REVIEW OF LITERATURE
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External eye is in continuation with the skin surface and directly exposed to the environment. Pathogens can get access to the cornea from the environment. In immunocompromised subjects even the commensals of the outer eye may become pathogenic and cause keratitis. Usually inoculation of the pathogen is done by damage of the protective covering of cornea, i.e. the epithelium by minor trauma but some pathogens can break the epithelial barrier and get access in the cornea.

2. TYPES OF ULCERATIVE KERATITIS:

Ulcerative keratitis may be bacterial, mycotic or viral in origin. Comparatively the ulcerative keratitis of viral origin is less common (Duke Elder, 1965). The commonest viruses are herpes simplex, vaccinia and varicella viruses. The major bacterial families that cause ulcerative keratitis are micrococcaceae, streptococcaceae, pseudomonaceae and enterobacteriaceae. The organisms belonging to these families are Staphylococcus aureus, Staphylococcus epidermidis, Micrococcus spp, Streptococcus haemolyticus, Streptococcus pneumoniae, Pseudomonas aeruginosa, Proteus spp., Klebsiella pneumoniae etc. Apart from these families other organisms also can cause ulcerative
keratitis, such as, *Haemophilus*, *Listeria*, *Moraxella* and
*Mycobacterium* etc. (Tabbara and Hyndiuk, 1986). The
organism causing keratomycosis commonly include
*Aspergillus* spp., *Fusarium* spp., *Curvularia* spp.,
*Penicillium* spp. and *Helminthosporium* spp. Since
bacterial and fungal origin of keratitis is much more
common, it has been discussed in detail.

2.1 Bacterial keratitis:

In spite of social awareness, better diagnostic
methods for detection of causative agents, and newer
broad spectrum antibiotics, bacterial diseases of the
cornea still remain an important health hazard either in
the form of impairment of vision or complete loss of
vision.

2.1.1 Historical background:

The term keratitis was first introduced by James
Wardrop (1782-1869) in his essays in the "Morbid anatomy
of the human eye". Earlier it was a general practice to
refer all cases of ocular infection as "Ophthalmias". Gasparrini (1893) and Unthoff and Axenfeld (1896)
discovered the majority of hypopyon ulcer cases caused
by pneumococci. Gifford (1896) published the description
of acute conjunctivitis and ulcer and demonstrated the
transmission of the infection by inoculation of the
secretion containing pneumococci.
For the first time, the bacteria were described and isolated as ocular pathogens during the late nineteenth century. Neisser, in 1879, discovered the first bacterial pathogen in the eye and thus the genus bears his name *Neisseria gonorrhoeae* from a patient with purulent urethritis and conjunctivitis, and his findings were confirmed by many clinicians in the clinical samples in 1881, and within a few years intracellular nature and toxins produced by the *N. gonorrhoeae* had been studied, thus establishing an early scientific basis for gonococcal infection (Duke-Elder’s et al., 1965).

In the year 1881, Hirchbenth described rod-shaped bacteria, that he found associated with trachoma and the organism was later placed in the genus *Haemophilus* and given the species name *aegyptius* because of its prevalence in Egypt, but later on more correctly classified as haemagglutinable variant of *Haemophilus influenzae*, bitype -3, (Kilian et al., 1976)

In 1896 Morax and independently Axenfeld identified a gram negative diplobacillus that caused chronic subacute conjunctivitis (Axenfeld, 1897 and Morax 1896). The organism was soon named the Morax-Axenfeld bacillus, but in 1899, Petit discovered a diplobacillus that he believed was different from the Morax-Axenfeld organism. Petit’s organism was associated with a central hypopyon
cornel ulcer, whereas the Morax-Axenfeld diplobacillus was only associated with chronic, subacute conjunctivitis and sometimes marginal ulcers (Fedukowicz, et al. 1953 and Petit, 1899). It has now been established that there is difference between the two organisms; thus the Morax-Axenfeld diplobacillus has been termed Moraxella lacunata and the diplobacillus of Petit has been designated as Moraxella lacunata subsp. liquefaciens. The liquefaciens variant differs in growing at room temperature and in its rapid liquefaction of gelatin. The variant can grow without the addition of natural animal protein. (Topley and Wilson, 1990)

The early history of Pseudomonas aeruginosa is interesting in that the pigment products of the organism were recognized long before the organism itself. Physicians recognized the poor prognostic implications of blue green pigment in a wound; and eventually blue green discolouration on surgical dressings was associated with the infection. Bacteriological proof, however, had to wait until 1882, when the organism originally Bacillus pyocyaneus was first isolated by Gessard (Gessard, 1882). The first case of corneal ulcer known to be caused by Pseudomonas was reported by Sattler at the International Congress of Ophthalmology in Germany (Sattler, 1881). The clinicians who reported
the first case of *Pseudomonas* corneal ulcer were impressed with the virulence of the organism, which caused extensive corneal necrosis.

2.1.2 Causative agents and their pathogenesis:

The bacteria isolated from ulcerative keratitis mainly belonged to four families, viz. micrococcaceae, streptococcaceae, pseudomonaceae and enterobacteriaceae. The genera *Haemophilus*, *Moraxella* and *Listeria* have been reported as the rare causes of ulcerative keratitis. (Hyndiuk, 1981; Gutiérrez, 1972; Duke-Elder, 1965; Fedukowicz, 1978; Laibson, 1972; Leibowitz, 1984; Liesegang, 1980; Ostler, 1978; Sigtenhorst, 1957; Vaughn, 1886).

The *Staphylococcus* is usually considered the most frequent cause of bacterial keratitis. (Leibowitz, 1984) The significance of staphylococci as ocular pathogens rests not only on their prevalence but also on their ability to develop antibiotic resistance and virulence. Many strains of staphylococci are beta-lactamase producing and are resistant to penicillins. The hospital personnel harbouring such strains in their throat and nose may act as a source of nosocomial infections.

Staphylococci are gram positive, non-encapsulated cocci of the family micrococcaceae. These are catalase positive. Coagulase positivity is an indication of
potential pathogenicity for the human host. As such the test is important to ascertain whether or not the strain in question is a human pathogen. The coagulase positive species of the genus *Staphylococcus* are *Staphylococcus aureus*, *Staph. intermedius* and *Staph. hyicus* (Leibowitz, 1984). The virulence of *Staphylococci* is related to the production and release of toxins and enzymes. Pathogenic staphylococci release a variety of exotoxins including haemolysin, leucocyte destroying leucocidin, alpha-and beta-toxins (Gemmell and Roberts, 1974; Heczko et al., 1974). The enzymes generated by the organism include hyaluronidase, lipase, nucleases and staphylokinase (Christie and Wilsor, 1941; Quie and Wannamaker, 1961; Arvindson, 1983). These toxins and enzymes render the staphylococci resistant to phagocytosis.

Jones has outlined the sequential events in bacterial keratitis (Jones, 1978) and Hyndiuk has emphasised the importance of initial adhesion in the pathogenesis of corneal infections (Hyndiuk, 1983). The sequential events are outlined as under

1. Adhesion
2. Entry of the organism
3. Multiplication of the organism
4. Spread of the organism
5. Host inflammatory response
6. Phagocytosis
7. Host immune response
8. Tissue damage

Adhesion and entry of the organism is facilitated by an epithelial defect and/or stromal injury. Direct corneal invasion through intact epithelium is not common. Haematogenous spread and microbial spread via the anterior chamber are also rare events. Host cells, especially the injured cells and bacteria may have surfaces that promote adhesion. Bacterial keratitis begins with adhesion of the organisms to injured epithelium and stroma. The organisms invade through areas of absent or damaged epithelium.

On entry, the organisms multiply. Chemotactic factors are released that stimulate migration of polymorphonuclear leucocytes (PMNs). Early in the infection, PMNs migrate via the tear film and later from the limbus to the area of infection. Organism and host generated toxins are released, resulting in tissue destruction.

Organism-generated chemotactic factors, including microbial substances, tissue factors, complement, immune complexes and immune cells stimulate PMNs. The PMN phagocytic response is the principal host defence in most bacterial keratopathies. Cell mediated immunity is
important only with regard to certain intracellular bacteria such as *Mycobacterium* and *Listeria*. Humoral immunity is generally a second line of defence in such cases.

Organisms are engulfed and contained within intracellular vacuoles known as lysosomes. Enzymes such as lysozyme are released into the lysosomes, and the organisms are digested. (Baggiolini, 1972). In addition, oxidising agents such as myeloperoxidase (MPO) and oxidative burst phenomena play an important role in bacterial destruction. (Babior, 1978). The normal action of PMN include release of destructive enzymes and oxidising agents, which are better tolerated in vascular tissues than in the cornea. These host lysosomal enzymes cause further destruction of the corneal collagenous tissue (Van Horn, 1958). Thus the PMNs so important to host defence, become a source of further destruction in the avascular corneal stroma.

The organism toxins versus host toxins play role in the pathogenesis of the corneal destruction, which depends on the virulence of the organism and the magnitude of the host response. Tissue damage is followed by repair and recovery. But in some cases the destruction continues and results into descemetocele formation or perforation. Even in the natural course of healing, there is scar formation which results into
nebular, macular or leucomatous types of opacification and thereby impairing vision to varying grades, causing blindness.

**Streptococcus**:

In older reported series *Streptococcus pneumoniae* was the most common cause of bacterial keratitis. Its prevalence as an aetiologic agent seems to have been superseded both by *Staphylococcus* and *Pseudomonas* in more recent times (Leibowitz, 1984). *Pneumococcus* continues to be isolated frequently from bacterial corneal ulcers, a finding that undoubtedly reflects its normal habitation of the upper respiratory tract, lacrimal drainage apparatus, and at times the conjunctiva (Leibowitz, 1984). Other species such as *Streptococcus pyogenes*, *Streptococcus viridans*, *Streptococcus faecalis*, infrequently cause bacterial keratitis (Tabbara, 1986). These are gram positive cocci that divide in one plane to form pairs or chains. Haemolysins, (streptolysin-O and streptolysin-S), streptokinase, nucleases, hyaluronidase etc. are the toxins secreted by the organisms.

*Streptococcus pyogenes* secretes proteases which degrade proteins of cornea and causes its destruction. The sequence of events is similar to one described earlier in the pathogenesis of ulcerative keratitis by
Pneumococci produce an intracellular haemolysin that is liberated by autolysis. Pneumococci can be leucotactic. Pneumococci also produce immunoglobulin Al protease. (Jones, 1978). It also produces a neuraminidase and a hyaluronidase. The various toxins and enzymes are responsible for the invasiveness of the organism. Its invasiveness and virulence is related to its ability to invade and multiply in tissues rapidly, which in turn is related to presence of distinct capsular antigens. The capsule also provides protection from phagocytosis. (Davis, et al., 1973; Joklik, et al., 1976; and Dubos, et al., 1965).

Streptococcus pneumoniae produces a central corneal ulcer associated with hypopyon that forms early in the course of infection. The hypopyon remains sterile unless corneal perforation occurs. The ulcer spreads irregularly and it is undermined along its edge. The organisms are found at the progressive border, while the trailing edge tends to heal (Duke-Elder, 1965; Tabbara, 1986). The ulcer is generally well circumscribed with grayish white infiltrates in the ulcer centre and somewhat adjacent to the ulcer, with the surrounding cornea often remaining clear. As the ulcer progresses the deeper layers are involved.
**Moraxella**: Infection with Moraxella group was the most common cause of bacterial corneal ulcer in the population of derelict alcoholics in New York from 1965 to 1968 (Baum et al., 1980). With improvement in nutrition and sanitation marked decline in the incidence of corneal ulcer caused by Moraxella spp. has been noted (Baum et al. 1980). It is now recognized that it may cause central or marginal ulcerative keratitis.

**Neisseria**: Incidence of infection due to Neisseria is very low now, but the infection is highly contagious. *N. gonorrhoeae* and *N. meningitidis* are established human pathogens. The organisms adhere to epithelial cells of conjunctival and genito-urinary tract mucosa and colonise to cause suppuration. Neisseria are gram negative diplococci found within the cells. Meningococcal keratitis is rare in absence of concurrent meningitis (Odegard, 1944). Neisseria may penetrate intact epithelium.

**Enterobacteriaceae**: Some members of the family enterobacteriaceae can cause fulminant corneal ulcers such as *Serratia* sp., *E. coli*, and *Klebsiella*. (Liesegang, 1980 and Wilson, 1971). These ulcers are rare and are found in persons who wear contact lenses or as secondary infections. Okumoto from Japan in his study has reported that Proteus is the most common species to cause bacterial ulcerative keratitis in human eyes.
Serratia marcescens is a common contaminant of hospital equipments, so infection of corneal ulcers due to these organisms occur mostly in hospitalised patients. It can occur also in contact lens wearers (Lass, et al., 1981).

Klebsiella pneumoniae, Enterobacter, sp., and Citrobacter sp. have also been reported in literature as cause of ulcerative keratitis (Liesegang, et al., 1980).

Haemophilus sp : In Gram’s stained smear these are observed as Gram negative cocco-bacilli. These are significant aetiological agents for ocular infections. The keratitis is reported as a frequent complication of seasonal Haemophilus conjunctivitis in tropical climates (Minton, 1945).

Pseudomonas aeruginosa : The organism as a significant cause of bacterial keratitis has been emphasised by many workers (Hyndiuk, 1981; Jones, 1979; Wilson, 1984). In some studies Pseudomonas has replaced Streptococcus pneumoniae, Staphylococcus aureus, and Streptococcus sp. as the most frequent cause of bacterial keratitis. (Jones, 1979; Liesegang, 1980). The rapidly progressive, severe nature of the infection coupled with the increased frequency of infection makes
**Pseudomonas** the most important cause of severe corneal ulceration. (Laibson, 1972). The slime envelope capsule known as glycocalyx is important with respect to its adherence properties (Hyndiuk, 1981; Hyndiuk, 1983). *Pseudomonas aeruginosa* has simple growth requirements, tolerance of wide temperature swings and its resistance to chemical disinfection, thus accounting for its presence as a contaminant in many eye-related products, irrigating solutions, contact lenses and their related preparations and cosmetics (Thygeson, 1948; Vaughan, 1986; Wilson, 1977).

*Pseudomonas* spp. produce a variety of extracellular products that contribute to the development of corneal ulceration (Fisher et al., 1958). Pigments, proteases, haemolysins, exotoxins and endotoxins are produced by these organisms. Proteases include elastase, pseudocollagenase, nonspecific collagenase and proteoglycanase (Morihara, 1964; Fisher et al., 1958; Liu, 1979; Brown, 1974). Phospholipase is the primary haemolysin elaborated by *Pseudomonas aeruginosa*. Three exotoxins, viz. A, B and C may be produced by the organism. Exotoxin has been shown to kill corneal epithelium and endothelial cells with resulting corneal oedema (Iglewski, 1977).

Disturbance in any of the protective mechanisms of the eye, i.e. smooth corneal surface, tear, lid movement
may lead to the break in the corneal epithelium. Nearly all bacteria are opportunistic pathogens and are unable to penetrate intact corneal epithelium. The notable exceptions are *Neisseria gonorrhoeae*, *Corynebacterium Diphtheriae*, *Haemophilus influenzae* (Tabbara, et al., 1986)

2.1.3. Predisposing factors

(i) The eye cosmetics... Mascara, eyeliner, and eye shadow have been found responsible for eye infections (Wilson et. al., 1971). *Pseudomonas aeruginosa* has been isolated from cornea of mascara using ladies and the same organism was isolated from the cosmetic solution. When purchased the cosmetic solutions are free from microbes but contamination occurs during use. The disease is a significant health hazard in the ladies using the eye makeups. Wilson, et al., 1971 cultured 428 eye cosmetic samples for bacteria. Bacterial contamination was noted in 43% of the samples.

(ii) Contact-lenses have been associated with a variety of bacterial corneal ulcers (Dohlman, 1973, Krachmer et al., 1978). Factors contributing to the development of contact lens associated corneal ulcers include bacterial contamination of the lens or of the solutions and associated but undefined foreign bodies.
Drugs such as antibiotics (Tabbara, 1986), corticosteroids (Mitsui and Hanabusa, 1955) and immunosuppressive agents (Tabbara, 1986) impede the host defenses by several mechanisms. These include inhibition of chemotaxis, inhibition of phagocytosis, blockage of degranulation, interference with lysosomal levels and reduction in the proportion of phagocytes (Jones, 1978). Corticosteroids suppress inflammation and many mask significant clinical signs, thus sometimes delaying recognition of infection (Mitsui and Hanabusa, 1955; Valenton, 1972).

In the treatment of the ocular inflammations, nowadays cortisones and hydrocortisones are widely used, particularly in allergic conditions, such as phlyctenular kerato-conjunctivitis, vernal conjunctivitis and scleritis with improvement of symptoms. But cortisone is a double edged sword, in spite of beneficial effects, it may also increase the susceptibility of an individual to microorganisms. Mitsui and Hanabusa et al. (1955) reported a case of hypopyon keratitis due to an infection of Pseudomonas aeruginosa induced by topical cortisone. Corneal infection by fungi and Pseudomonas sp. cannot easily be controlled by most of the antibiotics so far available to us and it presents a serious hazard to the safety of the eye. It may therefore be suggested to make
periodical bacteriological examinations of the conjunctiva before and during a course of cortisone treatment. If any dangerous sign appears, the application should be stopped.

(iv) Burns are also considered as the predisposing factors. Epithelial loss favours access, multiplication and invasion by the pathogens. Certain endogenous local conditions are predisposing, such as lid disorders, lagophthalmos, exophthalmos, entropion and blepharitis. Some conjunctival and lacrimal disorders also predispose bacterial keratitis such as dacryocystitis, trachoma, xerophthalmos. Trigeminal anaesthesia, bullous keratopathy and herpetic ulcers also favour the ulcerative keratitis (Tabbara, et al., 1986).

(v) Some systemic factors have been described as predisposing in the literature, like alcoholism, allergies, blood dyscrasias, coma, diabetes, immune disorders, prematurity, nutritional deficiency and psychosis. (Tabbara, 1986).

2.1.4 Epidemiology.

The isolation rate of the pathogens of bacterial ulcerative keratitis varies depending on the time period of the study, the geographic area and the patient population under study.
Das et al. (1955) studied the bacteriology of 100 cases of corneal ulcer at Amritsar. He has reported that the commonest pathogen responsible was \textit{Staphylococcus aureus} (31\%). Next in importance was \textit{Pseudomonas} (14\%). Pneumococcus and \textit{Streptococcus viridans} was isolated from 8 percent respectively.

Rotagi (1967) isolated \textit{Streptococcus} in 20 percent cases while Sood (1968) isolated \textit{Pseudomonas} in 21.4 percent cases of hypopyon corneal ulcers.

Rao (1972) studied ulcer scrapings from fifty-eight cases of corneal ulcers. In his study he found that \textit{Pseudomonas} was the most dominant bacterial pathogen.

Aurora et al. (1971) studied 167 corneal buttons following keratoplasty for corneal ulceration in the urban population of Northern India. Microbiological studies were conducted in 95 cases. Out of these 59.7 percent cases showed bacterial infection. \textit{Pseudomonas aeruginosa} was isolated in 37.1 percent cases, \textit{Streptococcus pyogenes} in 4.8 percent cases and coagulase negative \textit{Staphylococci} in 17.7 percent cases.

Valenton (1978) in his study of 250 corneal ulcer cases in Philippines showed that the commonest offending bacteria were \textit{Pseudomonas} and \textit{Pneumococcus}.

Du Niam Zu et al. (1979) in Japan studied the
microbiology of seventy cases of purulent corneal ulceration. Bacterial infection was found in 37.1% out of which Pseudomonas infection was found in 14.3% and Staphylococcus in 10% of cases.

Liesegang and Forster (1980) studied the microbiology of 663 corneal ulcer cases in South Florida over a period of nine years (Jan. 1969 to Dec. 1977). 35.9 percent cases were positive for bacterial culture. Staphylococcus aureus, 21.4 percent cases, Streptococcus pneumoniae, 7.6 percent cases and Pseudomonas, 31.1 percent cases were found as the commonest offenders.

Chaudhuri et al., (1982) studied the corneal scrapings of 32 clinically diagnosed bacterial corneal ulcers. Staphylococcus aureus, 28.1 percent cases, Pneumococci, 9.4 percent cases and Pseudomonas, 6.3 percent cases were the commonest among 53 percent growth positive cases.

Carmichael et al. (1985) studied the microbiology of 91 cases of moderate to severe central corneal ulcers at an urban African Hospital. Out of these 62 (67%) cases showed bacterial infection. Streptococcus pneumoniae was the most common bacteria isolated.

Maske et al. (1986) studied 45 clinically diagnosed bacterial corneal ulcer cases while bacteria could be
isolated from 64 percent cases. In his series of patients Staphylococcus aureus was found in 13.8 percent cases, Staphylococcus epidermidis in 31.0 percent cases, Pseudomonas in 10.3 percent cases and Pneumococci in 20.6 percent cases.

The most common organism isolated by Parks et al. (1993) in his study of bacterial ulcerative keratitis were Staphylococcus epidermidis from 36 percent, Staphylococcus aureus from 18 percent and Pseudomonas aeruginosa from 13 percent cases.

**Bacterial keratitis in relation to age:**

As regards age the bacterial keratitis is more prevalent in the fourth and fifth decade of life. (Parks, et al., 1993).

**Bacterial keratitis in relation to sex:**

The disease is more common in males as compared to the females. The male female incidence ratio has been reported by some workers to be 3:2 (Liesegang, et al., 1980). But some workers have reported equal incidence in both the sexes (Parks, et al., 1993). The bacterial keratitis leading to corneal ulcer is more prevalent in outdoor life as the cornea is exposed more to the environmental bacteria. Although the disease has been reported in house-wives also, in them corneal trauma
occurs during household work and some bacteria, specially the pathogenic ones adhere to the cornea and cause the disease. The contaminated objects also at times inoculate the bacteria in the eroded epithelium as a result of trauma.

**Bacterial keratitis in relation to occupation:**

Bacterial ulcerative keratitis is considered to be occupational hazard of farmers and labourers. Those working in the environment of iron or dust particles frequently sustain ocular injuries and bacteria get an access to the abraded cornea. (Tabbara, et al., 1986).

2.1.5. *Antibacterial treatment:*

The most commonly prescribed form of therapy is fortified concentrations of topical antibiotics, given in a liquid vehicle. The fortified topical antibiotics provide high corneal and anterior chamber levels and are highly effective agents for bacterial killing (Davis, et al., 1979 and Hyndiuk, 1981). With topical preparations alone mild, moderate and severe type of bacterial ulcers have been treated (Davis, et al., 1977).

In addition to topical antibiotics, subconjunctival antibiotic injection is also used in some cases associated with endophthalmitis, imminent perforation, scleral extension of ulcer and deep extension of ulcer.
The mode of administration of antibiotics has also been successfully tried in non-complicated cases and also when fortified topical antibiotics are not available.

Some workers have prepared ointment with higher concentrations of antibiotics. The trial has given promising results with, e.g. Gentamicin 10 mg/gram to 40 mg/gram effective in killing Pseudomonas even in severe keratitis (Skorich, et al., 1985). Use of fortified ointment has been proved beneficial not only in keratitis but also in other infection of the outer eye. The advantage over antibiotic solutions is that the frequency of applications is markedly reduced with such preparations.

Use of antibiotics by intravenous route is rarely in use now. But some infections are better treated by dual administration, e.g. in Neisseria and Haemophilus. The corneal ulcers involving sclera or those having danger of impending perforation are also treated with intravenously injected antibiotics.

Antibiotics to be administered by any of the routes are selected on the basis of initial Gram's stained smear results and the sensitivity pattern of the organism/organisms isolated from the ulcer (Tabbara, et al., 1986). But there is controversy on the use of antibiotics before getting the culture reports, some
authors base antibiotic therapy on Gram's stain results (Jones, 1979). The policy being single organism treated with single antibiotic, as determined by Gram's stained smear examination, whereas two or more than two organisms to be treated with a broad-spectrum antibiotic. Other workers are little bothered about the correlation between Gram's stained smear results and the culture results and cover all suspected bacterial ulcers with broad spectrum antibiotics until culture results are reported (Baum, 1979; Hynduik, 1981; Hynduik et al., 1983; Leibowitz, 1984). Those who advocate the second view are of the opinion that a short course of broad spectrum antibiotics carries low risk in view of the serious nature of the infection (Tabbara, 1986). Resistance to broad spectrum antibiotics as reported in France has not yet been encountered elsewhere (Gelender, et al., 1984). Broad spectrum coverage consists of a topically applied aminoglycoside (fortified Gentamycin or Tobramycin) plus a Cephalosporin or Bacitracin. Subconjunctival antibiotics are added depending on the serious nature of the disease as described previously. If Gram's negative organisms such as Pseudomonas are suspected, therapy is modified to include topical and subconjunctival aminoglycoside plus Carbenicillin. This combination is synergistic for gram negative organisms (Hynduik, et al., 1983). The therapy is modified depending on final culture and
sensitivity reports. It is further continued if a
positive response is observed. Culture negative ulcers
may not respond to the treatment. In these cases, the
therapy should be discontinued for 24 to 48 hours and
the corneal specimen recultured in an attempt to recover
a causative agent.

2.2 **Fungal Keratitis (Keratomycosis):**

Fungal corneal infection is a common ailment in
developing countries like India. It frequently results
in visual disability by development of corneal opacity
and perforation, thereby loss of the eye. Corneal
capacification is the second major cause of blindness
after cataract. Farmers and labourers, who work in the
fields especially in India, may develop fungal keratitis
subsequent to corneal trauma by vegetative matter or
soil. Rarely it may occur following surgery like
keratoplasty. Injudicious use of topical corticosteroids
and antibacterial agents for external ocular diseases
predispose keratomycosis. The increasing frequency of
mycotic corneal ulcer is of considerable concern in
present day ophthalmic practice in India. There are
reports of high incidence from various parts of the
country. In the vegetative matter or soil of the fields
fungal spores are widely distributed which are implanted
into the cornea at the time of injury (Gingrich, 1962
and Puttanna, 1967).
2.2.1 **Historical background:**

Leber in 1879 reported the first case of keratomycosis due to *Aspergillus*. By now more than forty genera of fungi have been described, which can cause keratomycosis. Thygeson et al. (1953) was the first to draw attention to the development of fungal infection of the cornea after the use of topical corticosteroids. This was substantiated by several convincing studies by Mitsui and Hanabusia (1955) and others (Ley et al., 1956; Hirose, et al., 1957; Anderson et al., 1959). A review of the literature shows a definite increase of fungal infections all over the world. Mycotic infections of the eye have assumed increasing importance in recent years in India (Pankaj Lakshmi, et al., 1989 and Talwar et. al., 1978).

In Indian literature fungus as a causative agent of corneal ulcer was first reported in 1962 (Agarwal, et al., 1962). Later on a detailed study was done by the same workers in 1967 (Agarwal, et al., 1967).

2.2.2 **Causative agents; their prevalence and pathogenesis:**

More than forty genera of fungi are reported to be associated with keratomycosis, but only a few are recognized as human pathogens. (De Voe, et al., 1976). Most of the organisms are saprophytic that cause opportunistic corneal infection in traumatised or
immunologically compromised eyes. Candida albicans is the most important species of yeast responsible for keratomycosis.

Aspergillus: In India Aspergillus is the commonest fungus that invades the cornea. This finding is supported by the Indian workers who isolated Aspergillus from majority of their series of corneal ulcer cases —— Puttanna et al. (1969) from 41.1 percent, Sharma et al., (1986) from 52.6 percent, Halder et al., (1992) from 50.7 percent and Chander et al. (1993) from 40.0 percent of the cases. Generally in Aspergillus keratitis, superficial layers of cornea are involved and typical ulcer is described as a raised circular grey plaque with well demarcated rolled-out edges. Cauliflower like lesions are also reported (Bothman and Crowe, 1947). Castrovejo and Munoz, in 1921, reported perforation of these corneal ulcers.

Primary aspergillus keratitis could be produced experimentally in animal cornea by injecting culture isolates from the fungus from the clinical cases (Agarwal, et al., 1963 and Puttanna, 1969). Experiments have further revealed that the inflammatory responses varied widely in the same species of aspergillus both in experimental animal cornea and in human lesions (Puttanna, 1969).
The species of *Aspergillus* isolated in different studies are *Aspergillus fumigatus*, *Aspergillus flavus*, *A. nidulans* (Puttanna, 1969), *A. glaucus* (Chander, et al. 1993), *A. terreus* (Koul, et al., 1975), but *A. fumigatus* is the commonest species isolated by these workers. It can be either primary or secondary invader. As a primary invader it causes superficial keratomycosis and the secondary infections have been reported to be associated with dendritic ulcers, ulcer serpens, eczematous pannus and inclusion catarrh (Fazakas, 1959).

**Fusarium** :- It is a soil saprophyte and has been reported as a cause of keratomycosis (Sigtenhorst, et al., 1957). It can be isolated from almost any soil or water sample. The infection is said to occur at the time of injury with vegetative matter. In India, *Fusarium* has been reported as an agent of keratomycosis by various authors (Puttanna, 1969; Dasgupta, et al., 1973; Grover, 1975; Sandhu et al., 1981; Patwardhan, 1991; Haldar, et. al., 1992 and Chander, et al., 1993). Animal experimental study has been conducted (Puttanna, 1969) and the fusarium keratitis was produced into rat's cornea after injection of culture material from a patient.

In America *Fusarium* is the predominant fungus isolated from clinical samples (Liesegang, et al., 1980). The incidence reported in India is variable, the
difference being on account of variable geographical conditions, e.g. in Bangalore 6% (Puttanna, 1969), in Pondicherry 13.3% (Dasgupta, 1973), in Nagpur, 7.2% (Grover, 1973) in Amritsar, 3.5% (Sandhu, 1981), in Aurangabad, 37.5% (Patwardhan, et al., 1991), in Darjeeling, 3.2% (Halder, 1992), in Chandigarh, 14% (Chander, et al., 1993). Soil is the home of the fungus and the fungi constitute micro-fauna of a place. The micro-fauna of different places is variable, so the diversity as noted above is self explanatory.

**Penicillium:**

It is a frequent isolate from keratomycosis. In Bangalore (Puttanna, 1969) 9% cases of keratomycosis showed presence of Penicillium as a causative agent. The culture isolate, when injected into rat's cornea, produced keratitis at the site of injection. Scrapings from the rat's cornea showed fungal hyphae. (Puttanna, 1960).

Incidence is variable in various parts of India. In Pondicherry Penicillium constitutes 2.2% of the fungal isolates from keratomycosis, (Dasgupta, et al., 1973). in Jaipur it is 8.7% (Kulshrestha, 1973), in Lucknow 4.5% (Koul, et al., 1975), in Nagpur the fungus has been isolated in 14.3% of cases (Grover et. al., 1975), in Darjeeling the incidence has been reported to be 6.3%
(Haldar, et al., 1992), and in Chandigarh 4% only (Chander, et al., 1993).

As reported by foreign workers in the U.S.A. (Liesegang, et al., 1980) and the U.K. (Jones, et al., 1968) the fungus constitutes only a small percentage of the fungal isolates in keratomycosis.

**Curvularia:**

It is pigmented filamentous fungus which belongs to family dematiaceae and the species reported in the literature include C. senegalensis, C. verruculosa, C. lunata (Liesegang, et al., 1980). Generally there is history of trauma by vegetative matter in such cases.

In India various workers have isolated the fungus from clinical samples. In Pondicherry (Dasgupta, et al., 1973) the percentage of Curvularia in the fungal isolates is 9.0% in Nagpur, the occurrence is only 7% (Grover, et al., 1975). In Amritsar it is 3.6% of all the fungal isolates from keratomycosis (Sandhu, et al., 1981). In Darjeeling 1.8% of cases are reported to be caused by Curvularia. Curvularia is the cause of 10% cases of fungal keratitis (Chander, et al., 1993).

**Helminthosporium:**

It is a saprophytic dematiaceous fungus commonly found in the environment (Forster, 1975). It is
infrequent cause of keratomycosis. Some workers feel that dematiaceous infections are less predictably progressive and destructive than the monaliaceous ones. (Forster, 1975). The infection is associated with corneal trauma by a vegetative matter or contact lens and it is predisposed by long use of corticosteroids (Krechmer, et al., 1978).

The fungus is found to be a more prevalent ocular pathogen than it was previously recognized. The incidence of the Helminthosporium corneal ulcers varies in different parts of India. In Pondicherry 4.4% incidence has been reported (Dasgupta, et al., 1973). In Amritsar, 2.9% cases of corneal ulcers have been reported to be Helminthosporium positive. In Florida the incidence is reported to be 6.3% (Forster et al., 1975). In Chandigarh the reported incidence is 1% (Kalder, et al., 1993).

**Phialophora:**

The pathogen belonging to this genus, Phialophora verrucosa was isolated for the first time in Florida (Jones, et al., 1963). They found it in 2.3% cases of keratomycosis. From the same place, later on, this dematiaceous fungus was isolated by other workers too (Forster, et al., 1975). It was isolated from 6% of corneal ulcer cases. In Indian literature the genus has
not been isolated from clinical samples of keratomycosis.

The fungus causes chromomycosis and so the lesion in the eye is expected to be black in colour on the cornea.

**Candida:**

*Candida albicans*, the causative agent of some cases of keratomycosis is an yeast like organism. This affects lids, conjunctiva, cornea, tear duct and uvea. Mendelblatt reported the first case of corneal involvement due to *Candida albicans* (Mendelblatt, 1953).

Corneal ulcers caused by this fungus are shallow and indolent with undermined and infiltrated edges (Birge, 1941). The floor of the ulcer is usually covered with a thin dry membrane which adheres to the corneal tissue. It is associated with iritis and hypopyon. Pathogenecity of the fungus has been proved by inoculation of culture obtained from a patient into rabbit's cornea experimentally (Graf, 1963). Corneal ulcer due to *Candida parapsilosis* also has been reported (Manchester, et al., 1959).

Incidence in various parts of India has been reported in the literature as follows: 12% from Bangalore (Puttanna, 1969), 13.3 percent from Pondicherry (Dasgupta, et al., 1973) 7.1 percent from Nagpur (Grover
et al., 1975), 0.7% from Amritsar (Sandhu et al., 1981). 0.8% from Patiala (Sharma, et al., 1986), 17.5 percent from Darjeeling, (Haidar et al., 1992). 10 percent from Chandigarh, (Chander, et al., 1993). Various species reported are Candida albicans C. tropicalis, C. guilliermondii, C. krusei.

**Cephalosporium**

The fungus was first isolated from a case of corneal ulcer sustained from an injury of a cow's tail (Bedell, A.J., 1946). A potent proteolytic enzyme has been extracted from Cephalosporium which experimentally produced corneal destruction in 2 to 4 hours in rabbit's cornea (Burda, et al., 1960) and it has been established that the proteinase may be responsible for the clinical picture of Cephalosporium keratitis.

Only a few cases of Cephalosporium keratitis have been reported in the literature. Puttanna from Bangalore has reported 6% cases (Puttanna, 1969). In Tennessee 4% incidence was reported (O'Day, et al., 1979) whereas in Amritsar Cephalosporium has been isolated only in 1.4% cases of keratomycosis.

**Rhizopus**

It is a rare isolate from cases of keratomycosis. From Bangalore the incidence of this isolate in fungal
keratitis is reported to be 6% (Puttanna, 1969) and the worker has described the mode of inoculation of fungus in the eye by instillation of herbal juice in the eye following trauma of the cornea as a part of native treatment. The worker had inoculated the culture isolate into rat's cornea, which showed keratitis 48 hours after inoculation. In a study from Jaipur the incidence of Rhizopus is reported in 13% cases of keratomycosis (Kulshrestha, et al., 1973). From Patiala it has been reported in 5% only (Sharma, 1981), but a slightly higher percentage (6.8%) has been reported in another series of patients (Sharma, et al., 1986).

 Cryptococcus :-  

 The fungus is a true yeast and it has been reported as a causative agent of keratomycosis (Fazakas, 1953). He described deep and very extensive corneal involvement by the fungus, which results into corneal opacity rather than perforation.

 Pathogenesis :-

 Histopathological findings include loss of corneal epithelium, Bowman's layer and variable amounts of corneal stroma. The secretion of enzymes such as phospholipase, protease, and pseudocollagenase causes coagulative necrosis with the loss of keratocytes and disruption of collagen lamellae (Burds, 1960 and
The surrounding inflammatory cell infiltrate is typically granulomatous reaction although chronic non-granulomatous and purulent inflammatory reactions may also occur (Yanoff, et al., 1975 and Zimmerman, 1963).

As in other forms of microbial keratitis, fungal invasion of the cornea commonly stimulates an outpouring of a sterile hypopyon. Descemet's membrane acts as a relative barrier to limit fungal invasion of the cornea; however the fungi may pass through an intact Descemet's membrane (Naumann, et al., 1967).

Invading fungal elements may be identified in histological preparations processed with special fungal stains. Since invading fungi lack specialized reproducing structures, they cannot be classified according to their histologic appearance (Naumann, et al., 1967). However, large round on oval budding forms on pseudohyphae suggest the presence of yeast.

In keratomycosis, fungi may be identified histologically throughout all levels of the cornea and may extend beyond boundaries of clinically recognised infection (Kaufman, et al., 1965 and Naumann, et al., 1967). Hyphal elements are commonly arranged parallel to collagen lamellae, but perpendicular orientation can be there which implies increased virulence (Naumann, et
Fungi are often absent from the ulcer base and superficial corneal stroma (Naumann et al., 1967). The fact is of clinical significance when performing a diagnostic scraping on keratectomy and may explain findings of negative cultures in cases of progressive fungal infection. In complete excision of infected tissues in lamellar keratoplasty may lead to subsequent fungal proliferation in the graft host - interface, with destruction of the graft (Kaufman et al., 1965; Naumann et al., 1967; Singh et al., 1974).

Peripherally located corneal micro abscesses reveal histological findings similar to the main ulcer and correspond to the satellite lesions noted clinically. Peripheral corneal ring abscesses are characteristic of fungal keratitis and are composed of collections of polymorphonuclear leucocytes, plasma cells and eosinophils around invading hyphae (Kaufman et al., 1965; Naumann et al., 1967 and Zimmerman, 1963). The ring abscess formation probably represents a host immune response to fungal antigen and corresponds to the immune ring of Wessely commonly seen clinically (Jones, 1978; Kaufman et al., 1967).

2.2.3 PREDISPOSING FACTORS:

The fungi get entry into the eye and to the cornea by direct vegetative matter injury, instillation of
herbal drops in the countryside, extension of infection from infected neighborhood organs as in fungal dermatitis, nasopharyngitis and sinusitis and lack of personal hygiene especially women suffering from fungal vaginitis and allied fungal disorders. (De Voe, 1951; Kaufman and Wood, 1965; Liesegang and Forster, 1980; Polack et al. 1971).

(i) Some Systemic diseases:

There is evidence to show that even non-pathogenic species under certain conditions assume pathogenic character and proliferate on and penetrate into tissue producing necrosis. This is specially seen in patients with low resistance as in debilitated individuals, diabetics, leukaemics and those affected by cancer. (Jones, 1981; Bancroft and Stevens, 1982; Thygeson and Okumoto, 1974)

(ii) The contributing role of corticosteroids and antibiotics has become much important in recent years. This is due to the fact that the normal balance that exists between bacteria and fungi has been disturbed and the opportunistic fungi gain the upper hand and proliferate readily. So prolonged therapy with antibiotics and corticosteroids favour fungal infections of the eye. (Anderson et al., 1959; Ellison and Newmark, 1973; Haggerty and Zimmeraman, 1958; Polack, et al., 1971; Thygeson and Okumoto, 1974; Mitsui and Hanabusa.
Mitsui and Hanabusa (1955) found fungi in the conjunctival sacs of 67% of patients who were using topical steroids but only in 18% controls, and that 50% of those who had negative cultures became fungus positive after three weeks of topical steroid therapy.

Corticosteroids may indirectly promote fungal replication and corneal invasion by interfering with the host's inflammatory response (Jones, 1978). Chronic topical corticosteroid therapy also increases human conjunctival colonization by fungi (Mitsui and Hababusa, 1955).

(iii) Trauma:

Any minor trauma of the cornea favours such invasion, because mycotic infections never occur spontaneously on an intact healthy cornea. Hence trivial injuries of the cornea, especially in the field, caused by a cow's tail while milking, twigs, thorns, inflorescence of paddy or rice and other field crops or dusty grains or saw dust which may contain the fungal elements can well be the cause of mycotic infections of the cornea. (Doughman, et al., 1982; Jones, et al., 1969; Liesegang and Forster, 1980; Polack, et al., 1971).
(iv) **Poor hygiene:**

Women suffering from fungal vaginitis may infect their eyes due to lack of personal hygiene (Puttanna, 1969). An investigation of fungal flora of female reproductive tract showed that 50% among the pregnant group and 29% among the non-pregnant group had no symptoms of vaginitis or cervicitis, yet showed positive cultures of fungi and the fungi were mostly the species of the genus *Candida*. Hence it is worthwhile to investigate the fungal flora of female reproductive tract in cases of hypopyon corneal ulcers. (Puttanna, 1969).

(v) **Herbal Juice:**

Puttanna has reported a case of corneal ulcer due to *Fusarium* after instillation of herbal juice used as native treatment. It is believed that the herbal juice or the grass leaf used to scratch the ulcer as a part of the native treatment caused Keratomycosis. (Puttanna, 1964).

(vi) **Eye makeups and contact lenses:**

428 eye cosmetic samples were cultured for bacteria and fungi. Fungal contamination was noted in 12% of the cases. Cultures from the outer eye of women with contaminated cosmetics yielded the same organism in a
significant number of cases. In one instance, the mascara of a woman with keratomycosis due to *Fusarium solani* yielded the same fungus. Fresh cosmetics were essentially free of microbial contamination. Representative fungal isolates were shown to assimilate cosmetic components such as paraffin oil, petroleum and isopropyl myristate. The study demonstrated fungi from environment may contaminate eye makeups and present potential hazard to ocular infections.

The source of these fungi is most probably skin yeasts and air-borne fungal spores. The common species of moulds isolated from eye cosmetics are ubiquitous in the environment, therefore, chances of contamination of cosmetics with these fungi is possible.

Extended use of contact lenses for cosmetic purposes also has a bearing in the causation of mycotic corneal ulcers. Contact lenses are used for cosmetic or therapeutic purposes such as aphakia. Liesegang et al., in their study (1980), have reported three cases with fungal ulcers who were wearing soft contact lenses at the onset of their disease; in two of the patients the lenses had been prescribed for therapeutic purposes. The fungal organisms isolated in these patients were *Candida albicans*, *Aspergillus flavus* and *Fusarium dimerum*. 41
2.2.4. Epidemiology:

Corneal ulceration caused by fungus has long been one of the main ophthalmic problems in countries such as India. In the last two decades, however, the incidence of keratomycosis has also increased considerably in western countries owing to the injudicious use of topical corticosteroids and antibiotics.

Incidence of fungal keratitis:

The aetiologic agents isolated from fungal corneal ulcers vary according to geographic location and climate.

The suppression of bacterial growth by antibiotics has an equally unfortunate effect on the proliferation of fungi (Roberts, 1957; Pannarale, 1958; Anderson, et al., 1959).

The rising incidence of keratomycosis has been well documented by Haggerty and Zimmeraman (1958) in their review of the records of the Register of Ophthalmic Pathology; there were 3 cases from 1933 to 1951 and 13 cases from 1952 to 1956.

Futtanna (1969) studied 301 cases of corneal ulcers, out of these 11.3 percent were positive for fungus. In this series 47 percent were Aspergillus, 8.8 percent Cephalosporium, 8.8 percent Penicillium, 11.8
percent Fusarium, 5.9 percent Rhizopus, 5.9 percent Lasio diplodia and 11.8 percent were Candida.

Dasgupta et al. (1973), studied 175 cases of ulcerative keratitis. Fungi were isolated from 25.7 percent cases only. Out of these, Aspergillus was isolated from 35.6 percent cases, Fusarium from 13.3 percent cases, Candida from 26.7 percent cases, Trichosporon from 4.4 percent Mucor and Penicillium were isolated from 2.2 percent of the cases and 2.2 percent cases remained unidentified.

Kulshrestha et al. (1973), studied 52 cases of corneal ulcers. 44.2 percent of these cases showed fungal growth. In this series, Aspergillus was isolated from 34.8 percent, Candida albicans from 13.1 percent, Penicillium from 8.7 percent, Trichophyton from 4.3 percent, Microsporum from 4.3 percent, Epidermophyton form 8.6 percent, Rhizopus from 13.1 percent, Mucor from 4.3 percent and yeast from 8.7 percent of the cases.

Grover et al. (1975) studied 17 cases of corneal ulcers, fungi were isolated from 82.4 percent of cases. Out of these, Aspergillus were isolated from 50 percent of cases, Penicillium from 14.5 percent, Candida, Fusarium, Curvularia, Trichosporon and Scopuloriopsis were each isolated from 7.1 percent cases.
Chaddah and Agarwal (1978) in their series of 20 cases of corneal ulcers isolated fungi from 90 percent cases. Out of these *Aspergillus* was isolated from 77.6 percent cases, *Penicillium* from 5.6 percent, *Hemispora* from 5.6 percent, *Epidermophyton* from 5.6 percent and *Fusarium* from 5.6 percent cases.

Dutta et al. (1981) studied 100 cases of corneal ulcers, 32 percent cases revealed fungal growths. They isolated *Aspergillus* from 68.7 percent cases, *Penicillium* from 18.8 percent cases, *Fusarium* from 9.4 percent cases and *Lasio diplodia* from 3.1 percent cases.

Sharma (1981) studied 100 cases of corneal ulcers, out of those fungi were isolated from 29 percent cases. Among those *Aspergillus* was isolated form 52.6 percent, *Mucor* form 21.0 percent cases, *Rhizopus* from 5.3 percent, *Penicillium* from 15.8 percent and one strain remained unidentified.

Prasad and Nema (1982) studied 60 cases of corneal ulcers. 20 percent cases showed fungal growth. Out of these, *Aspergillus* was isolated form 50.0 percent cases, *Penicillium* from 16.7 percent, *Curvularia* from 16.7 percent and *Candida* from 16.7 percent cases.

Sharma et al. (1985) studied 510 cases of corneal ulcers and fungal positivity was reported in 17.1
percent cases. They isolated **Aspergillus** from 42.5 percent, **Mucor** from 18.4 percent cases, **Penicillium** from 13.8 percent cases. **Rhizopus** and **Candida** each were isolated from 6.9 percent cases.

Haldar et al. (1992) studied 203 cases of corneal ulcers. Fungi were isolated in 31.0 percent cases. The strains included in this series were **Aspergillus** 50.8 percent, **Candida**, 17.5 percent, **Acremonium** 12.7 percent, **Fusarium** 3.2 percent, **Curvularia** 4.7 percent, **Penicillium**, 6.3 percent, **Aureobasidium** 1.6 percent; **Alternaria** and **Cladosporium** each were isolated from 1.6 percent cases.

Chander, et al. (1993), studied 632 cases of corneal ulcers and isolated fungi from 7.9 percent cases. They isolated **Aspergillus** from 40.0 percent; **Fusarium** from 14.0 percent; **Curvularia** from 10.0 percent; **Candida** from 10.0 percent; **Acremonium** from 8.0 percent; **Paecilomyces** from 60.0 percent, **Penicillium** from 4.0 percent; **Alternaria**, **Drechslera**, **Aureobasidium**, **Fonsecaea pedrosoi** each were isolated from 2.0 percent cases.

On a worldwide basis, the keratomycoses are most often caused by **Aspergillus** in about 50 percent, **Candida** in about 25 percent and in 25 percent cases by other fungi especially **Fusarium**. (Peyman et al. 1987).
Jones et al. (1969) isolated thirty eight cases of fungal keratitis from South Florida. 76.4 percent of the fungal isolates were Fusarium. Aspergillus and Candida constituted 7.9 percent and 5.3 percent respectively. Curvularia lunata, Macrophoma, sp. Penicillium sp. and Phialophora verrucosa each were 2.6 percent of the total fungi isolated.

Forster and Rebello (1975) isolated 76 cases of keratomycosis. Out of these Fusarium was 77.0 percent, Candida 11.5 percent, Curvularia, Alternaria, Drechslera each constituted 3.3 percent of the total fungal isolates. Lasiodiplodia was 1.6 percent.

Liesegang and Forster (1980) isolated fungi from 20 percent cases of ulcerative keratitis. They isolated Fusarium from 61.7 percent cases. Aspergillus from 4.5 percent. Cephalosporium and Penicillium, each from 3.1 percent cases. Paecilomyces and Allescheria boydii each from 1.5 percent cases. Candida from 10.5 percent cases. They isolated Curvularia from 5.3 percent cases. Lasiodiplodia and Alternaria each from 3.7 percent cases. Helminthosporium and Cladosporium each from 0.7 percent cases.

**Fungus keratitis in relation to sex:**

As males are more indulged in farming than the females, the male female ratio of keratomycosis is 2:1.
(Puttanna, 1967 and Koul and Pratap, 1975) and by other workers 3:2 (Dutta, et al., 1981), 3:1 (Chander, et al., 1993). Ratio in various reports has been found variable but males are more affected by keratomycosis, the cause being the style of life which is outdoor type predominantly in the males; with the same reason farmers and labourers are main sufferers of the disease, In the females it is more prevalent in the housewives coming from low socio-economic group having poor living conditions and bad personal hygiene.

**Keratomycosis in relation to age**:

Keratomycosis has been reported to be a disease of middle and old age, the most vulnerable age group as reported by various Indian authors from various parts of the country. From Bangalore 40-60 years. (Puttanna, 1969), from Lucknow 30-50 years (Koul, 1975), from Patiala, 40-60 years (Sharma, 1981), from Guwahati, 40-60 years, (Dutta, et al., 1981) from Darjeeling, 30-50 years (Haldar, et al., 1992) from Chandigarh, 40-60 years. (Chander, et al., 1993).

**Seasonal Variation**:

Seasonal variation in the incidence of keratomycosis has been reported in literature by Indian authors, from Bangalore maximum number of cases have been reported in harvesting i.e., June, July, August,
September and October. (Puttanna, 1969), in Amritsar, from August to October (Sandhu, et al., 1981). From Gauhati in harvesting season (Dutta, et al., 1981), in a study at Patiala the incidence of mycotic corneal ulcers was reported to be highest in the months of February to March and September to October (Sharma, 1981).

2.2.5. Antifungal Therapy:

The initial management of suspected microbial keratitis depends on history, clinical impression and results of corneal scrapings (Jones, 1980). The presence of hyphae, pseudohyphae or yeast cells in a smear prepared from a corneal ulcer dictates antifungal therapy (Jones, 1980). But it is desirable, rather mandatory and scientific to withhold treatment until the laboratory confirmation of the fungus and its sensitivity pattern is obtained (Jones, 1980).

Natamycin:

It is an antifungal antibiotic originally isolated from Streptomyces natalensis from a South African soil sample (Struyk, et al., 1958). It is a member of the polyene group of antibiotics. It is a complex molecule with four conjugated double bonds and it is the first polyene antibiotic to be known structurally. Now it is considered as the topical antifungal of choice in the
initial management of mycotic corneal ulcers. (Jones, 1981 and Struyk, et al., 1958). It is tetraene polyene antifungal agent (Jones, 1969; Jones, 1975). Natamycin suspension is viscous, adheres to the cornea in areas of epithelial defects and remains in the conjunctival fornix.

As a group, the polyenes are thought to act by binding to the sterol moiety present in the fungal cell membrane but not found in the mammalian cell. The polyene sterol complex alters the cell membrane permeability and causes cell death by disturbing intracellular electrolyte concentrations. (Zygmunt, et al., 1966).

Polyene + Sterol $\rightarrow$ Polyene - Sterol - complex

$$\text{Altered membrane permeability}$$

$$\text{Internal acidification \quad leakage of sugars, enzymes, K}^+$$

**Proposed method of action of polyene antibiotics:**

Natamycin is a broad spectrum antifungal agent that has been used effectively in the treatment of fungal keratitis caused by *Fusarium*, *Cephalosporium*, *Aspergillus* and *Candida* species (Ellison, et al., 1969;

**Amphotericin-B:**

Amphotericin-B was isolated in the year 1955 (Gold, et al., 1956) from a soil strain of Streptomyces originating in Venezuela. It has been used in the form of aqueous solutions with encouraging results (Jones, et al., 1972 and Wood, et al., 1976). It is a broad spectrum heptene polyene antifungal agent. It is a large molecule and poorly water soluble, the characteristics that limit its ocular tissue penetration. The medication may be gradually tapered after a favourable clinical response is noted. The drug has been successfully administered as 10% ointment and was found to be well-tolerated without ocular irritation (Chaddah, et al., 1978) or as 0.1 to 1% drops (Wood et al., 1976).

Sodium deoxycholate has been used to keep amphotericin-B in colloidal suspension (Jones, et al., 1972), but both of the compounds have been found to be tissue toxic and highly irritating (Forster, et al., 1981). Between the concentrations 0.5 to 1.5 mg/ml, the antifungal drug (amphotericin-B) is effective to appreciable degree and sodium deoxycholate is non-toxic (Wood, et al., 1985). Conjunctival injection is associated with delayed corneal epithelial wound healing.
and iridocyclitis. Subconjunctival injection is extremely painful and may cause conjunctival yellowing and nodule formation (Bell, et al., 1973). Intravenous administration of Amphotericin-B may be complicated by nephrotoxicity, hypoglycemia and thrombocytopaenia, and is rarely indicated for treatment of keratomycosis.

**Nystatin:**

It is a tetracycline polyene antifungal agent that was used in the past for the treatment of superficial keratomycosis caused by *Candida albicans*. It has poor penetrating power into the cornea and has limited activity against other fungi (Jones, 1975 and Polack, et al., 1971). The mechanism of action is similar to Natamycin and Amphotericin-B.

**Imidazole Compounds:**

This group of antifungal agents include Clotrimazole, Miconazole and Ketoconazole, and these possess some gram-positive antibacterial activity as well. Their mechanism of action is two-fold: interruption of fungus membrane sterol synthesis, and by binding fatty acids on the cell wall of susceptible organisms, creating an intracellular imbalance in electrolytes similar to that of polyene antibiotics. Experimentally, these agents have not been as effective.
as Amphotericin-B or Natamycin in treating *Candida albicans* keratomycosis. The agents have been advocated for the treatment of filamentous keratomycosis associated with deep corneal infiltration impending corneal perforation or fungal scleral abscess (Jones, 1981).

These agents have been successfully used in the treatment of corneal ulcers caused by *Alternaria*, *Rhodotorula*, *Penicillium*, *Aspergillus*, *Fusarium* and *Candida* species (Forster, 1981 and Jones, 1975). The drug demonstrates good ocular penetration following either topical or subconjunctival administration and is associated with no significant ocular toxicity (Forster, et al., 1979; Foster, 1981 and Foster, et al. 1981).

Ketoconazole is a new water-soluble Imidazole compound that is structurally related to Miconazole (Dixon, et al., 1978 and Thienpont, et al., 1979). It has a broad spectrum antifungal activity, and experimental studies demonstrate good concentration in cornea after oral administration (Dixon, et al., 1978). 300 mg/day orally has given encouraging results. (Foster, 1981 and Ishibashi, 1983).

**Flourocytosine**:  
It is a fluorinated pyrimidine. Its mechanism of
action is probably exerted through its incorporation into fungal transfer ribonucleic acid, producing faulty protein synthesis within susceptible fungal organisms. (Wood, et al., 1985). The drug may be used as adjunctive therapy in the treatment of keratomycosis caused by Candida species (Jones, 1975, Jones, 1981 and Richards, et al., 1969). The drug is well absorbed from the gastro-intestinal tract, and therapeutic levels have been found in the aqueous humour following oral administration of 200 mg/kg /day (Steer, et al., 1972) 5-Fluorocytosine (5 FC) is excreted unchanged in the urine and has few serious side effects (Steer, et al., 1972). Some workers have observed that susceptible fungi convert 5-FC to 5-fluorouracil, which interferes with thymidine synthesis (Steer, et al., 1972). The addition of another antifungal agent may prevent the emergence of fungal resistance to 5-FC during prolonged therapy (Richards, et al. 1969 and Steer, et al., 1972).

Hamycin:

Some workers have tried Hamycin in glycerine suspension for treatment of experimental keratomycosis and have reached the conclusion that the drug is effective in Candida infections and not with Aspergillus fumigatus infections. (Ahuja, et al., 1967).