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THE CLASSIFICATION AND NOMENCLA-TURE OF THE RAY FUNGI.

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THE classification and the nomenclature of the group of organisms of which the "ray fungus" associated with actinomycosis is the most prominent member have been far from uniform. A glance at the current textbooks of pathology and bacteriology will at once show that the nomenclature of these organisms is even now somewhat confused and the lists of synonyms have reached startling dimensions. A simplified nomenclature which would be generally acceptable was not to be expected until this and similar groups of microorganisms were subjected to extended, systematic botanical investigation. It is believed that the recent studies in this direction by Gasperini, Rossi Doria, Domec, Sauvageau and Radais, Berestneff and Lachner have thoroughly paved the way for a more satisfactory classification and intelligent nomenclature of these organisms whose number has rapidly multiplied.

In order to appreciate the significance and the relative importance of the reasons for the use of various terms and schemes of classification previously and now in vogue in connection with the organisms in question, it becomes necessary to briefly

review their history.

It may be said at the outset that the essential difficulty met with in the study of these organisms has been the question whether they were to be regarded as belonging to the so-called higher bacteria or cladothrix, to the simpler moulds, or should they constitute an intermediate group, the streptothrix group. It is also perhaps well to remind the reader that the

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principal morphological characteristic of these plants is a unicellular, branching mycelium which generally sends out spore-bearing threads.

STREPTOTHRIX AND CLADOTHRIX.

In 1874 Ferdinand Cohn discovered in water, and in 1875 he described, a colorless, microscopic plant with so-called false ramifications which he termed Cladothrix dichotoma (κλάδος, a sprout θριξ, a hair) and assigned to the higher bacteria. At the same time Cohn described and figured certain filaments in peculiar concretions from the tear-duct, given him by R. Förster. Such lachrymal concretions had been recognized by A. von Graefe, Hirschler, Grube and others, and regarded usually as masses of the fungus of favus; Waldeyer and others likened the threads of which they were largely made up to the filaments of Leptothrix buccalis. Cohn, however, found the organism in question to be composed of branching threads, called it Streptothrix försteri (στρεπτος, twisted; $\theta_{\rho \iota} \xi$, hair), and assigned it also to the higher bacteria with a question-mark. He could not study the organism as closely as necessary in order to give it a definite place in the system of botanical classification because he failed to obtain any cultures.

Cohn divided the thread-like bacteria (Nematogenae) into two groups, the unbranched and the ones with false branches, and he tentatively assigned both cladothrix and streptothrix to the latter. In his description, however, Cohn states the differences between cladothrix and streptothrix somewhat more

clearly:

"Cladothrix n. g. filamenta leptotrichoidea tenerrima, achroa, non articulata stricta vel undulata,

pseudichotoma.

"Streptothrix n. g. filamenta leptotrichoidea tenerrima, achroa, non-articulata vel anguste spiralia, parce ramosa."

Cohn's figures show very distinctly that the method of branching of his Streptothrix försteri has nothing in common with that of the cladothrix. Nevertheless there remained a degree of uncertainty in regard to their classification which undoubtedly helped much to bring about the confusion which later arose.

In 1877 Cienkowski studied the process of segmentation in Cladothrix dichotoma of Cohn, with whom he agreed as to principal characteristic, namely the mode of branching. He states that he has sometimes seen branches arising by true ramification, but he makes no mention at all of the Streptothrix försteri

and its peculiarities in this respect.

It is true that Marchand, in 1883, classified Streptothrix försteri as a distinct genus, separable from cladothrix because of its true branching, but both Zopf and Winter at about the same time regarded the two as wholly identical, Zopf designating Cohn's Streptothrix F. as Cladothrix F. Cladothrix is described as a fission fungus, composed of encapsulated cells, arranged in the form of threads which present a pseudo-dichotomy due to the divergence of two members of a thread and their further growth by cellmultiplication. Thus it came about that the streptotrices, of which many new species were described during the subsequent 8 or 10 years, were all, or nearly all, designated as cladothrix and classed with the bacteria, the fundamental difference between the modes of branching of streptothrix and cladothrix being wholly overlooked or wrongly interpreted.

Lachner calls special attention to the element of confusion which arose from the erroneous interpretation of the significance of dichotomy, or dichotomous branching, by writers, especially by pathologists and bacteriologists. To quote directly from Lachner:

"In every textbook of botany dichotomy is defined as a peculiar form of branching in which the mother axis becomes replaced by two daughter axes, of equal value, due to a symmetrical division of the stem into two limbs at the point of branching. In all other forms of branching, on the other hand, the mother axis runs on so that one or more branches of higher order appear below the point of growth. Now pseudo-dichotomy is used to signify that the above arrangement is only an apparent one, whereas in reality the mother axis runs on. Accordingly Cohn correctly described the branching of his cladothrix as a pseudo-

dichotomy. Many authors, however, wrongly regard dichotomy as meaning any branching which results in two limbs at the point of ramification, regardless of whether these are equivalent or whether one represents the mother axis, dichotomous branching being used as synonymous with one-sided branching. This is the sense in which dichotomy is described in connection with streptothrix and actinomyces (Affanassiew, Harz, Terni, Hesse, Hertwig, Rutimeyer, Bostroem, Th. v. Schroeder, v. Mosetig-Moorhof, Zopf and Winter classed streptothrix with cladothrix and as an exact examination is difficult on account of the minuteness of streptothrix, many authors referred to the latter also as being pseudodichotomous, which in the accepted significance of dichotomy means "false branching." Even W. Kruse fell into this error in his description of the streptotrices when he states that the threads branch dichotomously and then contradicts himself a few lines further on by saying: "This branching, which forms acute or (mostly) right angles, is a genuine one due to lateral budding. A lateral budding, however, excludes all dichotomy!"

The unification of streptothrix and cladothrix greatly hindered the knowledge of the former because the observations in regard to cladothrix were simply applied to streptothrix, and thus it came about that years passed after the discovery of actinomyces Harz, for instance, before it was recognized that it belonged

to the streptothrix group (Lachner).

During this time—1882–1892—many new varieties or species of streptothrix were described. In 1883 J. Miquel observed a branching organism in the air which he held to be cladethrix, but which, according to Rossi Doria, is closely related to streptothrix. In 1887 Edington described the Bacillus arborescens which he found in the scales of scarlet fever and which is a streptothrix. In 1888 B. Naunyn described a ferruginous organism in the endocardial vegetations and the leptomeninx of a case of chorea. Zopf declared the organism to be a cladothrix, whereas it concerned a typical streptothrix. Then Rabe described his Cladothrix canis, a branching mycelial organism

somewhat similar to the one already described by Vachetta and Rivolta as Discomyces pleuriticus or Pleuromyces canis familiaris. Next Nocard isolated and studied the organism of farcin du boeuf; he regarded it as cladothrix because he was inclined to believe the branching as pseudodichotomous, but Sauvageau and Radais later show that it concerns a genuine streptothrix. In the same year Macé still further confounded the confusion by describing under the name of Cladothrix dichotoma Cohn an organism which became better known under the later term of Streptothrix chromogena; Macé describes the branching correctly as due to the outgrowth of lateral buds, although be claims to have observed the formation of a septum at the base of the branch which thus became separated from the main axis. Macé also employed Cohn's original figures incorrectly.

In 1889 De Toni and Trevisan, who edited the part of Saccardo's Sylloge Fungorum relating to Schizomycetes, dropped the word cladothrix and in honor of Nocard's discovery of the organism of farcin du boeuf, they substituted the word nocardia for Streptothrix Cohn on the ground of the preexisting Streptothrix Corda. They included in nocardia the actinomyces of Harz and placed the group among the

bacteria.

In this year Eppinger described his Cladothrix asteroides isolated from a case of cerebral abscess

and meningitis—another streptothrix.

In 1890 Almqvist published some investigations comcerning "Certain bacteria with mycelia," describing cultures from three different sources. He recognized the branching of the unicellular threads as genuine; the formation of spores on air threads and the development of mycelium from the spores are described. Following Brefeld's system, Almqvist places the streptotrices, or microorganisms with true branches, midway between the moulds and the fissionfungi.

Then follows the description by Gasperini of a streptothrix, found in the air, under the name of Streptothrix försteri. Inasmuch as Cohn did not secure pure cultures of his Streptothrix försteri

Gasperini could have no basis for the establishment of the identity of these two forms and his organism subsequently became the Streptothrix alba, an apparently widely distributed organism described under various names by different observers. Gasperini expresses the opinion that these organisms belong among the hyphomycetes. Soon afterwards he describes another streptothrix from the air, the Streptothrix chromogena which is identical with Mace's Cladothrix dichotoma Cohn.

In 1891 Rossi-Doria described five new pigment-producing varieties from the air and, striking the road indicated by Cohn, he points out that many of the organisms heretofore described as cladotrices in reality are streptotrices and that the actinomyces of Harz is a streptothrix—the Streptothrix actinomyces; he accordingly arranges Nocard's and Eppinger's organisms in this division, renaming them Streptothrix farcinicus and Streptothrix eppingeri. The division forms a transition between the moulds and the bacteria

Sauvageau and Radais conclude their interesting and valuable study (1892) concerning the genera cladothrix, streptothrix and actinomyces as follows:

I. Cladothrix dichotoma is a bacterium; it possesses a sheath and has false ramifications.

2. Streptothrix of Cohn has no sheath; the conidia form by segmentation of simple or ramifying filaments, a little larger than the nonsegmenting mycelium.

3. The different species of streptothrix differ but little in microscopic characters, more in culture. All produce tufts in bouillon without clouding it.

4. The term streptothrix of Cohn should be dropped and this group should be included under Oospora Wallroth; they belong to hyphomycetes, genus mucedinae.

5. The actinomyces of Harz concords perfectly in all details with streptothrix of Cohn and should be called oospora bovis.

6. Diseases like farcin of Nocard, actinomycosis of Bollinger, pseudo-tuberculosis of Eppinger are not bacterial maladies, but are due to oospora.

Sauvageau and Radais dropped the name streptothrix for the same reason as did De Toni and Trevisan, namely its previous application to another fungus

by Corda.

In the same paper Sauvageau and Radais describe two new varieties: the Oospora metchnikovi and Oospora guignardi. The various morphologic and biologic characters of these two organisms are

thoroughly described.

Now follow in rapid succession the description by Hesse of Cladothrix liquefaciens; by Gruber of the Micromyces hofmanni; by E. Acosta and F. Grande Rossi of Cladothrix invulnerabilis; by Casabo of the Cladothrix rubra; by Vincent of Streptothrix madurae, the cause of the yellow variety of mycetoma; by Garten of Cladothrix liquefaciens No. 2; by Rullmann of Cladothrix odorifera; DuBois Saint-Séverin of Streptothrix aurea; by Kedzior of a thermophilic cladothrix; by G. Thiry of Cladothrix mordoré.

In the meantime Rossi-Doria, Gasperini and Terni in Italy were describing several varieties of similar organisms under the names of streptothrix or acti-

nomyces.

The indiscriminate use of the word cladothrix in the above list of names shows how deep-rooted the confusion of streptothrix with cladothrix had become.

Blanchard, in Bouchard's General Pathology, Vol. II, 1896, describes many of the pathogenic forms of these organisms under the term nocardia because this word had no special significance in regard to their botanical position, a question concerning which the conceptions were sure to be soon modified, as he thought.

ACTINOMYCES BOVIS ET HOMINIS.

In the foregoing, reference has repeatedly been made to the Actinomyces bovis and hominis, or the ray fungus of Harz. It is now in order to take up the consideration of the history of this organism and its relation to a transfer by a dead their

its relation to streptothrix and cladothrix.

As is now well known, Langenbeck, in 1845, was the first to see the actinomyces as it occurs in the lesions which it may produce. Lebert, in 1857, and Robin, in 1871, were the first to publish some of the details of certain cases, with more or less accurate

drawings of the as yet unrecognized organisms; Rivolta (1868 and 1875) and Perrocinto (1875) were the first to recognize the yellow granules in the affected tissues of cattle as parasites. Bollinger and Harz, in 1877, first used the term actinomycosis as the name of the disease this organism causes in cattle, Harz naming the parasite which he found in the masses, given him by Bollinger from a lumpy jaw, actinomyces bovis on account of its radiating appearance in the Then James Israel, in 1878, in an article entitled "Neue Beobachtungen auf dem Gebiete der Mykosen des Menschen," described several cases of multiple abscesses in man due to the same organism. He pointed out its similarity to Cohn's streptothrix, and this was confirmed by Cohn himself. Israel had no knowledge, however, of the previous observations referred to, except Langenbeck's, the notes of whose case and whose drawings were first published in Israel's paper. Finally, Ponfick, in 1880, showed that the bovine and human diseases were identical.

Harz did not succeed in cultivating the organism. He regarded it as a mould related to the genera Botrytis, or Monosporium. The peculiar grains or bodies in the actinomycotic lesions were held to constitute a single individual, composed of septate threads and a basal cell, which gave rise to dichotomously branching hyphae, ending in radially arranged knobs or clubs; the clubbed terminations Harz held to be conidia, which he noted sometimes contained small granules, cells,

and vacuoles.

Since then, a host of articles have appeared, bearing upon the morphology and biology of this organism. For the present, the interest is centered upon those which bear more directly upon its relation to cladothrix and streptothrix, and upon its position, concerning which there has been much controversy.

Oscar Israel, Boström, P. Ziegler, Affanassiew and Schulz, O. Bujwid, Protopopoff and Hammer, and others, described the organism as it occurred in pure

cultures.

Before long, it became apparent that similar clinical and anatomic processes are caused by different species of actinomyces, which differ in cultural and pathogenic properties, but only little or not at all morphologically. Thus the organism isolated and studied by Max Wolff and James Israel, differs so markedly from the ones studied by Boström and others, that Kruse definitively separates them into two species.

The formation of spores (segmentation) and the occurrence of degenerative fragmentation were recognized by Rutimeyer, McFadyean, and others, while the clubbed formations were variously interpreted, Babes and Bujwid regarding them as conidia, Boström, on the other hand, showing quite conclusively that they were due to a species of degeneration of the ex-

ternal membrane of the threads.

As stated, Harz regarded the actinomyces as a mould, and in this opinion Babes and Bujwid were inclined to agree, because they looked upon the clubs as representing conidia. Délepine also regarded it as a mould fungus, the clubs corresponding to asci, hence it belongs to the ascomycetes; family, pyrenomycetes. DeBary, James Israel, Affanassiew and Schulz, Boström, Wolff and Israel regarded actinomyces as a pleomorphous bacterium, or cladothrix, and this conclusion, strengthened by its appearances in the tissues, being recognized in the interior of grains or kernels as ramifying threads, as simple threads, as short rods, spirals, and cocci, found general acceptance among the authors of textbooks on pathology and bacteriology (Baumgarten, Flügge, Ziegler, Sternberg, De Bary, van Tieghem, and many others). Affanassiew and Schulz proposed the name of actinocladothrix, and Protopopoff and Hammer went so far as to make actinomyces synonymous with crenothrix, because they interpreted the branching appearances as due to a longitudinal division of the threads.

Israel, in 1878, and, later, Boström referred to the Streptothrix försteri as an organism, similar to the

actinomyces of Harz.

De Toni and Trevisan included actinomyces, together with other similar organisms usually described as forms of cladothrix, under the common name of nocardia.

Rossi-Doria, in 1891, placed the actinomyces among the streptotrices, which he thought belong midway between the moulds and the bacteria, and during 1892 there appeared a series of publications by Domec, Gasperini, and Sauvageau, and Radais, which contained corroborating observations which form the basis for a correct classification of the organism.

Domec describes the formation of spores and their development into threads as it could be followed in hanging-drop cultures. From the mode of branching, the appearance of the cultures upon acid and sugar media, from the limited resistance of the spores to heat and other peculiarities, he concludes that the actinomyces of Harz is a mould.

Gasperini also identified actinomyces as a streptothrix, and placed both among the moulds, and pointed out that the experiments with pure cultures indicate the existence of different varieties capable of produc-

ing actinomycosis.

Sauvageau and Radais also found that the actinomyces of Harz corresponds in all the principal morphologic and cultural details with other species of streptothrix, all of which they include under the oospora of the hyphomycetes.

It will thus be seen that, at the end of 1892, the actinomyces, or actinomycetes, were united with the streptotrices, and we are now in position to consider their place in the general system of classification.

Oospora, Streptothrix, Cladothrix, Actinomyces or Nocardia?

As was to be expected, the notion that actinomyces belong among the bacteria had to meet a hard and lingering death. F. Helme and Range, Birch-Hirschfeld, Migula and others still class it as a bacterium. Migula, who classes cladothrix, streptothrix and actinomyces as chlamydobacteria and applies the word streptothrix to unbranched mycelial forms (as do Helme and Range) has, however, according to Lachner, become convinced that the ray fungi belong to the moulds.

Contrary to the general belief still in vogue by virtue largely of tradition, Busgen in 1894 showed, as Winogradsky had already done, that Cladothrix dichotoma in reality is not pleomorphous and that it

should therefore no longer be held up as a model of an almost inexhaustible variability. For the purposes of multiplication, cylindrical cells separate from within the sheath of the pseudo-dichotomous threads or chains, develop cilia and swarm as conidia until they become stationary and grow out into new colonies. Neither cocci nor threads form stages in the development, and accidental changes in form are not to be considered as pleomorphous manifestations.

It is therefore quite evident that the continued misinterpretation of the relation between streptothrix, cladothrix and actinomyces, on the part of many pathologists and bacteriologists, depends largely upon a total disregard of the work of the investigators

cited.

In 1894 Gasperini reaches the conclusion that the organisms described as streptothrix, or which morphologically belongs to this type, should be classified taxonomically as actinomyces because the word streptothrix was used long ago by Corda to specify more complex and organized plant types. By actinomyces he would understand microorganisms, closely allied to bacteria, and which show a plain "dichotomy" and air-threads without special organs of fructification. The use of the words oospora, nocardia, cladothrix is not warranted by taxonomic rules. At the same time Gasperini describes 3 new varieties, the Actinomyces bovis albus, luteo-roseus, and citreus.

Terni, in describing the Actinomyces gruberi, also expresses his preference for the word actinomyces.

Eppinger believes that streptothrix, cladothrix and actinomyces belong to an order midway between hyphomycetes and schizomycetes—the streptotrices. He finds it difficult to give up the idea that the branching in organisms classed as cladothrix, such as actinomyces, is a false one, and claims that the segmentation of the threads and the development of the branches at the point of segmentation have not been sufficiently understood, and that here lies the difference between true and false branching, and the distinction between streptothrix and eladothrix, including actinomyces. In the latter two the study of hanging drops and of smear preparations, stained

with hematoxylin and eosin, will show, he insists, that the branches are not continuous with the cell protoplasm. In this view he has not been corroborated, but is contradicted by his own figures and by Sauvageau and Radais, Berestneff and others.

Kruse, Dammann and others arrange the actinomyces, streptothrix and cladothrix forms under the name streptotrices, which they regard, with Rossi-Doria, as occupying an intermediate place between

the moulds and the bacteria.

Blanchard discusses these organisms under the name of nocardia, introduced by De Toni and Trevisan; Déléarde, Neumann and Lehmann describe them as hyphomycetes under the name oospora, introduced by Sauvageau and Radais. In their large "Traité clinique de L'Actinomycose Humaine," Poncet and Bérard ascribe actinomycosis as due to a polymorphous fungus of the genus oospora or nocardia, designating the organism consistently as

Actinomyces bovis.

A. Fischer, professor of botany in Leipzig, in 1898, places the streptotrices among the true fungi, and denies that they really have anything in common with bacteria. This is an important statement because it is made by a botanist ex professione. The fructification by conidia, observed in "Streptothrix actinomyces," needs further study in comparison with that of other hyphomycetes, and Fischer does not fully believe that this, the most important streptothrix, completes its developmental cycle in artificial culture. He surmises that it grows as a mould upon grass and grain, through which the infection more frequently takes place. Behla expresses similar views.

Now the common morphologic characteristic of these plants is the branching unicellular mycelium; they multiply by acrogenous spores or conidia and are also reproduced from mycelial fragments. The clubshaped peripheral thickenings of certain actinomycetes, frequently mistaken for conidia or asci, are accidental formations under parasitic conditions and are practically absent in the artificial cultures as well as in some cases of actinomycosis. The confederation

of all streptotrices, or ray fungi, is therefore perfectly natural and, as stated, this may be regarded as fully accomplished in 1892. The question now is what shall this species be named and where classified?

In answering this question it seems that the arguments already set forth by Gasperini, Berestneff and especially Lachner are fully competent to satisfactorily

settle the matter.

As regards the name the principle of priority is clearly the determining factor. As already set forth, Cohn, in 1875, named one of these organisms streptothrix; in the course of years this was largely replaced by cladothrix; in 1889 De Toni and Trevisan introduced the term nocardia, and in 1892 Sauvageau and Radais placed all the organisms of this class under the oospora of Wallroth 1831, claiming that the organisms correspond best to this species of hyphomycetes. In 1877 Harz named the fungus of actinomycosis the actinomyces, and Gasperini, Terni and Berestneff have applied this name to the whole group. Many other names given individual varieties, such as Discomyces pleuriticus Rivolta 1884, Actinocladothrix Affanassiew 1888, etc., are, together with nocardia, so recent as compared with the others that they hardly require any further consideration at this time.

Cohn's streptothrix, the name given the first species discovered and which has been extensively used (Rossi-Doria, Kruse and others), would therefore be appropriate had it not already been introduced by Corda, in 1839, in order to designate an entirely different family of hyphomycetes, and, according to Saccardo, there are today four species with this name. As can be seen at once from Corda's original description, our fungi cannot possibly be classed with this

group:

"Streptothrix n. g.: flocci erecti, septato articulati, virgato ramosi, ramis ramulisque alternis, articulatis, spiraliter tortuosis sporis simplicibus, terminalibus apiculo suffultis, aut axillaribus sessibus hylo adfixis; nucleo furino; guttullarum olarum pleno; episporio crasso."

In modern mycology streptothrix in this sense would be one of the varieties of botrytis of the mould family ascomycetes.

It has already been made clear that it is utterly impossible to arrange the organisms in question under bacteria as a form of cladothrix. All botanists are agreed that the mode of branching of the streptotrices definitely exclude this possibility. As long ago as 1882 Zopf wrote that genuine branching is not possible for bacteria, and if he himself, as well as subsequent writers, had heeded this rule, much of the confusion which resulted would have been avoided. It may therefore be taken for granted that a plant, without chlorophyll, composed of a true branching mycelium with spore-bearing hyphæ is a true fungus and not a bacterium.

Thus there remain but two terms, namely oospora

and actinomyces, to choose from.

Sauvageau and Radais assigned streptothrix and actinomyces to the genus oospora of the mucedineae of the hyphomycetes largely because of the similarity in the verbal descriptions. Thus this section of the mucedineae are characterized by Saccardo in part as follows: "Conidia ex apice hypharum libere nascentia.—Conidia globosa, ellipsoides, suboblonga vel fusiformis," and in the description of oospora it reads: "Hyphae breves, subsimplices; condia globosa vel suboblonga."

Sauvageau and Radais note, however, that the streptotrices are distinguished from oospora by the narrowness of their filaments and the absence of

transverse septa.

Lachner shows that the only resemblance between oospora and the organisms in question consists in the production of chains of spores. The spores of the oospora are 20 times as large as the spores of the ray fungi, have a thick colorless wall with endosporium and exosporium as well as pigmented contents. The oospora are perfect giants as compared with the ray fungi and are formed of thick branching chains of rather short cells with double contured walls and a fatty and granular protoplasm. Such differences increase in importance when we consider that we are dealing simply with external appearances because a most important morphologic characteristic, namely the ascus-form, is absent and without this full identification is imposible.

Inasmuch as the fungi in question are not readily placed among any of the known hyphomycetes it becomes necessary for the present to consider them as a separate group, following the example of Rossi-Doria, Gasperini, Kruse, Lachner and others, and according to the principle of priority, to designate this group as actinomyces, as already carried out by Gasperini, Berestneff and Lachner. The term actinomyces has certain additional advantages over streptothrix: Though first introduced by Harz as descriptive of the radiating appearance of the actinomyces in the tissues, it is equally descriptive of the radiating arrangement of the threads of nearly all the actinomycetes as seen especially in pure cultures; and then it is particularly serviceable in the English language because so readily translatable into the equally significant and appropriate ray fungus.

Note.—Perroncito (Deutsche Zeitschr. f. Thiermed. und Veterinär-Pathologie, v, 35) states that Dr. Piretta declares that Mayen (Linnea, 1827, v, 433) used the word actinomyces to designate something else than here indicated, but I am not aware that the word has since been used in any other significance than ray fungus.

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