GENERAL SUMMARY
The fungi, which inhabit keratinic substances are of diversified characteristics, whether judged on the basis of morphology, physiological properties, nutritional characteristics or synthetic abilities. Most of these fungi are potentially pathogenic to man and animals. In the present investigation an effort has been made to determine the prevalence of keratinophilic fungi and related dermatophytes in the samples collected from the soils of various localities. Soil samples collected from forest litter, garden, crop field, cattle farm, poultry farm and cattle shed sweepings were baited for the isolation of keratinophilic fungi. Keratinic substances like animal horns, hooves, human hair, and nails and peacock feathers were used as baits for the isolation purposes.

A total of 111 isolates belonging to different groups were collected in this way. These isolates represent 43 species of 17 different genera in addition to 6 sterile forms. During the course of these studies, 18 morphologically different strains belonging to 3 important genera were recorded. These were, 9 of *Chrysosporium tropicum*, 6 of *Microsporum gypseum* and 3 of *Malbranchea pulchella*. Maximum number of fungi were recorded on hair, hooves and feathers. In all 29, 28 and 27 fungi were recorded on buried hair, hooves and feathers respectively. Colonization on buried horns was found somewhat
poor showing the presence of only 21 fungal species. Six fungi were also recorded from decomposing human nails. The following are the brief details of the collections (taxonomical):

(A) Genera reported for the first time from India

1. Auxarthon
2. Cladorrhinum

(B) Species reported for the first time from India

1. Auxarthon thaxteri
2. Chrysosporium mardarium
3. Curvularia lunata var. aerea (Keratinophilic)
4. Malbranchea albolutea
5. Malbranchea aurantiaca
6. Verticillium tenuipes
7. Cladorrhinum sp. (not described)

(C) Species to be reported as new ones.

1. Chrysosporium sp. (IMI 196824)
2. Chrysosporium sp. (IMI 196823) (IMI 196825)
3. Myrothecium sp. (IMI 196830)
4. Cladorrhinum sp. (Deposited at 'Centraalbureau Voor Schimmelcultures' Netherlands)

The strains of C. tropicum, M. fulvum and M. pulchella were tested for their identity. Detailed nutritional and
physiological studies of these molds have been carried out to find out the physiological differences and their response to various environmental conditions.

Four keratinophilic fungi, i.e., *Chrysosporium tropicum*, *Keratinophyton terreum*, *Trichophyton rubrum* and *Verticillium tenuipes* were selected for detailed physiological studies. The growth and sporulation of these fungi were tested in 10 different natural and synthetic media. Among natural media peptone dextrose broth and among synthetic media glucose asparagine were found to be the most suitable nutrient media for the optimum growth of these fungi. Maximum growth of *C. tropicum*, *K. terreum* and *T. rubrum* was recorded at 30°C. Optimum growth and sporulation in *V. tenuipes* was found at 35°C. When these fungi were grown in the Sabouraud's dextrose broth medium a gradual increase in the growth duration resulted in the increase of mycelial yield which was followed by a phase of no net growth or of autolysis. Maximum dry mycelial weight of all but *T. rubrum* was recorded after 7 days of incubation when grown in Sabouraud's dextrose broth medium. The optimum growth of *T. rubrum* was recorded after 13 days of incubation. The sensitivity of fungal mycelium and sporulation for some trace elements, viz. Cu, Zn, Mn, and Fe at their different concentrations have been evaluated. In most of the cases lower concentrations (0.1 to 5.0 ppm) of these microelements were found acceleratory for the growth of these
keratinophilic fungi but when the mycelial yield was compared to that of controls these elements were not found indispensable for them.

The nitrogen requirements of *V. tenuipes*, *K. terreum*, *C. tropicum* and *Malbranchea pulchella* were determined. Peptone supported the best growth of all the fungi it is concluded that peptone provides all the nutritional requirements of these fungi under test. The possibility of contamination of this natural product with some vitamins and trace elements has also been commented. Among the amino acids, asparagine supported the growth of most of the test fungi. *V. tenuipes* and *M. pulchella* could grow well in the medium containing inorganic nitrogen sources. These four fungi have also been classified according to their nitrogen requirements.

An emphasis was also given to the different phases of fungal growth in relation to the change in pH of the culture medium, production of indolic compounds and utilization of glucose from the medium. For this purpose glucose asparagine medium was used as the culture medium. A total of 86.11, 81.88, 52.27 and 47.69 per cent loss in the mycelial yield of *T. rubrum*, *K. terreum*, *V. tenuipes* and *C. tropicum* respectively was recorded in 30 days of autolysis. The higher degree of autolysis as observed in these fungi may be due to the cytoplasmic degradation as well as the autolysis of the cell wall. Synthesis of indolic compounds was recorded only in the
early days of autolysis, i.e., up to 10 days. In no case
20 days autolysed mycelium was found to synthesize indolic
compounds in the stationary cultures. The initial pH (6.5)
of the medium drifted towards alkalinity during growth of
these fungi. Utilization of sugars present in the basal
medium was recorded during the optimum phase or in the early
days of autolysis in the cultures of these fungi. The change
in free amino acid pool of C. tropicum, M. pulchella and V.
tenuipes during different incubation periods have been
determined and discussed.

Microbial production of indolic compounds has been a
subject of active interest. In the present study extracellular
production of these substances in the cultures of 6 typical
species of Nannizzia and 7 atypical forms of Microsporum
gypseum group have been evaluated. It is revealed that all
the test fungi were able to synthesize indoles in the cultures.
The plus strains (+) of N. gypseum, N. fulva and N. incurvata
were found to be less adoptive for the production of indolic
compounds in comparison to that of minus strains (-) of these
species. The higher production was found in cultures of
M. fulvum strain III. Another point worthy of note is that
the acid fractions usually contained more indoles than the
basic fractions.

The sensitivity of sulphadrugs, viz. sulphadiazine,
sulphadimethoxine, sulfamethoxazole and pentid sulfas was assayed against some keratinophilic fungi. Sulphadiazine and sulfamethoxazole were found most toxic for the mycelial growth of C. tropicum, K. terreum and M. pulchella only at their higher concentrations (500-1000 ppm). Pentid sulfas showed its toxicity against these fungi but in no case its fungicidal action was recorded in present study. The lower concentrations of sulfamethoxazole and sulphadiazine accelerated the growth in C. tropicum, V. tenuipes and M. pulchella.

A parallel experiment was also conducted to determine the effect of four antibiotics, i.e., aureofungin, griseofulvin, nystatin and lincocin on the growth of keratinophilic fungi. Except Lincocin most of the antibiotics were found quite effective against these fungi under test. Aureofungin showed a significant effect against the growth of V. tenuipes and C. tropicum. Nystatin and griseofulvin were found effective only when used in higher concentrations. Lincocin (Lincomycin hydrochloride) an antibacterial antibiotic showed inhibitory action only against V. tenuipes, on the other hand the lower doses of this antibiotic stimulated the growth of K. terreum, M. pulchella and C. tropicum.

The vapours emanating from some volatile substances were tested against the growth and sporulation of some selected keratinophilic fungi. For this purpose eight volatile substances, i.e., ethyl alcohol, n-butyl alcohol, isopropyl
alcohol, acetone, formic acid, pyridine, ethyl acetate and benzene were selected. Acetone was found almost toxic for the growth and sporulation of all the fungi. It showed maximum inhibition (92.3%) in the growth of V. tenuipes. Except acetone and pyridine all the volatiles accelerated the growth of K. terreum. Maximum growth stimulation, i.e., 228.7, 224.0 and 200.0% was noted when this organism was cultured in the atmosphere saturated with the vapours of benzene, ethyl acetate and formic acid respectively. N-butyl alcohol supported the best growth of C. tropicum. Ethyl alcohol, isopropyl alcohol, pyridine and benzene showed growth retarding property causing 36.9, 33.8, 33.8 and 27.6% growth inhibition respectively against M. pulchella.

The antifungal spectrum of some essential oils of medicinal plants and their components have also been studied. The acetates of eugenol and geraniol were found to be the most inhibitory constituents against most of the keratinophilic fungi. The essential oils of Anomum subulatum, Balsamodendron mukul, and Cinnamomum macrocarpum showed antifungal activity and inhibited the growth of most of the keratinophilic fungi and related dermatophytes. It is suggested that these oils can be employed as surface applicants as preventive measures for dermal diseases caused by various keratinophilic fungi.

In the present study keratin degrading capability of 34
isolates have been determined and discussed in term of their nature and relation to two different keratinic substances, i.e., pig hair and peacock feathers. The maximum keratinolytic activity was recorded in case of *V. tenuipes*, *Trichophyton mentagrophytes*, *I. equinum* and a total of 77, 72 and 70 per cent loss of feather keratin and 57, 56 and 50.4 per cent loss in the hair keratin was recorded after 20 days of incubation. All the typical and atypical isolates of *Chrysosporium tropicum*, *Microsporum fulvum* and *Malbranchea pulchella* were found keratinolytic in nature. The keratinolytic activity of *Malbranchea aurantiaca* was confirmed. It showed 67.6 and 64.2% loss of keratin when peacock feathers and pig hair were used as substrates. *Auxarthron thaxteri* degraded 42.4% and 32.0% feather and hair respectively in 20 days of its growth. The isolates of *Chrysosporium merdarium* and *Cladorrhinum* sp. did not hydrolyse keratin substances tested here but their growth was recorded over these keratinic substances, which confirms their keratinophilic nature.

Four keratinophilic fungi, i.e., *C. tropicum*, *K. terreum*, *I. rubrum* and *V. tenuipes* were taken for their detailed enzymological studies. The release of protein in the cultures during growth of these organisms on three different keratinic substances, i.e., pig hair, peacock feathers and buffalo horns was evaluated in terms of the activity and nature of the enzymes keratinases produced by these molds. Maximum keratinase
activity was recorded in the cultures of *V. tenuipes* when feathers were taken as keratinic substrate. A total of 309 mg protein release was recorded in the culture medium of this fungus after 30 days of incubation. After the same incubation period 303, 187 and 215 mg proteins per sample was recorded for *K. terreum, T. rubrum* and *C. tropicum* respectively. The enzymes produced by *K. terreum* failed to hydrolyse horn keratin. The release of protein was recorded more from the hair when *T. rubrum* was used as test organism. It is concluded that among the substrates used here feathers are the first for the action of enzymes produced by most of the keratinophilic fungi. Amylolytic capabilities of these fungi have also been tested. All the organisms have been found to be capable of producing extracellular amylase.