# REPORT

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By
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1925

EXFOLIATIVE DERMATITIS IN THE INDIAN RHINOCEROS (*Rhinoceros unicornis*), WITH DESCRIPTION OF A NEW YEAST SPECIES, PITYROSPORUM PACHYDERMATIS.

"Peggy," a nearly mature female specimen, estimated as being four years old, was received in the Garden in May, 1923. Nothing unusual was noted in respect to her hide until August, 1924. At this time it was noted that the skin was flaking off.

By September the condition had become so extensive as to be serious. The inflammation was worst over the back and flanks, but as it progressed there was no part of the body that escaped. Eventually, the skin became superficially fissured along lines that corresponded roughly to the normal more or less polygonal markings of the skin. After this it became detached and curled up along these lines; the central parts of the polygonal placque or scale remaining adherent for a considerable time. Eventually they would fall off, leaving a definitely congested surface. There was never any ulceration, pus formation or oozing of blood, although the color at freshly denuded places was rusty brown like that of skin into which diffuse hemorrhage had taken place. These were the most important features from the pathological and practical standpoint, but the most spectacular was the close and deep pitting which was seen when scales were freshly removed; the counterparts appearing on the underside of the detached scale in the form of closely placed hemispheres. The skin thus came to take on a honeycomb appearance. The pits were two or three millimeters in diameter, of about the same depth, and had smooth linings.

In the absence of any knowledge of the histology of normal rhinoceros skin, the explanation of these pits must remain tentative. Perhaps they correspond to the orifies of sabaceous glands which have become plugged with keratinous material; where this is the case, the plug may (in human cases at least) be so adherent to surface scale as to come away with it when removed. Or the pit may represent the summit of an especially exaggerated and acanthotic (and perhaps

FIG. 9.

FIG. 10.

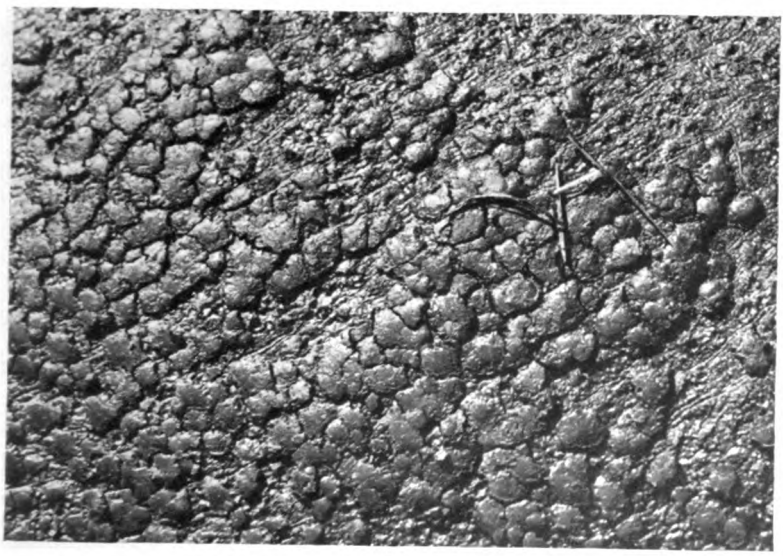


FIG. 11.

hyperkeratotic) interpapillary peg such as commonly develops in severe and chronic dermatitis and which may at times take on the form of the well known epithelial pearl. This likewise could be extracted from the deeper skin parts on removal of the more superficially lying scale.

The scales were largest over the back of the animal, say 2 or 3 cm. in diameter, and here, too, the pittings were most conspicuous. Elsewhere the scales were smaller, did not curl upwards so much at the edges, and remained adherent for a longer time.

The animal gave no indications of discomfort or constitutional disturbance. Her appetite and general behavior remained normal.

Laboratory Examinations. Direct examination of the scales (in 10% sodium hydrate solution). It was felt at first that the disease might be of animal parasite nature; this led to direct microscopic examination of the scales in sodium hydrate solution. In preparing the material, surprisingly fine lamellae were obtained by gently scratching the under side of a scale, for from the hide of a rhinoceros one would, *a priori*, expect to deal with a most tough and horny material. The latter was indeed the case on the immediate surface; but here on the under side of the scale the delicate flakiness must indicate that keratinization was not proceeding as normally, and that the cells were not becoming condensed and fused as normally. This could explain the detachment at this level.

No animal parasites were found in these scales under the microscope. Instead, there was an abundance of fungus elements. Two forms predominated.

The first was a minute yeast cell of very low type, close to the Bacteriaceae as will be seen from what follows. It measured 3 micra, was ovoid, structureless, contained no vacuoles or granules, and did not exhibit a double contour. Budding took place by constriction of one pole, during the course of which the cell would take on the form of an Indian club, beer bottle, or bowling pin. These cells were present, in places, in masses which extended over expansive areas. Individual cells could not be traced into connection with the mycelial filaments.

The mycelial filaments were likewise numerous, as mycelial filaments go in cutaneous material. They were broad, long, branched frequently, were hyaloid, did not show a double contour, and contained hyaloid arthrospores. The epidermal cells in the neighborhood of the larger mycelia were more faintly tinged than elsewhere, as though some of the blood pigmentary substances had been extracted in this position. Clusters of spores were present adjacent to this mycelium, but the grouping was not distinctive of Sporotrichum or other species.

## Cultures.

Altogether, three sets of cultures have been taken, the first during the height of the attack, the second toward its termination, and the third from a similarly affected animal at the New York Garden.

Set 1. In this set three blastomycetes species were isolated.

Species 1. This, which corresponded to the bottle-shaped cell that predominated in the scales, was only obtained once in culture, and then with much difficulty, because, being a slow grower, it was so speedily overrun by associated organisms. It did not make its appearance until after seven days. In twenty-five days it measured 8-10 mm. in diameter, and was highly heaped-up, extending fully 2 to 3 mm. above the level of the medium. The center was slightly acuminate—from it the sides sloped downward to a sharp, even, well-defined margin. It was dark

creamy yellow in color, and was thick, pasty, almost doughy in consistency. The surface was smooth, regular and even, and showed no trace of radial qualities or striations which might suggest that a mycelium was entering into its composition. It was drier than most cultures, lacking all moist and glistening qualities. The colony extended downward but a short distance (say 1 mm.) into the underlying medium. The qualities which were the most valuable for differential diagnostic purposes were the slow growth, dark yellow color, and the comparative dryness of the colony.

As seen in hanging drops mycelia never developed. The morphology of the cells was the same as described above in the scales, and again was found to take place by constriction of one pole. As the cells proliferated they tended to become arranged in short rows—something as the diphtheria bacillus does in culture; and eventually this led, in old preparations, to the accumulation of such cells into aggregates or clumps. Asci did not develop. This and the succeeding two species were tested in this respect by approved methods, *i. e.*, on beer wort agar and on Gorodkowa's medium, and after treatment by special staining methods.[1]

Species 2. This was a rapidly growing one, developing on glucose agar into a colony 2½ cm. in diameter in three weeks. By this time it was at its prime morphologically. It was fairly well mounded in the center, and had the moist glistening quality seen in many of the "wild yeasts." There were 30 to 40 incomplete radial furrows at the periphery of the colony: and delicate radial striations on the surface suggested the presence of mycelia. In the immediate center the colony was finely cerebriform or wrinkled. In color it was uniformly pale creamy yellow, and was semi-translucent. In the course of three or four weeks more the entire surface became similarly cerebriform and changed to a dirty brown color; at the same time the colony became more and more dry and homogeneous and took on a waxy character.

In fragments detached from old brown colonies, the mycelium was found to be broad, the segments sharply marked off, and their centers to contain numerous fine granules. Attached to the sides, or free in the surrounding fluid there were large spherical or subspherical yeast cells, some of which were budding, and from others of which mycelia were sprouting. Many had the "fish-eye" appearance exemplified in Monilia psilosis, *i. e.*, they contained the large, hyaloid, refractile nucleus of a faintly greenish-yellow color; other cells contained up to 5 or 6 small refractile granules, but no asci could be demonstrated,—even after employing the technique described for Species 1.

The cells in younger colonies appeared very different. In hanging drop colonies 5 days old, the mycelium was well developed, delicate, hyaloid and inclined to be varicose, *i. e.*, the side walls, which were sinuous, were not parallel to each other. Yeast cells were small, perfectly ellipsoidal and contained one or two very small vacuoles. The buds were at first perfectly spherical and attached to the parent cell by the most delicate sort of a neck. As it became older it also became ellipsoidal.

At first sight it is difficult to harmonize the extremes in morphology of these two forms of cells within the same genus. I have checked up on the matter of purity of culture, and can only explain the phenomenon on the basis of the marked variation that is known to occur among the yeasts, particularly when grown under the different circumstances that these were. Thus, the young forms were growing in glucose boullion in a hanging drop and the old ones on glucose agar

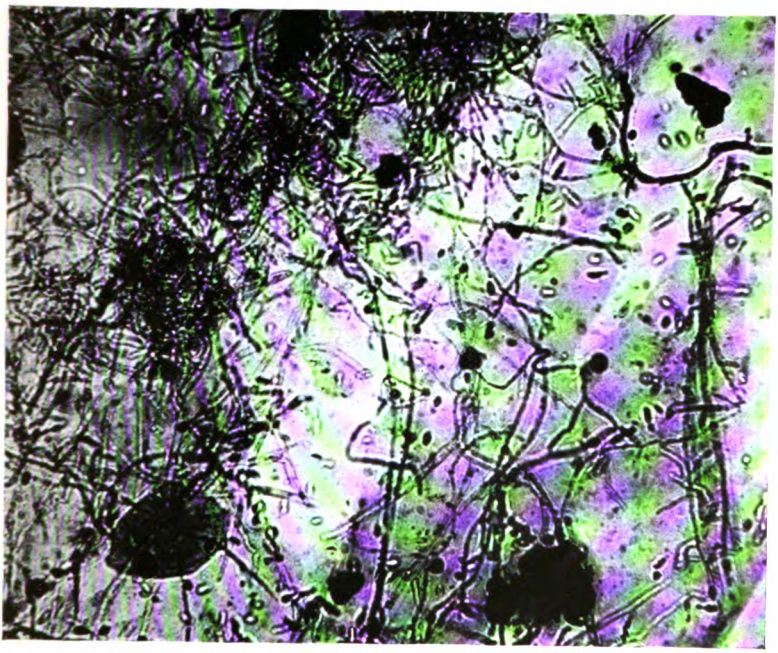


FIG. 12.

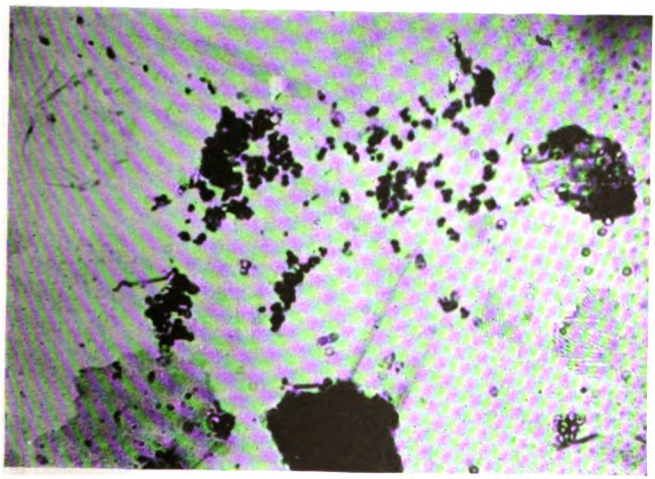
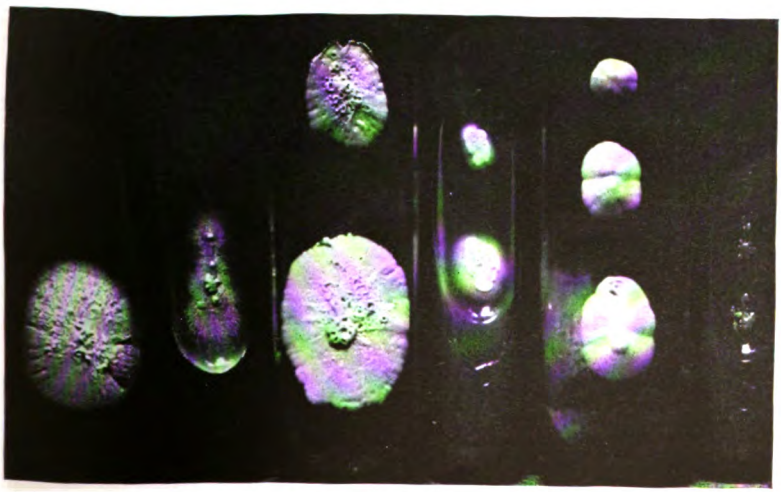


FIG. 13.

FIG. 14.



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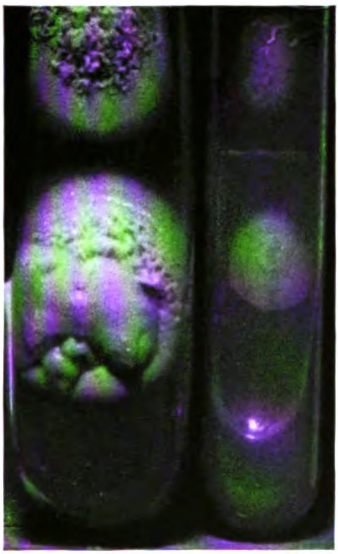

FIG. 16.

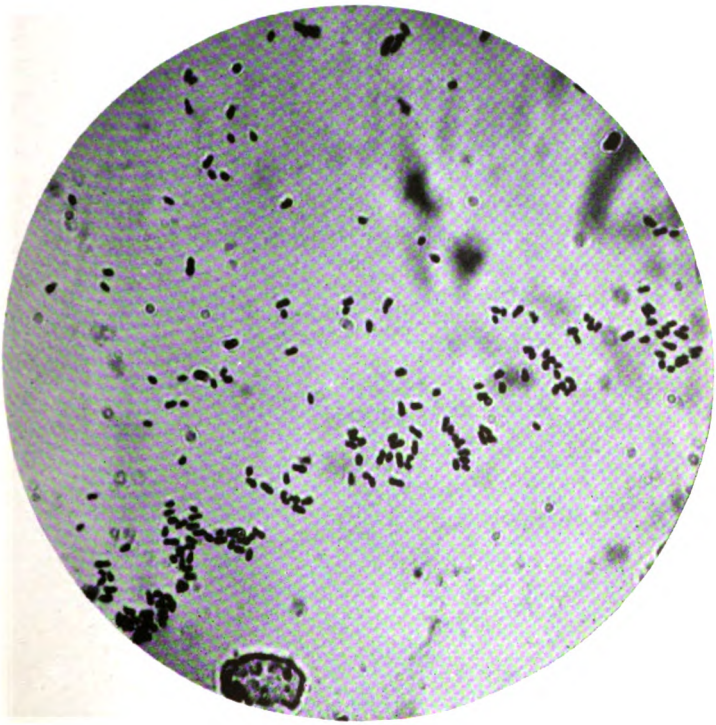

FIG. 17.

in a large test tube. One may well imagine the difference in aeration, moisture-supply, pabulum, etc., that obtained in the two containers, to say nothing of the factor of age.

Species 3. In 26 days this colony was 2 cm. in diameter, thin, flat, hugged the surface of the medium closely, was sharply marginated, and had a semi-translucent, pale yellow color. It did not have the thick, voluminous qualities of many "wild" yeasts. The surface was, in its most characteristic part, somewhat glistening. From time to time there were spread over it radial streaks or bands of a more luxuriant, pasty growth. There were no gross indications of the production of mycelium; the colony did not extend deeply into the underlying medium.

In the hanging drop the yeast cells were extremely small. They came to appear in two forms. First, a larger, spheroidal one which showed numerous buddings. Frequently three or four buds projected from one and the same parent cell. Occasionally one or two granules were present in the interior, but generally they were homogeneous and hyaloid. Vacuoles were rare. On search, examples were found where two short mycelia extended from the same parent cell from which spheroidal buds were projecting. This clinched the question of purity of our culture, and showed that this spheroidal form and the elongate one to be described next were of identical species.

The second form was a markedly elongated cell, chains of four or five came to resemble strings of sausage. In only one instance was a true lateral branch seen extending from one of these pseudo-mycelial arrangements. Frequent, and most distinctive, was the circumstance where two elongated buds projected from the same point on the parent cell. A double membrane was never seen on the "mycelium," and granules were extremely sparse in them. On the whole, this was a very primitive, hyaloid mycelium development indeed. Evidently mycelial formation took place in an irregular, splotchy fashion through different parts of the colony—this may explain the more luxuriant, pasty streaks and bands to which attention was called when describing the gross appearance of the colony.

In the culture attempts made during the subsidence of the disease none of the foregoing organisms were recovered. A parendomyces was obtained, but not seriously considered because no mycelia were found in the scales at this time.

*Identity of the above-described species.* I hesitate to name these organisms because the whole subject of yeast classification and species determination is so chaotic—particularly in respect to pathogenic forms; and when it comes to the yet more removed subject of wild animal mycology the usefulness of essaying it is most doubtful.

A short statement of the present situation *in re* classification will illustrate this, and prepare us for the details to follow.

At present there are three systems of fungus classification to be taken in account when dealing with animal parasitic yeast forms. The most widely known, that of Vuillemin,[2] has been modified by Hansen[3] and by Guilliermond *l. c.*,—none of these three authors was particularly interested in the animal pathogenic forms. They grouped all of the pathogens under the meaningless heading "Cryptococcus." Vuillemin's system has been followed by Castellani[4] and by Brumpt,[5] both of whom are parasitologists of the first order. Yet, in none of these is there provision for sufficient subdivision of the pathogenic forms; for, where so many are asporogenic they must fall into the very unsatisfactory genus Monilia. In order to cover the increasing

numbers of animal pathogenic forms, most of which are anascogenous and have not been studied as exhaustively as the ascogenous, and therefore were not provided for in systems extant, de Beurmann and Gougerot[6] erected the genera Atelosaccharomyces, Parendomyces and Zymonema. This was a useful service. Ota[7] has recently supplied an additional classification,—an appendix limited to the asporogenic *yeasts*, the essential departures of which are the construction of subgenera to accommodate certain of the cryptococci and torulae whose mycelia are arranged into tree-like branchings (Blastodendrion, new subgenus), or which send down "rootlet-like" mycelia in the depths of the medium (Mycelorrhizodes, new subgenus).

It will not be possible to enter into a detailed discussion of the affinities which the several rhinoceros species bear to all of the taxonomic groups above mentioned. For the present I believe it to be truly more scientific to describe the organisms and indicate their approximate systematic position than to align them into all niches possible.

Species 1. According to both Vuillemin and Ota's systems, this species with its utter absence of asci and even of mycelia formation would fall into the family Cryptococcaceae. In de Beurmann and Gougerot's system it would be an Atelosaccharomyces. It does not compare with any species which I can find described, the outstanding peculiarities being the small size of its cell and its permanence of form even in old cultures. In this respect it lies close to the bacteria, but is still removed from them by reason of the vacuole which it sometimes contains, and by the inequality of the two moities concerned in the transverse fission. It resembles Pityrosporum ovale Bizzozero, the reputed cause of dandruff, more than it does any other organism; differing from it, however, in the irregularity of form and size of the latter. Thus, P. ovale measures 3-18 micra as against 2 micra for the rhinoceros organism.

I believe that this is a new species, and propose the name *Pityrosporum pachydermatis* for this parasite of the rhinoceros.

Species 2. This would probably be classed in the heterogeneous genus Monilia under both Vuillemin and Ota's systems. By de Beurmann and Gougerot's system, Parendymyces would be more acceptable in view of the rather definitely formed mycelium which appears in old cultures; and the presence of at least large granules, if not arthrospores, with them. Grossly it resembles Hemispora rugosa, and to a less extent Oidium rotundatum, but not microscopically. While I am ready to believe that this is a new species, I cannot claim it in view of the faulty descriptions of already-named species. I would suggest, provisionally, Monilia ellipsoideus.

Species 3. According to Vuillemin's classification this organism would be classed as a Monilia, and probably also by Ota's. By de Beurmann and Gougerot, since the mycelium is not typical and since such typical yeast cell forms are present at the same time, this species might be considered as of the genus Zymonema. As it does not appear to be pathogenic, I have not accumulated sufficient data to justify naming it as a new species. I have carefully and sympathetically considered the possibility that these three forms are mutants, but am satisfied that Species 1 is quite apart from 2 and 3.

Culture Series 2. These cultures were made at a time when the animal had almost recovered, but although direct microscopic examination of the scales showed that both Species 1 and 2 were present, the former did not grow. Since salicylic acid ointment had been applied to the skin, traces of this antiseptic may have been sufficient to restrain the fastidious Species 1.

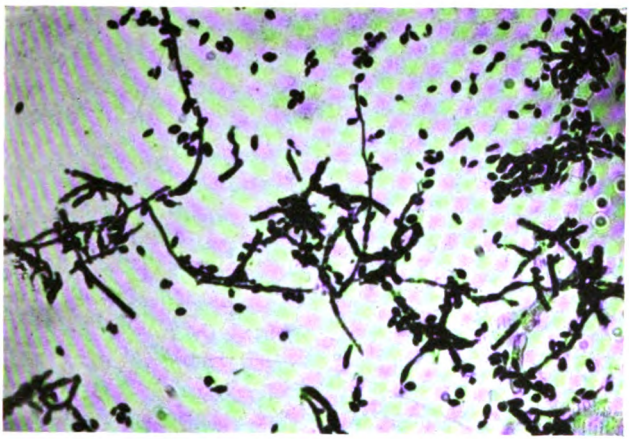

FIG. 18.

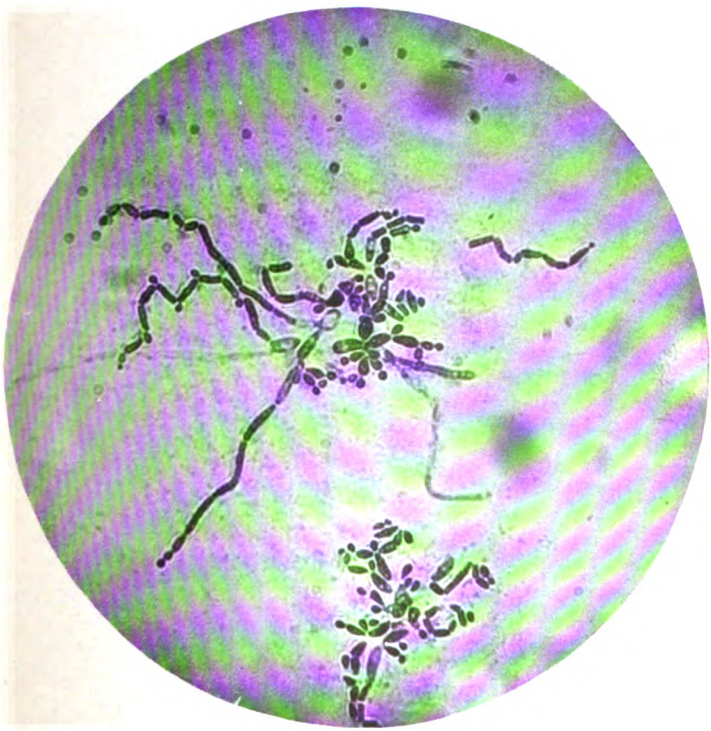

FIG. 19.

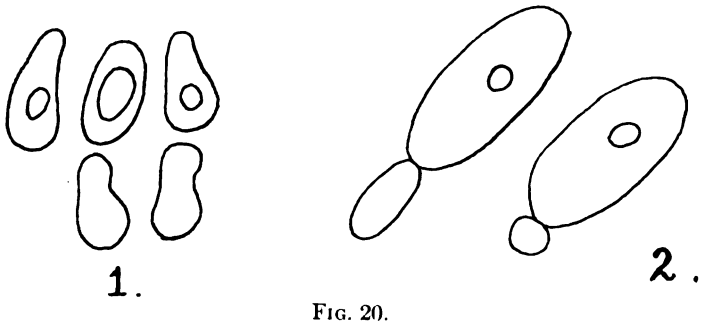


FIG. 20.

*Pathogenicity.* Taking the predominating organisms in the scales as the criterion, and in the absence of an appropriate animal in which to attempt to reproduce the disease, Species 1 and 2 are the ones to be suspected. Certainly the culture which I named Species 1 represents the minute budders seen in the scales, and, less certainly, Species 2 conforms in its mycelial characters to the mycelial forms seen in the scales. I could not find any organisms in the scales which resembled those in cultures of Species 3. As between Species 1 and 2, the odds are far away in favor of Species 1 as being the valid pathogen; it was present in large quantities in every scale examined on three occasions, *i. e.,* at the beginning and during subsidence of the outbreak in the Philadelphia rhinoceros, and on the one occasion when the New York rhinoceros was examined. Furthermore, it was difficult to culture on our artificial media, even though present in such great quantities in the material sown.

*Treatment.* Once the nature of the causative agent had been determined it was clear that salicylic acid—a standard ring-worm remedy—was the drug to be employed. This was applied in 1% strength in lard, anointing one-quarter of the animal's surface per day until all parts had been covered. At the end of a week it could be recognized that there was definite improvement, and this continued for about four weeks when it became stationary. A second course similar to the first was then given. Thereafter the skin progressively improved; in approximately two months it was quite normal and has remained so to date.

*Summary and Discussion.* Clinically, the disease corresponded to the "dermatitis exfoliativa" of human dermatology; *i. e.,* it was generalized, dry, exhibited marked exfoliation of scales and an inflam-

matory base. Being so generalized, there was nothing in its distribution to assist in relegating it to any of the more clearly defined entities; but after finding the yeast organisms a new face was put on the case. Now it became necessary to compare the disease with cutaneous "thrush" of human beings.

Doing so, we at once encounter the conflicts that arise in connection with a mongrel disease like thrush; one which may be caused, at least in the mouth, by no less than eighteen different species of fungi belonging to no less than six genera. Add to this the ridiculous difference in anatomy between rhinoceros and human skin and it must be realized that it will not profit us to compare it with thrush, and that we should advance no further than to consider this as an exfoliative dermatitis.

A further question that must be disposed of is that of normal exfoliation of the skin; whether shedding, for instance, may occur more or less periodically in the same way that the hair of some animals is shed. I have not been able to obtain convincing data on this point. That there is a certain amount of finer scaling the keepers seem to agree; likewise they agree that a periodic scaling to the extent seen in this dermatitic outbreak does not occur often, certainly not as a periodic affair. Beyond this they cannot help us, for naturally they cannot go further and pass on the matter of associated erythema; it cannot be expected that they should be able to discriminate between these cases of dermatitis with their subjacent erythema, and a physiological exfoliation which is free from erythema.

To come to the end of the story, I was at first inclined to believe that a marked exfoliation, unattended by inflammation, might occur as a normal, physiological process and that the yeast infection became superimposed; after the testimony of keepers, however, to the effect that marked exfoliation is

not common, I have concluded that the yeasts were the primary agents concerned in this dermatitis, and that it is not necessary to presuppose a preceding exfoliation.

THE NEW YORK RHINOCEROS. This animal was affected at the same time as the Philadelphia rhinoceros. While the disease was of the same type in the two animals, it was more severe in the New York one; thus, fissures and ulcers were present upon the legs, which symptoms the Philadelphia beast never showed at any time. In the scales from this animal the same kinds of yeast cells and mycelia were found as in the Philadelphia animal. Our Philadelphia Species 2 was cultivated from the New York scales, a circumstance which confirms the idea that it was one of the pathogens as well as Species 1.

Comparison with Dermatitis of African Rhinoceros (*Rhinoceros bicornis* 4256). The African animal died on March 28, 1917, with an acute pustular dermatitis and with acute changes in the internal organs, of which pseudolobar pnuemonia was the most important. The natural question in connection with our Indian rhinoceros is whether these two dermatites were the same. The keepers are of the opinion they were not, but this of course refers to appearances and not to cause or with reference to the course of the disease. Evidently, as will appear from the following abstract from the autopsy notes, the dermatitis of the African beast was the more outspoken of the two; suppuration was in the foreground.

During the winter the animal's hide was rough enough to arouse comment. On March 19, *i. e.*, only nine days before death, pustules appeared, particularly around the head, back of the ears, and to a less extent on the sides. These increased rapidly both in number and size, and in a week many had ruptured. Nine days after the appearance of pustules the animal died. At autopsy there were swellings on the skin 1-4 cm. in diameter and projecting above the general skin level for 5-15 mm. When incised, pus was found under the heavy covering of the skin, *i. e.*, comparatively deep. In some places there were ulcerations. Smears from the pus showed a short bipolar rod, and smears from the lung showed the same organism.

On analysis, it appears that the African beast had had its dermatosis for several months, and that the suppurative features were terminal ones; that is, that the disease which had lasted so many months became aggravated, was complicated by suppuration, and that this led to the animal's death—perhaps through a general septicemia as expressed in the pseudolobar pneumonia and other severely toxic internal changes. There were no yeast forms in the internal organs.

Treatment consisted in the application of neatsfoot oil, to which boric acid was added in the last week of the disease.

## The Female Genital Tract.

The following interesting conditions in the female genital tract have been observed:

Lioness (*Felis leo* 7617), fibromyoma of the uterus. The specimen consists of the genital tract which presents a large dense fibroid tumor, originating from the lower aspect of the uterine body and growing downward into the right broad ligament. The tumor consists of two nodules, the larger 8 cm. in diameter, the smaller 6 cm. The tumor is well encapsulated and richly vascularized. Microscopically the growth is a fibromyoma bearing a considerable amount of connective tissue scattered throughout the muscular bundles. There are several small fibroid nodules distributed about the uterine cornua. The right ovary is about twice the usual size and contains a follicular cyst.

Coypu (*Myocastor coypus* 7590), adenocarcinoma of uterus. At the upper pole of the right cornu is a mass grossly resembling a much enlarged placenta. No fetus was found. The other horn is empty and the ovaries are negative. Microscopically this growth consists of a markedly hyperplastic endo-

## DESCRIPTION OF ILLUSTRATIONS.

FIG. 1. The "Office."

FIG. 2. Specimen racks.

FIG. 3. A work corner.

FIG. 4. Arrangement of framed legends and card index describing pathological specimens on a given rack.

FIG. 5. Collection of normal organs.

FIG. 6. One of the card indices, showing scope of descriptions of individual specimens.

FIG. 7. Framed legends describing contents of racks in general.

FIG. 8. Intussusception of intestines of young African elephant "Mary."

FIG. 9. Head, showing adherent scales. The pittings were not as conspicuous here as on the back.

FIG. 10. Skin of back. Marked pitting. All scales have fallen off.

FIG. 11. Skin of back. Most of the scales have fallen off, leaving a pitted, honeycomb-like appearance between those that remain.

FIG. 12. Appearance of scales under microscope. Dense tangles of mycelia, with yeast cells in the meshes.

FIG. 13. Scales under microscope. Field selected to show what great numbers of *Pityrosporum pachydermatis* there were in the skin.

FIG. 14. Portion of scale under microscope. Both the mycelia at the left and the larger of the isolated cells at right represent elements of *Monilia ellipsoideus*.

FIG. 15. Cultures of the three fungi isolated from the Philadelphia rhinoceros. Larger tubes contain glucose agar, and the smaller ones plain peptone (conservation) agar; (both of Sabouraud's formula). First pair of tubes, *Monilia ellipsoideus;* second pair, undetermined Parendomyces; third pair, *Pityrosporum pachydermatis*. All are at their prime in respect to distinctive characteristics (4 to 6 weeks).

FIG. 16. Culture from the New York rhinoceros. Colonies 14 days old.

FIG. 17. Stained cells of *Pityrosporum pachydermatis* from a 4 weeks old culture. No mycelia ever developed.

FIG. 18. Stained cells of *Monilia ellipsoideus* in hanging drop (glucose boullion) culture.

FIG. 19. Young colony from margin of hanging drop culture (glucose boullion).

FIG. 20. Comparison as to form and size of yeast cells of *Pityrosporum pachydermatis* (1), and *Monilia ellipsoideus*.