between Groups	1174.361	8	146.795	48.483	.000
	Within Groups	190.750	63	3.028		
	Total	1365.111	71			

Post Hoc Tests

Homogeneous Subsets

Aspergillus terreus

Duncan[a]

VAR00001	N	Subset for alpha = .05			
		1	2	3	4
3.00	8	10.0000			
9.00	8	10.0000			
7.00	8	11.6250			
1.00	8		15.2500		
8.00	8			17.1250	
4.00	8			18.2500	18.2500
5.00	8			18.5000	18.5000
6.00	8			18.5000	18.5000
2.00	8				20.0000
Sig.		.082	1.000	.156	.070

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Blastomyces Dermatitidis

Duncan[a]

VAR00001	N	Subset for alpha = .05				
		1	2	3	4	5
3.00	8	10.0000				
9.00	8	10.0000				
7.00	8	11.6250				
1.00	8		15.1250			
8.00	8			17.1250		
4.00	8			18.2500	18.2500	
6.00	8			18.5000	18.5000	
2.00	8				20.0000	20.0000
5.00	8					20.5000
Sig.		.085	1.000	.145	.064	.572

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

T. violaceum

Duncan[a]

VAR00001	N	Subset for alpha = .05				
		1	2	3	4	5
3.00	8	10.0000				
9.00	8	10.0000				
7.00	8	10.8750				
1.00	8		15.1250			
8.00	8			17.2500		
4.00	8			18.2500	18.2500	
6.00	8			18.5000	18.5000	
2.00	8				20.0000	20.0000
5.00	8					20.5000
Sig.		.349	1.000	.181	.061	.568

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Epidermophyton Fluccosum	1.00	8	15.6250	1.1877	.4199	14.6320	16.6180	14.00	18.00
	2.00	8	17.5000	1.7728	.6268	16.0179	18.9821	16.00	20.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	17.7500	2.1876	.7734	15.9211	19.5789	15.00	20.00
	5.00	8	20.5000	2.5635	.9063	18.3569	22.6431	16.00	24.00
	6.00	8	18.5000	3.6645	1.2956	15.4364	21.5636	12.00	24.00
	7.00	8	12.0000	1.5119	.5345	10.7361	13.2639	10.00	14.00
	8.00	8	17.1250	.9910	.3504	16.2965	17.9535	16.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	15.4444	4.0694	.4796	14.4882	16.4007	10.00	24.00
Microsporum audouinii	1.00	8	15.1250	.6409	.2266	14.5892	15.6608	14.00	16.00
	2.00	8	20.0000	1.5119	.5345	18.7361	21.2639	18.00	22.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	18.2500	1.5811	.5590	16.9281	19.5719	16.00	20.00
	5.00	8	20.5000	2.5635	.9063	18.3569	22.6431	16.00	24.00
	6.00	8	18.5000	3.6645	1.2956	15.4364	21.5636	12.00	24.00
	7.00	8	11.3750	.9161	.3239	10.6091	12.1409	10.00	12.00
	8.00	8	17.1250	.9910	.3504	16.2965	17.9535	16.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	15.6528	4.3188	.5090	14.6379	16.6677	10.00	24.00
Penicillium citrinum	1.00	8	15.1250	.6409	.2266	14.5892	15.6608	14.00	16.00
	2.00	8	20.5000	2.0702	.7319	18.7693	22.2307	18.00	24.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	18.6250	1.5980	.5650	17.2890	19.9610	16.00	20.00
	5.00	8	21.0000	2.8284	1.0000	18.6354	23.3646	16.00	24.00
	6.00	8	18.5000	3.6645	1.2956	15.4364	21.5636	12.00	24.00
	7.00	8	14.6250	2.5036	.8851	12.5320	16.7180	10.00	17.00
	8.00	8	15.8750	1.5526	.5489	14.5770	17.1730	14.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	16.0278	4.3215	.5093	15.0123	17.0433	10.00	24.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Epidermophyton Fluccosum	Between Groups	947.528	8	118.441	32.691	.000
	Within Groups	228.250	63	3.623		
	Total	1175.778	71			
Microsporum audouinii	Between Groups	1135.194	8	141.899	47.269	.000
	Within Groups	189.125	63	3.002		
	Total	1324.319	71			
Penicillium citrinum	Between Groups	1064.444	8	133.056	32.055	.000
	Within Groups	261.500	63	4.151		
	Total	1325.944	71			

Post Hoc Tests

Homogeneous Subsets

Epidermophyton Fluccosum

Duncan[a]

		Subset for alpha = .05			
VAR00001	N	1	2	3	4
3.00	8	10.0000			
9.00	8	10.0000			
7.00	8	12.0000			
1.00	8		15.6250		
8.00	8		17.1250	17.1250	
2.00	8		17.5000	17.5000	
4.00	8			17.7500	
6.00	8			18.5000	
5.00	8				20.5000
Sig.		.050	.066	.194	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Microsporum audouinii

Duncan[a]

VAR00001	N	Subset for alpha = .05				
		1	2	3	4	5
3.00	8	10.0000				
9.00	8	10.0000				
7.00	8	11.3750				
1.00	8		15.1250			
8.00	8			17.1250		
4.00	8			18.2500	18.2500	
6.00	8			18.5000	18.5000	
2.00	8				20.0000	20.0000
5.00	8					20.5000
Sig.		.139	1.000	.139	.060	.566

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Penicillium citrinum

Duncan[a]

VAR00001	N	Subset for alpha = .05			
		1	2	3	4
3.00	8	10.0000			
9.00	8	10.0000			
7.00	8		14.6250		
1.00	8		15.1250		
8.00	8		15.8750		
6.00	8			18.5000	
4.00	8			18.6250	
2.00	8			20.5000	20.5000
5.00	8				21.0000
Sig.		1.000	.253	.067	.625

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
T. rubrum	1.00	8	13.7500	1.5811	.5590	12.4281	15.0719	12.00	16.00
	2.00	8	15.5000	2.5635	.9063	13.3569	17.6431	10.00	18.00
	3.00	8	10.5000	1.4142	.5000	9.3177	11.6823	10.00	14.00
	4.00	8	17.3750	2.9731	1.0511	14.8894	19.8606	14.00	24.00
	5.00	8	20.5000	2.5635	.9063	18.3569	22.6431	16.00	24.00
	6.00	8	17.0000	3.5456	1.2536	14.0358	19.9642	12.00	22.00
	7.00	8	11.6250	2.7223	.9625	9.3491	13.9009	10.00	18.00
	8.00	8	16.2500	2.7124	.9590	13.9824	18.5176	10.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	14.7222	4.0602	.4785	13.7681	15.6763	10.00	24.00
T. soudanense	1.00	8	14.7500	1.4880	.5261	13.5060	15.9940	12.00	16.00
	2.00	8	14.2500	2.2520	.7962	12.3673	16.1327	10.00	16.00
	3.00	8	11.2500	3.5355	1.2500	8.2942	14.2058	10.00	20.00
	4.00	8	18.5000	2.7775	.9820	16.1780	20.8220	16.00	24.00
	5.00	8	20.5000	2.5635	.9063	18.3569	22.6431	16.00	24.00
	6.00	8	17.0000	3.5456	1.2536	14.0358	19.9642	12.00	22.00
	7.00	8	12.3750	2.6152	.9246	10.1886	14.5614	10.00	18.00
	8.00	8	16.1250	2.6424	.9342	13.9159	18.3341	10.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	14.9722	4.0732	.4800	14.0151	15.9294	10.00	24.00
M. ferrugineum	1.00	8	14.2500	2.2520	.7962	12.3673	16.1327	12.00	18.00
	2.00	8	15.5000	2.5635	.9063	13.3569	17.6431	10.00	18.00
	3.00	8	10.7500	2.1213	.7500	8.9765	12.5235	10.00	16.00
	4.00	8	18.7500	2.6049	.9210	16.5722	20.9278	16.00	24.00
	5.00	8	20.5000	2.5635	.9063	18.3569	22.6431	16.00	24.00
	6.00	8	17.0000	3.5456	1.2536	14.0358	19.9642	12.00	22.00
	7.00	8	12.3750	2.6152	.9246	10.1886	14.5614	10.00	18.00
	8.00	8	16.2500	2.7124	.9590	13.9824	18.5176	10.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	15.0417	4.1092	.4843	14.0761	16.0073	10.00	24.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
T. rubrum	Between Groups	793.694	8	99.212	16.590	.000
	Within Groups	376.750	63	5.980		
	Total	1170.444	71			
T. soudanense	Between Groups	754.694	8	94.337	14.042	.000
	Within Groups	423.250	63	6.718		
	Total	1177.944	71			
M. ferrugineum	Between Groups	805.000	8	100.625	16.095	.000
	Within Groups	393.875	63	6.252		
	Total	1198.875	71			

Post Hoc Tests

Homogeneous Subsets

T. rubrum

Duncan[a]

		Subset for alpha = .05				
VAR00001	N	1	2	3	4	5
9.00	8	10.0000				
3.00	8	10.5000				
7.00	8	11.6250	11.6250			
1.00	8		13.7500	13.7500		
2.00	8			15.5000	15.5000	
8.00	8			16.2500	16.2500	
6.00	8				17.0000	
4.00	8				17.3750	
5.00	8					20.5000
Sig.		.216	.087	.057	.168	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

T. soudanense

Duncan[a]

VAR00001	N	Subset for alpha = .05				
		1	2	3	4	5
9.00	8	10.0000				
3.00	8	11.2500				
7.00	8	12.3750	12.3750			
2.00	8		14.2500	14.2500		
1.00	8		14.7500	14.7500		
8.00	8			16.1250	16.1250	
6.00	8			17.0000	17.0000	
4.00	8				18.5000	18.5000
5.00	8					20.5000
Sig.		.088	.088	.055	.088	.128

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

M. ferrugineum

Duncan[a]

VAR00001	N	Subset for alpha = .05					
		1	2	3	4	5	6
9.00	8	10.0000					
3.00	8	10.7500					
7.00	8	12.3750	12.3750				
1.00	8		14.2500	14.2500			
2.00	8			15.5000	15.5000		
8.00	8			16.2500	16.2500	16.2500	
6.00	8				17.0000	17.0000	
4.00	8					18.7500	18.7500
5.00	8						20.5000
Sig.		.077	.139	.136	.264	.062	.166

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
P.aschersonia	1.00	8	15.2500	.8864	.3134	14.5089	15.9911	14.00	16.00
	2.00	8	19.7500	1.6690	.5901	18.3546	21.1454	18.00	22.00
	3.00	8	10.7500	1.0351	.3660	9.8846	11.6154	10.00	12.00
	4.00	8	18.2500	1.5811	.5590	16.9281	19.5719	16.00	20.00
	5.00	8	20.5000	2.5635	.9063	18.3569	22.6431	16.00	24.00
	6.00	8	18.5000	3.6645	1.2956	15.4364	21.5636	12.00	24.00
	7.00	8	12.1250	.8345	.2950	11.4273	12.8227	11.00	14.00
	8.00	8	19.0000	2.1381	.7559	17.2125	20.7875	16.00	22.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	16.0139	4.2740	.5037	15.0095	17.0182	10.00	24.00
Unknown O	1.00	8	15.0000	.7559	.2673	14.3680	15.6320	14.00	16.00
	2.00	8	17.0000	2.3905	.8452	15.0015	18.9985	12.00	20.00
	3.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	4.00	8	17.7500	1.6690	.5901	16.3546	19.1454	16.00	20.00
	5.00	8	20.5000	2.5635	.9063	18.3569	22.6431	16.00	24.00
	6.00	8	18.5000	3.6645	1.2956	15.4364	21.5636	12.00	24.00
	7.00	8	11.6250	1.3025	.4605	10.5361	12.7139	10.00	14.00
	8.00	8	17.1250	.9910	.3504	16.2965	17.9535	16.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	15.2778	4.0706	.4797	14.3212	16.2343	10.00	24.00
Penicillium spp	1.00	8	16.6250	1.5980	.5650	15.2890	17.9610	14.00	18.00
	2.00	8	20.0000	1.5119	.5345	18.7361	21.2639	18.00	22.00
	3.00	8	11.1250	.9910	.3504	10.2965	11.9535	10.00	12.00
	4.00	8	18.2500	1.5811	.5590	16.9281	19.5719	16.00	20.00
	5.00	8	20.5000	2.5635	.9063	18.3569	22.6431	16.00	24.00
	6.00	8	18.5000	3.6645	1.2956	15.4364	21.5636	12.00	24.00
	7.00	8	12.7500	1.0351	.3660	11.8846	13.6154	12.00	14.00
	8.00	8	17.1250	.9910	.3504	16.2965	17.9535	16.00	18.00
	9.00	8	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	72	16.0972	4.0565	.4781	15.1440	17.0505	10.00	24.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
P.aschersonia	Between Groups	1070.111	8	133.764	37.144	.000
	Within Groups	226.875	63	3.601		
	Total	1296.986	71			
Unknown O	Between Groups	954.194	8	119.274	33.810	.000
	Within Groups	222.250	63	3.528		
	Total	1176.444	71			
Penicillium spp	Between Groups	955.694	8	119.462	35.396	.000
	Within Groups	212.625	63	3.375		
	Total	1168.319	71			

Post Hoc Tests

Homogeneous Subsets

P.aschersonia

Duncan[a]

		Subset for alpha = .05				
VAR00001	N	1	2	3	4	5
9.00	8	10.0000				
3.00	8	10.7500	10.7500			
7.00	8		12.1250			
1.00	8			15.2500		
4.00	8				18.2500	
6.00	8				18.5000	18.5000
8.00	8				19.0000	19.0000
2.00	8				19.7500	19.7500
5.00	8					20.5000
Sig.		.432	.152	1.000	.155	.057

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Unknown O

Duncan[a]

VAR00001	N	Subset for alpha = .05			
		1	2	3	4
3.00	8	10.0000			
9.00	8	10.0000			
7.00	8	11.6250			
1.00	8		15.0000		
2.00	8			17.0000	
8.00	8			17.1250	
4.00	8			17.7500	
6.00	8			18.5000	
5.00	8				20.5000
Sig.		.107	1.000	.151	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Penicillium spp

Duncan[a]

VAR00001	N	Subset for alpha = .05				
		1	2	3	4	5
9.00	8	10.0000				
3.00	8	11.1250	11.1250			
7.00	8		12.7500			
1.00	8			16.6250		
8.00	8			17.1250		
4.00	8			18.2500	18.2500	
6.00	8			18.5000	18.5000	
2.00	8				20.0000	20.0000
5.00	8					20.5000
Sig.		.225	.082	.066	.076	.588

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 8.000.

Appendix F: One way analysis of variance (ANOVA) of the growth studies on different media

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Trichophyton concentricum strain A (day 12)	1.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	2.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	3.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	4.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	5.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	6.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	Total	36	11.3333	1.3939	.2323	10.8617	11.8049	10.00	14.00
Trichophyton concentricum strain A (day 14)	1.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	2.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	3.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	4.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	5.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	6.00	6	15.5000	.5477	.2236	14.9252	16.0748	15.00	16.00
	Total	36	13.0833	1.4015	.2336	12.6091	13.5575	12.00	16.00
Trichophyton concentricum strain A (day 16)	1.00	6	16.0000	.0000	.0000	16.0000	16.0000	16.00	16.00
	2.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	3.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	4.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	5.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	6.00	6	18.5000	.5477	.2236	17.9252	19.0748	18.00	19.00
	Total	36	15.0833	1.7300	.2883	14.4980	15.6687	14.00	19.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Trichophyton concentricum strain A (day 12)	Between Groups	62.000	5	12.400	62.000	.000
	Within Groups	6.000	30	.200		
	Total	68.000	35			
Trichophyton concentricum strain A (day 14)	Between Groups	61.250	5	12.250	49.000	.000
	Within Groups	7.500	30	.250		
	Total	68.750	35			
Trichophyton concentricum strain A (day 16)	Between Groups	103.250	5	20.650	413.000	.000
	Within Groups	1.500	30	5.000E-02		
	Total	104.750	35			

Homogeneous Subsets

Trichophyton concentricum strain A (day 12)

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
2.00	6	10.5000		
3.00	6	10.5000		
4.00	6	10.5000		
5.00	6	10.5000		
1.00	6		12.0000	
6.00	6			14.0000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton concentricum strain A (day 14)

Duncan[a]

VAR00001	N	Subset for alpha = .05			
		1	2	3	4
2.00	6	12.0000			
4.00	6	12.0000			
5.00	6	12.0000			
3.00	6		13.0000		
1.00	6			14.0000	
6.00	6				15.5000
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton concentricum strain A (day 16)

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
2.00	6	14.0000		
3.00	6	14.0000		
4.00	6	14.0000		
5.00	6	14.0000		
1.00	6		16.0000	
6.00	6			18.5000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Trichophyton tonsurans strain A (day 12)	1.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	2.00	6	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	3.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	4.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	5.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	6.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	Total	36	10.6667	.7559	.1260	10.4109	10.9224	10.00	12.00
Trichophyton tonsurans strain A (day 14)	1.00	6	12.5000	.5477	.2236	11.9252	13.0748	12.00	13.00
	2.00	6	11.0000	.0000	.0000	11.0000	11.0000	11.00	11.00
	3.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	4.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	5.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	6.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	Total	36	12.5833	1.1307	.1885	12.2007	12.9659	11.00	14.00
Trichophyton tonsurans strain A (day 16)	1.00	6	14.5000	.5477	.2236	13.9252	15.0748	14.00	15.00
	2.00	6	14.5000	.5477	.2236	13.9252	15.0748	14.00	15.00
	3.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	4.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	5.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	6.00	6	16.0000	.0000	.0000	16.0000	16.0000	16.00	16.00
	Total	36	14.5000	.7746	.1291	14.2379	14.7621	14.00	16.00
Trichophyton mentagrophyes var. quinckeanum (day 12)	1.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	2.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	3.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	4.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	5.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	6.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	Total	36	11.9167	1.6279	.2713	11.3659	12.4675	10.00	14.00
Trichophyton mentagrophyes var. quinckeanum (day 14)	1.00	6	16.0000	.0000	.0000	16.0000	16.0000	16.00	16.00
	2.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	3.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	4.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	5.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	6.00	6	16.0000	.0000	.0000	16.0000	16.0000	16.00	16.00
	Total	36	14.0000	1.6562	.2760	13.4396	14.5604	12.00	16.00
Trichophyton mentagrophyes var. quinckeanum (day 16)	1.00	6	18.0000	.0000	.0000	18.0000	18.0000	18.00	18.00
	2.00	6	15.5000	.5477	.2236	14.9252	16.0748	15.00	16.00
	3.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	4.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	5.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	6.00	6	18.0000	.0000	.0000	18.0000	18.0000	18.00	18.00
	Total	36	15.5833	1.8264	.3044	14.9654	16.2013	14.00	18.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Trichophyton tonsurans strain A (day 12)	Between Groups	14.000	5	2.800	14.000	.000
	Within Groups	6.000	30	.200		
	Total	20.000	35			
Trichophyton tonsurans strain A (day 14)	Between Groups	31.250	5	6.250	13.889	.000
	Within Groups	13.500	30	.450		
	Total	44.750	35			
Trichophyton tonsurans strain A (day 16)	Between Groups	18.000	5	3.600	36.000	.000
	Within Groups	3.000	30	.100		
	Total	21.000	35			
Trichophyton mentagrophyes var. quinckeanum (day 12)	Between Groups	88.250	5	17.650	117.667	.000
	Within Groups	4.500	30	.150		
	Total	92.750	35			
Trichophyton mentagrophyes var. quinckeanum (day 14)	Between Groups	84.000	5	16.800	42.000	.000
	Within Groups	12.000	30	.400		
	Total	96.000	35			
Trichophyton mentagrophyes var. quinckeanum (day 16)	Between Groups	115.250	5	23.050	461.000	.000
	Within Groups	1.500	30	5.000E-02		
	Total	116.750	35			

Post Hoc Tests

Homogeneous Subsets

Trichophyton tonsurans strain A (day 12)

Duncan[a]

		Subset for alpha = .05	
VAR00001	N	1	2
2.00	6	10.0000	
1.00	6	10.5000	
3.00	6	10.5000	
4.00	6	10.5000	
5.00	6	10.5000	
6.00	6		12.0000
Sig.		.092	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton tonsurans strain A (day 14)

Duncan[a]

		Subset for alpha = .05			
VAR00001	N	1	2	3	4
2.00	6	11.0000			
3.00	6		12.0000		
1.00	6		12.5000	12.5000	
4.00	6			13.0000	
5.00	6			13.0000	
6.00	6				14.0000
Sig.		1.000	.207	.233	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton tonsurans strain A (day 16)

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
3.00	6	14.0000		
4.00	6	14.0000		
5.00	6	14.0000		
1.00	6		14.5000	
2.00	6		14.5000	
6.00	6			16.0000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton mentagrophyes var. quinckeanum (day 12)

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
3.00	6	10.5000		
4.00	6	10.5000		
5.00	6	10.5000		
2.00	6		12.0000	
1.00	6			14.0000
6.00	6			14.0000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton mentagrophyes var. quinckeanum (day 14)

Duncan[a]

		Subset for alpha = .05			
VAR00001	N	1	2	3	4
4.00	6	12.0000			
3.00	6		13.0000		
5.00	6		13.0000		
2.00	6			14.0000	
1.00	6				16.0000
6.00	6				16.0000
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton mentagrophyes var. quinckeanum (day 16)

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
3.00	6	14.0000		
4.00	6	14.0000		
5.00	6	14.0000		
2.00	6		15.5000	
1.00	6			18.0000
6.00	6			18.0000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Trichophyton tonsurans strain B (day 12)	1.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	2.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	3.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	4.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	5.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	6.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	Total	36	11.0000	.8281	.1380	10.7198	11.2802	10.00	12.00
Trichophyton tonsurans strain B (day 14)	1.00	6	13.5000	.5477	.2236	12.9252	14.0748	13.00	14.00
	2.00	6	12.5000	.5477	.2236	11.9252	13.0748	12.00	13.00
	3.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	4.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	5.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	6.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	Total	36	13.0000	.9258	.1543	12.6867	13.3133	12.00	14.00
Trichophyton tonsurans strain B (day 16)	1.00	6	15.5000	.5477	.2236	14.9252	16.0748	15.00	16.00
	2.00	6	14.5000	.5477	.2236	13.9252	15.0748	14.00	15.00
	3.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	4.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	5.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	6.00	6	16.0000	.0000	.0000	16.0000	16.0000	16.00	16.00
	Total	36	14.6667	.8619	.1436	14.3750	14.9583	14.00	16.00
Trichophyton rubrum (day 12)	1.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	2.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	3.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	4.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	5.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	6.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	Total	36	11.0833	.8742	.1457	10.7875	11.3791	10.00	12.00
Trichophyton rubrum (day 14)	1.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	2.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	3.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	4.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	5.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	6.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	Total	36	13.0000	1.0142	.1690	12.6568	13.3432	12.00	14.00
Trichophyton rubrum (day 16)	1.00	6	16.0000	.0000	.0000	16.0000	16.0000	16.00	16.00
	2.00	6	14.5000	.5477	.2236	13.9252	15.0748	14.00	15.00
	3.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	4.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	5.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	6.00	6	16.0000	.0000	.0000	16.0000	16.0000	16.00	16.00
	Total	36	14.7500	.9373	.1562	14.4329	15.0671	14.00	16.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Trichophyton tonsurans strain B (day 12)	Between Groups	18.000	5	3.600	18.000	.000
	Within Groups	6.000	30	.200		
	Total	24.000	35			
Trichophyton tonsurans strain B (day 14)	Between Groups	15.000	5	3.000	6.000	.001
	Within Groups	15.000	30	.500		
	Total	30.000	35			
Trichophyton tonsurans strain B (day 16)	Between Groups	23.000	5	4.600	46.000	.000
	Within Groups	3.000	30	.100		
	Total	26.000	35			
Trichophyton rubrum (day 12)	Between Groups	16.250	5	3.250	9.286	.000
	Within Groups	10.500	30	.350		
	Total	26.750	35			
Trichophyton rubrum (day 14)	Between Groups	24.000	5	4.800	12.000	.000
	Within Groups	12.000	30	.400		
	Total	36.000	35			
Trichophyton rubrum (day 16)	Between Groups	29.250	5	5.850	117.000	.000
	Within Groups	1.500	30	5.000E-02		
	Total	30.750	35			

Post Hoc Tests

Homogeneous Subsets

Trichophyton tonsurans strain B (day 12)

Duncan[a]

		Subset for alpha = .05	
VAR00001	N	1	2
2.00	6	10.5000	
3.00	6	10.5000	
4.00	6	10.5000	
5.00	6	10.5000	
1.00	6		12.0000
6.00	6		12.0000
Sig.		1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton tonsurans strain B (day 14)

Duncan[a]

		Subset for alpha = .05			
VAR00001	N	1	2	3	4
4.00	6	12.0000			
2.00	6	12.5000	12.5000		
3.00	6		13.0000	13.0000	
5.00	6		13.0000	13.0000	
1.00	6			13.5000	13.5000
6.00	6				14.0000
Sig.		.230	.257	.257	.230

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton tonsurans strain B (day 16)

Duncan[a]

		Subset for alpha = .05			
VAR00001	N	1	2	3	4
3.00	6	14.0000			
4.00	6	14.0000			
5.00	6	14.0000			
2.00	6		14.5000		
1.00	6			15.5000	
6.00	6				16.0000
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton rubrum (day 12)

Duncan[a]

		Subset for alpha = .05	
VAR00001	N	1	2
3.00	6	10.5000	
4.00	6	10.5000	
5.00	6	10.5000	
2.00	6	11.0000	
1.00	6		12.0000
6.00	6		12.0000
Sig.		.191	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton rubrum (day 14)

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
4.00	6	12.0000		
5.00	6	12.0000		
2.00	6		13.0000	
3.00	6		13.0000	
1.00	6			14.0000
6.00	6			14.0000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton rubrum (day 16)

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
3.00	6	14.0000		
4.00	6	14.0000		
5.00	6	14.0000		
2.00	6		14.5000	
1.00	6			16.0000
6.00	6			16.0000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Trichophyton soudanense (day 12)	1.00	6	12.5000	.5477	.2236	11.9252	13.0748	12.00	13.00
	2.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	3.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	4.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	5.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	6.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	Total	36	11.2500	1.2507	.2085	10.8268	11.6732	10.00	14.00
Trichophyton soudanense (day 14)	1.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	2.00	6	12.5000	.5477	.2236	11.9252	13.0748	12.00	13.00
	3.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	4.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	5.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	6.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	Total	36	13.0833	1.2734	.2122	12.6525	13.5142	12.00	16.00
Trichophyton soudanense (day 16)	1.00	6	15.5000	.5477	.2236	14.9252	16.0748	15.00	16.00
	2.00	6	14.5000	.5477	.2236	13.9252	15.0748	14.00	15.00
	3.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	4.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	5.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	6.00	6	17.0000	1.0954	.4472	15.8504	18.1496	16.00	18.00
	Total	36	14.8333	1.2306	.2051	14.4170	15.2497	14.00	18.00
Epidermaphyton fluccosum (day 12)	1.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	2.00	6	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	3.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	4.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	5.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	6.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	Total	36	10.7500	.8409	.1402	10.4655	11.0345	10.00	12.00
Epidermaphyton fluccosum (day 14)	1.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	2.00	6	11.5000	.5477	.2236	10.9252	12.0748	11.00	12.00
	3.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	4.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	5.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	6.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	Total	36	12.7500	1.1052	.1842	12.3761	13.1239	11.00	14.00
Epidermaphyton fluccosum (day 16)	1.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	2.00	6	12.5000	.5477	.2236	11.9252	13.0748	12.00	13.00
	3.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	4.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	5.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	6.00	6	16.0000	.0000	.0000	16.0000	16.0000	16.00	16.00
	Total	36	14.2500	1.1802	.1967	13.8507	14.6493	12.00	16.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Trichophyton soudanense (day 12)	Between Groups	41.250	5	8.250	18.333	.000
	Within Groups	13.500	30	.450		
	Total	54.750	35			
Trichophyton soudanense (day 14)	Between Groups	43.250	5	8.650	19.222	.000
	Within Groups	13.500	30	.450		
	Total	56.750	35			
Trichophyton soudanense (day 16)	Between Groups	44.000	5	8.800	29.333	.000
	Within Groups	9.000	30	.300		
	Total	53.000	35			
Epidermaphyton fluccosum (day 12)	Between Groups	14.250	5	2.850	8.143	.000
	Within Groups	10.500	30	.350		
	Total	24.750	35			
Epidermaphyton fluccosum (day 14)	Between Groups	23.250	5	4.650	7.154	.000
	Within Groups	19.500	30	.650		
	Total	42.750	35			
Epidermaphyton fluccosum (day 16)	Between Groups	41.250	5	8.250	33.000	.000
	Within Groups	7.500	30	.250		
	Total	48.750	35			

Post Hoc Tests

Homogeneous Subsets

Trichophyton soudanense (day 12)

Duncan[a]

		Subset for alpha = .05	
VAR00001	N	1	2
2.00	6	10.5000	
3.00	6	10.5000	
4.00	6	10.5000	
5.00	6	10.5000	
1.00	6		12.5000
6.00	6		13.0000
Sig.		1.000	.207

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton soudanense (day 14)

Duncan[a]

		Subset for alpha = .05			
VAR00001	N	1	2	3	4
3.00	6	12.0000			
5.00	6	12.0000			
2.00	6	12.5000	12.5000		
4.00	6		13.0000		
1.00	6			14.0000	
6.00	6				15.0000
Sig.		.233	.207	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton soudanense (day 16)

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
3.00	6	14.0000		
4.00	6	14.0000		
5.00	6	14.0000		
2.00	6	14.5000		
1.00	6		15.5000	
6.00	6			17.0000
Sig.		.159	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Epidermaphyton fluccosum (day 12)

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
2.00	6	10.0000		
3.00	6	10.5000	10.5000	
4.00	6	10.5000	10.5000	
5.00	6	10.5000	10.5000	
1.00	6		11.0000	
6.00	6			12.0000
Sig.		.191	.191	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Epidermaphyton fluccosum (day 14)

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
2.00	6	11.5000		
4.00	6	12.0000	12.0000	
1.00	6		13.0000	13.0000
3.00	6		13.0000	13.0000
5.00	6		13.0000	13.0000
6.00	6			14.0000
Sig.		.291	.057	.057

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Epidermaphyton fluccosum (day 16)

Duncan[a]

VAR00001	N	Subset for alpha = .05			
		1	2	3	4
2.00	6	12.5000			
3.00	6		14.0000		
4.00	6		14.0000		
5.00	6		14.0000		
1.00	6			15.0000	
6.00	6				16.0000
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Trichophyton violaceum (day 12)	1.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	2.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	3.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	4.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	5.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	6.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	Total	36	11.4167	1.4015	.2336	10.9425	11.8909	10.00	14.00
Trichophyton violaceum (day 14)	1.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	2.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	3.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	4.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	5.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	6.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	Total	36	13.3333	1.5119	.2520	12.8218	13.8449	12.00	16.00
Trichophyton violaceum (day 16)	1.00	6	17.0000	1.0954	.4472	15.8504	18.1496	16.00	18.00
	2.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	3.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	4.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	5.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	6.00	6	17.0000	1.0954	.4472	15.8504	18.1496	16.00	18.00
	Total	36	15.1667	1.5399	.2567	14.6456	15.6877	14.00	18.00
Microsporum audouinii (day 12)	1.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	2.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	3.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	4.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	5.00	6	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	6.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	Total	36	10.9167	1.2042	.2007	10.5092	11.3241	10.00	14.00
Microsporum audouinii (day 14)	1.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	2.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	3.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	4.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	5.00	6	11.5000	.5477	.2236	10.9252	12.0748	11.00	12.00
	6.00	6	11.5000	.5477	.2236	10.9252	12.0748	11.00	12.00
	Total	36	12.8333	1.4832	.2472	12.3315	13.3352	11.00	16.00
Microsporum audouinii (day 16)	1.00	6	17.0000	1.0954	.4472	15.8504	18.1496	16.00	18.00
	2.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	3.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	4.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	5.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	6.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	Total	36	14.3333	1.6213	.2702	13.7848	14.8819	12.00	18.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Trichophyton violaceum (day 12)	Between Groups	46.250	5	9.250	12.333	.000
	Within Groups	22.500	30	.750		
	Total	68.750	35			
Trichophyton violaceum (day 14)	Between Groups	56.000	5	11.200	14.000	.000
	Within Groups	24.000	30	.800		
	Total	80.000	35			
Trichophyton violaceum (day 16)	Between Groups	65.000	5	13.000	21.667	.000
	Within Groups	18.000	30	.600		
	Total	83.000	35			
Microsporum audouinii (day 12)	Between Groups	34.250	5	6.850	12.455	.000
	Within Groups	16.500	30	.550		
	Total	50.750	35			
Microsporum audouinii (day 14)	Between Groups	50.000	5	10.000	11.111	.000
	Within Groups	27.000	30	.900		
	Total	77.000	35			
Microsporum audouinii (day 16)	Between Groups	68.000	5	13.600	17.000	.000
	Within Groups	24.000	30	.800		
	Total	92.000	35			

Post Hoc Tests

Homogeneous Subsets

Trichophyton violaceum (day 12)

Duncan[a]

		Subset for alpha = .05	
VAR00001	N	1	2
3.00	6	10.5000	
4.00	6	10.5000	
5.00	6	10.5000	
2.00	6	11.0000	
1.00	6		13.0000
6.00	6		13.0000
Sig.		.370	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton violaceum (day 14)

Duncan[a]

		Subset for alpha = .05	
VAR00001	N	1	2
3.00	6	12.0000	
5.00	6	12.0000	
2.00	6	13.0000	
4.00	6	13.0000	
1.00	6		15.0000
6.00	6		15.0000
Sig.		.085	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Trichophyton violaceum (day 16)

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
3.00	6	14.0000		
4.00	6	14.0000		
5.00	6	14.0000		
2.00	6		15.0000	
1.00	6			17.0000
6.00	6			17.0000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Microsporum audouinii (day 12)

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
5.00	6	10.0000		
3.00	6	10.5000	10.5000	
4.00	6	10.5000	10.5000	
6.00	6	10.5000	10.5000	
2.00	6		11.0000	
1.00	6			13.0000
Sig.		.296	.296	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Microsporum audouinii (day 14)

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
5.00	6	11.5000		
6.00	6	11.5000		
2.00	6		13.0000	
3.00	6		13.0000	
4.00	6		13.0000	
1.00	6			15.0000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Microsporum audouinii (day 16)

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
5.00	6	13.0000		
6.00	6	13.0000		
3.00	6	14.0000	14.0000	
4.00	6	14.0000	14.0000	
2.00	6		15.0000	
1.00	6			17.0000
Sig.		.085	.076	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Microsporum ferrugineum (day 12)	1.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	2.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	3.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	4.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	5.00	6	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	6.00	6	10.0000	.0000	.0000	10.0000	10.0000	10.00	10.00
	Total	36	10.4167	.6492	.1082	10.1970	10.6363	10.00	12.00
Microsporum ferrugineum (day 14)	1.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	2.00	6	11.5000	.5477	.2236	10.9252	12.0748	11.00	12.00
	3.00	6	11.5000	.5477	.2236	10.9252	12.0748	11.00	12.00
	4.00	6	11.5000	.5477	.2236	10.9252	12.0748	11.00	12.00
	5.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	6.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	Total	36	11.4167	1.0522	.1754	11.0607	11.7727	10.00	14.00
Microsporum ferrugineum (day 16)	1.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	2.00	6	12.5000	.5477	.2236	11.9252	13.0748	12.00	13.00
	3.00	6	12.5000	.5477	.2236	11.9252	13.0748	12.00	13.00
	4.00	6	12.5000	.5477	.2236	11.9252	13.0748	12.00	13.00
	5.00	6	11.5000	.5477	.2236	10.9252	12.0748	11.00	12.00
	6.00	6	11.5000	.5477	.2236	10.9252	12.0748	11.00	12.00
	Total	36	12.5833	1.3390	.2232	12.1303	13.0364	11.00	16.00
Microsporum nanum (day 12)	1.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	2.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	3.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	4.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	5.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	6.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	Total	36	10.8333	.9103	.1517	10.5253	11.1413	10.00	12.00
Microsporum nanum (day 14)	1.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	2.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	3.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	4.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	5.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	6.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	Total	36	12.6667	.9562	.1594	12.3431	12.9902	12.00	14.00
Microsporum nanum (day 16)	1.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	2.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	3.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	4.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	5.00	6	13.5000	.5477	.2236	12.9252	14.0748	13.00	14.00
	6.00	6	13.5000	.5477	.2236	12.9252	14.0748	13.00	14.00
	Total	36	14.5000	1.1339	.1890	14.1163	14.8837	13.00	16.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Microsporum ferrugineum (day 12)	Between Groups	4.250	5	.850	2.429	.058
	Within Groups	10.500	30	.350		
	Total	14.750	35			
Microsporum ferrugineum (day 14)	Between Groups	25.250	5	5.050	11.222	.000
	Within Groups	13.500	30	.450		
	Total	38.750	35			
Microsporum ferrugineum (day 16)	Between Groups	49.250	5	9.850	21.889	.000
	Within Groups	13.500	30	.450		
	Total	62.750	35			
Microsporum nanum (day 12)	Between Groups	2.000	5	.400	.444	.814
	Within Groups	27.000	30	.900		
	Total	29.000	35			
Microsporum nanum (day 14)	Between Groups	8.000	5	1.600	2.000	.107
	Within Groups	24.000	30	.800		
	Total	32.000	35			
Microsporum nanum (day 16)	Between Groups	18.000	5	3.600	4.000	.007
	Within Groups	27.000	30	.900		
	Total	45.000	35			

Post Hoc Tests

Homogeneous Subsets

Microsporum ferrugineum (day 12)

Duncan[a]

		Subset for alpha = .05	
VAR00001	N	1	2
5.00	6	10.0000	
6.00	6	10.0000	
2.00	6	10.5000	10.5000
3.00	6	10.5000	10.5000
4.00	6	10.5000	10.5000
1.00	6		11.0000
Sig.		.201	.191

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Microsporum ferrugineum (day 14)

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
5.00	6	10.5000		
6.00	6	10.5000		
2.00	6		11.5000	
3.00	6		11.5000	
4.00	6		11.5000	
1.00	6			13.0000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Microsporum ferrugineum (day 16)

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
5.00	6	11.5000		
6.00	6	11.5000		
2.00	6		12.5000	
3.00	6		12.5000	
4.00	6		12.5000	
1.00	6			15.0000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Microsporum nanum (day 12)

Duncan[a]

VAR00001	N	Subset for alpha = .05
		1
5.00	6	10.5000
6.00	6	10.5000
1.00	6	11.0000
2.00	6	11.0000
3.00	6	11.0000
4.00	6	11.0000
Sig.		.431

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Microsporum nanum (day 14)

Duncan[a]

VAR00001	N	Subset for alpha = .05
		1
5.00	6	12.0000
6.00	6	12.0000
1.00	6	13.0000
2.00	6	13.0000
3.00	6	13.0000
4.00	6	13.0000
Sig.		.097

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
T.concentricum strain B (day 12)	1.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	2.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	3.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	4.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	5.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	6.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	Total	36	11.3333	1.3939	.2323	10.8617	11.8049	10.00	14.00
T.concentricum strain B (day 14)	1.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	2.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	3.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	4.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	5.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	6.00	6	15.5000	.5477	.2236	14.9252	16.0748	15.00	16.00
	Total	36	13.0833	1.4015	.2336	12.6091	13.5575	12.00	16.00
T.concentricum strain B (day 16)	1.00	6	16.0000	.0000	.0000	16.0000	16.0000	16.00	16.00
	2.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	3.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	4.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	5.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	6.00	6	18.5000	.5477	.2236	17.9252	19.0748	18.00	19.00
	Total	36	15.0833	1.7300	.2883	14.4980	15.6687	14.00	19.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
T.concentricum strain B (day 12)	Between Groups	62.000	5	12.400	62.000	.000
	Within Groups	6.000	30	.200		
	Total	68.000	35			
T.concentricum strain B (day 14)	Between Groups	61.250	5	12.250	49.000	.000
	Within Groups	7.500	30	.250		
	Total	68.750	35			
T.concentricum strain B (day 16)	Between Groups	103.250	5	20.650	413.000	.000
	Within Groups	1.500	30	5.000E-02		
	Total	104.750	35			

Post Hoc Tests

Homogeneous Subsets

T.concentricum strain B (day 12)

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
2.00	6	10.5000		
3.00	6	10.5000		
4.00	6	10.5000		
5.00	6	10.5000		
1.00	6		12.0000	
6.00	6			14.0000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

T.concentricum strain B (day 14)

Duncan[a]

VAR00001	N	Subset for alpha = .05			
		1	2	3	4
2.00	6	12.0000			
4.00	6	12.0000			
5.00	6	12.0000			
3.00	6		13.0000		
1.00	6			14.0000	
6.00	6				15.5000
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

T.concentricum strain B (day 16)

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
2.00	6	14.0000		
3.00	6	14.0000		
4.00	6	14.0000		
5.00	6	14.0000		
1.00	6		16.0000	
6.00	6			18.5000
Sig.		1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Microsporum nanum (day 16)

Duncan[a]

VAR00001	N	Subset for alpha = .05	
		1	2
5.00	6	13.5000	
6.00	6	13.5000	
1.00	6		15.0000
2.00	6		15.0000
3.00	6		15.0000
4.00	6		15.0000
Sig.		1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Absidia corymbifera (day 3)	1.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	2.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	3.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	4.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	5.00	6	12.5000	1.6432	.6708	10.7756	14.2244	11.00	14.00
	6.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	Total	36	11.7500	1.9030	.3172	11.1061	12.3939	10.00	16.00
Absidia corymbifera (day 4)	1.00	6	12.5000	.5477	.2236	11.9252	13.0748	12.00	13.00
	2.00	6	17.0000	1.0954	.4472	15.8504	18.1496	16.00	18.00
	3.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	4.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	5.00	6	13.5000	.5477	.2236	12.9252	14.0748	13.00	14.00
	6.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	Total	36	13.8333	1.8898	.3150	13.1939	14.4728	12.00	18.00
Absidia corymbifera (day 5)	1.00	6	14.5000	.5477	.2236	13.9252	15.0748	14.00	15.00
	2.00	6	19.0000	1.0954	.4472	17.8504	20.1496	18.00	20.00
	3.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	4.00	6	17.0000	1.0954	.4472	15.8504	18.1496	16.00	18.00
	5.00	6	15.5000	.5477	.2236	14.9252	16.0748	15.00	16.00
	6.00	6	13.5000	.5477	.2236	12.9252	14.0748	13.00	14.00
	Total	36	15.7500	1.9911	.3318	15.0763	16.4237	13.00	20.00
Aspergillus terreus (day 3)	1.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	2.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	3.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	4.00	6	15.5000	.5477	.2236	14.9252	16.0748	15.00	16.00
	5.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	6.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	Total	36	12.2500	1.8574	.3096	11.6215	12.8785	10.00	16.00
Aspergillus terreus (day 4)	1.00	6	13.5000	1.6432	.6708	11.7756	15.2244	12.00	15.00
	2.00	6	14.0000	1.0954	.4472	12.8504	15.1496	13.00	15.00
	3.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	4.00	6	17.0000	1.0954	.4472	15.8504	18.1496	16.00	18.00
	5.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	6.00	6	12.5000	.5477	.2236	11.9252	13.0748	12.00	13.00
	Total	36	14.0000	1.7566	.2928	13.4056	14.5944	12.00	18.00
Aspergillus terreus (day 5)	1.00	6	26.5000	1.6432	.6708	24.7756	28.2244	25.00	28.00
	2.00	6	17.0000	1.0954	.4472	15.8504	18.1496	16.00	18.00
	3.00	6	16.0000	.0000	.0000	16.0000	16.0000	16.00	16.00
	4.00	6	20.0000	1.0954	.4472	18.8504	21.1496	19.00	21.00
	5.00	6	15.5000	.5477	.2236	14.9252	16.0748	15.00	16.00
	6.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	Total	36	18.1667	4.2995	.7166	16.7119	19.6214	14.00	28.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Absidia corymbifera (day 3)	Between Groups	92.250	5	18.450	16.043	.000
	Within Groups	34.500	30	1.150		
	Total	126.750	35			
Absidia corymbifera (day 4)	Between Groups	104.000	5	20.800	29.714	.000
	Within Groups	21.000	30	.700		
	Total	125.000	35			
Absidia corymbifera (day 5)	Between Groups	116.250	5	23.250	31.000	.000
	Within Groups	22.500	30	.750		
	Total	138.750	35			
Aspergillus terreus (day 3)	Between Groups	95.250	5	19.050	22.412	.000
	Within Groups	25.500	30	.850		
	Total	120.750	35			
Aspergillus terreus (day 4)	Between Groups	75.000	5	15.000	13.636	.000
	Within Groups	33.000	30	1.100		
	Total	108.000	35			
Aspergillus terreus (day 5)	Between Groups	620.000	5	124.000	137.778	.000
	Within Groups	27.000	30	.900		
	Total	647.000	35			

Post Hoc Tests

Homogeneous Subsets

Absidia corymbifera (day 3)

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
1.00	6	10.5000		
6.00	6	10.5000		
3.00	6	11.0000		
4.00	6	11.0000		
5.00	6		12.5000	
2.00	6			15.0000
Sig.		.469	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Absidia corymbifera (day 4)

Duncan[a]

		Subset for alpha = .05			
VAR00001	N	1	2	3	4
6.00	6	12.0000			
1.00	6	12.5000	12.5000		
3.00	6	13.0000	13.0000		
5.00	6		13.5000		
4.00	6			15.0000	
2.00	6				17.0000
Sig.		.058	.058	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Absidia corymbifera (day 5)

Duncan[a]

VAR00001	N	Subset for alpha = .05			
		1	2	3	4
6.00	6	13.5000			
1.00	6	14.5000	14.5000		
3.00	6		15.0000		
5.00	6		15.5000		
4.00	6			17.0000	
2.00	6				19.0000
Sig.		.055	.067	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Aspergillus terreus (day 3)

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
1.00	6	11.0000		
5.00	6	11.0000		
6.00	6	11.0000		
3.00	6	12.0000	12.0000	
2.00	6		13.0000	
4.00	6			15.5000
Sig.		.095	.070	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Aspergillus terreus (day 4)

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
6.00	6	12.5000		
5.00	6	13.0000	13.0000	
1.00	6	13.5000	13.5000	
2.00	6		14.0000	
3.00	6		14.0000	
4.00	6			17.0000
Sig.		.128	.141	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Aspergillus terreus (day 5)

Duncan[a]

VAR00001	N	Subset for alpha = .05				
		1	2	3	4	5
6.00	6	14.0000				
5.00	6		15.5000			
3.00	6		16.0000	16.0000		
2.00	6			17.0000		
4.00	6				20.0000	
1.00	6					26.5000
Sig.		1.000	.369	.078	1.000	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Emericella nidulans (day 3)	1.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	2.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	3.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	4.00	6	15.5000	1.6432	.6708	13.7756	17.2244	14.00	17.00
	5.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	6.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	Total	36	12.0833	1.8727	.3121	11.4497	12.7170	10.00	17.00
Emericella nidulans (day 4)	1.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	2.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	3.00	6	14.5000	.5477	.2236	13.9252	15.0748	14.00	15.00
	4.00	6	18.0000	2.1909	.8944	15.7008	20.2992	16.00	20.00
	5.00	6	12.5000	1.6432	.6708	10.7756	14.2244	11.00	14.00
	6.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	Total	36	14.1667	2.2233	.3705	13.4144	14.9189	11.00	20.00
Emericella nidulans (day 5)	1.00	6	16.0000	1.0954	.4472	14.8504	17.1496	15.00	17.00
	2.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	3.00	6	16.0000	.0000	.0000	16.0000	16.0000	16.00	16.00
	4.00	6	20.0000	2.1909	.8944	17.7008	22.2992	18.00	22.00
	5.00	6	15.0000	2.1909	.8944	12.7008	17.2992	13.00	17.00
	6.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	Total	36	16.1667	2.2615	.3769	15.4015	16.9318	13.00	22.00
Mucor racemosus (day 3)	1.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	2.00	6	13.5000	1.6432	.6708	11.7756	15.2244	12.00	15.00
	3.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	4.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	5.00	6	10.5000	.5477	.2236	9.9252	11.0748	10.00	11.00
	6.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	Total	36	11.5000	1.4041	.2340	11.0249	11.9751	10.00	15.00
Mucor racemosus (day 4)	1.00	6	14.0000	1.0954	.4472	12.8504	15.1496	13.00	15.00
	2.00	6	15.5000	1.6432	.6708	13.7756	17.2244	14.00	17.00
	3.00	6	14.0000	1.0954	.4472	12.8504	15.1496	13.00	15.00
	4.00	6	14.5000	.5477	.2236	13.9252	15.0748	14.00	15.00
	5.00	6	12.5000	.5477	.2236	11.9252	13.0748	12.00	13.00
	6.00	6	13.5000	1.6432	.6708	11.7756	15.2244	12.00	15.00
	Total	36	14.0000	1.4343	.2390	13.5147	14.4853	12.00	17.00
Mucor racemosus (day 5)	1.00	6	16.0000	1.0954	.4472	14.8504	17.1496	15.00	17.00
	2.00	6	18.0000	1.0954	.4472	16.8504	19.1496	17.00	19.00
	3.00	6	16.0000	1.0954	.4472	14.8504	17.1496	15.00	17.00
	4.00	6	16.5000	.5477	.2236	15.9252	17.0748	16.00	17.00
	5.00	6	16.5000	2.7386	1.1180	13.6260	19.3740	14.00	19.00
	6.00	6	15.5000	1.6432	.6708	13.7756	17.2244	14.00	17.00
	Total	36	16.4167	1.6279	.2713	15.8659	16.9675	14.00	19.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Emericella nidulans (day 3)	Between Groups	91.250	5	18.250	17.381	.000
	Within Groups	31.500	30	1.050		
	Total	122.750	35			
Emericella nidulans (day 4)	Between Groups	122.000	5	24.400	14.353	.000
	Within Groups	51.000	30	1.700		
	Total	173.000	35			
Emericella nidulans (day 5)	Between Groups	113.000	5	22.600	10.273	.000
	Within Groups	66.000	30	2.200		
	Total	179.000	35			
Mucor racemosus (day 3)	Between Groups	36.000	5	7.200	6.545	.000
	Within Groups	33.000	30	1.100		
	Total	69.000	35			
Mucor racemosus (day 4)	Between Groups	30.000	5	6.000	4.286	.005
	Within Groups	42.000	30	1.400		
	Total	72.000	35			
Mucor racemosus (day 5)	Between Groups	22.250	5	4.450	1.894	.125
	Within Groups	70.500	30	2.350		
	Total	92.750	35			

Post Hoc Tests

Homogeneous Subsets

Emericella nidulans (day 3)

Duncan[a]

		Subset for alpha = .05	
VAR00001	N	1	2
2.00	6	11.0000	
5.00	6	11.0000	
6.00	6	11.0000	
1.00	6	12.0000	
3.00	6	12.0000	
4.00	6		15.5000
Sig.		.141	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Emericella nidulans (day 4)

Duncan[a]

		Subset for alpha = .05		
VAR00001	N	1	2	3
5.00	6	12.5000		
2.00	6	13.0000	13.0000	
6.00	6	13.0000	13.0000	
1.00	6	14.0000	14.0000	
3.00	6		14.5000	
4.00	6			18.0000
Sig.		.077	.077	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Emericella nidulans (day 5)

Duncan[a]

VAR00001	N	Subset for alpha = .05	
		1	2
2.00	6	15.0000	
5.00	6	15.0000	
6.00	6	15.0000	
1.00	6	16.0000	
3.00	6	16.0000	
4.00	6		20.0000
Sig.		.307	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Mucor racemosus (day 3)

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
5.00	6	10.5000		
1.00	6	11.0000	11.0000	
3.00	6	11.0000	11.0000	
6.00	6	11.0000	11.0000	
4.00	6		12.0000	
2.00	6			13.5000
Sig.		.459	.141	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Mucor racemosus (day 4)

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
5.00	6	12.5000		
6.00	6	13.5000	13.5000	
1.00	6	14.0000	14.0000	14.0000
3.00	6	14.0000	14.0000	14.0000
4.00	6		14.5000	14.5000
2.00	6			15.5000
Sig.		.052	.191	.052

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Mucor racemosus (day 5)

Duncan[a]

VAR00001	N	Subset for alpha = .05	
		1	2
6.00	6	15.5000	
1.00	6	16.0000	16.0000
3.00	6	16.0000	16.0000
4.00	6	16.5000	16.5000
5.00	6	16.5000	16.5000
2.00	6		18.0000
Sig.		.323	.050

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Descriptives

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
						Lower Bound	Upper Bound		
Penicillum citrinum (day 3)	1.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	2.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	3.00	6	12.0000	.0000	.0000	12.0000	12.0000	12.00	12.00
	4.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	5.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	6.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	Total	36	12.0000	1.6562	.2760	11.4396	12.5604	10.00	16.00
Penicillum citrinum (day 4)	1.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	2.00	6	14.0000	.0000	.0000	14.0000	14.0000	14.00	14.00
	3.00	6	13.5000	.5477	.2236	12.9252	14.0748	13.00	14.00
	4.00	6	16.5000	1.6432	.6708	14.7756	18.2244	15.00	18.00
	5.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	6.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	Total	36	13.8333	1.5946	.2658	13.2938	14.3729	12.00	18.00
Penicillum citrinum (day 5)	1.00	6	14.5000	.5477	.2236	13.9252	15.0748	14.00	15.00
	2.00	6	16.0000	.0000	.0000	16.0000	16.0000	16.00	16.00
	3.00	6	15.5000	.5477	.2236	14.9252	16.0748	15.00	16.00
	4.00	6	16.5000	4.9295	2.0125	11.3268	21.6732	12.00	21.00
	5.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	6.00	6	15.5000	.5477	.2236	14.9252	16.0748	15.00	16.00
	Total	36	15.5000	2.0494	.3416	14.8066	16.1934	12.00	21.00
Penicillum aschersonia (day 3)	1.00	6	11.5000	.5477	.2236	10.9252	12.0748	11.00	12.00
	2.00	6	12.0000	1.0954	.4472	10.8504	13.1496	11.00	13.00
	3.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	4.00	6	15.5000	.5477	.2236	14.9252	16.0748	15.00	16.00
	5.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	6.00	6	11.0000	1.0954	.4472	9.8504	12.1496	10.00	12.00
	Total	36	12.0000	1.8516	.3086	11.3735	12.6265	10.00	16.00
Penicillum aschersonia (day 4)	1.00	6	13.5000	.5477	.2236	12.9252	14.0748	13.00	14.00
	2.00	6	14.0000	1.0954	.4472	12.8504	15.1496	13.00	15.00
	3.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	4.00	6	17.0000	1.0954	.4472	15.8504	18.1496	16.00	18.00
	5.00	6	12.0000	2.1909	.8944	9.7008	14.2992	10.00	14.00
	6.00	6	13.0000	1.0954	.4472	11.8504	14.1496	12.00	14.00
	Total	36	13.7500	1.9911	.3318	13.0763	14.4237	10.00	18.00
Penicillum aschersonia (day 5)	1.00	6	16.0000	1.0954	.4472	14.8504	17.1496	15.00	17.00
	2.00	6	16.0000	1.0954	.4472	14.8504	17.1496	15.00	17.00
	3.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	4.00	6	19.0000	1.0954	.4472	17.8504	20.1496	18.00	20.00
	5.00	6	15.5000	.5477	.2236	14.9252	16.0748	15.00	16.00
	6.00	6	15.0000	1.0954	.4472	13.8504	16.1496	14.00	16.00
	Total	36	16.0833	1.6797	.2800	15.5150	16.6517	14.00	20.00

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Penicillum citrinum (day 3)	Between Groups	72.000	5	14.400	18.000	.000
	Within Groups	24.000	30	.800		
	Total	96.000	35			
Penicillum citrinum (day 4)	Between Groups	56.000	5	11.200	10.182	.000
	Within Groups	33.000	30	1.100		
	Total	89.000	35			
Penicillum citrinum (day 5)	Between Groups	15.000	5	3.000	.682	.641
	Within Groups	132.000	30	4.400		
	Total	147.000	35			
Penicillum aschersonia (day 3)	Between Groups	93.000	5	18.600	20.667	.000
	Within Groups	27.000	30	.900		
	Total	120.000	35			
Penicillum aschersonia (day 4)	Between Groups	89.250	5	17.850	10.818	.000
	Within Groups	49.500	30	1.650		
	Total	138.750	35			
Penicillum aschersonia (day 5)	Between Groups	67.250	5	13.450	12.810	.000
	Within Groups	31.500	30	1.050		
	Total	98.750	35			

Post Hoc Tests

Homogeneous Subsets

Penicillum citrinum (day 3)

Duncan[a]

		Subset for alpha = .05	
VAR00001	N	1	2
1.00	6	11.0000	
5.00	6	11.0000	
6.00	6	11.0000	
2.00	6	12.0000	
3.00	6	12.0000	
4.00	6		15.0000
Sig.		.092	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Penicillum citrinum (day 4)

Duncan[a]

		Subset for alpha = .05	
VAR00001	N	1	2
1.00	6	13.0000	
5.00	6	13.0000	
6.00	6	13.0000	
3.00	6	13.5000	
2.00	6	14.0000	
4.00	6		16.5000
Sig.		.150	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Penicillum citrinum (day 5)

Duncan[a]

VAR00001	N	Subset for alpha = .05
		1
1.00	6	14.5000
5.00	6	15.0000
3.00	6	15.5000
6.00	6	15.5000
2.00	6	16.0000
4.00	6	16.5000
Sig.		.156

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Penicillum aschersonia (day 3)

Duncan[a]

VAR00001	N	Subset for alpha = .05	
		1	2
3.00	6	11.0000	
5.00	6	11.0000	
6.00	6	11.0000	
1.00	6	11.5000	
2.00	6	12.0000	
4.00	6		15.5000
Sig.		.112	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Penicillum aschersonia (day 4)

Duncan[a]

VAR00001	N	Subset for alpha = .05		
		1	2	3
5.00	6	12.0000		
3.00	6	13.0000	13.0000	
6.00	6	13.0000	13.0000	
1.00	6	13.5000	13.5000	
2.00	6		14.0000	
4.00	6			17.0000
Sig.		.073	.228	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.

Penicillum aschersonia (day 5)

Duncan[a]

VAR00001	N	Subset for alpha = .05	
		1	2
3.00	6	15.0000	
6.00	6	15.0000	
5.00	6	15.5000	
1.00	6	16.0000	
2.00	6	16.0000	
4.00	6		19.0000
Sig.		.141	1.000

Means for groups in homogeneous subsets are displayed.

a. Uses Harmonic Mean Sample Size = 6.000.