

The Natural Focality of Adiaspiromycosis (Adiasporosis)

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Abstract. Adiasporosis occurs in nature in foci from which it may spread from case to case in the cold season of the year and under certain ecological conditions to animals reared by man (especially laboratory animals) and even to man himself. Hence this disease is a natural focus infection of a non-transmissible type occurring in the northern and southern part of the temperate zone. Individual components of the natural focus and characteristic features of natural focality of this systemic mycosis are discussed in detail and its dependence upon the substrate of burrows in the cold season and upon the maintenance hosts in the warm season is pointed out.

In a previous paper (DVOŘÁK, OTČENÁŠEK, ROSICKÝ 1966) the probable life cycle of *Emmonsia crescens* Emmons et Jellison, 1960 in nature was outlined and the term adiasporosis recommended. *E. crescens* apparently can have the following cycles in nature: a) terrestrial (extra-animal) cycle and b) animal-terrestrial cycle. Diseases of laboratory animals have been also observed (DVOŘÁK, OTČENÁŠEK, RAŠÍN 1966) and an occurrence of the infection in man has been described (DOBY—DUBOIS et al. 1964).

Our aim in this paper is to analyze the hitherto known facts about this disease, in order to find out whether adiasporosis is a typical disease with a natural focus character in the sense of PAVLOVSKY'S theory (1940, 1964). Of course, we must bear in mind that this infection is not of transmissible character (i.e. it is not transmitted by arthropods), although the theory of natural focality has been extended and applied by its founder also to diseases of non-transmissible type. Taking as a basis the results of our field investigations and laboratory experiments (DVOŘÁK, in press) we present the known and most recent facts and assess them in the light of the natural focality phenomenon. Under the term natural focality we understand a general biological phenomenon, where the pathocergont, being a component of an ecosystem formed in the course of its evolution or due to human activities and circulating in this ecosystem independently from the organism "at risk",*) spreads to the species at risk by various routes under favourable ecological conditions (ROSICKÝ, in press).

*) In the concept formulated by Audy (1965).

1. The pathoergont

In our case the pathoergont (sensu PAVLOVSKY 1965) is the fungus *Emmonsia crescens* belonging to the *Deuteromycetes*. The adiaspores identified were undoubtedly those of *E. crescens*. The mycelial phases of the individual *E. crescens* strains are indistinguishable at present, because differentiating methods are not known. On the other hand, differences between some strains seem to exist in the morphology and in the size of mature (largest) adiaspores. In this connection the similarity of our Czechoslovak strains to the Scandinavian strains (JELLISON et al. 1960) will be pointed out. Strains of both countries produce thin-walled, relatively large adiaspores in the lung of the host. Experimentally produced lung adiaspores of Czechoslovak strains were visible with the naked eye no sooner than after 20 days of infection.

It was reported (DVOŘÁK, OTČENÁŠEK, PROKOPIČ 1967) that the infection elements — aleuries — are produced by the mycelial phase of *E. crescens* even at +4 °C. Such low temperatures seem to be more advantageous for the saprophytic life of *E. crescens* in nature. The moulds commonly present in the soil, which at room temperature rapidly grow to suppress *E. crescens*, are remarkably inhibited by such low temperatures as observed in our laboratory experiments. Moreover, two as yet unidentified antagonists of the mycelial phase of *E. crescens* ascertained by us (a mite and a nematode) move and consume the developing mycelium with aleuries at a slower speed when exposed to these low temperatures than, for example room temperature. It remains to be seen what the conditions are for the growth of the saprophytic phase of *E. crescens* in nature.

We have little information on natural conditions (temperature, humidity etc.) in the nests of small terrestrial mammals. Most useful and in our opinion most authentic data were obtained by DANIEL (1964, 1965). He carried out continuous measurements of temperature about 10 cm below the ground surface including the litter layer of artificial nests containing the young of *Clethrionomys glareolus*. In summer, up to the 24th day after the young were born, the temperature in the nest ranged between 17° and 19 °C, but later it reached 18° to 21 °C. The temperatures measured outside the nest had a greater range, from 12° to 28 °C. The nest functioned as a perfect thermostat. The mentioned summer temperatures in the nest of *C. glareolus* correspond with room temperature, i.e. the temperature at which the growth of *E. crescens*, as our laboratory experiments showed, is suppressed by other moulds.

On the other hand, the same continuous measurements conducted in winter showed (DANIEL 1964, pers. comm.) that the nest temperature during the frosty spell ranged between +3° to +5 °C. This fact indicates that there are portions of time during the cold period of the year which promote the development of aleuries of *E. crescens* (i.e. the terrestrial cycle), as the species is well adapted to lower temperatures than to those existing in the soil in the summer period. Our laboratory experiments on the growth of *E. crescens* at the temperature of +4 °C

prove this convincingly. Outside the actual nest chamber and the vicinity of burrows the temperatures are much lower and do not allow further growth of the fungus. Most rodents and insectivores which have been identified as maintenance hosts of the fungus usually build their nests at a depth of 10—30 cm. Adiaspores were hardly ever found in nonfossorial animals (DVOŘÁK, OTČENÁŠEK, PROKOPIČ 1965).

The properties and conditions under which the only infectious elements of the fungus, the alcuries, can grow, make the pathoergont dependent upon certain soil sections (burrows and nests of terrestrial mammals), promote the infection of small mammals and lead to the seasonal occurrence of the infection etc. All these criteria point to the natural focality in the occurrence of the fungus.

2. The maintenance hosts

The vertebrates which are involved in diseases characterized by a natural focality phenomenon, are usually designated as reservoirs of infection and the pathoergont, as a rule, multiplies in them. In our case, however, we have proved (Dvořák, in press) that *E. crescens* does not multiply in its host after the latter became infected with alcuries and that the infection does not spread from animal to animal. The adiaspores, i.e. the parasitic stages, reach the soil only after the death of the host.

In spite of the fact that the fungus does not multiply in the organisms of the maintenance hosts, the small terrestrial mammals are an inseparable link in its life cycle, because they facilitate its survival during the warm period of the year, as well as its dissemination in nature (DVOŘÁK, OTČENÁŠEK, ROSICKÝ 1966). The biology of the fungus, the favourable environment of nests and the population dynamics of small terrestrial mammals combined together promote the existence of the fungus in nature. This is demonstrated by our host material and by analysis of the population dynamics of *Microtus arvalis*.

From January 1, 1963 to April 30, 1964 a total of 1465 small mammals belonging to 9 species were collected in East Bohemia (Nový Bydžov) and the finding of *E. crescens* in four species of rodents (*Apodemus flavicollis*, *A. sylvaticus*, *Clethrionomys glareolus* and *Microtus arvalis*) was reported (PROKOPIČ, DVOŘÁK, OTČENÁŠEK 1965). Later the fungus has been repeatedly isolated (OTČENÁŠEK, DVOŘÁK, PROKOPIČ 1965). The subsequent studies undertaken on this collection showed that the monthly distribution of adiaspores was remarkably irregular. Only the animals captured in March, April and May were found to be infected. Therefore we continued our investigations in the course of a whole year (1964) in the same locality. We conducted observation on *A. flavicollis*, *C. glareolus* and *M. arvalis*, occurring in sufficient numbers in the locality mentioned above. A total of 2619 animals (150 *A. flavicollis*, 204 *C. glareolus* and 2265 *M. arvalis*) was collected and their lungs were carefully examined macro- and microscopically for the presence of adiaspores. Four specimens of *M. arvalis* captured as early as in February were infected, but the highest incidence of infection was found in the animals captured in April and May,

similar to the incidence of the preceding year (DVOŘÁK, OTČENÁŠEK, PROKOPIČ 1967).

In our further studies of findings reported from other countries a very interesting reference attracted our attention. In the vicinity of Hamar (Norway), an immature female of *Microtus* sp. infected with adiaspores was captured on April 7, 1959 (JELLISON, VINSON, HOLAGER 1960). Unfortunately, the specific name of the vole is not mentioned, but as the mode of life of all species of the genus *Microtus* is relatively similar, the lacking identification of the species is not important. This finding permits to estimate approximately the date of acquisition and perhaps also the source of infection.

An analysis of seasonal changes which took place in the population of *M. arvalis* in the spring (March—April) indicates that the population was composed of individuals which were born in mid-summer or some later period of the previous year and which were relatively developed (PELIKÁN 1959). As the living conditions were improving gradually during the spring, these old individuals which had overwintered rapidly developed to maturity and the first young animals were added to the population. In this period masses of aleuries were probably inhaled into the lungs of the animals and subsequently developed into adiaspores. On the basis of our findings we can presume, that some individuals became infected the previous autumn when they first inhaled aleuries from mycelia developed from adiaspores which had been carried into the burrows by individuals infected in spring.

We may presume that at least 2 to 3 F_1 generations and 1 F_2 generation become infected in spring, depending on local conditions. The females of *M. arvalis* reach sexual maturity when they are about one month old (PELIKÁN 1959). Approximately for the first three weeks of their life the animals live mainly in the burrows. On an average the pregnancy lasts 19—21 days and the female can become pregnant again immediately after giving birth. Thus the female may produce four litters in three months (FRANK 1956). This high reproductive rate, however, is characteristic mainly for the common vole. Other species of voles (for example *Microtus oeconomus*) have a lower breeding rate; KRATOCHVÍL and ROSICKÝ (1955) reported only two litters produced by parental generation by the end of May. Hence it follows that in spring there are numerous individuals in nature, especially those of *M. arvalis*, which may become infected with aleuries at the beginning of spring. In summer the population grows rapidly and young individuals are predominant. Due to worsening living conditions the old individuals born in the spring (and among them single old individuals that have overwintered) die off. In this way the adiaspores are returned to the soil and to the burrows (completion of the animal—terrestrial cycle) and with the increasing cold weather the mycelia start to develop. Towards the end of autumn and at the beginning of winter the old population disappears altogether and all adiaspores are returned to the soil. During the warm season of the year the adiaspores survive in the lungs of host. However, only a small number of hosts in the whole population survives until autumn.

The inclusion of the parasitic phase in the host is therefore an important factor for the existence of *E. crescens* in the biocenosis of the natural focus.

3. The habitat

The burrow of the small mammal, with its nest and other frequently used areas, is the biotope (habitat) proper of the mycelium of *E. crescens*. The relatively closed space of nest probably facilitates the aspiration of aleuries. On the other hand, it is very probable that in the warm period the mycelial phase disappears or is reduced in burrows by the action of antagonists stimulated by higher terrain temperatures.

Moreover, the faeces of small terrestrial mammals may have a similar effect on *E. crescens* as bird droppings have on *Histoplasma capsulatum*. Avian faeces stimulate the production of viable small aleuries of *Histoplasma capsulatum*. Only spores up to 5 μ in size have a chance to enter deeply the respiratory tract and to be fixed there (FURCLOW 1965).

The fungus is maintained in a certain biotope by a complicated dynamic mechanism resulting from the two-phase property of the pathoergont. This property of alternating saprophytic and parasitic phases makes the pathoergont in a certain ecosystem dependent upon a certain substrate (burrow) as well as upon the host and vice versa, the host creates a suitable environment for the existence of the saprophytic phase. It stands to reason that such intricate relationships become fixed into certain ecosystems (geobiocenosis—ROSICKÝ, in press) and are indicative of an ancient evolutionary process.

We assume that the burrows are the actual places of the occurrence of *E. crescens* in winter while in summer the life of the hosts is concentrated there. The nests are therefore basic elements of the maintenance and further dissemination of adiasporosis. In the natural focus the burrows may be regarded as certain elementary foci (ROSICKÝ 1962a), although we are well aware that burrows in an ecological zone of small terrestrial mammals represent a communication system.

Natural foci are peculiar to certain geographic complexes consisting of biocenoses which have developed in the course of a long evolution and in which the pathoergont circulates. In our case, we think, the infection is related to both parts of the temperate zone, occurring in the ecological zone of small terrestrial mammals. We cannot be sure, however, to which type of vegetation (forest, steppe etc.) *E. crescens* belongs because our knowledge of its distribution is still incomplete.

4. The seasonal incidence

DVOŘÁK, OTČENÁŠEK, PROKOPIČ (1967) studied the monthly frequency of the infected animals (*Apodemus flavicollis*, *Clethrionomys glareolus* and *Microtus arvalis*) captured at Nový Bydžov (East Bohemia) in 1964. In the case of *Microtus arvalis*, the highest incidence of infected animals was stated to occur in April. Thus it is clear that the mentioned female of *Microtus* sp. found in Norway was infected in a cold period of the year, probably from a source occurring in the burrows. Among 150 spe-

circens of *Apodemus flavicollis* and 204 specimens of *Clethrionomys glareolus* captured in the course of the whole year, the infection was diagnosed only in those animals which were captured in May. Therefore it might be possible that these animals were also infected in the cold period of the year.

The occurrence of adiasporosis in small mammals reaches a pronounced peak in spring. Later in summer, however, the composition of the host population changes and the occurrence of infected individuals decreases both absolutely and relatively because they are members of generations which either over-wintered or were born in early spring. New generations gradually appear which cannot become infected and clearly predominate in the population. The infected animals seem to die only in some cases, probably when numerous large adiaspores are aspirated into their lungs. It may be expected that some infected animals survive summer. E. g. our investigations showed that towards the end of August in the population of small mammals in South Bohemia a mass infection in *Arvicola terrestris* (in 1 out of 14 specimens captured) could be traced.

Especially in the autumn cannibalism among *Microtus* individuals is not exceptional. It may be assumed that the adiaspores, like the elements of some other fungi (e.g. HEJTMÁNEK, HERODEK 1958), remain viable after having passed through the alimentary tract of the animals and thus infect the burrow.

Like other natural focus diseases, adiasporosis is characterized by definite seasonal dynamics, a fact which plays a significant role as far as the infection of organisms at risk is concerned. The findings in laboratory animals and in man also indicate the seasonal dynamics of the disease.

5. The dissemination from natural foci

As mentioned above, the dissemination of adiasporosis to organisms at risk is of non-transmissive character, i.e. without vectors. At the time of the development of aleuries the nest (including the adjoining system of burrows and other nest area where there are favourable conditions for the saprophatic phase of fungus) provides an environment in which every warm-blooded organism encroaching it may become infected. *Emmonsia crescens* exists there independently on the organism at risk (sensu AUDY 1965). This means, that apart from the inhabitant of the burrow, all other mammals which enter such a burrow, are included in the circulation. From these burrows (elementary foci) the infection may then spread to animals reared by man and to man himself.

Diseases of laboratory animals which were caused, according to the epizootologic analysis, by providing the animal cages with soil containing the remnants of burrows of free living small mammals together with aleuries, are a convincing proof of such a dissemination of the disease. The infection of laboratory mice and rabbits was observed by DVOŘÁK, OTČENÁŠEK, RAŠÍN 1966.

The observed phenomenon is in harmony with the theoretical suppositions

concerning the spread of natural focus diseases to laboratory animals, as formulated by ROSICKÝ (1962b).

In another case the infection occurred in man (DOBY—DUBOIS et al. 1964). Cases of potential infection cannot be ruled out either, because man is often handling soil during the excavation work (digging of ditches, forest work etc.) in the cold season of the year in which the rodents reached high numbers. Thus adiasporosis may be regarded as a potential disease of man—*E. crescens* being a recently discovered human pathogen—characterized by the natural focality phenomenon and it should be included in the medical science as a new disease in the sense of PAVLOVSKY'S theory.

The routes by which the infection may spread to the organism at risk are also in accordance with the natural focality theory. We suppose in our case that we are confronted with an example par excellence of a non-specific pathoergont which, under certain ecological conditions, may reach man and domestic animals.

CONCLUSIONS

On the basis of our analysis we may conclude that adiasporosis occurs in natural foci from which it may spread from case to case in the cold season of the year and under definite ecological conditions to laboratory animals and even to man himself. Hence it follows that this disease is a natural focus infection of non-transmissible type occurring in the northern and southern parts of the temperate zone. According to the present knowledge its saprophytic form, the mycelium, occurs in burrows of small terrestrial mammals and produces aleuries which are inhaled by the animals. The aleuries in the lungs of the animals develop later into adiaspores, which represent the parasitic phase of the fungus. This phase facilitates the fungus to survive the warm period of the year when the mycelium in soil is subject to the antagonism of other moulds. In most cases the presence of adiaspores in its lungs does not endanger the life of the host which carries them until it dies. Carriers of adiaspores are primarily population individuals born in the spring season. The cold weather in the autumn period causes the high die-off of the population of small terrestrial mammals and consequently the return of adiaspores to the soil in the burrows, where the mycelia grow again.

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