Histopathologic Aspects of the Pathogenesis of Some Opportunistic Fungal Infections, as Exemplified in the Pathology of Aspergillosis and the Phycomycetoses

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The concept of microbial opportunism as an important occasional factor in the etiology of infectious diseases has been generally recognized and its validity has been accepted. Essentially, it amounts to the premise that the pathogenic capacity of certain microorganisms that ordinarily do not readily or frequently cause progressive disease may be so enhanced by circumstantial modification of a patient's resistance to their attack that they are able to set up an infection of overwhelming severity.

Much has been learned during the past decade and a half about the types of infection that, in particular circumstances, may be regarded as opportunistic, and about the conditions that predispose to their occurrence. Opportunistic infections by bacteria, viruses, and protozoa are known, in addition to the opportunistic fungal infections which are the subject of this symposium. The factors that predispose to opportunistic infections, whatever the nature of the microorganisms concerned, are similar—they are certain types of diseases and certain types of therapeutic procedures which, alone or in combination, alter the capacity of the patient's natural defenses to prevent or overcome incipient infection by these organisms. Many of the organisms, under ordinary circumstances, are of restricted or negligible pathogenicity, and a few of them would even be regarded as nonpathogenic.

It is in the nature of these dangerous, and very frequently mortal, opportunistic infections that they attract much attention, for their development is often attributable, at least in part, to side effects of modern therapeutic procedures which should, instead, have prolonged or saved the life of the patient. Yet, in spite of the wide interest that opportunistic infections have occasioned, understanding of their pathogenesis remains very largely in the realms of hypothesis and of the shrewd guess. Etiologically, it is at least clear that certain types of opportunistic infection are particularly associated with certain types of predisposing disease or certain types of therapy.

The opportunistic fungal infections fall into two large groups—those in which the causative organism occurs in the infected tissues in the form of mycelium, and those in which the organism occurs in the tissues in a yeastlike form.

THE "MYCELIATE OPPORTUNISTS"

The most frequently occurring of the opportunistic infections that are caused by myceliate fungi are aspergillosis,[6] candidiasis,[38] the phycomycetoses,[10] and nocardiosis.[19]. However, even those infections that are extreme rarities, such as septicemic trichophytosis, have an importance to the individual that is altogether out of proportion to their absolute incidence, for the prognosis of all opportunistic deep-seated mycotic infections is very grave, whatever the identity of the fungus responsible. All these fungi are sufficiently prevalent among the population, or free in nature, to be a potential hazard to any patient who is suffering from one of the diseases or is under treatment with one of the agents that are known to predispose to opportunistic infections. Thus, trichophytosis has a worldwide prevalence as an infection of the epidermis. Occasionally, under circumstances that are very incompletely understood, the trichophytic infection extends into the dermis, either producing the classic form of Majocchi's trichophytic granuloma or appearing as the nodular perifolliculitis described by

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Wilson, Plunkett, and Gregersen.[37] It is but one step farther for the infection to extend to the regional lymph nodes,[4] and thence to the blood stream; in fact, however, there are only two cases of septicemic trichophytosis on record[22, 29]—one of these was undoubtedly an instance of opportunistic infection.[29] What determines the extreme rarity of hematogenous dissemination of Trichophyton infection is as problematical as the explanation of the fact that it should occur at all.

Aspergillosis, candidiasis, the phycomycetoses, and nocardiosis may occur opportunistically in a variety of clinicopathologic forms. *Septicemic infections* by the causative organisms of any of these mycoses may complicate treatment with cytotoxic drugs, x-rays, or corticosteroids. Antibiotics, too, may predispose to this complication, but it seems likely that the importance of these drugs as predisposing factors was overstated during the period when the probability of drug-induced predisposition to infections was first attracting attention.[1] The septicemic forms of these four mycoses may also be seen as complications of disease: most typically and frequently, it is the diseases that are characterized by granulocytopenia—agranulocytosis, aplastic anemia, leukoerythroblastic anemia, and the late phases of any variety of leukemia—that predispose to these septicemic mycoses; however, the systemic diseases of the lymphoreticular system, which lower resistance to infection by depressing both the humoral and the cellular defense responses, may also do so, although in such cases it is particularly difficult to apportion the etiologic responsibility equitably between the disease and its treatment.

Orbitocerebral phycomycetosis is, typically, a complication of a severe metabolic disorder; poorly controlled diabetes mellitus is the classic predisposing disease,[7] but oliguric renal failure[13, 33] and hyperemetic acidosis may also be responsible. The same syndrome of phycomycetic infection may be seen, but very exceptionally, in the absence of any recognized predisposing disease. Interestingly, an identical sequence of mycotic sinusitis and orbital cellulitis is a rare manifestation of aspergillosis;[36] when an Aspergillus is the cause, there seems to be no evidence of any general disease as a predisposing factor (Figs. 1 and 2).

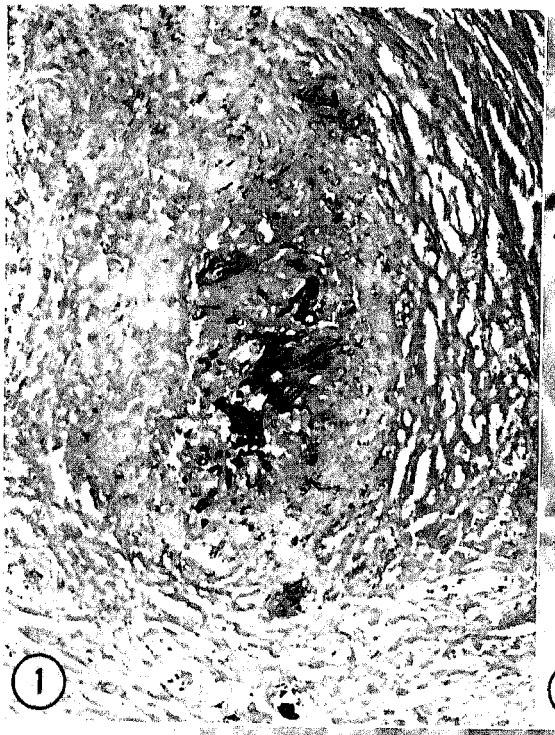
The *necrotizing mycotic pneumonia* that may be associated with infection by an Aspergillus (usually *Aspergillus fumigatus*) or by one of the phycomycetes (usually species of Mucor or Rhizopus) may be the result of colonization of pulmonary infarcts by the fungi. Alternatively, the fungi may themselves be responsible for the necrosis of the lung tissue, either through a direct necrotizing action of toxic substances that they evolve or because of mycotic invasion of blood vessels, with resulting thrombosis and consequent ischemia and infarction.

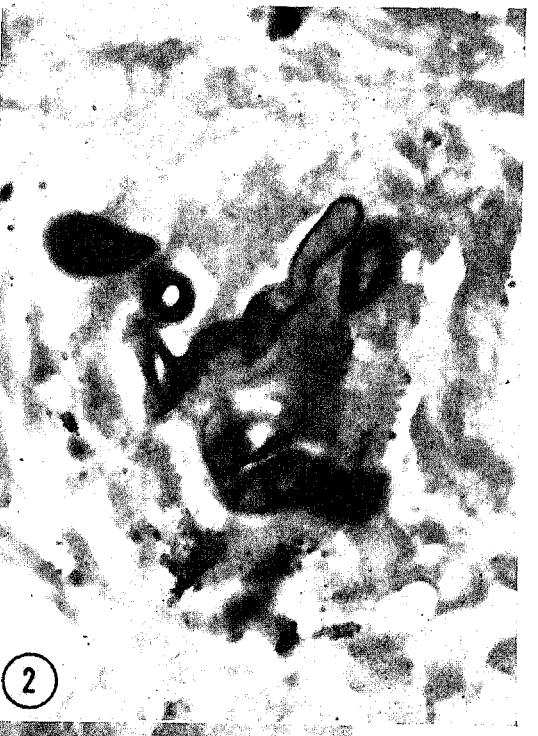
A much rarer example of an opportunistic infection of necrotic tissue is cerebral chromoblastomycosis ("cladosporiosis"),[32] which,

FIG. 1. Chronic orbital aspergillosis. The picture shows a collection of hyphae in the organized thrombus filling the lumen of a blood vessel in a biopsy specimen. The patient was an otherwise healthy Chinese man who was on a visit to England from Hong Kong when he began to develop unilateral proptosis. When he first came to hospital, 3 months later, the ocular protrusion was clearly seen to be due to a brawny induration of the orbital tissues. X-ray examination showed thickening of the mucosa of the antrum on the same side. Biopsy of the orbital mass yielded a pure culture of *Aspergillus flavus*; a heavy growth of this mold was also obtained regularly from specimens collected during lavage of the antrum. Considerable improvement in the condition took place, without any treatment, during an observation period of several weeks; the patient then left England, and nothing is known about the subsequent course of the infection (methenamine silver, × 150).

FIG. 2. Same orbital biopsy specimen as in Figure 1, showing cluster of hyphae cut in various planes (methenamine silver, × 1300).

FIG. 3. Hyphae in the granulation tissue at the margin of an acute ulcer in the fundus of the stomach. The condition presented with hematemesis, necessitating emergency gastrectomy. *Aspergillus fumigatus* was isolated in pure culture from swabs taken from the ulcerated area immediately after the operation. The patient was an undernourished but otherwise healthy Negro girl, aged 9 years, who had just arrived in Britain from the West Indies. There has been no further evidence of illness during the 3 years since the operation (Gridley's stain, × 1300).





FIGS. 1 TO 3

in a significant proportion of cases, is a complication either of cerebral infarction due to thrombosis or embolism, or of a cerebral hemorrhage. In one of my cases the Cladosporium infection developed in a necrotic malignant glioma;[33] as is usual in this mycosis, the portal of entry of the infection was not known.

THE "YEAST-FORM OPPORTUNISTS"

The infections that are included in the second group of opportunistic mycoses are predisposed to particularly by the systemic diseases of the lymphoreticular system (Hodgkin's disease, lymphosarcomatosis, lymphatic leukemia, and sarcoidosis). The most important member of the group, because the most frequent, is cryptococcosis. A considerable proportion of the cases of cryptococcosis that occur in association with lymphoreticular diseases is of the otherwise comparatively rare, generalized, hematogenous form.[39] The lymphoreticular diseases also predispose to widespread, hematogenous dissemination of the infection in cases of histoplasmosis and coccidioidomycosis; the grave prognosis of this disseminated form of these mycoses contrasts strikingly with their benign nature in the overwhelming majority of cases occurring in the absence of any associated, resistance-lowering disease. In comparable fashion, but with much less frequency, the systemic diseases of the lymphoreticular system may encourage generalized dissemination of the infection in cases of North American blastomycosis and South American blastomycosis (paracoccidioidomycosis).[30]

THEORETICAL CONSIDERATIONS RELATING TO THE PATHOGENESIS OF OPPORTUNISTIC FUNGAL INFECTIONS

It is clear that infections by the "myceliate opportunists" and by the "yeast-form opportunists" are predisposed to by different circumstances. These circumstances are by no means fully defined, nor are the pathogenetic mechanisms yet fully understood. A wide variety of possible mechanisms could, theoretically, contribute to the alteration in the patient's defenses that enables an opportunistic infection to develop. The field for speculation is almost unlimited, and there is little objective evidence with which to temper the views that are put forth.

The fundamental condition underlying the occurrence of an opportunistic infection is the failure of resistance to the attack by the organism. The failure might be mediated in various ways, alone or in combination: (1) by interference with the defensive activity of the lymphoreticular system by the diseases that are peculiar to this system, or by destruction or inhibition of its cells by drugs or irradiation; (2) by interference with the defensive activity of the neutrophilic leukocytes, due to failure of leukopoiesis, of whatever cause: (3) by alterations in the body's microflora, occasioned by administration of courses of antibacterial chemotherapy or of antibiotics, and resulting in conditions that favor the growth of fungi, such as *Candida albicans*, with consequently increased opportunities for these organisms to establish themselves as tissue invaders should they gain entry to the tissues; (4) by the inhibitory effect of therapeutically administered corticosteroids on humoral or cellular defensive mechanisms; (5) by the comparable effects of certain endocrine diseases, particularly those that present the clinical picture of Cushing's syndrome;[27] or (6) by changes in the biochemical environment of the tissues, induced by disorders of metabolism, such as diabetes mellitus and uremia. Indeed, the list of hypotheses can be enlarged far beyond the scope of those outlined here.

In contrast to this abundance of theoretical considerations, remarkably few factual observations have been published that throw light on the means by which the opportunistic organisms gain access to the tissues. It is with the hope of drawing attention to some of the histopathologic aspects of the pathogenesis of the opportunistic deep-seated fungal infections that this paper is presented. The two forms of mycoses that I have been asked to speak about particularly, aspergillosis and the phycomycetoses, illustrate peculiarly well some of the possible pathogenetic mechanisms. Both these mycoses are caused by fungi of which a number of distinct pathogenic species has

been recognized. Pathogenic aspergilli, especially *A. fumigatus*, have commonly been found in the sputum of persons who have had no symptoms indicative of any mycotic disease;[23, 26] pathogenic Phycomycetes have also been found in such circumstances, although with much less frequency.[26] Both types of fungi may also be isolated from a small proportion of specimens obtained by lavage of the maxillary antra in cases of banal, chronic, nonmycotic sinusitis. The fact that these organisms may occur in the respiratory tract as saprophytes indicates the probable route of infection in most cases of aspergillosis and phycomycetosis. Occasionally, either of these mycoses may develop in the mucosa of the alimentary tract (Fig. 3)[28, 35] or in the skin at the site of a local injury.[33] In exceptional circumstances, Aspergillus infection, like candidiasis, has been observed as a complication of open heart surgery, either through direct infection of the operation site in the heart or great vessels, or because of infection of the blood stream as a result of contamination of the apparatus used to maintain the extracorporeal circulation.[8]

Aspects of the pathology of aspergillosis and of the phycomycetoses that are specially pertinent to the problems of opportunistic infections are considered in the following sections of this paper.

ASPERGILLOSIS

By far the most frequent and most important site of Aspergillus infection is the lungs, including, of course, the bronchial tree. It is not necessary to deal here with aspergillosis of other parts of the body, except insofar as its occurrence is a manifestation of opportunistic infection.

Forms of Bronchopulmonary Aspergillosis

There are three main forms of bronchopulmonary aspergillosis: (1) infection of the bronchial tree, with or without the development of clinical and pathologic manifestations attributable to allergic sensitization to the fungus;[9] (2) intracavitary aspergilloma;[14] and (3) Aspergillus infection associated with pulmonary necrosis, including infarction.[6, 9]

It is noteworthy that pulmonary aspergillosis, in common with aspergillosis of other tissues, is very rarely, if ever, a primary disease.

Pulmonary Aspergilloma

The intracavitary aspergilloma is invariably a complication of a preexisting lesion[34]—a cavity due to pulmonary tuberculosis or sarcoidosis or to bronchiectasis, or a chronic lung abscess, an emphysematous bulla, or any other chronic cavitary lesion. Even cavities that are the result of infection of the lungs by another fungus, as in certain cases of chronic histoplasmosis, may become the site of an Aspergillus ball.[24] In a sense, therefore, an aspergilloma is a form of opportunistic infection; but it is a very slowly developing, chronic condition, which remains confined to the site of the antecedent pulmonary lesion and does not immediately or necessarily threaten the patient's life. It is an opportunistic infection that is germane to this symposium only insofar as it might, on occasion, establish the portal of entry of the fungus into the body should circumstances develop that predispose to the acute, mortal, septicemic form of aspergillosis.

Relation between Aspergillus Infection and Necrosis of Lung Tissue—Necrotizing Mycotic Pneumonia

An Aspergillus may colonize dead lung tissue under two circumstances: following infarction, and when toxic products of the fungus itself have caused a "diffusion necrosis."

Infarction. A pulmonary infarct that is invaded by an Aspergillus may have resulted from such ordinary causes as embolism or thrombosis. Alternatively, in the condition that is sometimes referred to as primary mycotic infarction, the organism itself, by invading the tissues and gaining access to the blood vessels, is believed to occasion the vasothrombosis that is the cause of the infarction.

Aspergillus infection of an infarct that has developed independently of the infection is

essentially an example of saprophytosis—the growth of the organism on dead organic material—and, in the majority of cases, it is an incidental postmortem finding; there is no evidence of a true parasitic invasion of the lung tissue. Only when saprophytic colonization of an infarct occurs under circumstances in which the patient's resistance is lowered by the drugs or diseases mentioned above (see discussion under "The 'Myceliate Opportunists' ") is the development of a true Aspergillus infection likely to supervene; in such cases a fulminating septicemia would be the usual outcome. It may be difficult, or impossible, to be sure that this is the sequence of events, for infected pulmonary infarcts could, in fact, develop as a complication of an Aspergillus septicemia: however, when there is a recognizable cause of pulmonary infarction other than the Aspergillus itself, it is often obvious that the likelier sequence has been infarction followed by infection, and not the reverse.

Pulmonary Necrosis Possibly Caused by Toxic Products of Aspergilli (So-Called "Diffusion Necrosis"). In many cases in which an Aspergillus is found in necrotizing pulmonary lesions there is no absolute evidence that the lesions are ischemic in origin. Gowing and Hamlin[6] have indicated that the fungus is probably able to cause pulmonary necrosis directly. They noted that it could reach the lung substance either by way of the air passages to the alveoli, thence spreading into the surrounding parenchyma, or by direct invasion through the walls of the bronchi. There is some evidence that aspergilli, including *A. fumigatus*, are a source of soluble products that have a pronounced necrotizing action.[21] It has been suggested that such substances may diffuse from a colonized site, killing the tissues and thereby furthering the invasion by the fungus.[6] It is possible, even, that the fungus, growing within the lumen of the bronchial tree, might, by means of such noxious products, destroy parts of the mucosa, thus enabling the mycelium to grow into the tissues, a spreading zone of necrosis advancing through the bronchial wall and the surrounding lung in an ever widening field as the fungus penetrates farther into the necrotic mass. It is debatable whether these products are evolved by the growing Aspergillus or are released in the process of disintegration of those parts of the fungal colony that have died. Again, it is uncertain whether there are particular strains of the fungus that are particularly likely to produce these substances; some strains are undoubtedly more toxigenic *in vitro* than others, but the pathogenic importance of such differences is questionable. There is little doubt, however, that deficiencies in host resistance, due to disease or to inhibitory effects of therapeutic procedures, must be an important factor in preparing the tissues for the initial damage by the fungus or its products.

If the hypothesis that the fungus itself may cause necrosis of lung tissue is correct (and the evidence in support of this is considerable), the frequency of necrotizing pulmonary foci as a manifestation of opportunistic infection by aspergilli is more readily understandable. However, the largest lesions, which may be several centimeters in their longest dimensions, seem to be true infarcts, secondary to embolism or to nonmycotic vasothrombosis; their general configuration and situation are consistent with this interpretation, and it is a significant observation that the thrombus within the main artery to the affected part may be altogether free from infection by the fungus, or, when invaded, may be no more heavily invaded than the smaller vessels that lie within the colonized parts of the infarct. In contrast to this, pulmonary infarcts associated with phycomycetosis are much oftener the result of a vascular occlusion that is due to thrombosis actually caused by invasion of the pulmonary vessels by the fungus; in these cases, mycelium is very abundant throughout the thrombus in the main vessel of the infarcted part.

Invasion of the Tissues by Aspergilli

It is generally said that under ordinary conditions there is no mycotic invasion of the tissues in cases of bronchial or intracavitary aspergillosis. Thus, in the allergic form of the disease the organism grows in the plugs of thick, mucous secretion within

the lumen of the bronchi; similarly, in the aspergillomatous form, the fungal ball usually lies completely free within the cavity. However, in some of the cases of nonallergic bronchial aspergillosis and of aspergilloma that I have examined, it has been possible to demonstrate sparse but definite penetration of the organism into the tissues, even when the infection has not been accompanied by any of the circumstances that predispose to opportunistic extension of the mycosis. It is possible that such observations indicate the portal by which the Aspergillus enters the tissues in that majority of cases of opportunistic septicemic aspergillosis in which there has been no other apparent route of invasion, such as a saprophytically colonized infarct. In noting these findings, I would stress that their importance and their frequency are quite uncertain.

It is generally considered that the fungus in cases of bronchial aspergillosis is virtually a saprophyte, growing within the lumen of the bronchial tree in the mixture of bronchial secretion, low grade inflammatory exudate, and desquamated epithelial cells that is characteristic of chronic catarrhal bronchitis. Conidiophores are often developed by the fungus when growing in the well aerated environment of the larger airways. Histologically, unless the sections are studied with particularly meticulous attention, the impression is often obtained from an examination of hematoxylin-eosin or periodic acid-Schiff preparations that the fungus in these cases is entirely without connection to the bronchial wall: the mycelium seems to lie wholly free in the exudate covering the surface of the bronchial lining. In contrast, silver preparations, and specially those designed to demonstrate reticulin fibers, show very clearly that the fungus sometimes has a distinct attachment to the basement membrane of the chronically inflamed mucosa. The basement membrane of a healthy bronchus appears as a very fine, intensely argyrophilic structure in reticulin preparations; in cases of chronic bronchitis it may become greatly thickened, even as much as 20-fold, and it then appears as a homogeneous, more or less uniformly broad, hyaline membrane, which has little affinity for silver and takes a golden brown color in-

sections prepared by one of the Foot-type methods. In cases of bronchial aspergillosis, sections show that some of the hyphae end in single, vesicle-like expansions, 5 to 10 μ in diameter, which may be situated in contact with the thickened basement membrane, or even wholly within the substance of the latter (Fig. 4). The number of hyphae that end in these bulbous, intramembranous structures may be large or small in relation to the number of hyphae in the lumen of the bronchus at the same level. The junction between a hypha and its vesicle must be fragile, for very frequently the two are separated, and a considerable search may be necessary before continuity between hyphae and intramembranous vesicles can be demonstrated (Fig. 5).

Exceptionally, it is found that a mycelial thread has penetrated into the lamina propria of the mucosa; these rare, penetrating hyphae seem sometimes to originate in one of the intramembranous vesicles (Fig. 6), and sometimes, in contrast, to pass directly through the membrane from the bronchial lumen (Fig. 7).

Such observations may indicate a potential pathway of infection in cases of opportunistic aspergillosis. It should be stressed that they were made in cases in which there was neither an established opportunistic infection nor any disease or therapy recognized as predisposing to the development of opportunistic infections. The tissues concerned did not show necrosis of the bronchial wall, which, as Gowing and Hamlin[6] noted, may accompany opportunistic Aspergillus infection.

In assessing the significance of the superficial penetration of Aspergillus hyphae into the tissues, consideration has to be given to the possibility that this occurrence is merely a manifestation of saprophytic extension of the mold, following the patient's death or following surgical removal of the affected part of the lung. *A. fumigatus*, like other species of Aspergillus, can grow with remarkable speed on animal tissues that have just died, and it is quite possible that its mycelium could penetrate into the bronchial wall if there were sufficient delay before inhibition of fungal growth by refrigeration or by exposure to tissue fixa-

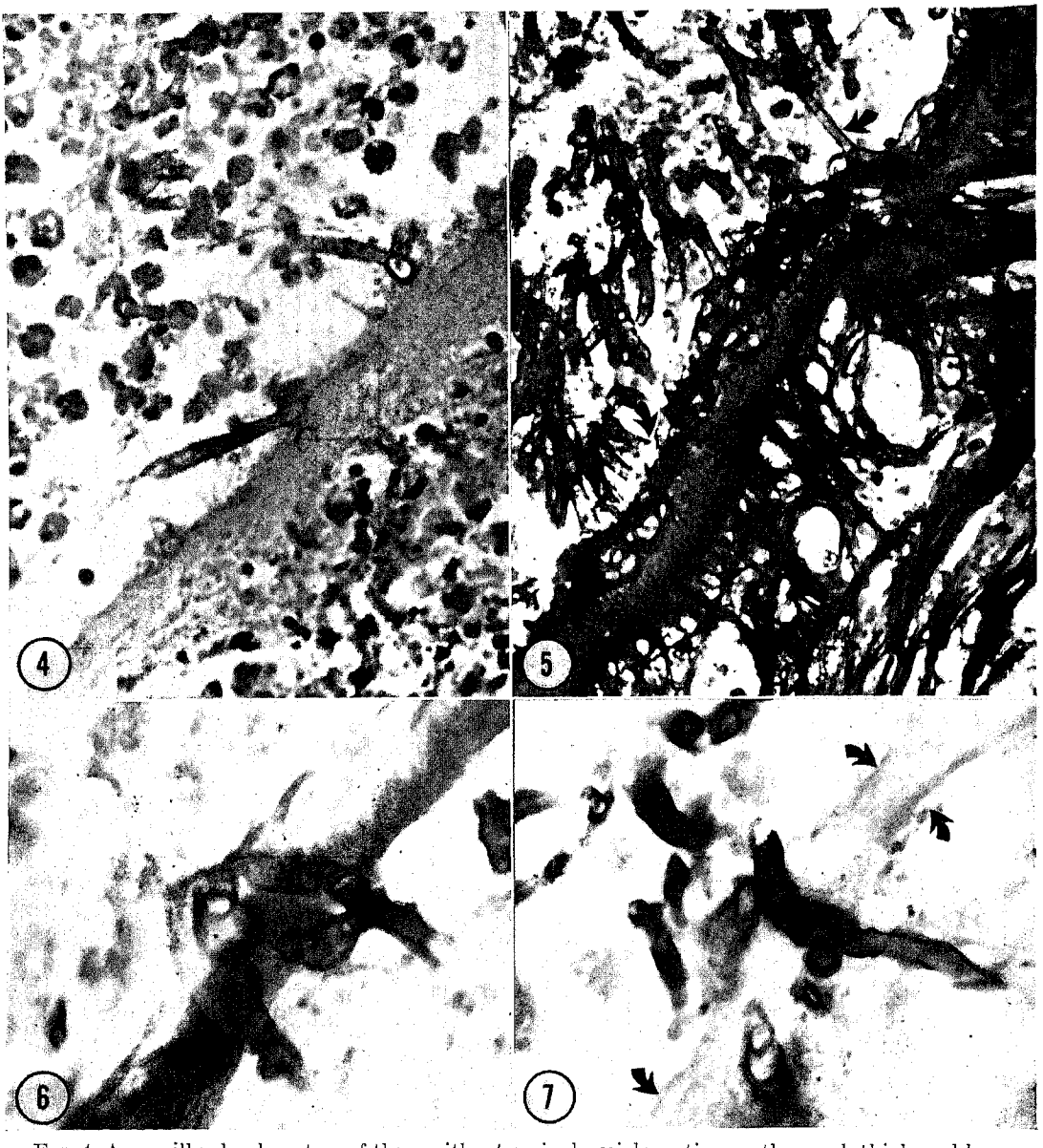


FIG. 4. Aspergillus hyphae, two of them with a terminal vesicle resting on the much thickened basement membrane of the bronchial mucosa (chronic nonspecific bronchitis accompanying bronchiectasis). The bronchial lumen lies above the membrane in the picture (hematoxylin and eosin, × 450).

FIG. 5. Bronchial aspergillosis. Numerous hyphae are present in lumen of bronchus (above the thickened basement membrane), and well developed vesicles within the basement membrane. *Arrows* indicate a hypha in continuity with its intramembranous vesicle, and other hyphae that have been separated by artifact from their vesicles (Robb-Smith modification of Foot method for silvering reticulin fibers, × 450).

FIG. 6. A pair of Aspergillus hyphae invading the lamina propria of the bronchial mucous membrane in a case of chronic nonspecific bronchitis. At least one of these hyphae appears to have originated from the vesicle that can be seen within the thickened basement membrane. The lumen of the bronchus is above the basement membrane in the picture (methenamine silver, × 1300).

FIG. 7. Aspergillus hyphae passing directly through the thickened basement membrane from the lumen of the bronchus (*above*); same specimen as shown in Figure 6. The outline of the basement membrane is *arrowed* (methenamine silver, × 1300).

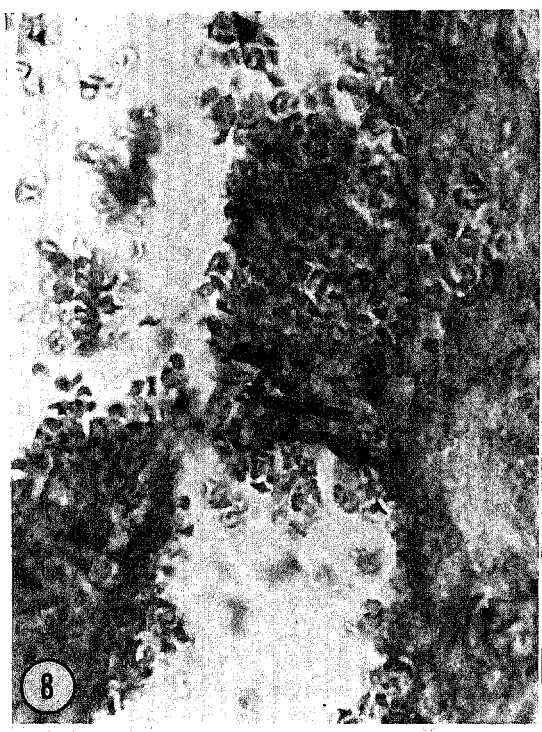
tives. It is, therefore, significant that hyphal invasion of the bronchial mucosa may sometimes be found in surgical specimens that have been fixed *immediately after the operation* by vascular and bronchial perfusion with formol saline (Figs. 8 to 11).

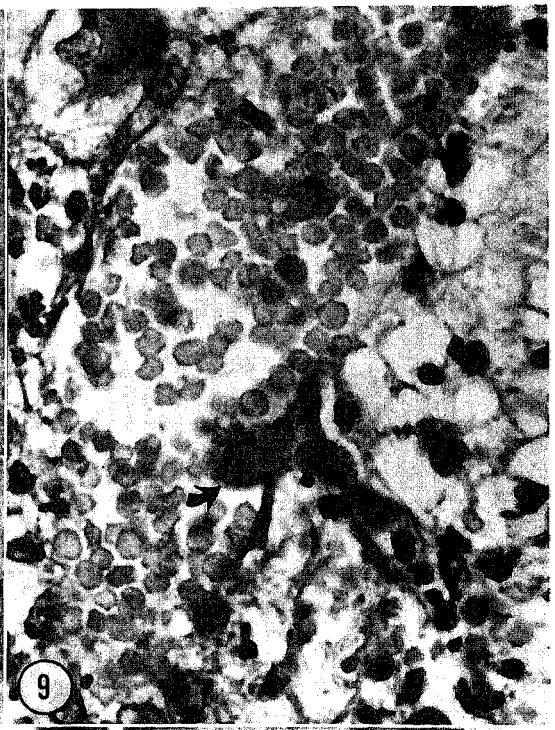
Similarly, and with the same type of difficulty of interpretation, it is sometimes possible to demonstrate that the Aspergillus has penetrated into the superficial zone of the granulation tissue that may form part of the lining of a pulmonary cavity in which an aspergilloma has developed. Foci of necrosis may be found at the surface of the granulation tissue in some of these cavities; these foci are probably a result of pressure by the fungal ball or of other trivial, nonspecific circumstances—their banal appearance, lack of abrupt demarcation, and shallow extent make it very unlikely that they are caused by tissue-damaging substances produced by the fungus. If histologic preparations are examined that include the fungal ball, or part of it, *in situ*, it may be found that the aspect of the ball that has been apposed to a necrotic focus is coated with fresh fibrin threads, enmeshing leukocytes and red blood cells. Although it is technically difficult to display the fungal ball histologically in its natural position in relation to the wall of the cavity, there seems to be no doubt that in certain circumstances it may, at least temporarily, become bound to the wall by fibrinous exudate; at such sites the fungus may be able to extend for a short distance into the tissues (Fig. 12). Oftener, however, the corresponding area of the surface of the ball is thickly covered by an intensely eosinophilic, amorphous deposit of fibrin, which forms clublike expansions over the tips of the hyphae (Fig. 13) comparable to those seen at the surface of colonies of various other pathogenic myceliate fungi, such as *Actinomyces israelii* and *Streptomyces madurae*. Whether this fibrinous deposit has a protective function is speculative; it may tend to limit the growth of the colony, and it may, perhaps, hinder invasion of the tissues by the hyphae. The fibrin-enclosed hyphae may be distinctly attenuated, and their staining reactions may be modified—they may fail to give a positive periodic acid-Schiff reaction, and they may not blacken in Grocott-Gomori methenamine (hexamine) silver preparations.

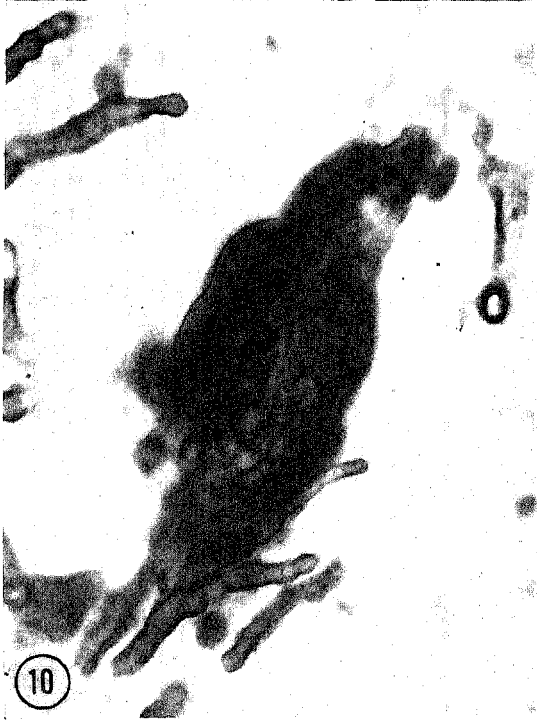
Elsewhere in some of these aspergilloma cavities, clusters of the vesicle-like hyphal expansions may be seen embedded in the surface of the granulation tissue (Fig. 14); sometimes, small masses of mycelium are seen in a similar situation (Fig. 12). These vesicles and hyphae are free from any fibrinous coating, and appear to be quite healthy. Sometimes, only sparse, solitary mycelial filaments are to be found in the granulation tissue lining the cavity (Fig. 15), and, equally often, there may be no evidence at all of invasion by the fungus. I have not observed fungal penetration of the wall of the cavity in any case in which the latter has retained an intact epithelial lining. In those instances in which there has been invasion of granulation tissue, I have not found the hyphae ever to have reached a depth greater than a few hundred microns beyond the surface of the tissue. As in the cases of bronchial aspergillosis mentioned above, it is an unanswered question whether this invasion by the fungus occurs while the tissues are alive, or only after their death.

An Important Reservation Regarding These Observations

In order to underline the hypothetical nature of the suggestion that these findings indicate a possible pathway by which the Aspergillus enters the tissues, it is important to note that neither serious invasion of the lung tissue nor septicemia has been reported as a complication of aspergilloma, even when the patient has been under treatment with corticosteroids and other drugs that are believed to predispose to these serious manifestations of opportunistic aspergillosis. Moreover, it has never been proved that intrabronchial Aspergillus saprophytosis has been the origin of septicemic infection. Nevertheless, there is good reason to consider carefully the possible significance of the observations described here, for it is quite reasonable to believe that fungal penetration of the tissues in the conditions noted might prove to be the starting point of the more serious, opportunistic forms of this mycosis. It would seem advisable, therefore, to regard any patient who is known to have an As-







FIGS. 8 TO 11

pergillus infection as exposed to a greater than average risk, should he develop one of the diseases that predispose to opportunistic aspergillosis, or require treatment with drugs that carry the same hazard, or with x-rays.

PHYCOMYCETOSES

If there is very little factual knowledge about the pathogenesis of opportunistic Aspergillus septicemia, there is even less about the corresponding form of phycomycetous infections. The phycomycetoses, like aspergillosis, have become very much more frequent during the past decade and a half.[10, 18] It is pertinent to recall that it is as recently as June 1956 when Baker,[2] discussing mucormycosis in the United States, was able to entitle his address, "Mucormycosis—A New Disease?" Nowadays, septicemic phycomycetoses, like the septicemic forms of aspergillosis and candidiasis, are known, at least by repute and all too often from actual clinical experience, in every clinic and hospital in which cytotoxic drugs, corticosteroids, or x-rays are in frequent use in the treatment of cancer. Leukemia in particular seems to be specially liable to complication by opportunistic phycomycetoses.[1]

Most of the phycomycetes have a special affinity for blood vessels, causing thrombosis (Fig. 16) and colonizing the thrombi, and spreading thus within the vasculature. This is the means of spread of the infection from the nasal sinuses to the orbit, and thence to the meninges and brain, in cases of *naso-orbitocerebral phycomycetosis*, the opportunistic syndrome to which diabetes mellitus and certain other metabolic disorders predispose.[7] The infection in cases of this syndrome is believed to originate in the upper respiratory tract. It is relevant that other conditions characterized by destructive lesions involving the nose or the nasal sinuses may be complicated by phycomycetosis; this has been recorded in cases of severe facial burns,[25] and I have observed it as a terminal event in the course of a very longstanding, untreated, basal cell carcinoma of the face, in which the maxillary and frontal sinuses had been laid open by the cancer.[33] Ulcerative *cutaneous phycomycetosis* is rare on other parts of the body surface; there is always doubt in these cases whether the mold is to be regarded as the initial infecting agent or as a secondary, opportunistic invader. In some cases the lesion has developed in the course of diabetes mellitus, and there has been no history of a preceding injury to the skin;[12] in others, the lesion has been traumatic in origin (Figs. 17 and 18), and in these cases there may be no associated disease.[33]

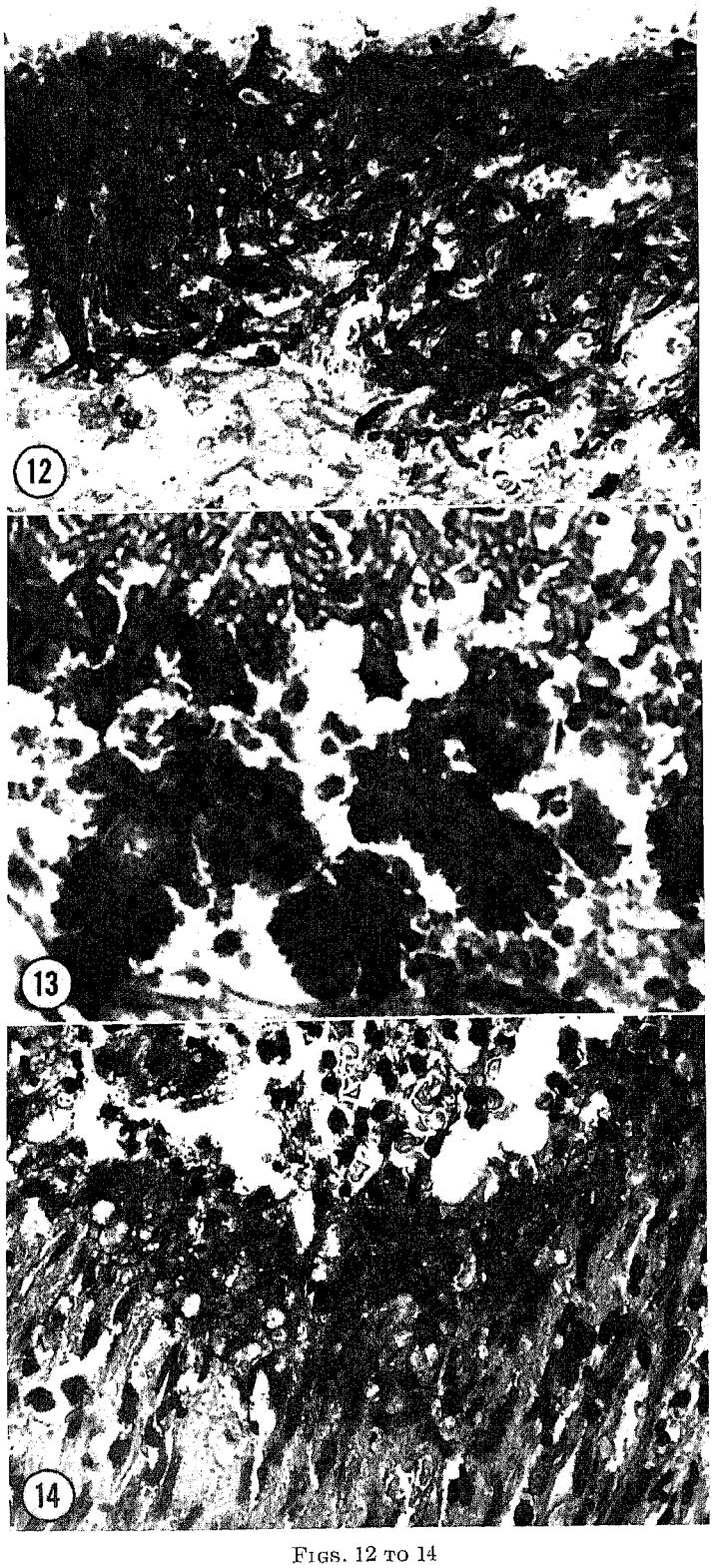
Gastrointestinal phycomycetosis may present as solitary or multiple ulcers (Fig. 19), which may occur in any part of the alimentary canal.[20] Malnutrition is an important

FIG. 8. Aspergillus hyphae in a freshly formed mural thrombus in a vein in the bronchial mucosa The specimen had been fixed by intrabronchial infusion of formol saline immediately after lobectomy (the operation was undertaken because of localized, "dry" bronchiectasis, which had been the cause of repeated hemoptysis). The patient had no other disease, and the only drug therapy given was a course of tetracycline as a prophylactic cover for the surgical procedure. The bronchi contained a small quantity of mucous secretion and exudate; the Aspergillus was sparsely present in this, and had penetrated the mucosa in some of the bronchiectatic areas (Gridley's stain, × 400).

FIG. 9. The *arrow* points to a homogeneous mass of fibrin formed around an Aspergillus hypha that has penetrated a venule in the lamina propria of the bronchial mucosa; same specimen as shown in Figure 8 (Gridley's stain, × 600).

FIG. 10. A large, compact deposit of fibrin around Aspergillus hyphae within the lumen of a vein in a lobectomy specimen in a case of bronchial adenoma. The specimen had been fixed by perivascular and intrabronchial perfusion of formol saline at a pressure head of less than 20 cm. There was heavy Aspergillus colonization of the mucous plugs within the dilated bronchi peripheral to the obstructive lesion, and the mold had invaded parts of the chronically inflamed, collapsed lung tissue. *Aspergillus fumigatus* was isolated from the mucous plugs (methenamine silver, × 1000).

FIG. 11. A pair of attenuated hyphal fragments in the blood within a pulmonary venule in the specimen that is also illustrated in Figure 10. The patient's convalescence from the lobectomy was uneventful, and she has had no symptoms of illness during the 9 years that have passed since the operation. Aspergilli were not found in her sputum at any time after the operation (hematoxylin and eosin, × 900).



FIGS. 12 TO 14

predisposing factor. In some cases, the mold invades ulcers caused by other types of infection, such as amebiasis.

Pulmonary Phycomycetoses[3]

Like the aspergilli, the phycomycetes may invade infarcted lung tissue; more frequently, they may themselves be the cause of infarction, as a consequence of vascular occlusion by infected emboli or by thrombosis secondary to invasion by the mold. It is also possible that toxic products of phycomycetes may cause a "diffusion necrosis" comparable to that accompanying some Aspergillus infections. Whatever its pathogenesis, pulmonary phycomycetosis has a very high mortality. It is most commonly seen as the terminal development in the course of leukemia or of comparable diseases in which the combined effect of the underlying disease and its treatment is so to lower the patient's resistance that the fungus can set up infection of the lung tissue. As in the case of aspergillosis, the precise portal by which the fungus enters the tissues is uncertain; in contrast to aspergillosis, however, there is no evidence of invasion of tissues except as a manifestation of serious, progressive infection. This important difference may well be a reflection of the fact that phycomycetoses are not known to occur in any benign, chronic, nonprogressive forms comparable to the various forms of bronchopulmonary aspergillosis.

Septicemic Phycomycetoses

Any of the foregoing manifestations of ulcerative or visceral phycomycetosis may be the starting point of a generalized dissemination of the infection through the blood stream, and, indeed, this must be regarded as the natural outcome of an infection that is caused by fungi that have such a remarkable proneness to invade blood vessels.

It is a characteristic of the generalized hematogenous disease that widespread vasothrombosis may occur ("septicothrombotic phycomycetosis") (Figs. 16 to 18). There are two clinicopathologic forms of this condition that deserve special mention. One, which is relatively uncommon, is *phycomycetic hepatorenal failure*, in which rapidly progressive jaundice and uremia, due to widespread thrombosing mycotic invasion of the arteries and veins of the liver and kidneys, develop as a complication of leukemia or of other malignant disease under intensive treatment with corticosteroids, cytotoxic agents, or x-rays. The second, and commoner form, is characterized by the development of multiple foci of mycotic infarction in the lungs and brain.

Subcutaneous Phycomycetosis[16]

Subcutaneous phycomycetosis is peculiar among the phycomycetoses because its course is benign, its lesions are confined to the subcutaneous tissues, its geographic distribution is restricted, and its causative organisms are members of the family Entomophthoraceae (*Basidiobolus ranarum*, and possibly other species). The entomophthoraceous fungi appear not to be opportunistic organisms, in contrast to the mucoraceous fungi (mainly species of Mucor and Rhizopus), which are the causes of the more usual clinicopathologic varieties of phycomycetoses. In fact, subcutaneous phycomycetosis is a distinct entity, perhaps no more closely related to other phycomycetoses than, say, nocardial mycetoma is related to tuberculosis. The disease was first recognized in Indonesia, by Lie Kian Joe and his colleagues, in 1956,[15] and has since

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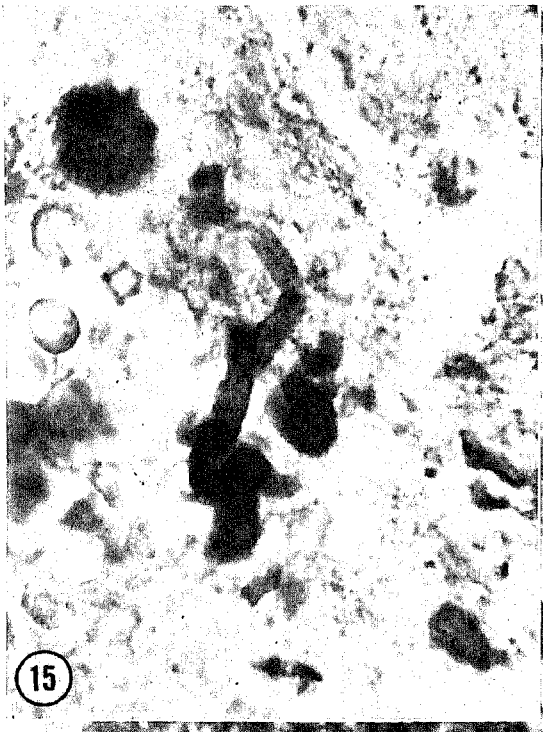
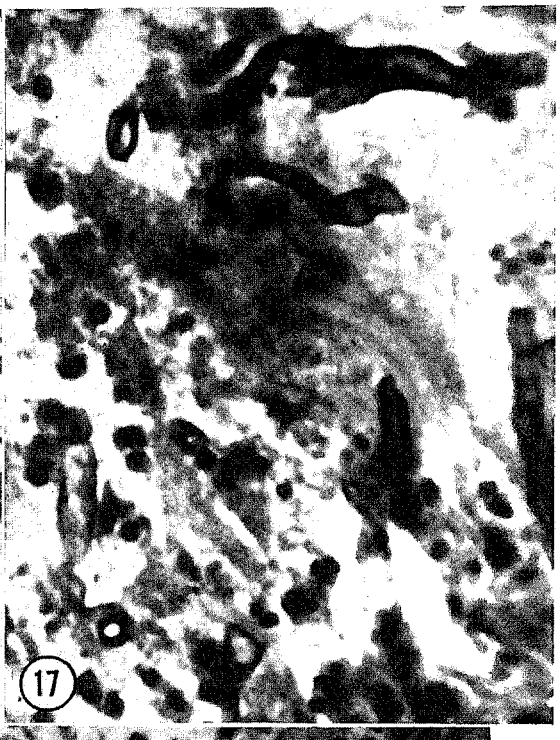
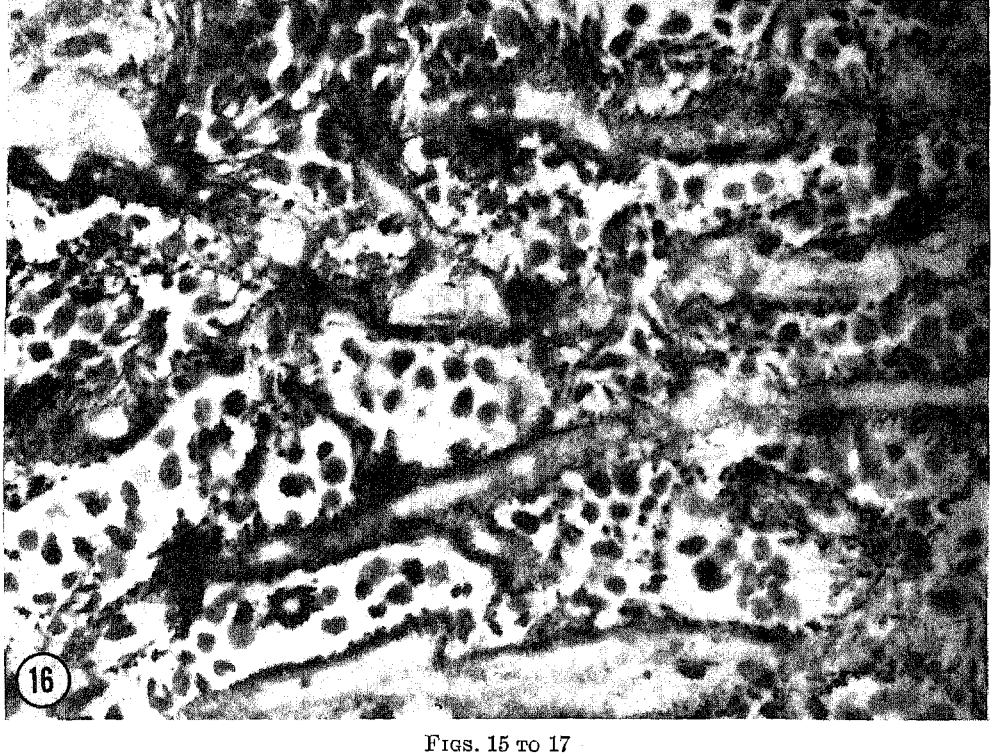
Fig. 12. Superficial invasion of eroded granulation tissue by a feltwork of Aspergillus mycelium at the site of ulceration of the lining of a cavity containing an aspergilloma (Gridley's stain, × 350).

Fig. 13. Clublike structures resulting from deposition of fibrin around some of the hyphae at the surface of an aspergilloma. With care, the outline of the hyphae can be made out, even within the large, dense, fibrinous masses in the lower half of the picture (periodic acid-Schiff and light green, × 550).

Fig. 14. A cluster of Aspergillus vesicles is seen at the margin between the organized granulation tissue and the necrotic surface of the lining of the cavity in which an aspergilloma had formed. The specimen was obtained by lobectomy; the cavity was regarded as of bronchiectatic origin. There was no evidence of any other disease, and the patient had not had any drug therapy (hematoxylin and eosin, × 400).







FIGS. 15 TO 17

been reported from parts of Africa.[5, 11, 17] In my report of a case seen in Britain in a Dutch child from Indonesia,[31] I mentioned that there was thrombosis of some of the blood vessels in the infected tissue. At the time of writing that paper, an exhaustive study of serial sections had failed to show actual invasion of these vessels by the fungus, but this has since been achieved (Figs. 20 and 21). Ordinarily, subcutaneous phycomycetosis is regarded as exceptional among the phycomycetoses because of the absence of a mycotic angiitis; the involvement of the vessels in my case, although on such a very small scale, indicates the possibility of a potentially dangerous extension of this mycosis should the patient's resistance be lowered, for instance by the ill-advised use of drugs such as corticosteroids in treatment of the infection itself, or by their use against some other disease with which the infection happened incidentally to be associated.

SOME COMPARISONS AND CONTRASTS BETWEEN ASPERGILLOSIS AND PHYCOMYCETOSES

Although, etiologically, aspergillosis and the phycomycetoses have much in common (see introductory discussion, above), there are differences in the relative frequency with which they are liable to follow the various types of predisposing circumstances. For instance, diabetes mellitus is frequently a predisposing factor in cases of opportunistic phycomycetoses, whatever clinicopathologic form the infection may take; in contrast, aspergillosis seems to occur no more frequently in cases of diabetes than could be explained by chance association. Again, opportunistic pulmonary infections are caused much oftener by aspergilli than by phycomycetes, and the reverse is true of the opportunistic infections of the gastrointestinal tract. The explanation of this differential incidence may prove to lie in the differences in habitat of the fungi; *A. fumigatus* and other Aspergillus species are not uncommon in sputum, whereas phycomycetes appear to be rarely found in the lower parts of the respiratory tract.[26] I know of no investigations into the relative frequency of aspergilli and phycomycetes in the alimentary tract. There is presumptive evidence that phycomycetes may be harbored more frequently than aspergilli in the nose or nasal sinuses; aspergillosis is a rare infection in these parts, whereas their liability to be the site of opportunistic phycomycetous infections is well marked.

Little can be said about the possible pathogenetic similarities of the two diseases. The potential significance of the localized, superficial invasion of the tissues in cases of chronic or saprophytic bronchopulmonary aspergillosis has been discussed above. In contrast, the organisms that cause the opportunistic phycomycetoses are not known to cause chronic localized infections, comparable to those of aspergillosis; tissue invasion by these phycomycetes is, in fact, a manifestation of an already established, and

FIG. 15. A solitary Aspergillus hypha in the granulation tissue lining an ulcerated area in a cavity in which there was an aspergilloma. There had been repeated episodes of hemoptysis, and eventually the patient died from hemorrhage from an eroded pulmonary artery. The necropsy was performed an hour after death, and the specimen was fixed immediately in formol saline. Pulmonary fibrosis and cavitation were attributed to sarcoidosis; the patient had not been treated with any drugs except for a broad spectrum antibiotic during the terminal stage of his illness (Gridley's stain, × 1300).

FIG. 16. Extensive phycomycetous colonization of a blood vessel in the lung in a case of "septicothrombotic phycomycetosis" complicating severe, poorly stabilized diabetes mellitus of only 5 weeks' known duration. The patient was a schoolgirl, aged 14. She died of renal and hepatic failure, due to extensive hemorrhagic necrosis of the kidneys and the liver; the necropsy also showed multifocal hemorrhagic infarction of the lungs. Note the striking, fibrillar deposition of fibrin on the surface of the hyphae. No attempt was made to isolate any organisms in cultures (phosphotungstic acid-hematoxylin, × 600).

FIG. 17. Hyphae of a phycomycete in necrotic tissue in a cutaneous ulcer that had developed as a result of pressure by an ill-fitting, rough plaster-of-Paris cast. The patient, a middle-aged man, sustained a Pott's fracture while he was under treatment with corticosteroids for chronic lymphatic leukemia. He died of hepatorenal failure; necropsy showed "septicothrombotic phycomycetosis," with extensive renal and hepatic necrosis (see Fig. 18) (periodic acid-Schiff and hematoxylin, × 600).

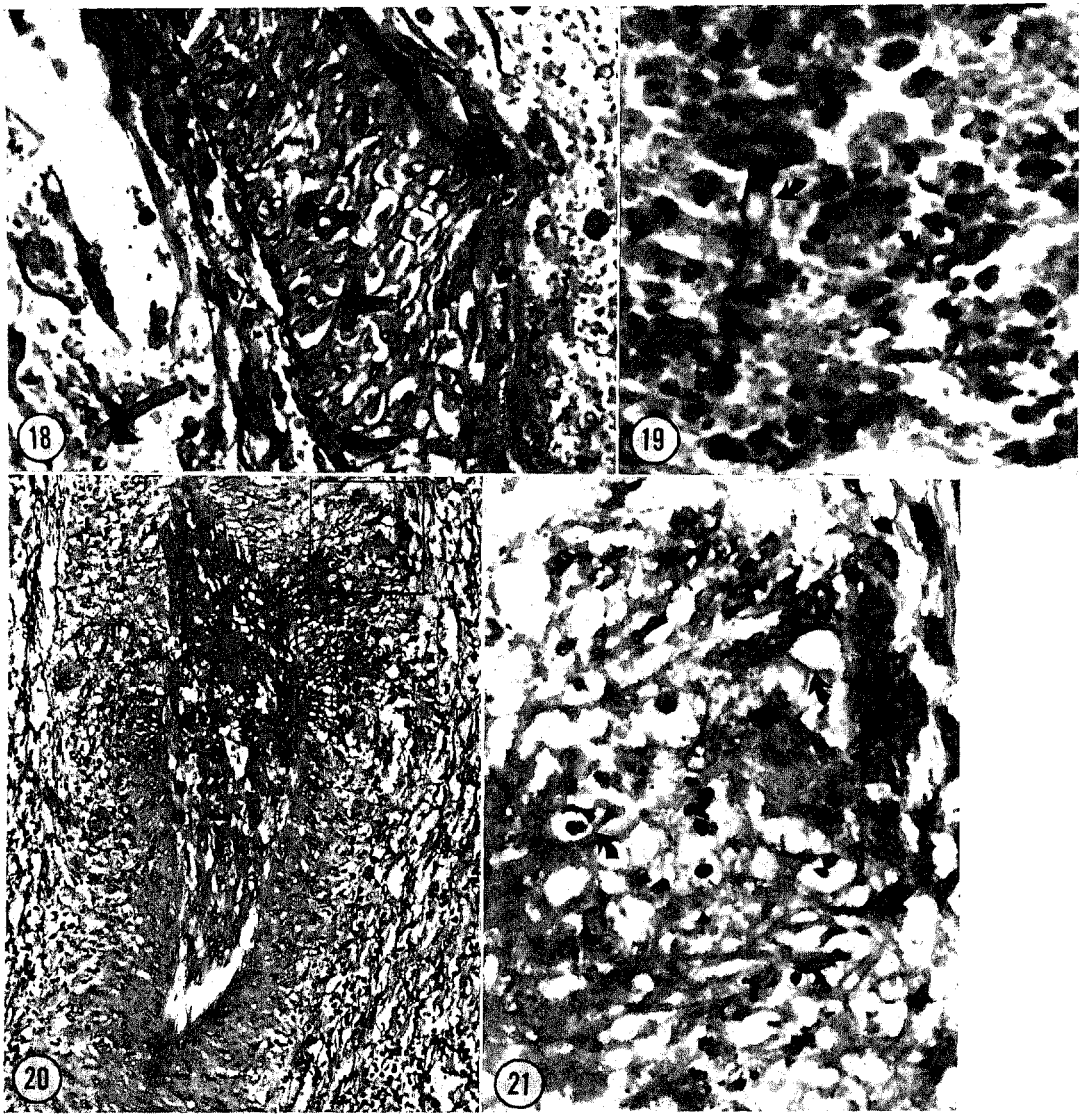


FIG. 18. Invasion of the wall of a branch of a renal artery by hyphae of the phycomycete (same case as illustrated in Figure 17). The vascular lumen is on the right of the picture; there is thrombosis over one area, where the internal elastic lamina has disappeared. It should, however, be noted that a breach in the elastic lamina is not necessary for penetration of the vessel wall by the fungus (methenamine silver, × 290).

FIG. 19. The *arrows* point to hyphae of a phycomycete in the granulation tissue of an acute gastric ulcer. The patient had aplastic anemia, and died of staphylococcal pneumonia. The gastric lesion was the only evidence of phycomycetosis found at necropsy (periodic acid-Schiff and hematoxylin, × 600).

FIG. 20. The wall of this thrombosed artery in the affected tissues in a case of subcutaneous phycomycetosis (previously reported[31]) is invaded by the fungus. The area outlined is reproduced at a higher magnification in Figure 21 (periodic acid-Schiff and hematoxylin, × 115).

FIG. 21. This field, which is the area enclosed within the rectangle marked in Figure 20, includes two cross-sections through hyphae of the phycomycete (*arrowed*) (periodic acid-Schiff and hematoxylin, × 480).

usually opportunistic, progressive infection. The striking affinity of the phycomycetes for the blood vessels must, however, indicate the likelihood that their initial access to the blood stream in cases of opportunistic septicemia is usually through the vasculature in some focus of mucosal infection complicating local damage, of whatever nature.

SUMMARY

Aspects of the pathology of aspergillosis and of the phycomycetoses are reviewed, with particular reference to the pathogenesis of the opportunistic septicemic forms of these infections.

Attention is drawn to the invasion of the tissues, and eventually of the vasculature, that may occur locally in cases of Aspergillus bronchial saprophytosis and of intracavitary aspergilloma. Comparable vascular invasion may be seen in some localized lesions of candidiasis. Presumably, under ordinary conditions, penetration of these fungi into the blood vessels does not lead to septicemia, the infection being successfully held in check by the body's defenses. When these defenses are weakened, either by the diseases that predispose to generalized infections by these fungi, or by therapeutic agents that have the same effects, opportunistic infection is liable to follow, with the development of mycotic septicemia.

Similarly, the opportunistic phycomycetoses develop when predisposing conditions so interfere with the defensive mechanisms that the characteristic tendency of the causative phycomycetes to invade the blood vessels leads on unhindered to the establishment of septicemia. No observations relating to the possible portal of entry of the phycomycetes into the tissues can be made, beyond noting that the nose and nasal sinuses, and ulcerative lesions of the gastrointestinal tract, may be the most frequent sites of the initial development of the infection.

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