

**Parasitological investigations upon vegetable organisms found in measles, typhus exanthematicus, typhus abdominalis, small-pox, kine-pock, sheep-pock, cholera, etc / by Ernst Hallier.**

**Contributors**

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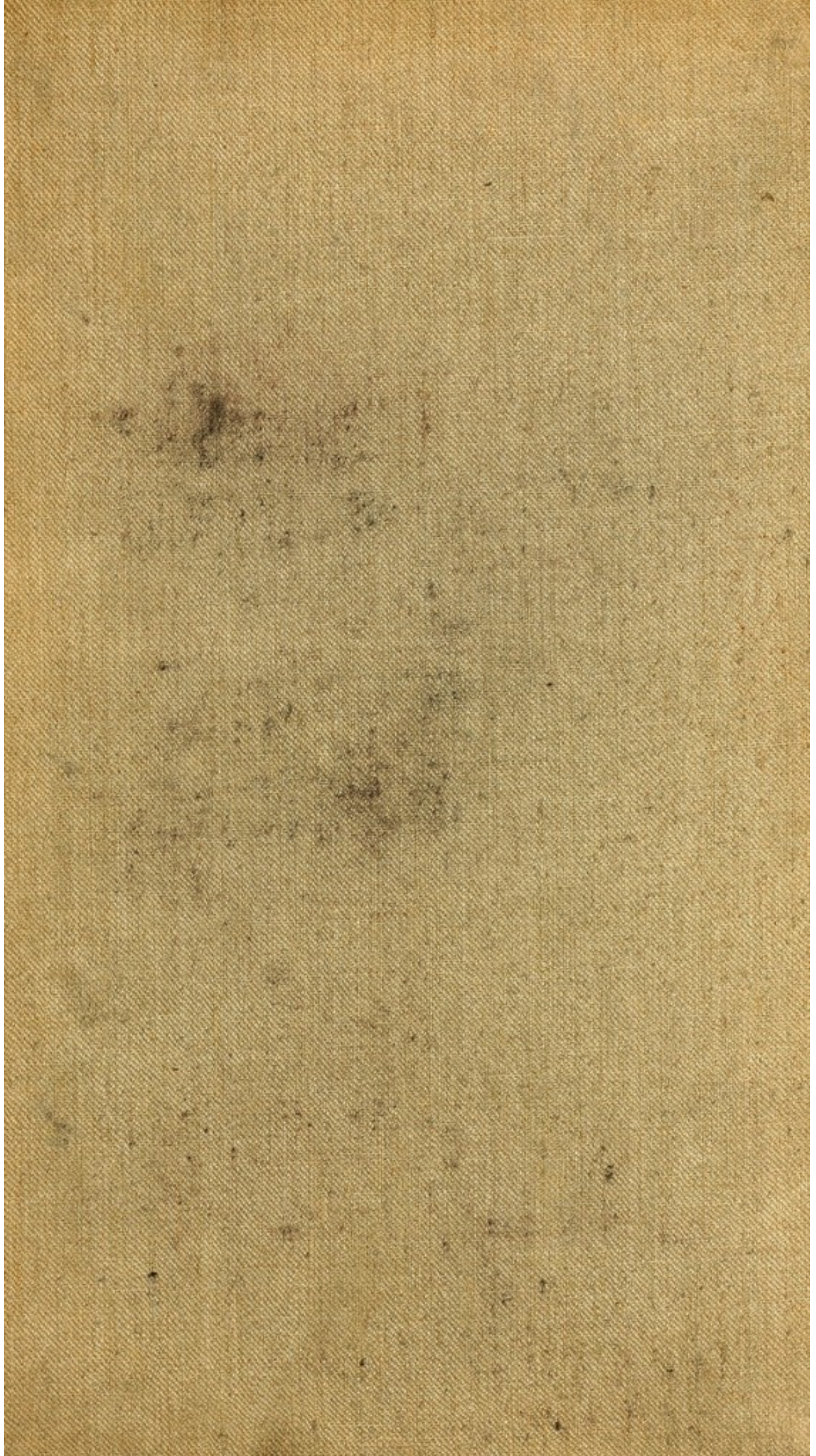
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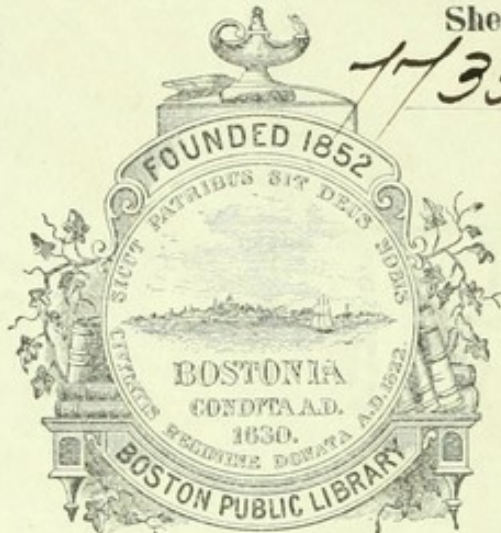


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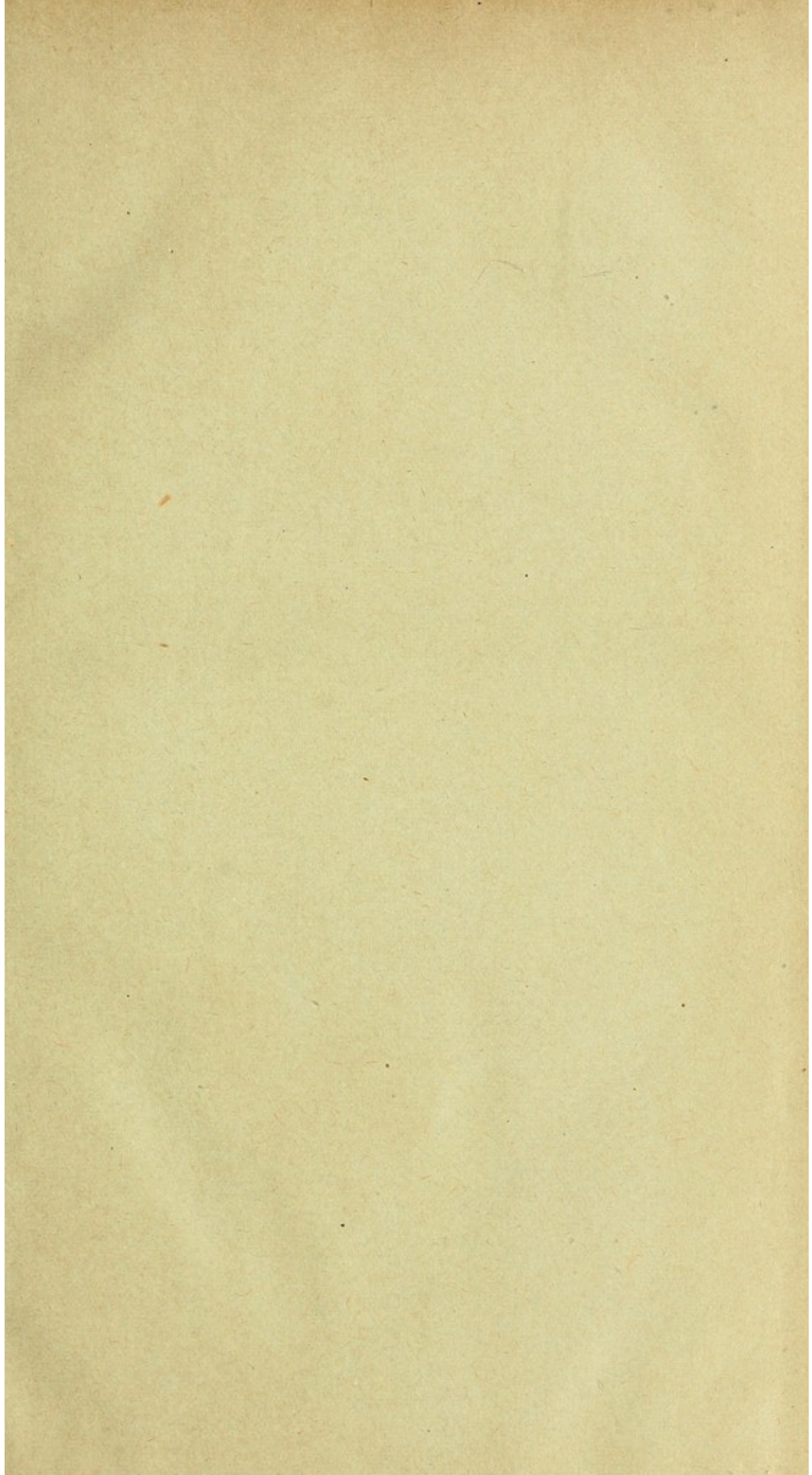
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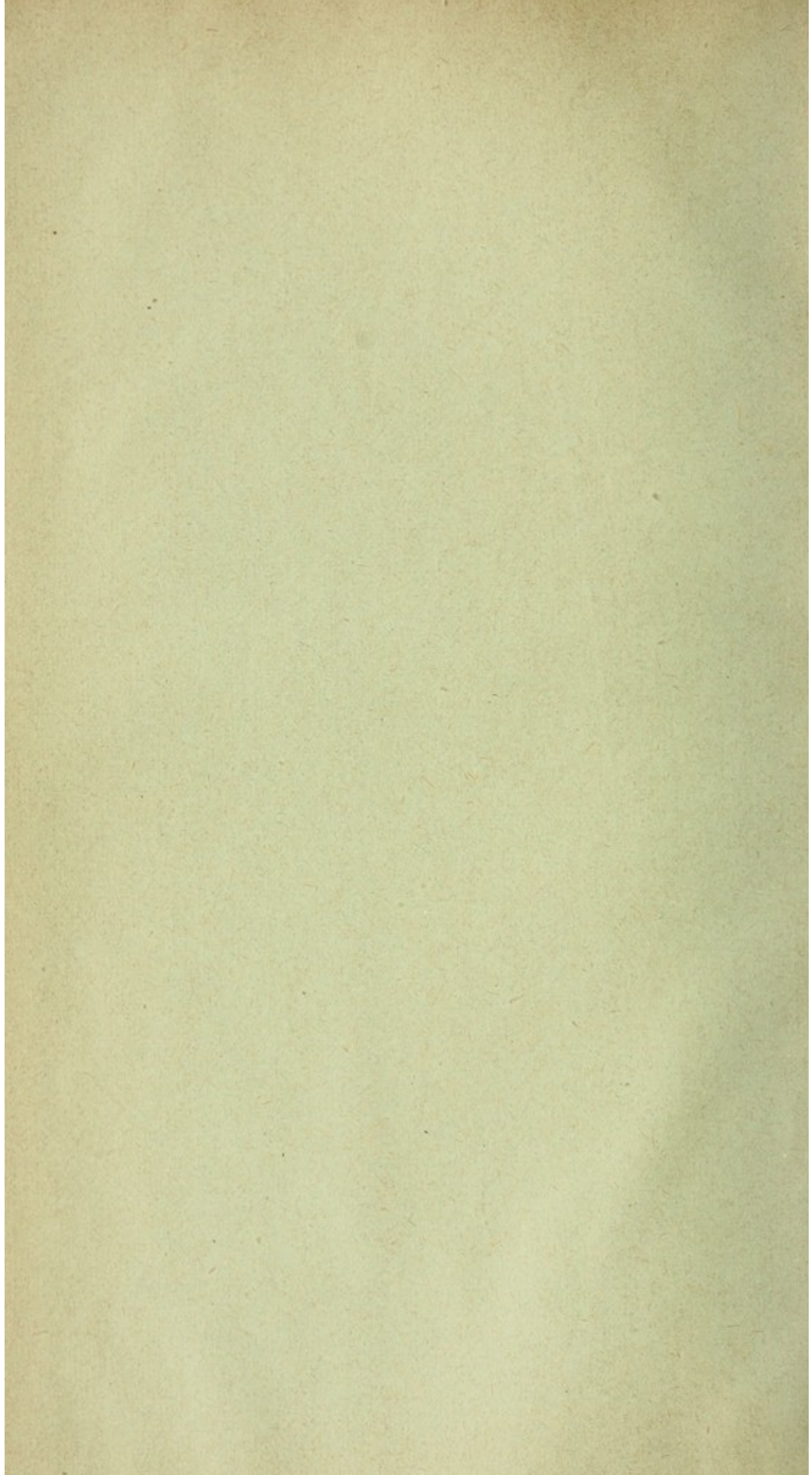


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FOUND IN

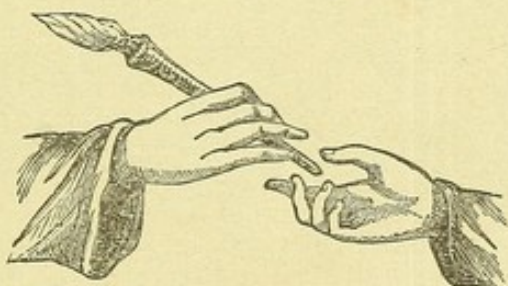
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MEASLES, TYPHUS EXANTHEMATICUS, TYPHUS ABDOMINALIS, SMALL-POX, KINE-POCK, SHEEP-POCK, CHOLERA, &c.

BY DR. ERNST HALLIER,  
PROFESSOR IN JENA.

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*Translated from the German,*  
WITH AN APPENDIX,  
By HENRY C. PERKINS, M.D.  
NEWBURYPORT.



ΑΑΗΛΙΑ ΕΧΟΝΤΕΣ ΔΙΑΔΟΣΟΥΣΙΝ ΑΛΛΗΛΟΙΣ.

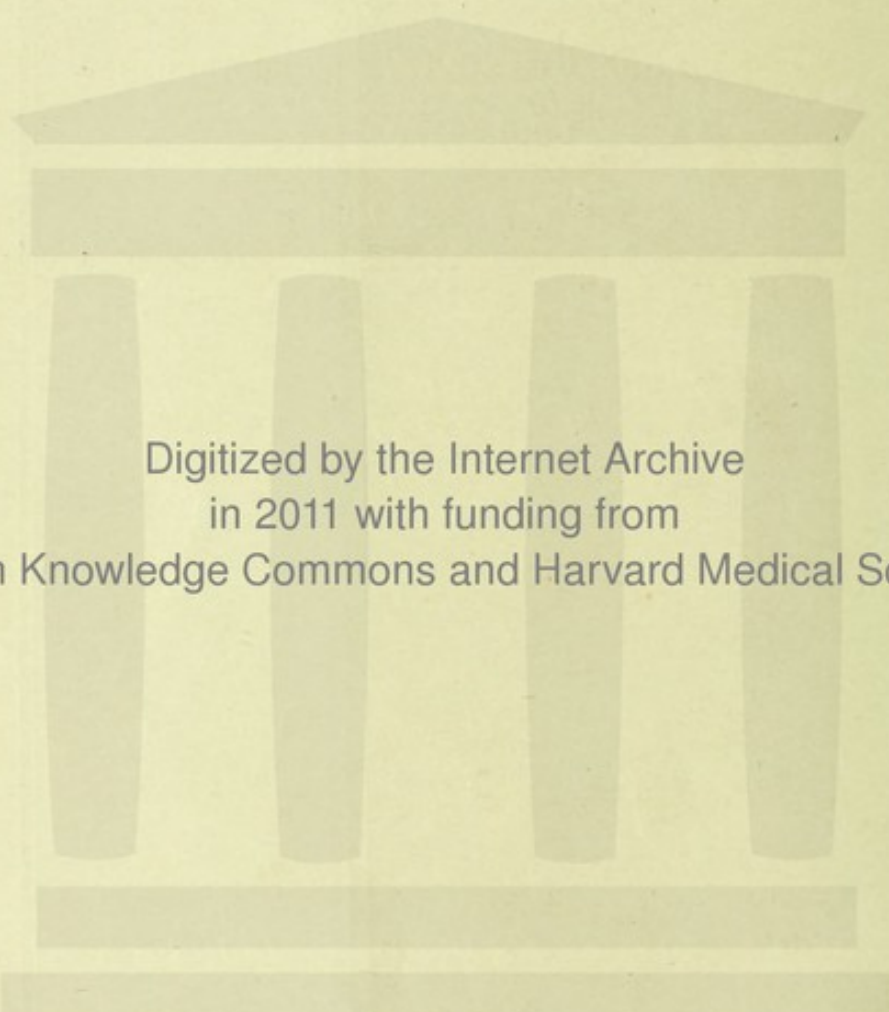
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PARASITOLOGICAL INVESTIGATIONS  
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TO THE FELLOWS,

OF THE

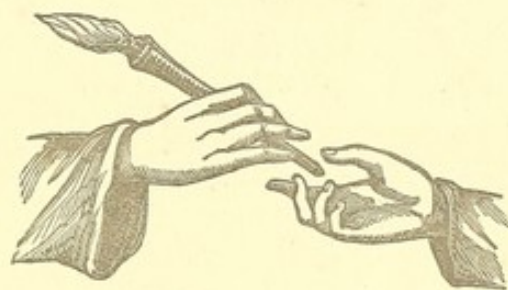
Massachusetts Medical Society,

THIS TRANSLATION OF PROF. HALLIER'S BOOK ON THE "GERMS" OF  
CERTAIN DISEASES,

Is respectfully submitted, by

A Fellow.

*Estate of Henry C. Perkins*  
*Aug. 2. 1895*



ΛΑΜΠΑΔΙΑ ΕΧΟΝΤΕΣ ΔΙΑΔΩΣΟΥΣΙΝ ΑΛΛΗΛΟΙΣ.





## INTRODUCTION,

BY THE TRANSLATOR.

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My attention was first directed to the subject of "Disease Germs" by a work of Dr. Beale's bearing this title. After its perusal, which left the matter still open, the work of Prof. Hallier on "The Vegetable Organisms found in Sheep-pock, Kine-pock, Small-pox, Measles, &c.," was kindly put into my hands by a friend, Mr. Carl Meinerth. It was in an unknown tongue; but the interest felt in the subject led to the study of so much of the German language as would enable me to understand its contents.

I had already for some months enjoyed the acquaintance of a young German student, Mr. CARL CASTELHUN, who had been educated at Heidelberg, and who had made the study of the lower vegetable organisms a pastime in connection with his professional studies, and who besides was familiar with the use of the microscope. His services therefore were solicited and cheerfully granted in this new investigation. To him I am indebted for the correct rendering of the text.

The microscope used for our purpose was got up by Mr. Edwin Bicknell, microscopist at the Zoölogical Museum, Cambridge. Its optical properties have proved satisfactory, its powers ranging from 35 to 2750 linear diameters.

Some liberties have been taken with the original in omitting the translation of certain parts which were of an historic and polemic nature, or which had no important bearing upon the subject. All else has been translated, and I have thought it advisable to add, from another work of Prof. Hallier, a



description of his Culture apparatus, that such as may feel disposed to repeat his cultures may profit by his experience.

The hypothesis that there was something invisible and intangible which occasioned contagious and miasmatic diseases, it is well known, had been started in earlier times; and until now all medical men have believed and declared, that the causes of certain diseases, as measles, scarlatina, &c., were conveyed through the medium of the air. This doctrine, restored by Hallier and others, not only brings the bodies above referred to before us, but assumes to define for each different form of zymotic disease, the micrococcus (or yeast) of a specified fungus as the miasm or contagion.

This revived hypothesis, the doctrine of ferment, has met with an exceedingly favorable reception in Germany, and wrought a change in the views of many distinguished observers and physicians.

It remains now to test its truth, not by speculative reasoning, but by experiment and observation. Under direction of the British Government the initiative has been taken, and it is to be hoped the example may be followed, if not by other nations, by individuals willing to aid in discovering the truth in so far as relates to other diseases incident to man and brute animals, and the plants upon which they feed, and which are used as medicines.

τ I would only add, that in order to understand my author's theory, his work on "Fermentation" was imported, and such parts studied as appeared necessary. Its translation must be left to some younger person.

## ABSTRACT OF PREFACE.

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IN this work a division and contribution of labor took place which I have so often desired, and which is more necessary for an epidemico-logical than for any other question in Pathology. The previous history of Parasitism and Epidemiology proves this difficulty, that no inquiry into these two subjects can be satisfactorily carried on from the medical side solely; the co-operation of Botany, Zoölogy and Chemistry is required. \* \* \* \*

Dr. Zürn undertook the pathologico-anatomical part of this investigation, and myself the botanical, &c. A constant mutual interest and control was kept up by daily discussion. The publications are kept apart, so that herein only the vegetable organisms and their effects appear. Dr. Zürn will hereafter report upon his part of the labor.

Dr. Reiter, of Munich, most obligingly and faithfully assisted me by furnishing the material of cow-pock and that taken from the human subject. What I could find in it is given in the following pages. Furthermore, I add the result of the most recent investigations upon the contagion of cholera, for the material of which, from five different cases of Cholera asiatica and one of Cholera nostras I thank Prof. J. Vogel, of Halle.

Whereas, in cholera the chief argument, that fungus and contagion are identical, is with great difficulty supported directly, because so few cases are offered for examination, and the nature of the disease itself presents obstacles—so there remains only the second way of proving indirectly, that always the same organisms accompany the cholera. This method I have pursued, and as will be seen at the end of the treatise, not without happy results. The dejections of seven different patients have yielded in my cultures precisely the same results, viz., that in the cysts (fruits) micrococcus is formed, which must be considered as the contagion.



The proof of this proposition was more necessary, as pathologists who labor without the aid of botany have recently again confounded the fruits of fungi with organic concretions. This is not to be wondered at, inasmuch as they are not provided with sufficient botanical knowledge, hence they pronounce the real fruits which they know only from illustrations or preparations, inorganic forms, just as the London Medical Society did twenty years ago. To a botanist well acquainted with fungi, such a mistake could not happen. An essential support and perhaps the most important of all for my view of cholera contagion, was afforded by my rice cultures with choleraic dejections, wherefrom it results, that, in fact, the cholera fungus produces a disease in the leaf of the rice plant, which reproduces the same micrococcus from cysts as appear in the dejections of patients sick with cholera. \* \* \* \*

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## PARASITOLOGICAL INVESTIGATIONS.

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### I.

#### SOLUTION OF THE PROBLEM AND THE METHOD OF INQUIRY.

It is already known that the assertion has been made that in the measles-fluid vegetable organisms are met with. Already some persons have asserted that the small cells found in the fluid of pock, together with the tender-jointed threads, are of a vegetable form. Nearest to solving the problem, was Dr. Bender of Camburg, on whose very ably executed investigations upon the vegetable organisms in vaccine lymph I have to report. Shortly before the discovery of the vegetable forms in sheep-pock and in vaccine lymph, by Dr. Zürn and myself, it was in a certain manner (so to speak) foretold by Dr. M. Popper, of Prague :—"that measles is a fermentative process ; that the cause of the fermentation is probably organized ; that near proximity of marsh-water favors the appearance of this disease ; that the poison from the marshes is introduced into the system through the lungs and eliminated by the skin ; and that it can cling to the body and be destroyed by heat." Gorup Besanez had compared the conditions under which albumen and gelatine furnish tyrosine and leucine, and those occurrences which take place at the formation of these combinations in the animal body, with fermentative processes ; but it was already known that

in the measles-pustule an albuminous stuff, leucine, fat and salts, particularly muriate of soda, were contained; that the blood of those sick with measles contains less sugar and urea than is usual; that in their urine may appear albumen, sulphuretted hydrogen and valerianic acid, and that the latter was from the decomposition products of leucine.

Schönbein, in the year 1865, had made the important discovery, that lymph in dilute hyperoxide of hydrogen causes the development of oxygen gas, and in the same way as platina resolves the hyperoxide into water and oxygen, as he had before shown with organized exciters of fermentation.

Coze and Veltz, by an investigation of the blood of those sick with measles and of that of living animals inoculated with measles-matter and measles-blood, have essentially supported the view of Schönbein, inasmuch as they found in their blood masses of quiet bacteria.

Finally Wertheim showed that here likewise the influence of the soil, under certain circumstances, appears as a favoring force, and that the height of the marsh-water has not less influence. This last statement, therefore, is of great importance, because from our investigations result the great influence of moisture upon the organisms coming here in question. Küchenmeister further shows, that the entrance of the pock-poison takes place through the lungs.

One of the most convincing facts, arguing the material nature of contagion, Prof. von Hessling has furnished, which I give in his own words:—"Some years since a man had the measles. According to the rules of the hospital, he was kept in a separate room, where he died. The bed and bedding were destroyed. Fourteen days after, the mason scraped and re-whitewashed the walls. The mason also sickened with the measles and died, while throughout the town there was not another case of measles."

There is, then, here, an important fact shown: that the



contagion of measles adhered to stationary bodies ; in this instance to the walls of the room.

The botanical questions here to be answered are therefore the following :—

First, whether in the sheep-pock, cow-pock, and small-pox, vegetable organisms always appear ; secondly, of what nature and descent are they ; thirdly, where do they naturally exist, and what action do they exert upon animal matter ? When these questions can be definitely answered — whence and in what manner men and animals are infected with pock-contagion — then the way and the manner of diffusion of the contagion in the animal body become subjects of inquiry, as also the production of the infection by the entrance of the microscopic organisms. This part of the labor, by far of the most practical importance, requires, if its solution should not be continued for years, rooms and capital, which a private individual, much less a professional man, cannot afford. Thence this part of the labor will be advanced very slowly, as long as Government are unwilling to furnish one hundred sheep, ten or twenty cows, and provide necessary rooms for the investigation ; an imputation which probably, through the uncommonly large practical consequences of this question, can be justified. A large number of animals for inoculation, and the introduction of the contagion by other methods, is for many reasons an indispensable condition for the success of the work ; then, likewise, when one would not at least disregard the essential fitness of the animal, as also that every one of these organisms coming in question belonged to some morphisms (generation and its forms), we should take advantage from each of such morphisms to experiment upon and raise the yeast-form constituting the contagion. But even the soil to be used will in no way be indifferent. The substratum can greatly influence the physiological nature of the yeast which is derived therefrom.

With a larger number of individuals for experiment, the probability of a quick result is very great; with a smaller number, much less.

The means, which for the solution of the first question necessarily constitute the botanical part of the problem, I have already made known in my work upon "Fermentation." I have now to remark upon some improvements in my apparatus and method of pursuit.

The culture and isolating apparatus which I have described and figured in my work on "Fermentation," are still used in my later investigations. The large isolating apparatus has answered, throughout, its purpose. The air-pump is to be preferred to the aspirator for various practical reasons. The aspirator requires distilled water for the renewal of the air. If we would be secure from soiling, a large quantity of distilled water is needed to keep the apparatus in order, because the water which has once been used is no longer to be regarded as pure. But it is impossible that the water in such an apparatus can be free from air and dust, so that it does not answer the purpose of the perfect isolation of the culture. I say nothing of the difficult management of the apparatus.

All these inconveniences are avoided by the use of the air-pump. One obtains by it a complete isolation, and has control of the conveyance of the air. At the conclusion of each culture, the air-pump must be perfectly cleansed, and proved before the commencement of a new culture. Culture in the isolating apparatus, besides, serves for control over the result obtained in the culture apparatus; hence it should be first opened at the conclusion of the labor: especially is it the best criterion of the exactness of the obtained results, if all the cultures agree.

At present, I usually provide the culture apparatus with a bell-glass, through whose air-tight open mouth a many times bent tube is passed and secured so as to effect a slow inter-



change of the air. The cleansing of all apparatus I have lately attended to with much greater caution. For instance, all glass apparatus, as bells, saucers, &c., after having been carefully washed with water, are rinsed in a solution of permanganate of potash, 10 grains to water  $\frac{3}{4}$  vi., then washed again with water, and finally rinsed in alcohol. The corks were submerged for half an hour at least in a solution of permanganate of potash. The water used in the culture apparatus was daily disinfected by means of the same solution, so that the culture might not be vitiated by the admittance of any spores of fungi. This method of proceeding has still the greater advantage, that with a dangerous contagion, as measles, for instance, it is not likely that the pestilence can be conveyed through the air by means of the fung-element. Besides, the air of the culture-rooms was daily disinfected by chlorine. For the benefit of the operator, a gargle of dilute alcohol was daily or oftener used, and the most scrupulous attention given to personal cleanliness.\*

Whoever attentively continues later investigations upon contagion, or studies the vegetable parasites on animals and man, or investigates the cause of a parasitic disease, will find that there are every where the smallest and most simple yeast-formings which I have called micrococcus, and which have more or less influence. Much more remarkable is it that in cholera, sheep-pock, kine-pock, the silk-worm disease, spleen-rot, yes, to all appearance in intermittent fever, typhus and measles, the micrococcus of a well-defined fungus (or alga) is diffused in diseased parts of the body.

Nearly two years since, I advanced (in the scientific appendage to the Leipsic newspaper) the hypothesis that all

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\* The chapter containing a description and figures of the culture and isolating apparatus, from Prof. Hallier's work on Fermentation, may be found in chapter xi.



contagion and miasm proceed from the micrococcus of algæ and fungi, and until now, this hypothesis has gained additional strength with each new step in the investigation. Likewise with the diseases of plants, it appears that very frequently the micrococcus is the penetrating and decomposing principle, and is the contagion. Especially were all decomposing rots, as the potato-rot, the turnip-rot, according to M. Willkomm, the rot of living and dead wood, introduced by means of the micrococcus of a certain fungus. Shall the micrococcus, when derived in great numbers from the human body, be wholly inert? Such a conclusion would have something very contradictory in it; much more is the supposition enforced by the mode of action of the yeast, that this, when it appears in large quantity, likewise produces a mighty change in the tissues and secretions of the body. Thus should one adopt the opinion that these micrococcus cells, always belonging to a certain fungus, and appearing in certain diseases and in certain organs in great number, are wholly adventitious and of no importance for the causation of the disease, he would do most atrocious violence to nature and sound reason.

Thus it was preliminary for all the following labors upon contagion, as the leading maxim, the postulate must come in practice, to find the micrococcus and to raise from it the fungus to which it belongs.

Perhaps algæ furnish contagion or probably miasm; this appears more probable since Cohn has shown that the genuine vibrios belong to the Oscillatorix, and that they contain phykochrome. What I earlier advanced upon bare conjecture, this has Cohn with remarkable acuteness and through exact inquiry shown—that with algæ as with fungi, an independent self-development of plasma-nuclei, that is, a micrococcus-forming takes place, which I have for a year observed, and which is evident from the labors of Cohn and

Itzigsohn. Thus the algæ have yeast-forming analogous to the fungi, and it is possible that these may become destructive to human beings.

In whatever manner one may reflect upon contagion and miasm, this is certain, that if contagion and miasm are of vegetable nature, they can be nothing else than micrococcus. The evidence thereupon lies simply herein, that the micrococcus is the only vegetable structure which can pass through the most delicate capillaries. Even the smallest spores of algæ and fungi are much too large thereto; but from my admeasurement of the micrococcus of various fungi it is evident, that these small bodies of some fungi have a diameter much less than that of the most delicate vessels.

But I have further shown, that the micrococcus of certain fungi can be taken in through the lungs, can advance to the mammæ, while earlier I have shown that they occasionally appear in the blood of beasts and likewise of man.

This agrees throughout with Küchenmeister's statement, that the measles-poison is taken into the system through the lungs and can be thrown out by the skin.

I cannot close this paragraph without stating, that in the first half of this century clear-sighted men have again and again maintained the vegetable nature of contagion, more especially Prof. Frank X. von Gietl; while Dr. Cowdell in the autumn of 1849 first advanced the idea, that the cholera was produced by means of a small microscopic fungus, an opinion which was in the same year confirmed through direct observation by Budd, Swayne and Brittan. Dr. Gietl, from the year 1831, had repeatedly maintained the organic nature of cholera and typhus, and invited the microscopists to examine the dust and walls of the rooms where the sick were confined:—that infection was conveyed by material bodies, the observations upon which were to be made from all points



converging to this opinion. But are the diseases producing decomposition yeast-forming processes—they can only be introduced by means of micrococcus.

As all my studies upon vegetables appearing in contagious diseases are based upon my yeast-doctrine, it is evident, that these investigations as well as the earlier, are wholly unintelligible without an accurate knowledge of my writings upon vegetable parasites and yeast-formation. Whoever has not my yeast doctrine in mind, must take in hand both works and appropriate their contents. Whoever omits to do this, is not to be pitied for inability to understand me, or for lack of knowledge.

In reference to the nomenclature of yeast-forming some modifications have been recently introduced, of which I will give the most important, because heretofore they have appeared only in the journals.

First, I have designated, conformably to the proposition of the Professors Richter of Dresden, all those forms belonging to one fungus-species as morphisms. Generation I call the chief form, which is distinguished from the other generations by essentially varying spore-forming. Thus, for example, are *Mucor racemosus* Fres. and *Penicillium crustaceum* Fres. different generations; because the *Mucor* develops theca-spores, but the *Penicillium* acro-spores. But the varying forms in which the *Penicillium* appears, as for example, on the rice, Cladospore-like, as common mould in the form of normal *Penicillium*, &c., are only morphisms, not generations, because the spore-forming is here the same. So is it with the various forms of yeast, the micrococcus, cryptococcus and arthrocooccus; further, their transition form into aërophytic forms, as for example, the *Mycothrix* chains (*Leptothrix* auct.), whose remnants were so often called *Bacteria*, *Hormiscium*, *Oidium lactis*, the *Torula aceti*, &c. are regarded altogether as morphisms.



The *Leptothrix* chains, that is to say, the micrococcus, which under the influence of the air on the surface of a liquid remains connected in chains, I have, according to Itzigsohn's suggestion, called *Mycothrix*. I have been often reproached concerning the name *Leptothrix*, but very improperly; for first, I was not the originator of the new name in this sense, inasmuch as Remak and others had introduced it: secondly, I do not call it, as Algologists do, a species, but a morphism, which appears in very many, perhaps in all fungi. As a specific name *Leptothrix* must probably be dropped, because this Alga-species is mixed up with the formations of various Algæ, not independent formations of different Algæ. But it is proper to give to the associated chain-forming micrococcus cells the name *Mycothrix*, to the analogous formations of an Alga the name *Leptothrix*. The genuine vibrios are plainly *Oscillariæ*, and the whole group of these consist of the yeast-like morphisms of the higher Algæ.

The mystery of the discovery of vegetable contagion consists herein, that one can raise from the micrococcus, the only vegetable form in the diseases of beasts and men produced by contagion and miasm, the higher generations and morphisms of the fungi.

This culture succeeds in a two-fold manner. Have we a strong soil whereon a certain fungus thrives, the micrococcus cells sprout in very delicate threads, which combine and strengthen themselves through multiplied anastomosis and soon develop fruit-hyphens. This furze we can call *Mycothrix* furze. It consists in a certain sense of very delicate *Sclerotium* formations. If the soil is very moist or liquid, these *Mycothrix*-chains have no fructifying furze, but every micrococcus cell swells slowly under the influence of a small draught of air, and sprouts, after it has increased from 10—20 diameters. These sprouting cells are of the form and sig-

nification of spores, and certainly of acro-spores ; I hence name them Sporoids.

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## II.

### VEGETABLE ORGANISMS IN SHEEP-POCK.

Early in October, 1867, we found, under a magnifying power of 500 diameters, in the sheep-pock matter taken, in small glass tubes and hermetically sealed, from various individuals who had the epidemic, whirling cells, clustering swarms, which at once were recognized as swarming micrococcus like those of fungi. Fig. 4 shows them as seen under the highest power of one of Zeis's microscopes. They were of a dark-brown color. I at once undertook their culture.

FIRST CULTURE. On sweetened water I put some of the sheep-pock matter from the glass tube, and exposed it in a culture apparatus to the room-temperature of Oct. 11, 1867. After some days there were found on the surface of the liquid micrococcus cells from two to six jointed, so that small Mycothrix chains, known as the Bacteria of the earlier nomenclature, were formed, as shown in Fig. 6. Numerous swarms of micrococcus cells were found at rest on the day after the sowing (about twenty hours after) ; many of them had formed chains. In each link of the chain I saw very distinctly a dark nucleus (Fig. 6.), probably for a new swarm. Such chains with a nucleus in each joint I found at different times in the lymph.

On the 16th of October, i. e. in six days, there were to be seen at the boundary of the liquid from swelling micrococcus, heaps of larger olive-colored, round, and at last lancet-shaped cells with one or more nuclei (Fig. 7), which on the following day had become larger and partly sprouted (Fig. 7), so as to exhibit sporoids. While these sporoids



were sprouting at the brink of the liquid, they had developed micrococcus under the surface of the liquid, so that on the 14th day after the sowing I found in the liquid numerous burst spores (Fig. 8), by the side of which lay a heap of quiescent micrococcus and other spores not yet empty, in whose plasma numerous cells appeared, instead of the original central nucleus (see Fig. 8).

At the brink of the fluid in the vessel sprouting sporoids were seen; at the end of each irregularly divided sprout were chains of spores (Fig. 9), at first blunt and broad lanceolate, at last globular. These sprouting Monilia-like bodies were at first pale or colorless, at last the spore became olive-colored, dark-brown, compact, like a Cladospore (Fig. 10), and the more so, as the lower links of the chain were becoming narrower and longer, like Monilia spores (in Fig. 10), and at last divided by septa (*cl*, Fig. 10). Some of the Monilia spores became very large, of a pale-brown color (*m*, Fig. 10), and closely resembled the well known Monilia cinerea of Bonorden. The developed Cladospore is not to be distinguished from Cladosporium herbarum auct.; however, I should hesitate to say that it was the shape or form which would enable me to identify it in the absence of other facts for its identification. Before many weeks had passed, upon the inner surface of the cork, by whose means the downward-bent glass tube was fastened into the receiver, a frail vegetation of Penicillium crust. Fr. was plainly seen, while at the same time in the liquid appeared sterile threads of the same fungus. Further off was found in the liquid, particularly at the bottom of the vessel, the mycelium of Cladospore, not a chain of spores, but single, large, yellowish spores not as yet forming sprouts, but mostly Tilletia spores forming micrococcus (see Fig. 14).

This culture was continued up to January, 1868, or for three or more months, and without the appearance of any other than the anticipated forms.



SECOND CULTURE. From a tube containing the lymph of sheep-pock a portion was blown upon the white of an egg, and placed in the culture-apparatus at the room-temperature of Oct. 18, 1867.

There was seen, on the day after, a change like that in the first culture, to wit, *Mycothrix* forms of the micrococcus cells which had come to rest, then appeared at the brink of the liquid in the vessel, later in the midst of them, a crop of sporoids (Fig. 12) out of the slowly swelling micrococcus. These sporoids were of a brownish color, and sprouted as soon as they had attained their full size (Fig. 12). Already early in November among the sporoids heaps of sprouts appeared, a part of which fructified in the form of very fine and strong specimens of *Cladosporium herbarum* Link. (Fig. 11); at the brink of the liquid, also, from which the fungus, by and by, spread out on the wall of the vessel. Amid the white of egg, at least close under the surface, the sprouts from the sporoids extended, but they formed large, nucleated, short, and at last globular joints and yellow spores (see Figs. 13, 25), which were single or connected in small chains at the ends of the branches (Fig. 13), forming by degrees a cross-barred or reticulated epispore, and by this means was formed a *Tilletia*-spore (Fig. 13). These cross-barred or reticulated spores are different from those of *Tilletia caries* Tul., in particular by their golden yellow color.

I would attach but little importance to the size. As we shall hereafter see, they belonged probably to *Tilletia lolii* Tul. These spores do not sprout on the white of egg, but they form micrococcus, which they leave after the irregular bursting of the epispore (Fig. 14). The micrococcus formation is essentially different from that of *Tilletia caries* Tul., but analogous. Out of the sprouts or branches of the sporoids come Cladospore-growths, which advance higher along the surface of the vessel and very soon produce, be-

side the Cladospore-chains, the Sporidesmus-fruits which are sometimes single, sometimes in chains, already known through Tulasne. Whoever compares the figures with those of Tulasne will have no reason to question their identity. In this opinion they will be confirmed by those communications which I shall hereafter give.

The culture-apparatus remained unopened for several months. The consequence was, that the varied formed fruit of Sporidesmium had dissolved their walls and had formed masses of black micrococcus. These were in such masses and so dark that the wall of the culture-apparatus had become very much obscured.

The following is in short the result of the second culture. From the micrococcus of sheep-pock sporoids formed, from whose sprouts came, in the air, Cladosporium-Sporidesmium plants, which appertained to Pleosporium herb. Tul. Within the substratum form male sprouts out of large globular joints, which, as is the case with *Ustilago carbo. Tul.*, are formed by the division of lyrate cells, and which probably belong to *Tilletia lolii Tul.*

THIRD CULTURE. On paste moistened with a solution of tartrate of ammonia, sheep-pock matter was blown from a tube and exposed in the culture-apparatus to the room temperature of Oct. 12, 1867.

On the first day after the planting, micrococcus, after resting, were extraordinarily multiplied, and formed a brownish layer on the paste. At the brink of the liquid in the culture-apparatus sporoids were to be seen, which brought forth Sporidesmium-plants, exactly as in the white-of-egg culture. Arising from the midst of the paste were single sprouts from sporoids with *Tilletia* fruit, but fewer in number than in the white-of-egg culture. These brought forth a Thecaspore-fungus, which could not be very closely defined, but reminded one of imperfectly fruited *Rhizopus*



nigricans Ehr. That this imperfectly developed fungus is really no other than imperfectly developed *Rhizopus*, the following culture demonstrates. The fungus commenced, as seen, from the sprouts of *Tilletia*, which were often covered with very tender epispores very similar to those of *Mucor racemosus* Fres., having the appearance of *Oidium* (see Figs. 27, 32), when the spores, or more truly *Macroconidia*, appeared in pale chains. But the *Macroconidia* of the *Mucor racemosus* are at first quadrilateral, oblong, broad-ovate, and at last globular. They were in the above named culture, as of *Rhizopus*, broad-oblong and blunt lancet-shaped, at last globular (Fig. 32).

At the brink of the liquid in the culture-vessel appeared, also, among the sporoids from *Cladosporium*, *Penicillium* crust. Fr. of the usual form. It is very probable that some of the micrococcus cells contained in the lymph or serum belonged to *Penicillium*; and, as I have shown, that now and then in the blood of healthy men and animals, as in milk and colostrum, micrococcus-cells belonging to *Penicillium* appear, so this might have happened in the sheep-pock and be nothing strange. These only accidental and exceptional micrococcus-cells of *Penicillium*, have in all probability no connection with the cause of this disease in the animals.

After the culture had been continued a week longer, there appeared at the boundary or edge of the paste, numerous purely vegetative mycelium-threads of *Cladospore*, which had long, continuous joints (Fig. 22); each of these joints had a long row of shining nuclei.

Upon the dryer surface of the vessel were to be seen the same threads; here, however, very slender (Figs. 23-24), of a pale purple color, and with strangely coiling branches (Fig. 23). At the spot, where such twisted branches terminated, was found a large, many-celled ball, at first reddish,



but later of a brown color. Thus there appeared to be here the forming of fruit, perhaps a genuine fructification. The final product, the result of the year's culture, unfortunately was not obtained, which is to be regretted, as nothing certain is known of the impregnation of Pleospore. It is possible that here is presented the first unfolding of Pycniden or Peridien. This last view, viz., that here the first condition of the real Pleospore-fruit is formed, may be regarded as very evident.

By a more excellent method of the remainder of this culture, the connection between *Cladosporium* and *Tilletia* was established; for at the edge of the substratum the genuine Cladospore-spores came out from one and the same mycelium-thread (Fig. 25), beside lyrate cells (*l*, Fig. 25), out of which the Macroconidia (*m*, Fig. 25), and further off within the substratum Macrospores (*Tilletia* spores) were found. High above the paste arose out of the micrococcus-cell, on the entirely dry surface of the vessel, no sporoids, but a crop of *Mycothrix furze*, from which very delicate Cladospore-plants sprung up.

FOURTH CULTURE. — On a slice of a peeled lemon was placed, on the 11th October, 1867, a quill charged with sheep-pock serum, which was put into a culture-apparatus. Eight days after the sowing, there was to be seen on the surface stout specimens of *Rhizopus nigricans* Ehrenb. (Fig. 28), and on the whole of the furze were arranged groups of a brownish-colored vegetation of *Cladosporium herbarum*. On the more moist parts of the fruit, there were at some places much larger spores of *Cladosporium*, which, as in the first culture, took the form of *Monilia*-spores, and indeed of the spores of *Monilia cinerea* Bon. (*m*, Fig. 27); in this manner they were passing over to Macroconidia. In the following week, some specimens of *Penicillium crustaceum* Fr. came to light, whose growth at first covered the cut-surface

and overrun the two other fungi. The culture was now set aside. For the rest, there appeared beside the *Penicillium*, in particular on the surface of the slice, still other *Penicillium* with colorless spores which I call *Penicillium grande*, an account of which will be found hereafter.

FIFTH CULTURE. — A small portion of sheep-pock matter, in particular from the gland-follicles, which was abundantly supplied with micrococcus, was, on the 20th of October, put into a culture-apparatus on paste prepared with a solution of tartrate of ammonia.

On the somewhat thin part of the paste *Cladosporium* was developed as in culture No. 3, besides, *Oidium lactis* and *Arthrocooccus lactis*. On opening the apparatus, the substratum had a very acid reaction.

SIXTH CULTURE. — On the same paste was put in a larger isolating apparatus (for the purpose of confirmation) some of the sheep-pock matter, and the apparatus left unopened until the 25th of December, during which time fresh filtered air was daily supplied by the air-pump. On opening the apparatus the result was the same as in the preceding culture.

SEVENTH CULTURE. — Starch paste, with some white of egg and a bit of sheep-pock matter, was put into a culture-apparatus on the 20th of October. The result, after fourteen days, was the same as in the preceding culture, with this difference, that some plants of *Penicillium crustaceum* Fr. of a normal development were obtained, in consequence probably of the more dry soil.

EIGHTH CULTURE. — On the first of November, on a slice of peeled pear, was put a quill charged with sheep-pock matter, and placed in the culture-apparatus. Fourteen days after, the cut surface of the pear was covered with a vegetation of *Monilia cinerea* Bon. (Figs. 27, 32), whose spores (*Macroconidia*) put forth vigorous specimens of *Rhizo-*



pus nigricans (Fig. 28). This culture was repeated because of its importance, and with exactly the same result.

NINTH CULTURE. — Some of the Cladosporium-sporidesmium-plants from culture No. 2 on the sliced pear, were put into the culture-apparatus. Fourteen days after the sowing the pear was covered with Rhizopus nigricans.

### *Results of these Cultures.*

From these nine cultures, made with the matter from various sheep affected with the epidemic, it appears to be clearly proved —

1. That among sheep-pock matter always appeared the micrococcus of Pleospora herbarum Tul.

2. That Pleospora stands in the same relation with Rhizopus nigricans, and with a Tilletia, Tilletia lolii Tul.

The questions now to be answered are—Whence came the Pleospora herbarum? and how were the sheep infected with this fungus?

The answer to the first question is very indefinite. Pleospora herbarum Tul. comes as a disease (Russ-brand) on the green parts of many plants; further, as I might show, on the wood of the vine, on the dry spots of the peel of the apple, pear and plum, and in particular of the lemon. I examined the fruit of the Pleospora and found it to be closely conformable with Tulasne's description. A more perfect knowledge of the generations and morphisms is desirable; for which I undertook the following cultures.

TENTH CULTURE. — On the first day of November, 1867, I sowed on the cut surface of a peeled pear some spores of Cladosporium herbarum Lk. from the peel of another pear. The sprouts were at the commencement like Cladosporium (Fig. 32), yet most of the spores were larger, broad, and blunt-lanceolate, toward the end of the chain globular

and often very large (Fig. 32, *m*), sometimes divided like Puccinia-spores (*p*, Fig. 32). The large spores (Macroconidia) produced vigorous specimens of *Rhizopus nigricans* Ehr. in eight days after sowing. Likewise the pale lanceolate *Monilia* spores sprouted, and brought forth *Penicillium grande* (*m*, Figs. 30, 33), which I have alluded to, with pale, large acrospores and oppositely branched pencils. This *Penicillium*, in a certain manner the unripe fruit of *Botrytis* (Fig. 36), sometimes resembled vigorous specimens of *Penicillium crustaceum* Fr., from which they are discriminated by the more opposite ramifications of the pencils, by the coarse-nucleated and shining plasma in the hyphen, and by the spores with the shining nuclei, and their distance from the including membrane. I did not see that this perhaps too rich culture produced any specimen of *Botrytis*.

ELEVENTH CULTURE. — On the third of November, 1867, *Monilia cinerea* Bon. was sown on the juicy peel of a sliced apple. On the ninth day after the sowing, a crowd of *Cladosporium herbarium* Lk. was found on the peel. On the cut-surface small specimens of *Rhizopus nigricans* Ehr. appeared. This *Rhizopus* arose (as more exact inquiry shows) from the sprouts of Macroconidia, i. e., from unripe *Tilletia* spores, which were not on the surface, but within the fruit-pulp. It is therefore here, as with *Penicillium-Tilletia-Mucor*, the Mucor-fruit (*Rhizopus*), a product of anaërophytic generation (*Tilletia*), either in a ripe state (Macrospore or *Tilletia*), or in an unripe condition (Macroconidia).

TWELFTH CULTURE. — On the third of November, 1867, on the cut surface of a succulent pear, *Monilia cinerea* Bon. taken from a plum was sown. The *Monilia* grew and multiplied rapidly; in a few days were seen the sprouts in the form of *Penicillium grande* (Figs. 32, 33), which at last became brownish, and took closely the form, mode of branching and fructification of *Botrytis elegans* Corda (Fig. 36).



The *Botrytis* is therefore a true acrospore-plant. It is seen very frequently in company with *Rhizopus*, and it stands to *Rhizopus* in a similar relation as *Sporodinia* to the *Syzygites* of Schacht, or rather as *Penicillium crust Fr.* to *Mucor racemosus Fres.*

I repeated the preceding culture many times, not always with success; frequently on too moist soil, the fungus appeared only in the unripe form of *Penicillium grande*. This fact was confirmed by every culture — that *Rhizopus* never appears on a surface which is exposed to the air, but that its first mycelium, as the product of *Macroconidia*-sprouts, issues forth from the inner part of the fruit-pulp; hence it was not often at the place of the sowing, but at some distance from it.

THIRTEENTH CULTURE. — On the seventh of November, 1867, the above-mentioned *Botrytis* was conveyed from the peel of a pear to a peeled slice of another pear. On the fourth day after came forth from the sprouts *Monilia cinerea Bon.*, in beautiful specimens. From this culture, however, no fruit pencil developed. We find *Botrytis* on the peel of fruit, but only in a particular moist condition, which is with difficulty arranged and kept for any length of time. The yeast-formation, which is with so much difficulty avoided, increased the amount of moisture in the soil, and by this means the culture was interrupted.

On several spots on the pear *Rhizopus* appeared, and the same thing happened in another culture.

FOURTEENTH CULTURE. — *Rhizopus nigricans Ehr.* was sown on paste moistened with tartrate of ammonia; the sprouts of *Rhizopus* (Fig. 34) formed interstitial *Macroconidia*, like those of *Mucor racemosus Fres.*, varying in color from a yellowish-red up to a rose-red, and mostly larger. I did not again succeed in raising, from the *Rhizopus*, *Botrytis* or *Pleospora*, because I did not succeed in forming the

necessary condition. Rhizopus was reproduced during a longer time, as indeed is the case with Mucor, when it falls on propitious soil. I had now to examine whether *Tilletia lolii* has any relation to *Pleospora herbarum*. In October I carefully examined numerous spikes of *Lolium perenne* L. I found the *Tilletia* already scattered in dust, but strangely all those flowers most plentifully attacked with *Pleospora*, which the sprouting *Tilletia* spores showed in greater numbers on the glumes, &c. This relation should be closely examined by the sowing of *Tilletia lolii* Tul.; as, however, the *Pleospora* forms an anaërophytic spore-form, which is readily distinguishable from *Tilletia lolii*, as further the *Tilletia* is almost always to be found in those *Lolium* plants, which are attacked by *Pleospora* at all periods of the shoots, which J. Kühn has so clearly described and figured, so at least it is very probable that the *Tilletia lolii* Tul. was the parent of the Pleospore; and so consequently that the *Lolium* plants had infected the sheep with *Pleospora*. I now sought *Pleospora herbarum* Tul. on the pear-slice used for cultivation, and had the pleasure to find always, after about eight days, that Rhizopus was reared, as if after the sowing of the fruit of *Pleospora*.

This result corresponds remarkably with the opinion of veterinary physicians as to the way in which sheep are infected with the pock. It is the general opinion of the most distinguished physicians and breeders of animals, that spoiled hay is the cause of pock-disease. As the *Pleospora* is known to reproduce itself as long as it finds a suitable soil, so it can be diffused from the *Lolium* plants to the grasses or other green parts of plants. Here, however, in Thuringia, I am sorry to say, are *Lolium*-borders not only allowed but even protected to the ruin of the corn-fields.

As I have already shown, the *Lolium*-borders supplied the fields with (Mutterkorn) smut, but supplied the sheep with



Pleospora. If the micrococcus is identical with the contagion of sheep-pock, so that we are permitted to draw therefrom the most practical inference, it is possible to succeed in preventing this disease. To understand what has been said, an accurate knowledge of the yeast-formation of Pleospora is indispensable. We have above seen that the micrococcus of sheep-pock largely multiply in nitrogenous soil (Fig. 18), and that they form sporoids on wet, and *Mycothrix* fungus on dry soil. On sowing the micrococcus of sheep-pock on the succulent slice of pear, also in glycerine, from the swollen micrococcus-cell first came *Cryptococcus* in globular sprouting cells (Fig. 19); on the liquid's becoming sour, *Arthro-coccus*, as Fig. 19 shows in a young state; in Fig. 20 it was in its ripe condition. Quite the same appearance takes place on the glumes of *Lolium*-grass, when it is covered with Pleospora and becomes wet. Figure 31 shows a small fragment with a vegetation of Pleospora of all possible forms. At *t*, empty *Tilletia* spores; at *p*, those of *Pycniden* lay around. As soon as the cell-texture becomes moist, the spores by degrees swell and discharge their contents as micrococcus, which always covered the glume in great numbers after moist weather (Fig. 31).

From these micrococcus we can very early raise, on cooked fruit-pulp, *cryptococcus* and *arthrococcus*. It must therefore be, that wet hay necessarily infects the atmosphere with micrococcus and Pleospore, and sheep which are exposed to moist hay provided with Pleospore, must inhale them. Thus they become diffused through the body and are thrown out by the skin.

A very important fact, in a pathological point of view, I must not omit to mention. During my labors I experienced, from the repeated opening of the apparatus, and the frequent sowings of the sheep-pock lymph, a severe bronchial catarrh with a painful cough. In the tough expectoration

I found large numbers of the brown-colored micrococcus, as they appeared in the sheep, which must have greatly multiplied on the mucous membrane. I cultivated them on paste moistened with a solution of tartrate of ammonia, and with pleasure saw within 14 days, from the sprouting micrococcus, that a vegetation of *Cladosporium*, &c. covered the paste, and afterwards *Monilia* in large numbers appeared within the paste, while *Tilletia* were developed.

From the entire labor it follows, therefore, with absolute certainty, that the micrococcus always appearing in sheep-pock matter belongs to *Pleospora herbarum* Tul. These results are of importance for the pathology of the disease. \* \* \* \*

To give all in a short resumé, the *Pleospora*, in the *Cladosporium* form, produces on soft nitrogenous soil *Macroconidia* and *Macrospore* (*Tilletia*), the first on more moist soil sprouts and generates *Rhizopus*; the second exhibits more durable spores, from which, after longer repose, *Rhizopus* is brought forth. On the surface there forms from the *Cladospore*-form, *Monilia*-*Botrytis*.

Thus we get the following parallelism :

- |                            |                                      |                             |
|----------------------------|--------------------------------------|-----------------------------|
| 1. Acrospores.             | 2. Thecaspores.                      | 3. Anaërophytic generation. |
| <i>Penicillium</i>         | <i>Mucor</i>                         | <i>Tilletia</i>             |
| crust. Fr.                 | <i>racemosus</i> Fres.               | <i>caries</i> Tul.          |
| 4. Generation descent.     | 5. Arthrospores and Schizosporangia. |                             |
| <i>Achlya</i> .            | <i>Cladosporium</i> and cysts.       |                             |
| <i>Fungus Ascophorus</i> . | <i>Cladosporium Sporidesmium</i> .   |                             |
| * * *                      | * * *                                | * * *                       |
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## III.

## VEGETABLE ORGANISMS IN VACCINE LYMPH.

For the material of these investigations I am indebted in part to the institution for vaccination at Hamburg, but mostly to the kindness of the central vaccinator, Dr. Reiter, of Munich.

There was found in the kine-pock lymph from both places large masses of micrococcus-cells and Mycothrix chains. The micrococcus is so very small, that under a magnifying power of 1000 linear it appeared still punctiform (Fig. 4). In the lymph I found the micrococcus mostly quiescent, but sometimes swarming.

The cultures gave the following results :

FIRST CULTURE. On paste made with phosphate of ammonia, I put, on the 16th Nov., 1867, the contents of a vaccine tube from the Munich institution. In the first hours after sowing an extraordinary number of micrococcus were to be seen. After twenty hours, the starch granules looked as shown at Fig. 44, *a*, *b*, *c*. The much swollen paste granules were seen with distinct cracks or fissures, which extended from the centre to the circumference. In these fissures (Fig. 44, *a*,) the micrococcus-cells were considerably multiplied, and crowding together in the less dense strata of the nucleus (Fig. 44, *b*, *c*), where they formed concentric zones.

On the following days the micrococcus-cells had swollen by degrees to sporoids, which sprouted (Fig. 45). These most delicate sprouts, at first by degrees becoming stouter, furrowed and perforated the starch body, which sieve-like, or more often carious, fell to pieces and dissolved entirely.

Meyen and Martius first intimated that the starch granules are perforated by small fungous threads ; later Schacht has succeeded in proving this. From observations on the

potato-rot I succeeded in showing that the fungus, as here, attacks the starch granule by the micrococcus advancing to the kernel (Korn) and growing into sporoids.

The sprouts of the sporoids represent moderately thick, irregularly branched fungous threads, which, after about fourteen days, brought forth pencils of *Penicillium crust. Fr.* Naturally these *Penicillia* are not from the micrococcus of kine-pock, but from such as had entered in with the breath and were blown through the tube. The *Penicillium* pencils, however, at first were not normal, but showed exactly those variations which I have previously described and figured as a bastard-form between *Penicillium* and *Aspergillus*. There is in fact the mould-covering, which settled on the sowing, of which we spoke, composed of *Penicillium*, *Aspergillus* and the bastard between them. As in all such cases, where the soil is moist, *Penicillium* very soon perfectly suppresses *Aspergillus*, so here also, the *Penicillium* suppressed the *Aspergillus* and the bastards in a short time.

Bail regards them only as the transition from *Penicillium* to *Aspergillus*. I have not surmised, but noticed this fact—that here a bastard production takes place. When it is seen that a typical specimen of *Penicillium* and a typical specimen of *Aspergillus* fuse, and the hyphens, sprouting from the united specimens, are seen bearing that kind of pencil that we can regard them as neither *Penicillium* or *Aspergillus*, then it is proved, that here is a bastard formation, not a transition. The transition of one fungus species into another has not as yet been observed by any one. From *Ustilago carbo Tul.*, *Aspergillus* can easily be raised, when a proper soil is selected, as it is easy, on a proper soil, for *Mucor racemosus* and *Penicillium crust. Fr.* to be reared from *Tilletia caries Tul.* But from no soil can one raise *Penicillium* from *Ustilago*, or from *Tilletia* an *Aspergillus*, so long as the culture remains pure. But as



soon as either fungus simultaneously appears, there is formed at the points of contact a fusion, and now come to light, immediately, bastard-pencils. Nowhere will one sooner be convinced of the exact limit of plant species than in the lower groups of fungi. To get, if possible, in the same culture other generations or morphisms, I put under the culture-apparatus a cork, because *Aspergillus*, as it is well known, loves a dry soil. The cork was well prepared, as in all similar cases, in the following manner: it was kept for half an hour in a solution of permanganate of potash; afterwards, for the same length of time, in alcohol, before being used in the culture. After about fourteen days there was to be seen on it a tender mould, which under the lens proved to be the smallest *Mucor* I had ever seen, by the naked eye scarcely to be seen at all. It is represented in Fig. 46, T. 1, as seen by the aid of a strong lens. The fruit bearers are mostly dichotomous, seldom single.

An *Oidium* fruit precedes *Mucor*, which could with difficulty be distinguished from *Torula rufescens* Fres. To this identity or diversity I shall return, and will here only remark, that in my earlier careful investigations of *Torula* I was reminded of the constant association of these fungi with *Aspergillus*. I have said already that the *Torula* was an *Oidium*; that *Oidium* belonging to *Mucor* shows at first small, globular, pale, reddish-brown conidia, very like those of *Torula rufescens*; these later became larger (*Macroconidia*), appearing as well interstitially as at the termination (Figs. 47, 48, T. 1, and Fig. 1, T. 2). I am not able to distinguish the interstitial conidia of the *Mucors*, which, as far as I know, was first done by Bail, from the terminating *Macroconidia*, which brings about the intermediate forming of *Acrospore* generations (*Penicillium*, *Aspergillus*, *Botrytis*), into the *Thecaspor*e generations (*Mucor*

racemosus, *Mucor mucedo*, *Rhizopus*). In all three cases there come forth from the germination of the interstitial Macroconidia, as well as from the terminating Macroconidia, Thecaspore-plants.

Bail observed the interstitial Macroconidia which he calls Gonidia, while others called them Gemmen in *Mucor racemosus*. With *Rhizopus* they are not less abundant and are mostly embedded in the soil. The Macroconidia of our *Mucor* on the cork form slender threads without septa, single or in chains, showing a highly remarkable peculiarity, viz., that of a Plasmodium to melt together when it finds others like itself. It is very tender, without a plain membrane, filled up with granular plasma, at least at spots (T. 2, Fig. 1, pl.). These form at such places larger or smaller swellings. These swellings are especially where two or more branches of shining threads melt together and connect laterally in Oidium-chains (Macroconidia-chains), which come to development in the thread. These lay together and are irregularly folded (T. 2, Fig. 1, v), in a slimy sac rich in plasma.

The fruit-bearers sprout from the cast-off as well as from the Macroconidia connected with the threads, as also from the above mentioned intumescence of the threads rich in plasma. The fruit is globular (T. 1, Fig. 48). Sometimes I saw in the bearer, enclosed Macroconidia (T. 1, Fig. 48, m). In the ripe fruit one does not see the basal cell (Columella), but after the casting off of the small, globular, colorless spores (T. 1, Fig. 48, sp), the basal cell appears as a round bladder (T. 1, Fig. 49), separated from its bearer by a septum, and on which some spores still are seen (T. 1, Fig. 49, sp).

I saw the tender thread of the Macroconidian-plant springing out of a very mis-shapen Sterigma of a bastard *Aspergillus* (T. 1, Fig. 47). The fruit-bearer, before the ripening of



the spores, is filled with plasma (T. 1, Fig. 48). After discharging them it is always empty (T. 1, Fig. 49), wherefrom it follows, that here also, the Columella, as with *Rhizopus*, comes to development as a real basal cell, at the time of the ripening of the spores. After discharging them the wall of the bearer shows plainly a striated appearance (T. 1, Fig. 49).

**SECOND CULTURE.** The contents of a vaccine tube from the institution at Munich, was put on a slice of peeled lemon and placed in the culture-apparatus on the 15th of November, 1867. Four days after the micrococcus had appeared in large numbers on the cut surface of the slice. Small sporoids on the dry part of the substratum (T. 1, Fig. 50) were sprouting (T. 1, Fig. 50, *k*), and at spots had already brought forth long threads, and surely bastards between *Aspergillus* and *Penicillium*. The *Penicillium* had increased so much at the end of the week that I thought it best to leave the culture and commence another.

Only on the white furze of the fruit, at some spots, *Aspergillus* on the first days remained nearly pure and exhibited, as always under such circumstances, the form of *Oidium albicans* Auct., i. e. Soor, or apthæ-fungi. The form, as I have repeatedly shown, is no other than the unripe Conidien-form of a Cladospore belonging to *Aspergillus-Ustilago*. Here also in the lemon was to be seen very early ripe Cladospore-spores, at first long, fusiform, divided by a septum, then shorter, one-celled and at last globular. I have so fully described and figured these Cladospores in my work on, "Parasites," that I would for convenience refer to that work, being content to delineate in Fig. 52, T. 1, some spores with their characteristic appendices. As of the greatest importance, I must not omit mentioning, that for some time Dr. Bender, of Camburg, experimented with, and raised from vaccine-lymph the *Oidium albicans* Auct. Thus he is the

first who discovered that in the lymph of kine-pock vegetable organisms constantly appear. When now, two inquirers, independently, and at different times and with different material, i. e. from different sources, experiment and come to the same conclusion, so is the security in the correctness of their judgment which but few labors can possess.

The threads of *Oidio-Cladosporium* crowded together deeper in the lemon; then there were to be seen *Ustilago*-spores, which never ripened, but were changed into large, globular *Macroconidia* (T. 1, Fig. 51). These strongly resemble what, in the above work concerning *Sclerotium*, I saw coming forth at the ends in the specimens of *Torula rufescens* Fr., and which I compared with the spores of a *Peronospora*. I called them "*Macroconidien*" on account of their resemblance to those forms from *Mucor*, and indeed, one would certainly, on comparing my Figs. 14, 15, 16, in the above work, conclude that they are no other than the *Macroconidia* of a *Mucor*.

On the sliced lemon had come forth those *Macroconidia* and perhaps a *Mucor*, which resembled *Mucor mucedo*, but it was somewhat stouter than that in the First culture.

THIRD CULTURE. Upon the white of an egg, on the third of November, 1867, the contents of a kine-pock tube from the Institution in Hamburg was placed in the culture-apparatus.

The micrococcus multiplied abundantly and put the albumen, as it were, into a violent fermentation. There were early developed very perfect specimens of *Oidium albicans* (Tab. 2, Fig. 2), whose spores ripened into *Cladosporium* (*cl*). The older spores, or more exactly, the short globular joints of the thread, had the appearance of *Ustilago*-spores, which immediately grew luxuriantly, as is the case in the culture with *Ustilago*.

Fermentation hindered the further development of



these Cladospores or any other morphisms. The culture was set aside for more than a month. Upon the coming of *Cladosporium* from micrococcus I have something more to add—that the forming of sporoids and their sprouting (T. 1, Fig. 53, *k*) is plainly to be followed.

Remarkable, and for a correct judgment of the origin of kine-pock, this fact is of the greatest importance, viz., that the white of the egg is colored of a beautiful wine-red by the reddish-brown micrococcus; they are somewhat lighter colored than the spores of *Torula rufescens* Fr.

FOURTH CULTURE. As I knew that the *Aspergillus* loved a dry soil, I put, on the 15th January, 1868, some kine-pock lymph from Munich, on a cork disinfected by the permanganate of potash and alcohol. On the 22d, there was to be seen on the cork, in the apparatus, a gorgeous typical specimen of *Aspergillus glaucus* Lk (T. 2, Fig. 3). The spore-heads were snow-white and became greenish on the following days. The white spores, when dry, showed their well known spicular surface (T. 2, Fig. 3, *sp*); in alcohol they seemed smooth and presented a large shining nucleus (T. 2, Fig. 3, *as p*). The fruit-bearer proceeds perpendicularly from the tender empty threads, and these mostly formed at their connection a horse-shoe shaped foot or root (T. 2, fig. 3, *s*). The bearing threads are here and there septate, but only at more delicate prolongations, mostly pretty far from the pencil-bearer. This stands in open communication with a part of the thread. Hence, Corda, when he described the bearer as much divided, must have had another plant in view. On the 6th of February, there was to be seen on the cork the first bright specimen of *Eurotium herbariorum*, which on the following day had developed to a larger mass. \* \* \* \*

FIFTH CULTURE. On the 6th of January, 1868, the contents of a vaccine-tube from Munich was put on milk

which had been boiled an hour. On the 20th of January, the cork bore a vegetation of *Aspergillus glaucus* Lk. and of bastards between *Aspergillus* and *Penicillium* (T. 1, Fig. 54). On the surface of the fluid appeared a rich *Arthrocooccus* vegetation, on which we can easily study the development of *Arthrocooccus lactis* from the micrococcus (T. 1, Fig. 55), and observe it in all its intermediate stages.

About four weeks after the sowing on the cork, began the fruitage of tender *Aspergillus* threads, and the development of brilliant typical specimens of *Eurotium herbariorum*.

SIXTH CULTURE. On the 23d of January, 1868, a small number of micrococcus on the white of an egg (from Culture No. 3) was placed on a disinfected cork, which lay in a porcelain vessel in the culture-apparatus.

Fourteen days after the apparatus was opened, micrococcus cells in all stages of sporoid-forming and sprouting were to be seen. The sporoids sprouted to beautiful typical specimens of *Torula rufescens* Fr. In this fully developed condition this fungus is no other than *Botrytis Jonesii*, from which already Itzigsohn and De Bary have shown, that it is a conidien-form of *Mucor mucedo* Fres. The spores, at the commencement, are broad-lanceolate, brownish-red, and arranged irregularly in chains. The mycelium is creeping.

Thus the plant here furnished a typical *Torula rufescens*, as one may find figured by myself in the Botanical paper (1866, T. vii.). Later, the hyphens raise and bear panicularly-arranged branches, with globular, mostly pale, but often fox-red spores, which Itzigsohn, in an autograph letter, has described. This is the *Botrytis Jonesii* of authors. The barren branch-ends, which one occasionally finds described and figured, probably rest upon an erroneous observation. I always found, upon careful management of my preparations, the branches covered with conidia, but they fall off easily, and thus are found some single barren



terminations. This same *Torula* bears, beside small conidia, much larger, globular *Macroconidia*. These come, as seen, with the conidia of *Botrytis*, and often at the same time and on the same thread with *Sporangiola* (*Ascophora elegans*, or *Thamnidium*), of Itzigsohn and De Bary; yet there is often seen merely large, single, globular *Macroconidea* at the end of the branches.

The *Macroconidea* sprouted and developed beautiful specimens of *Mucor mucedo* Fres. This fungus is wholly different in its form as well as its mode of living from *Mucor racemosus*.

While the *Macroconidea* of *Mucor racemosus* Fres. loves a wet nitrogenous soil, the *Mucor mucedo* Fres. develops on nearly dry soil. One gets on milk *Torula rufescens* Fr., but only on the perfectly dry milk, hence the cultivation requires some months. Further, the *Torula* and its highest form of development, the *Botrytis Jonesii*, love darkness, as Dr. Itzigsohn wrote me, and I find his opinion confirmed throughout. *Aspergillus* and *Eurotium* also thrive best in the dark. These facts are of the greatest interest in connection with the knowledge obtained in England, that small-pox heals in the dark without leaving scars. Mr. Bulmerincq agrees with me in the matter of fact, that sunlight in a short time renders the kine-pock lymph inefficacious. No *Mucor* is so variable as regards the size of the spores and sporangia as *Mucor mucedo* Fres. The largest specimens, from almost dry nitrogenous soil, equal in size the spores of *Rhizopus nigricans* Ehr. They show, also, a more tender form of mycelium, so that one might, without the lens, take *Mucor mucedo* in this form for *Rhizopus nigricans*, when he does not consider the kind of ramifications and the somewhat transparent, never wholly black and opaque condition of the sporangia.

As Itzigsohn and De Bary have correctly delineated, the

smaller sporangiola (*Thamnidium*), which are crowded on richly ramified branches, are one-celled, while the larger and even the smaller sporangia which are standing single at the ends of the longer branches possess, at least after the strewing of the spores, mostly a clavate protruding basal cell (*Columella*). Itzigsohn has described in his manuscript notice the beginning of the *Columella* as follows, which I can confirm.

"Through the hyphen a canal goes to the young sporangium, which is visible only when dry. Through this canal the plasma is always running to the young sporangium, for the forming of the spore-mass. When sufficient plasma is carried to the sporangium and thereby its figure becomes round, then the point of the hyphen becomes closed near the lumen of the sporangium by means of a vaulted wall, and by this means the *Columella* appears, which hence is seen only in the larger sporangium, while it is wanting in the smaller."

Therefore this occurrence is strictly the same, as I have already described concerning *Rhizopus*, only that with *Mucor mucedo* Fres. the basal membrane is in general earlier formed. The *Mucor mucedo* Fres. has sometimes, *Rhizopus* perhaps never, a proper basal cell, while I found this to be the case more frequently with *Mucor racemosus*. The genuine *Mucor mucedo* is readily to be known by the color and form of the spores. These, to wit, are never, as is always the case with *Mucor racemosus* Fres., truly globular, and colorless when seen single, but always oval, stretched out in length, and in a ripe condition always of a slate-grey or violet color. But these observations Itzigsohn has already made, whose *Mucor* studies I regret are not as yet published. Sometimes the color of the spores is of a splendid violet.

The color of the sporangia to the naked eye, as also under



the lens, is from a light grey-brown to a blackish-brown, always with an admixture of violet and often of a pure violet. The sporangium membrane, as some say, subsequently decays into a small granule, which neither Itzigsohn or myself have as yet noticed. Oftener, the sporangium-wall, irregularly torn in pieces, remains in the liquid; it may be torn off near the base, or the pieces still remain in connection with the base of the columella.

The ramification of the hyphen is very different. From the corymbose, richly ramified hyphen of the *Ascophora elegans* (Sporangiolen-form) up to the wholly branchless single *Mucor*-head, are to be seen all possible gradations, which depend on the quality and degree of moisture of the soil. On liquid soil, there form only sporangiola and very rich cymose and loosely ramified sporangia on tender web-like and prostrate threads; on the other hand, on strong and nearly dry soil, a single or slightly ramified hyphen, some millimetres high, appears.

The hyphen possesses one characteristic which never fails, viz., the appearance of transverse membranes. *Mucor racemosus* has generally only a few of these transverse septa in the mycelium-thread; never are they found in the hyphen-branch bearing a sporangium. Never in *Rhizopus*, does the transverse membrane appear in the fruit-bearing thread, very rarely in the mycelium. The fruit-bearing thread of *Mucor mucedo* Fres. very seldom is devoid of the membranes or septa, and for the most part these are very rich, as Fresenius and De Bary have already figured.

The Macroconidia of *Mucor racemosus* Fres., as I have often shown, are nearly always globular; those of *Mucor mucedo* Fres. are generally oblong, often fusiform. The interstitial macroconidia (Gemmen or Gonidia) are always quadrangular, as Itzigsohn has observed. They are mostly very numerous. No species of *Mucor* wants these forma-

tions, which have for every one a characteristic form. That they do not germinate is a mistake. They sprout very readily and bring forth new sporangia.

EIGHTH CULTURE. On the 27th of January, 1868, a small portion of the micrococcus from the sowing on the white of egg (No. 3) was put into the culture-apparatus on a sliced lemon. After 14 days, on the furze of the lemon *Cladosporium* (*Oidium albicans* auct.) appeared, among which, spores like those of *Ustilago* had formed confused chains. From these blackish chains the curious fruit, which I have figured (in T. 2, Fig. 7), appeared here and there, and which I maintain to be Pycniden of *Eurotium herbariorum*. Thus there are here, first of all, large globular-cells (T. 2, Fig. 7, *u u*), which are much divided by septa: frequently they are attached on long-celled threads, and are by these septa, *Sporidesmium* or *Stemphylium*-fruit (T. 2, Fig. 7, *s p t*). Sometimes there are in these cells kidney-shaped sporidia, without previous division, which I maintain to be analogous to the Pycniden of the nearly related *Erysibe*-species.

#### RESULT OF THE CULTURES WITH VACCINE LYMPH.

The Fungi-forms which come from the *Micrococcus* of kine-pock matter, by means of sporoid-forming and sprouting of sporoids, belong altogether to one species, and form the generations:

- |                                                 |                                  |
|-------------------------------------------------|----------------------------------|
| 1. Of Acrospore,                                | 2. Of Thecaspore, <i>Mucor</i>   |
| <i>Aspergillus glaucus</i> Lk.                  | <i>mucedo</i> Fr.                |
| 3. Anaërophytic spores,                         | 4. Fructification,               |
| <i>Ustilago carbo</i> Tul.                      | <i>Eurotium herbariorum</i> .    |
| 5. Pycniden, and its subordinate Morphisms:     |                                  |
| <i>Oidium lactis</i> .                          | <i>Torula rufescens</i> Fres.    |
| <i>Ascophora elegans</i> Corda.                 | <i>Botrytis Jonesii</i> Berkley. |
| <i>Oidium albicans</i> ( <i>Cladosporium</i> ). |                                  |



Compare with these the next related fungi-forms.

- |                                   |                             |
|-----------------------------------|-----------------------------|
| 1. Of Acrospores,                 |                             |
| <i>Penicillium crustaceum</i> Fr. |                             |
| <i>Botrytis elegans</i> Corda.    | 4. Fructification.          |
| 2. Of Thecaspores,                | <i>Achlya. prolifera.</i>   |
| <i>Mucor racemosus</i> Fres.      | Prings.                     |
| <i>Rhizopus nigricans</i> Eh.     | <i>Pleospora herb.</i> Tul. |
| 3. Anaërophytic spores,           | 5. Pycniden.                |
| <i>Tilletia caries</i> Tul.       |                             |
| <i>Tilletia (lolii)</i> Tul.).    |                             |

That the morphism shows also a resembling parallelism is evident.

But the question is, which generation or morphism furnishes the *Micrococcus* of kine-pock? First of all, one affirms, with some assurance, that *Mucor mucedo* and *Pycniden* have nothing to do with these *Micrococci*, because they do not spring up directly from them, not even on a favorable soil: nevertheless they readily appear sometimes as soon as the *Micrococcus* has vegetated on nitrogenous soil, and after that on dry soil (cork and lemon). *Aspergillus* and *Eurotium* hardly furnished kine-pock *Micrococcus*, for they form, in liquid, as I could often show for *Aspergillus*, very active *Micrococcus*, and closely, as with *Penicillium*, by means of the continued division of spore-nuclei and bursting of the spore-wall; but this *Micrococcus* is quite colorless, not from wine-red to reddish-brown color as that from kine-pock. *Torula rufescens* Fres. forms a reddish colored *Micrococcus*. I have for a long time shown this fact, but yet the matter would be better proved by new experiments; and therefore *Torula rufescens* was first sown on various liquids and then on boiled human excrement and boiled neat's-flesh.

The forming of *Micrococcus* was energetically going on upon nitrogenous soil. The discharge of the manifoldly divided nucleus is, as was above figured, concerning *Aspergillus*. The *Micrococcus* is deep-red, especially when seen

in mass. From the hard nitrogenous soil the *Torula* sprouted and developed *Macroconidia* and *Mucor mucedo* Fres. with sporangia, sporangiola and panicked *Botrytis*.

Further, I sowed *Ustilago carbo* Tul. on the same substances.

The *Micrococcus* I have already described, which have no resemblance with them in color, but are always dark brown.

On the flesh and faeces the sprouts formed *Torula* chains, afterwards *Macroconidia* and *Mucor mucedo* Fres. Thus it appears evident that the *Torula rufescens* Fres. furnishes the *Micrococcus* of kine-pock. Now it is of the greatest interest, that this fungus appears very abundantly in milk, perhaps always in colostrum. I have shown in the case of swine, that the *Micrococcus* in colostrum for the most part belongs to *Torula rufescens* Fres. Rarely if ever does primary kine-pock show itself upon oxen, and in cows it is mostly confined to the udder; hence the conclusion that the cow becomes infected from its own milk, which conclusion is strengthened by the fact, that the disease most frequently occurs upon the cow's calving.

It is now my duty to state, as I learned by correspondence with Dr. Bender, of Camburg, that this distinguished observer, in 1859, had seen the vegetable organisms in small-pox; that he arranged the first culture and that his experiment was rewarded with success.

He wrote me Nov. 3, 1867, as follows: "As in the year 1859 compulsive revaccination was ordered, the peculiar form which was observed, brought me to the view, that in and with small-pox matter there must be a vegetative process; and therefore I commenced microscopic observations of the lymph, which I had in abundance. Commonly I found, beside the usual morphologic elements, as epithelium-cells, flakes of filamentary stuff, little hairs, &c., fragments of sharply-defined hyaline threads of .009 m. in breadth and



of variable length; twice there were seen larger balls, in great numbers, in the matter of the small-pox pustule, which on the employment of a solution of potash, ether and ammonia, proved to be spores.

"A small drop of vaccine lymph with sweetened water was placed in warm air, which had been filtered through cotton; four days after, a multitude of quickly moving points appeared, whose motion was stopped by acetic acid. Fourteen days after, I succeeded in raising threads which greatly resembled the *Oidium* of aphthæ."

Dr. Bender, nine years ago, obtained a morphism of small-pox fungi by culture, and the entire conformity of results from experiments instituted at various times, certainly speaks well for their certainty and correctness. For Dr. Bender, from his cultures, obtained no other thing than *Cladosporium* belonging to *Aspergillus-Ustilago-Eurotium*, and *Mucor mucedo* Fres.

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#### IV.

##### VEGETABLE ORGANISMS IN HUMAN SMALL-POX.

For the material of this labor I am indebted to Dr. Reiter, of Munich. It was enclosed in a small tube and used for sowing as before.

FIRST CULTURE. A sowing upon the white of egg, in the culture-apparatus, was made on the 9th of December, 1867. In March, 1868, the *Micrococcus* was greatly multiplied. Higher fungi-forms did not develope. The white of egg, at first golden-yellow colored, became at last of a brownish-yellow.

SECOND CULTURE. On the 6th of January, 1868, the contents of a small tube was sown on boiled milk, and put

into the isolating apparatus. It was provided with a glass tube bent downwards, and the apparatus remained unopened until the 24th; when, on the surface of the milk, was found a large number of *Arthrocooccus lactis* (T. 2, Fig. 4), in all stages of development, the articulated joints mostly somewhat curved, resembling exactly those found in the vaccine lymph sown on milk (T. 1, Fig. 55). At the cork was found a beautiful vegetation of typical *Aspergillus glaucus* Lk. (Fig. 3, T. 2). Also coming from the small-pox matter on the cork, in the milk culture. Likewise a bastard between *Penicillium* and *Aspergillus* was developed, and thus (T. 2, Fig. 5) produced a spore-head of *Aspergillus*, which bears some branches, with chains.

*e* THIRD CULTURE. On the 8th of January, 1868, the contents of a small tube, on a peeled and sliced lemon, was put into the culture-apparatus. In the second week after the sowing there was to be seen, on the white part, a small furze of a typical *Cladosporium*, as is shown (in Fig. 6, T. 2) under a strong lens. There was here the same *Cladosporium* coming from *Oidium albicans*; but it did not remain so plainly as at the sowing of the kine-pock on the same soil. The *Ustilago*-spores developed, in part, imperfectly, as Fig. 2 shows, but in a larger part (T. 2, Fig. 7, *u*) they form a dense plasma and become independent. Chains of such cells form a dense fur (T. 2, Fig. 6), from which was produced very numerous branchlets, partly barren (T. 2, Fig. 7, *f*) partly bearing chains of *Cladosporium* (T. 2, Fig. 7, *c l*). Other analogous threads bear still other fruit-forms. These threads are either a little branched or not at all, but terminate in a single, large, at first globular cell (T. 2, Fig. 7, *p*). Frequently on the end of this cell there is another mitre-formed (T. 2, Fig. 7, *p m*). This cell has two different forms of development, to wit, more or less divided in one or two directions (T. 2, Fig. 7, *s p t*),



and becomes by that means *Sporidesmium-Stemphylium*-fruit, already known by me as *Aspergillus-Ustilago*. At some spots the terminating cell was not divided, but contained about eight free reniform spores, or rather sporidia (T. 2, Fig. 7, *sp, pp*), resembling those of *Erysibe* and *Pleospora*, as have become known through Tulasne.

These forms, which indeed were imperfectly developed with their twin-nucleated sporidia, resemble, in part, the Pycniden of *Erysibe*, particularly the very young Pycniden of *Erysibe Tuckeri* Berk: so much so, that I did not doubt concerning the close relation of the genera *Erysibe* and *Eurotium*, or hesitate to regard them as analogous. Possibly the Pycniden of *Eurotium* wants the enveloping cell-membrane, which distinguishes that of *Erysibe*, and still more that of *Pleospora*; but yet, possibly, it came only on this soil to imperfect development.

Thus it follows from these cultures, that the micrococcus of small-pox is derived from one and the same species as that of cow-pock, but that it comes from another Generation, viz., from *Stemphylium-Pycniden* plants. I did not succeed in oft-repeated cultures with kine-pock, by means of *Micrococcus* on lemon, to beget Pycniden plants. I saw standing out within the lemon reddish-yellow mycelium with cells containing drops of oil, without observing that any fruit had developed.

FOURTH CULTURE. On the 7th of January, 1867, (probably '68) there was put on starch-paste, prepared with acetate of ammonia, the contents of a small tube from Dr. Reiter, which was placed in the culture-apparatus. There appeared on the 27th of January, bastards between *Penicillium* and *Aspergillus*, and from the inner part of the substratum *Mucor* threads, which at the middle of February had not fructified.

FIFTH CULTURE. On the 7th of January, 1868, the

contents of a small tube was put on sweetened water, with phosphate of ammonia, and placed in the culture-apparatus.

There developed, at the brim of the culture-vessel, a rich vegetation of bastard *Penicillium-Aspergillus*. On the cork appeared, at the end of the third week from sowing, a fine typical *Aspergillus*, which at the middle of February covered the entire surface of the cork. Within the fluid there began at this time the forming of younger spores of *Ustilago carbo Tul*, while the fructification of *Eurotium* had commenced at the cork.

For verification, I put a slice of well disinfected cork across the mouth of the culture-vessel. Eight days after there were to be seen small white flocks on the surface of the cork. They originated from *Micrococcus*-cells which had fallen on the slice. These greatly multiplied and formed circular *Sclerotium*, like those seen on hair and which Dr. Beigel sent me, which heretofore were taken for *Algæ*. I have named it *Sclerotium Beigelianum*.

I have a *Sclerotium* from small-pox culture, marked Fig. 8, T. 2, as it appears in a young state. Later the cells were seen to be enlarged; those on the wall of the vessel germinated and produced a fine typical specimen of *Aspergillus-Eurotium*.

SIXTH CULTURE. <sup>was put</sup> On a well disinfected cork ~~was put~~ *micrococcus from* the white of egg on the 23d of January, 1868. Fourteen days after there was to be seen on the cork *Torula rufescens* Fres., beside the macroconidia of young specimens of *Mucor mucedo* Fres.

#### RESULT OF THE CULTURES WITH MICROCOCCUS OF SMALL-POX.

The result of the above cultures, nearly the same as that from the lymph of the vaccine disease, is remarkable, viz. :



that under the same circumstances, and from the same soil, the same generations or their morphisms appeared; i. e. on a dry disinfected plant soil, stand *Aspergillus* and *Eurotium*; on the fruit-peel of the lemon, *Cladosporium-Stemphylium*; on dry soil rich in nitrogen, *Torula rufescens* Fres. with macroconidia from which come *Mucor mucedo* Fres. But we cannot immediately, on the fruit rind of the lemon, get *Pycniden* from the *Micrococcus* of kine-pock; it is to be first cultivated on a nitrogenous soil (white of egg), and then transplanted on the lemon.

While the infection with the vaccine-fungus probably belongs to *Torula rufescens* Fres., which is to be considered as the younger progeny of *Ustilago carbo*, the *Micrococcus* so abundant in milk and even in colostrum, that of the small-pox probably proceeds from the *Micrococcus* which is developed from *Schizosporangia* (*Sporidesmium-Stemphylium*), and which always makes its appearance in company with *Pycniden*.

This result is therefore of the greatest practical importance, because the action of vaccination is perhaps thus best transmitted, when kine-pock and small-pox are derived from the same fungus; to guard against the small-pox is nothing else than to infect with the same disease, if the *Micrococcus* is the contagion. The great mystery of vaccination is thus explained—that one who has had the small-pox cannot have it a second time.

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## V.

## VEGETABLE ORGANISMS OF MEASLES.

For the material of this labor I am indebted to the kindness of Prof. Gerhardt and his assistant, Dr. Schneider. On February 3d, 1868, I received the sputa, and on the 14th the blood of those sick with measles.

Investigation showed in the sputa tender Micrococcus in abundance, and, among these, other spore-forms, resembling fungus-cells, some globular, some long, of the size of a Penicillium spore and larger. From what fungus the colorless spores and Micrococcus came, could not be determined. Fungus-cells and Micrococcus were always found in the sputa, though not always in so great number. Fresh blood contained similar Micrococcus cells, though not in great numbers.

By arranged cultures the following facts were obtained :

FIRST CULTURE.—Some of the sputa of a patient, sick with measles, on a peeled and divided lemon, on the 3d of February, was put into the culture-apparatus. There was evolved from the sprouting spores (later by the sprouting of sporoids), on the 7th of February, a vegetation of Penicillium crust. Fr. beside a Mucor-like fungus, which, however, was entirely suppressed by the Penicillium. The appearance of these fungi was not to be wondered at, but rather to be anticipated, because the Micrococcus and often also the spores were always to be found in the sputa.

SECOND CULTURE.—The sputa of measles with white of egg, on the 3d of February, was put into the culture-apparatus. On the 14th of February, at the edge of the white of egg, fine typical specimens of *Oidium albicans* Auct., developed to ripe Cladospores, from the Micrococcus swollen into sporoids, which sprouted and formed these Cladospores. To ascertain whether these Cladospores actually belonged



to *Aspergillus-Eurotium-Ustilago-Mucor*, &c., I placed on the same day a slice of peeled apple in the culture-vessel, and had the pleasure, on the 18th, to find a brilliant vegetation of *Mucor mucedo* Fres. and its preceding forms. e

**THIRD CULTURE.** On the 3d of February, some of the sputa on paste, prepared with phosphate of ammonia, was placed in the culture-apparatus. On the 10th the surface showed a vigorous vegetation of *Mucor mucedo* Fres.

**FOURTH CULTURE.** For verification the blood of one sick with measles, with paste and phosphate of ammonia, was put into a large isolating apparatus on the 4th of February. On the 10th I saw a vigorous vegetation of *Mucor mucedo* Fres. I remark here that not a trace of *Penicillium* or any other mould-fungus was to be seen until the opening of the apparatus, which, certainly, was very good evidence of the perfect closeness of the apparatus for experiments of this kind.

Thus the *Micrococcus* of *Mucor mucedo* Fres. is found in the blood and sputa of measles-patients, and the nurslings of this generation come directly through cultivation only, if we except the insignificant morphe of *Oidium albicans*. There is here also later evidence that the *Micrococcus* of a fungus retains throughout its own specific peculiarities; that from it that species only, yes, often that generation only, can be directly produced, which developed it. Naturally it has still its own action upon the substratum, and there is throughout no botanical contradiction in embracing the opinion, that the *Micrococcus* of *Eurotium-Pycniden* produces the small-pox; and on the other hand, that the *Micrococcus* of *Mucor mucedo* Fres. begets measles.

Concerning the spot where the infection through measles-fungus takes place, we can say but little, because the *Mucor mucedo* Fres., which is less abundant than *Mucor racemosus* Fres., appears on various decaying substances not too poor

in nitrogen. On human excrement, as well as on that of different mammalia, it is not rare. It appears on milk, but not so abundantly. It can be artificially raised on fruit, but it is not so vigorous as on a soil rich in nitrogen. It is found spontaneously produced and very abundantly upon all kinds of fruit, but mostly on cherries and plums. Therefore it is very probable that the infection with measles-fungus takes place in the privy, for the infection requires a copious evolution of the *Micrococcus* of *Mucor mucedo* Fres., which takes place in the highest degree in nitrogenous substances, and which is inhaled in great numbers. The *Micrococcus* of *Mucor mucedo* Fres. appears to be very abundant in human excrement, so that the opinion is probable, that by this means the atmosphere of pernicious decomposition must operate, and that the infection is introduced into the system from the air of privies.

I have, for a year and a half, publicly proclaimed, that it is our duty in all cases to disinfect the sinks and drains, because I was convinced that the *Micrococcus* of various fungi causes various contagious diseases, and that the contagion, i. e. the *Micrococcus*, are introduced by inhalation or by the alimentary canal.

This, my present opinion, was to be verified step by step, because every new detected vegetable appearance found in a contagious disease was the *Micrococcus* of a fungus, and indeed of a well defined (or specific) fungus; so that here is the place more than any where else for the above cautionary remark—that, at all times and in all places, to say nothing of cholera, disinfectants must be used if we would prevent or at least lessen the danger of infection with noxious *Micrococcus* from the privy or potable water.

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## VI.

## VEGETABLE ORGANISMS IN HUNGER-TYPHUS.

*Typhus exanthematicus, or Petechialis.*

For the material used in this investigation, I am likewise indebted to the kindness of Mr. Gerhardt and Dr. Schneider. It was in blood taken on the 7th of February, at the temperature of 32° R.

The precursory investigation showed that brownish Micrococcus existed in the blood in great numbers, which here and there formed small brown Mycothrix-chains. Also larger spore-like fungus cells, whose origin and signification I do not know. Probably they are the articulations of Oidium-chains of the respective fungi which came from the Micrococcus in the blood.

The cultures had the following results.

FIRST CULTURE.—On the 7th of February, on a piece of peeled and divided orange some blood was put and placed in the culture-apparatus. Already on the eighth day after, I saw sporoids forming and sprouting. The sprouts on the tenth day had taken the form of *Monilia cinerea* Bon. (T. 1, Fig. 32). Beneath the surface of the fruit-pulp macroconidia were formed, which on the thirteenth brought forth vigorous specimens of *Rhizopus nigricans* Ehr. (T. 1, Figs. 28, 40, 42).

SECOND CULTURE.—As on the 7th of February a sowing on a slice of apple was undertaken. Already till the 12th of February there was developed, as on the orange, from sprouting sporoids, the *Monilia cin.* Bon. with macroconidia, and from these came *Rhizopus nigricans* Ehr.

THIRD CULTURE.—A portion of blood, on the 7th of February, was put in sweetened water with phosphate of ammonia and placed in the culture-apparatus. At the end

of the month stout *Micrococcus* had multiplied, but no other sporoids appeared, as fermentation always hinders sprouting.

FOURTH CULTURE.—On the 7th of February a mixture of paste, prepared with sugar and phosphate of ammonia, after the addition of white of egg, and some blood from a patient sick with Typhus exanthematicus, was put into the large isolating apparatus. Until the 19th there were formed in the substratum yellow spots, which exhibited under the microscope a rich vegetation of *Monilia cinerea* Bon. and macroconidia. The *Rhizopus* came only feebly.

#### RESULT OF THE CULTURES.

There is thus found in the blood of Hunger-Typhus the *Micrococcus* of *Rhizopus nigricans* Ehr., which can be easily raised on suitable soil from sprouting *Micrococcus* swollen to sporoids. The infection with *Micrococcus* comes by means of rotten fruit, decaying succulent vegetables of every kind, and also through faecal deposits. Therefore, the infection from the privy is possibly, but more probably through the use of spoiled vegetable food of various kinds, and this throughout conforms with experience concerning infection with Typhus.

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### VII.

#### VEGETABLE ORGANISMS IN INTESTINAL-TYPHUS.

(*Ileo-typhus*, or *Typhus abdominalis*.)

For the material of this labor I am indebted to Mr. Gerhardt and his assistant Dr. Schneider, as well as to Dr. Gietl, physician to the King of Bavaria.



On the 14th of February, I received, from the clinic at Jena, some blood of one sick with Typhus abdominalis. It contained exceedingly small Micrococcus cells, mostly single, more rarely united in chains. On the 16th, some of the intestinal contents of the same patient was sent me. They contained an unusually large number of slender, partly swarming and partly quiescent, Micrococcus, with here and there large balls of cohering Micrococcus-cells, and more distantly, single, colored-spores mostly of the size of Penicillium spores.

On the 18th of February, I received from Munich, by the kindness of Dr. Gietl, the excrements of another patient who had typhus. These contained almost solely masses of Micrococcus, frequently adhering together in the form of balls. Spores and other fungus-cells were more rarely found than in similar material from Jena. They are in general of immaterial importance.

Small crystalline or crypto-crystalline, brownish balls appeared very abundantly in the Munich material, single or in a heap (T. 2, Fig. 9), often joined together, and then tolerably like the cholera-fungus, or easily distinguished by their physical or chemical properties. These crypto-crystalline forms I often saw in the intestines, and always in cases where the blood was undergoing decomposition, as in cholera-stools, ulcerated intestines, but never have I seen them in such abundance or in such masses as in Typhus fever.

#### THE CULTURES EXHIBITED THE FOLLOWING RESULTS.

FIRST CULTURE.—On the 14th of February, 1868, I sowed in the culture-apparatus, on paste prepared with phosphate of ammonia, blood from a patient with Typhus in Jena. On the 18th there appeared a tender furze, which, on the 22d, had formed a dense vegetation of Penicillium

crust. Fr., with entirely normal fructification. The sprouts were developed from sporoids, as I confirmed on the 18th.

On the 25th of February, there was to be seen at separate spots, exactly in the middle of the drop of blood, a specimen of a large and mostly dichotomous and ramified *Penicillium grande*, the same as is shown (in Fig. 30, T. 1) to belong to *Rhizopus nigricans* Ehr. Within the substratum also were formed macroconidia, and some specimens of *Rhizopus*.

SECOND CULTURE.—On the 14th of February, a small drop of the same typhus blood was put on a peeled apple and placed in the culture-apparatus. On the 19th a vigorous and normal vegetation of *Penicillium* crust. Fr. shot forth. Only at one place there arose from the blood a small spot of *Cladosporium herbarium* Lk. Within the fruit-pulp macroconidia were formed, which by degrees increased upon the surface in the form of a sickly pencil of *Penicillium grande* (T. 1, Fig. 30), which was soon overrun by *Penicillium crustaceum*. Till the 27th the macroconidia generated vigorous normal specimens of *Rhizopus*.

THIRD CULTURE.—On the 14th of February, some typhus blood on the white of egg was put in the culture-apparatus. *Micrococcus* multiplied largely in the albumen. At the surface of the vessel was to be seen in eight days an *Oidium* vegetation, which throughout resembled *Favus-fungus*, *Achorion Schönleinii*.

The swarming *Micrococcus* were put under an immersion lens 1-18 of Merz, which appeared as T. 2, Fig. 20, shows. The cells are round-pyriform, colorless and prolonged into a cilium (or tail?) which simply oscillated here and there. The cells sometimes exhibited a spiral and revolving motion. Also the large yellowish-brown *Micrococcus* of *Rhizopus* were shown, but these appeared in the culture only sparsely, without doubt because of the large number of *Penicillium*.



FOURTH CULTURE.—On the 16th of February, some typhus-excrement with lemon was put in the culture-apparatus. On the 22d the lemon was adorned with a fur of fructifying *Penicillium* crust. Fr. At some places there appeared more vigorous and taller fruit pencils with oblong-oval spores, as I have frequently observed with bastard specimens between *Penicillium* crust. Fr. and an *Acrospore* morphism belonging to *Rhizopus*.

FIFTH CULTURE.—On the 16th of February, was placed in the culture-apparatus, typhus excrement with the white of egg. The *Micrococcus* of *Rhizopus* and *Penicillium* rapidly multiplied, and this last named fungus, in perhaps fourteen days, was preponderant.

SIXTH CULTURE.—On the 20th of February, some excrement from a patient slightly sick with the Munich-typhus was placed on lemon in the culture-apparatus. On the 26th was found on the lemon, a normal, but as if from sour soil, a somewhat softer furze of fructifying *Rhizopus nigricans* Ehr., and also some *Penicillium* crust. Fr.

#### RESULT OF THE CULTURE WITH MICROCOCCUS OF TYPHUS.

The result is remarkable, and is different from all the investigated contagions till now examined. In all the other contagious diseases thus far investigated, the *Micrococcus* was derived from one and only one fungus. Here two fungi constantly appeared, viz., *Rhizopus nigricans* Ehr., and *Penicillium* crust. Fr. Moreover the relation of one to the other was very remarkable: while the *Micrococcus* of *Rhizopus* was sparse in blood, the small cells of *Penicillium* appear in far greater number; in the contents of the intestine, the case was the reverse, the *Micrococcus* of *Rhizopus* appearing in excess.

It is clear that the investigation concerning the vegetable

organisms of Intestinal-typhus cannot be regarded as conclusive, but in so far as any conclusion could be drawn from these cultures it was :— that the far larger celled Micrococcus of Rhizopus in the intestine is the first and true cause of the infection with the fungus. This Micrococcus makes way for the far smaller celled of Penicillium in the vascular system, whither it can itself advance only in less numbers on account of the larger size of the cell. But the smallest of the cells of the Micrococcus from Rhizopus, remarkable for their brownish color, one finds very sparsely in blood, while the very small-celled, colorless Micrococcus of Penicillium here preponderates.

The fact, in all probability, is this, that the Rhizopus with its spores come in the intestines ; that it forms Micrococcus, which produce a similar destruction in the tissue ; that the small celled Micrococcus of Penicillium, which is always abundant in the intestine, possibly can enter into the vascular system. Thus the cause of Ileo-typhus is to be distinguished from Hunger-typhus, not in the specific nature of the fungus, but in the manner of its inception. In Ileo-typhus the Micrococcus of Rhizopus comes in the intestine, where it will cause destruction ; in Typhus exanthematicus, it enters by the lungs and is carried to the blood. In Ileo-typhus, the decomposition of the blood is the product of the action of the Micrococcus of Penicillium ; in Hunger-typhus, of Rhizopus.

If we compare these results with clinical observation, we become convinced that Ileo-typhus is occasioned principally by the filthiness of culinary or potable water and by the food mingled with the Micrococcus of Rhizopus, and that these are diffused through the water by the imperfection of the drains and canals.

I think the privy, through evaporation, must be credited not indirectly as participating in the production of typhus-



contagion : in Typhus exanthematicus the evaporation from human excrement, bad food, decomposing vegetables and decaying bodies of all kinds, engenders the infection.

We find the influence of potable water upon the infection with Intestinal-typhus, particularly subjected to examination in the excellent work of Gietl. The water of some pumps in Munich was found, according to Vogel, to be so strongly impregnated with organic matters, that for its disinfection, a litre of water to ten milligrammes of the permanganate of potash was required.

Gietl shows conclusively that the impurity of the water in the Munich pumps depended on the imperfect condition of the drains, &c., and that the infection of some of the pump-water with organisms and organic substances was the cause of the disease in houses thence supplied. He comes, therefore, to this conclusion, viz.—That Typhus in Munich is quite independent of climate, as also of the soil, and the water in its natural condition. How just is this conclusion; the surprising abatement of Typhus since the arrangement and adoption of new water works, shows. \* \* \* \*

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## VIII.

### VEGETABLE ORGANISMS IN CHOLERA.

In my work\* devoted to this subject, I have shown, that in Intestinal cholera appears one and the same closely definable fungus, whose yeast brings forth in the form of Micrococcus the greatest destruction and commotion in albuminous substances. The Micrococcus is at no time wanting in the discharges of the cholera-patient; indeed, it is present in

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\* Hallier. Das Cholera-Contagion. Leipzig, 1867.

large masses in the dejections. On the other hand, the occurrence of developed fungi, and, in particular, of their fruit, which I have compared to the fruit of an *Urocystis* (*Polycistis*), are somewhat incidental; which, no matter what we may think of the fungus question, appears not to be in a necessary condition with the cholera-process. The *Micrococcus* is at any rate in that position to be multiplied indefinitely, so long as the necessary support is furnished it under favorable surroundings: the fungus fruits might thence, in consequence of their multiplication of the *Micrococcus*, aggravate the diseased condition of the intestinal wall; at any rate the latter sufficed to induce dangerous decomposition in the tissues. It is probable, in regard to the so seldom appearing fruits of the fungi, that they occur on the wall of the intestines; yet the attacking point of the *Micrococcus* and its action upon the human organism is not to be discussed upon the botanical, but only on the *Pathologico-anatomical and Pathological side*.

I have only shown, that the *Urocystis oryzae* and its *Micrococcus* are present in the intestines of persons sick with cholera, and that we can, at a high temperature and on nitrogenous soil, bring forth therefrom the fructifying fungus-form. But how the *Micrococcus* gets into the intestines, whether here is the point of attack, or whether its first appearance is in the blood, or whether it directly produces the disease, or constantly attends it, which is scarcely credible:—all these are questions which I cannot undertake to solve.

I have to add some facts which may favor the probability of the opinion that *Micrococcus* and contagion are identical; which is to be argued—first, from my investigations and cholera-stool cultivation of the epidemic at Halle in the latter part of the summer of 1867, whose results are closely like those made known in my work on “Cholera-contagion;” and, secondly, from the result of the rice-cultures, which



were supplied with the cholera-dejections, and which furnished the result, that a brand-fungus developed on the rice, which killed the plant, and that the fungus was the same whose fruit sometimes came in the cholera-stools, and which formed the *Micrococcus* of the dejections.

But before I enter upon this double series of investigations, it may be well to give a survey of what was already suspected of the nature of contagion, and which was accepted on the ground of remarkable facts.

One of the first promulgators of the opinion of the vegetable nature of cholera-contagion, perhaps the first, was Professor Gietl.

He, already, in the year 1831, at the first spreading of the cholera over Europe, advanced this opinion, which has since been developed more clearly and precisely in his writings. His investigations upon this subject began in the year 1831, at Berlin, and were continued during the epidemic at Breslau, Ratibor, Troppau, Olmütz, Brünn, Vienna; in the year 1832 in other regions of Bohemia, and in 1836-1854 in Munich. Upon the epidemic at Munich, in the year 1854, he prepared a special report.

Already the chronological order of the breaking out of the cholera in the city hospital at Munich, indicated the transport of the disease from person to person. Clearly was this doctrine found advanced in the paragraph upon the "Cause and essence of cholera."

"The cause of cholera is a specific poison, which is of an organized nature proceeding from the dejections, which perhaps go through a peculiar process of fermentation, and spread from the dejections through the air among the population of a town and district."

"This poison grows exuberantly in the mucus of the intestines, particularly the small intestines and the stomach, and begins to produce decomposition and decay. The bodies

of the sick and their corpses have throughout no poison or infection. Only the evacuations of cholera-patients convey the seeds of the disease where they are allowed to remain. This poison is always introduced, and never appears spontaneously. From its action it is evident that it is exotic, and its essence has nothing similar in known pathology. But the poison, in its mode of propagation, has a resemblance to the poisonous cause of dysentery and typhus (Typhus abdominalis or nervous fever). Concerning the dysentery, it is a well known fact that the infection comes from the dejections: concerning stationary typhus, I am convinced that it is a poisonous sickness, and that the poison may be developed and propagated by the evacuations and modified places (Decubitus); probably, also, the excretions can strengthen the virulence of the poison, according to the different stages of the disease, and by the process of decay."

Furthermore, we read at p. 8—"What is the condition of that which is suspended in the air and coming from the evacuations, is unknown. All sorts of hypotheses are advanced thereon: but one idea cannot be relinquished, that it is an infinitely small organic body, the spores of cryptogams, mould-like, and is imperceptible to our senses aided in every possible way."

Further, at another place, page 9:—"We come to a conclusion concerning the nature of the poison, only from its action upon the human body: wherefrom we see, that this organic-stuff, this sad visitant, has a certain duration of life, is not always possessed of the same strength, and that there must be certain conditions for it to ripen, and for its action to take place. Perhaps not immediately upon the discharge of the contents of the intestine does the poison present itself; but it requires still a certain time for it to be propagated. This organic body contained in the cholera-poison appears to exist a long time, and under certain circumstances (as moisture and warmth) it can again be set free."



Highly interesting is the return of typhus symptoms, when the patients are attacked with cholera, as Gietl remarks pp. 10, 11.

Further, toward forming a correct judgment upon the vegetable nature of contagion, the communication which Gietl has made, in confirmation of the investigations of others upon the predisposing circumstances, is of the greatest importance. He says—"All that irritates or weakens the mucous membrane of the stomach or intestines, or that puts the intestinal capillaries into diseased action, let it be through the medium of nourishment, foreign matters, changes of temperature, individual conditions or mental influences, favors and supports the reception of the poison and its efficacy in various degrees. Evidently all sip in whatever of poison is diffused through the air, which produces, in the greater number, the gastric symptoms present,—as oppression at the stomach, flatulence in the bowels, disturbance of the digestive process and modification of the fæces, and with a smaller number produces the more advanced grade of the disease."

In relation to the cholera disposing circumstances, the most conclusive and convincing is the experiment, which Dr. Ehrlich in Breslau and other physicians undertook, in swallowing cholera dejections. The support of this heroic experiment remains very sound. Had they, beforehand, contracted the intestinal catarrh, the result would have been very different.

The botanical hypothesis that the contagion of cholera is nothing else than the *Micrococcus* of rice-brand fungus, renders a predisposition necessary; did not the predisposition exist, or were it the same with all persons, every one living amid the cholera atmosphere would become sick, and the disease introduced from Asia would not disappear. In general, every fungus which has to attack and destroy animal

tissue, must find it predisposed, and this in a particular degree can be maintained in relation to parasitic diseases of the skin.

The Favus, it is well known, is strongly contagious, but the artificial transplanting by no means always succeeds. In experiments upon my own person I succeeded but once, and then only imperfectly.

The Favus, imperfectly developed, healed wholly spontaneously, without the aid of any parasiticide. With some persons the experiment easily succeeds, while with others it fails entirely.

It is the same with regard to the mucous membrane. In diphtheria and croup there are fungi-forms, whose spores go to the bodies of the healthy without harm. With such they never come to development on the mucous membrane. On an inflamed or weakened mucous membrane, on the other hand, they sprout and bring forth masses of fruit and yeast. These formations certainly are not inconsiderable for the course of the disease: not without foundation does one bring a parasiticide into use. Just as surely for the development of the disease is a mucous membrane predisposed by catching cold or any other cause. Closely so is it with intestinal diphtheria. Should cholera and typhus be differently suppressed?

I cannot leave this idea, without calling attention to the following diseases which occasionally follow the cholera:

Uræmia.

Cholera-typhoid.

Bright's disease.

Diphtheria.

Pyæmia.

Cholera exanthem, &c.

\* \* \* \* From my work on "Cholera-contagion," I must regard it as proved that in cholera dejections appears the Micrococcus of a certain fungus not a native of Europe.



I must explicitly state, that a great number of cultures which I made with the material of Halle, under the same circumstances, have furnished the same results. Very important is the question as to the distinction between the origin of Asiatic cholera and common cholera (nostras).

\* \* \* \* These fungi-generations were probably introduced with the wheat-culture. Upon the wheat is found only the blight or wheat-brand, *Tilletia caries* Tul. These appear with three generations, to wit, *Penicillium crust. Fr.*, *Mucor racemosus Fr.*, and *Achlya prolifera*, as they were described by Pringsheim. All these we can, by a change in the physical and chemical qualities of the matrix, bring up from *Tilletia*.

While now the wheat, as is well known, is not of European but of Asiatic origin, so it is in conformity with the Darwinian hypothesis, that the *Tilletia* is introduced with the wheat and furnishes *Penicillium*, *Mucor* and *Achlya*.

The question now to be decided is of the greatest importance, viz., whether, at the usual summer temperature, the germinating *Micrococcus* of our cholera, was brought forth from something like the *Urocystis oryzae* which I found in the Asiatic cholera. Therefore it was of the greatest value to me, that Prof. Vogel presented me with the dejections from some cases of PSEUDO-CHOLERA which could be used for culture.

It thus turns out, that at a certain summer temperature, 20 deg. R. or thereabouts, the cholera fungus does not come to maturity. The *Micrococcus* certainly sprouts, but produces normal specimens of *Penicillium crust. Fres.*, and *Mucor racemosus Fres.*, according to the matrix.

Still more cultures must be made before the question upon the distinction between the cause of Cholera-Asiatica and Cholera-nostras can be decided; but the result, meanwhile, is important, because it shows, that in the case in

question only native Fungi, not of the Asiatic form, but nevertheless Generations of the same species, appear in the intestines.

~~That~~ Here the Micrococcus, probably of Tilletia, acts, we may suppose, as contagion. The difference in its mode of action is only this, that the Micrococcus of Asiatic-cholera, i. e., of the *Urocystis oryzae*, acts more energetically and destructively and in smaller quantity. On this ground the Micrococcus of Cholera can act easier than the Micrococcus of Tilletia by one inspiration of the faecal emanations, while repeated inhalations of the Micrococcus of Tilletia from the cloaca, or their introduction by potable water, is necessary for the production of Cholera.

For either disease an investigation of the eventual vegetable forms in the blood is indispensable. This will furnish pure culture, and thence with greater certainty furnish an answer to the fungus question.

The question upon the causal force which lies at the foundation of Cholera (nostras) and Asiatic-cholera, leads naturally to the question of the predisposing force. That individual predisposition is present, has long since been proved by medical investigations. Wherein this consists, cannot be the subject of my discussion; much more have I to leave the investigation of these diverse and complicate conditions to pathology and pathologic-anatomy. But beside the individual predisposition, there is a general disposition present, and this for Cholera-Asiatica and Cholera (nostras) is one and the same, and is of the greatest importance. How remarkable is the graphic representation in the ninth report, where the curve representing the diarrhoea almost always rises and falls with that of Cholera.

Already, by this means alone, it has been settled, that Asiatic cholera is exotic, that it must differ from our cholera, which is very prevalent in years when the Asiatic cholera



does not appear. But sure is it, that the same miasm (pardon the word) which produces and enhances our cholera, intensifies also the disposition to Asiatic-cholera. Here, however, two different things are to be discriminated: (1) the real cause of our cholera, vegetable miasm or contagion itself; (2) the living condition of this contagion, whereunto warmth belongs. Hence the curve rises or falls more or less with the temperature. That a perfect parallelism cannot be found between the curve of temperature and the continuance of the disease is perceptible; then the temperature is not the cause, but only the means of its vitalization. That the cholera is not necessarily extinguished in winter needs no particular assertion; then always in the intestine is to be found the temperature necessary to the increase of the Micrococcus; not only in the heated rooms but in the heated houses in Russia the temperature suffices to keep alive the Micrococcus and to afford means of infection through the least filth. While now in summer, amid circumstances favorable to life-engendering, the Micrococcus of Tilletia in great numbers mix with the air and soil, so this Micrococcus, which probably is the cause of our cholera, must intensify the disposition to Cholera-Asiatica, for every reinforcement of fermentative precedents in the intestine must likewise strengthen the disposition to the cholera.

I am too far off to follow the distinguished PETTENKOFER'S investigations *pro et contra* in all their particulars, but this much I adopt as entirely conformable with my investigations:—that the conveyance of Micrococcus from the privy into the potable water and into the air and soil, not simply of Asiatic cholera but of our cholera, diarrhœa, and probably of other diseases, and which appear to be present in masses from Penicillium in healthy human intestines, and the Micrococcus from decaying substances in general; all furnish a disposition to Cholera-Asiatica. The Micrococcus

of *Urocystis oryzae* passes through the entirely healthy intestine without mischief, but if fermentation in the intestinal contents is already advanced to an unusual degree, then the *Micrococcus* finds a favorable soil for germination.

By far the most important result of my later investigations upon Cholera-fungus, is the success in the culture on *Oryza sativa*. Already at the appearance of my "Cholera-contagion," I could show how the *Micrococcus*, scattered through the soil by the rice-water stools, enters the sprouting rice-plant, and passes through its tissues with a slender mycelium. Likewise, that the *Micrococcus* mostly swells before sprouting, forming sporoids.

The culture was continued until the end of October, 1867, and gave surprisingly favorable results. The rice covered with the cholera-dejections in the course of the summer appeared wholly abnormal under the influence of the mycelium spreading through the tissues. The leaves remained pale, chlorotic, and scarcely attained half their healthy breadth. Cotemporary rice sown without admixture or with *Penicillium* spores brought forth healthy sprouts and normal plants.

There appeared at last on the rice-leaves a blackish stripe, which proceeded from the point to the base of the leaves. Here the fungus fructified. I have designated it *Urocystis oryzae*, placing it provisionally among the *Urocystis*, since the characteristics of this genus (*Polycystis* or *Urocystis*) throughout suits it. In the mean time it must be remarked, that this family is slightly known, and that fungi are arranged in it which have only slight relations with each other that can be confirmed by Generation changes. \* \* \* \*

Thus the vegetable nature of the small bodies in cholera-stools, is in every way confirmed and illustrated. It is only necessary, now, to investigate the *Ustilaginia*, which in India appears on the rice, in order to ascertain from experi-



ments, whether the *Urocystis oryzae* exist in greater numbers and whether they generally appear. From a notice in an English newspaper it would appear that the cholera-fungus is found in India; but as the notice is anonymous, it cannot be regarded as scientifically authentic.

Lately various plausible opinions have been advanced in relation to the spontaneous motion of the small organisms of cholera-stools. This difference of opinion does not now exist, but rested entirely on a misunderstanding. That the *Micrococcus* in cholera dejections was self-moving, was indeed doubted by no one. Already, B. Paccini had spoken of small infusorial-like bodies. But it is an entirely different question, whether one has to regard as essential those provided with tails, or those yellowish and motionless *Micrococcus* cells which occur in great number. I am myself mostly in favor of the latter view. The swarming *Micrococcus* of the cholera-stool belongs to *Penicillium crust.*; the faeces never lack them, although they are not always in great number, while the yellowish and motionless *Micrococcus* cells, without exception, are most abundant.

But I have more striking, yes, convincing evidence for my assertion—that not the swarming, colorless, but the quiescent, mostly yellow *Micrococcus* cell is the contagion, if there is generally a vegetable contagion. In the rice-culture, with *Urocystis oryzae*, there appeared from the spores of cysts (*Schizosporangia*) set free by means of a gelatinous swelling of the spore-wall, a motionless *Micrococcus*, and this it is which introduces the profound destruction.

I will not deny the possibility, that this *Micrococcus* can take the swarming condition under circumstances unknown to me. In every case, the swarming is, as with every other swarmer, only a temporary condition. There always appears, before the sprouting or dividing, a quiescent condition, and the tails vanish away.



Perhaps some remarks concerning remedies and disinfectants will not be unwelcome. Unfortunately I have had no time to multiply my earlier experiments upon the destruction of the *Micrococcus* of cholera-dejections; especially do I regret that it was not possible for me to cultivate the *Urocystis* in the presence of etherial oil, because this, as stated by others, had so great practical results. When the etherial oil is present, the development of the mould-fungus is hindered. But still it does not follow that this would be the case with every fungus, as cholera-fungus: the effect of every etherial oil upon every fungus must first be studied, before we can reach a complete induction upon this point.

In my work ("Cholera Contagion") I have reported a fine result upon the cholera-fungus with the solution of sulphate of quinine, acidulated with sulphuric acid, and can now with pleasure add, that I have received from several physicians notices where the efficacy of the quinine is confirmed. Dr. Methner of Breslau and Dr. Leib of Vienna, in particular, report favorably upon the use of medicines where quinine was the chief ingredient.

I am convinced that a more important result is yet to be attained, when the introduction into the system of the acidulated solution of the quinine is effected simultaneously by the stomach, by the bowels and subcutaneously: the dose being apportioned according to the stage and severity of the case. When used internally, either by the mouth or by the bowels, the dose should be from 2 to 5 grs.

In disinfecting the close-stool, I maintain, after my experiments, that the sulphate of iron is the best disinfectant. I have already shown in my work on the "Phenomena of Fermentation," that Mr. Pettenkofer, by the introduction of this substance, has made a happy hit, since disinfection depends less upon *destroying* the fungus vegetation than *preventing* the *Micrococcus* fermentation. Also, at a later



time, some inquirers, through experiments, disbelieved the efficacy of the sulphate of iron, inasmuch as in its solution and even in that of arsenical and cupreous salts, yes, even in a solution of corrosive sublimate, delicate fungus-mycelium may not only form but continue to vegetate. But this mycelium is entirely harmless, as it neither normally fructifies or produces any yeast, so that from it nothing unhealthy can be produced. When, therefore, such persons obtain from their experiments such mycelium, no serious objection to the use of acid disinfectants can be founded thereon. It is true, we can with this mycelium in a fluid liable to fermentation slowly excite the fermentative process, but so long as the mycelium remains in the vitriolic solution it is entirely harmless. Mycelium appears in a weak alkaline as well as in a weak acid solution. For the entire destruction of all vegetation, which however is wholly superfluous, large quantities of alkalies or acids are necessary.

To disinfect fluids, the mixture of Mr. Severn, of Halle, is most highly recommended. This, as is well known, consists of lime 100 parts, coal-tar 10 parts, and chlorate of magnesia 10 parts. I mixed a liquid strongly inclined to fermentation with 24 drops of cholera-stool from Halle, and added 24 drops of Severn's mixture. After some months the liquid in the isolating apparatus was wholly clear and free of fungi. The incipient vegetation was enveloped in this disinfecting medium and settled on the matrix. Hence in all cases where the contents of the close-stool can be kept in a liquid condition, this (Severn's) mixture is to be recommended as a superior disinfectant, and in such quantity as to cover the contents of the close-stool for some inches, for if only a small part remains uncovered the fungus will continue to vegetate. \* \* \* \*

## IX.

YEAST-FORMING IN THE INTESTINAL CONTENTS AND  
FROM THE MUCUS-MEMBRANE.

On May 13th, 1867, I examined, in Berlin, the vegetable-forms appearing in the excrements of a monkey (*Cercopithecus*).

There were found, besides the usual discharges, vegetable remains, plant-hairs, vascular cells, epithelial cells, &c., large masses of *Micrococcus* (T. 2, Fig. 10), which, as I have established by many investigations, are always found in the human intestines and introduce the decaying decompositions of the fæces; yes, in every digestion are of essential importance. In the stool of the monkey the *Micrococcus* often appears in conglomerate balls (T. 2, Fig. 10, *d*), that is to say, the parent cell was still scarcely dissolved. Likewise delicate sprout threads (T. 2, Fig. 10, *a*) were here and there to be found, pretty frequent spores of *Mucor* (T. 2, Fig. 10, *c*), less often articulations of an anaërophytic fungus-form (T. 2, Fig. 10, *b*), that is, of a brand-fungus, quite exceptional spores and cells of various brand and rust-fungi. Never did I find the spores sprouted, as is the case in the human intestine, only when in an unhealthy condition. The *Micrococcus* showed, for the most part, the swarming motions, partly, it was found, at rest and in the act of forming articulations which often remained connected together as *Mycothrix*-chains, as normally appearing in the healthy human intestine.

On the same day the monkey was fed on cake made of meal, eggs, sugar and water, with the addition of *Rhizopus*' spores.

On the 14th the remnants of the discharges were still the same, although the monkey had received nothing else than



the cake and milk as nourishment; plant-hairs, in particular, appeared in great numbers. No trace of starch could be detected, therefore the meal-cake (after twenty-four hours) had not reached the intestine. The fungus-forming in the morning still showed little change. In the evening there were found the *Mycothrix*-chains (T. 2, Fig. 11, *a*) far more numerous.

The *Micrococcus* was frequently still enclosed in the parent-cell in various stages of forming (T. 2, F. 11, *b c*). Spores or conidia, as T. 2, Fig. 11, *d*, shows, were numerous and often in the first stage of sprouting. These changed their color but little on the addition of iodine. By means of iodized chloride of zinc they, as well as the fragments of chains and the *Micrococcus*, became of a light greenish yellow-brown color.

Still on the third day were found plant-hairs (*Steinzellen*), fragments of plant-cuticle, &c. Traces of starch could be detected by iodine; thus three days were required for these to pass through the alimentary canal, while the monkey seemed quite well.

Not until the fourth day after the first meal did I find the spores of *Rhizopus* (T. 2, Fig. 15, *a-c*) in great numbers. These had partly lost their granular contents (T. 2, Fig. 15, *b*), partly the epispore was torn and empty. Besides the spores appeared numerous conidia (T. 2, Fig. 15, *d e*); as usual they appear in the midst of a pulpy substance on the sprout threads of *Rhizopus*. On these could be followed the gradual conversion, by subdivision, of the simple nucleus into *Micrococcus* in all stages (T. 2, Fig. 15, *d e*). Besides these appeared *Torula* chains of lanceolate fungus cells. Starch granules, colored blue by iodine, were present in great numbers. The articulated chains, conidia and spores were colored by means of iodized chloride of zinc from a burgundy-red to a violet, as Fig. 15 shows. The

odor of the monkey's feces was, during these digestions, very similar to that of human feces.

A second series of feedings was undertaken with the same monkey, after a time, when, in the cake, instead of *Rhizopus*' spores the spores of *Tilletia caries* Tul. were put, which furnished a wholly analogous result. On the third day could be followed the entire development of the *Micrococcus* from spores and conidia, by continuous subdivision of the nuclei, examples of which I have given in T. 2, Fig. 19, *a-d*, which, on comparison with my history of the development of *Micrococcus* in *Tilletia caries*, is readily understood.

It was remarkable, that during the feeding with *Tilletia* spores, the monkey's excrements exhibited a grayish (not yellow-brown) color, of a pasty and ropy consistence, evolving a cheese-like odor.

With a cake of the first kind (with *Rhizopus*) a man likewise was fed. He exhibited, by stool, after the sixteenth hour the spores of fungi and *Micrococcus*, changing gradually to balls (Fig. 16, *a-c*) of *Arthrocooccus*. This individual was constipated before the feeding.

As a consequence of preparing the cakes of the first series, I suffered under diarrhœa, while *Micrococcus* formations appeared in great numbers in my evacuations, as I have shown in T. 2, Fig. 12, *b-d*. Conidia, spores and tender sprout-threads also were found.

Still interesting was the appearance of what came from the mucous membrane of the mouth and throat. Here, by repeated inspiration of the spores, *Micrococcus* formed in unusual numbers, while I suffered a not inconsiderable catarrh.

The *Micrococcus* exhibited all the stages of sporoid-forming (T. 2, Fig. 13), so that I considered it necessary to destroy them by a parasiticide, and to guard against their sprouting.



The epithelial cells amid the mucus showed in the most perfect manner the outgrowth of the attacking *Micrococcus* cells into longer and shorter *Mycothrix*-chains (T. 2, Fig. 14).

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## X.

### LATEST INVESTIGATIONS UPON THE NATURE OF MICROCoccus.

That the *Micrococcus* is developed in the manner specified, viz., from the contents of the spores and vegetable cells of determined fungi by means of the repeated subdivision of the nucleus or nuclei, no one after my numerous labors upon this subject can any longer doubt; especially so, since a conscientious observer, provided with a first-class microscope, can confirm what has been said thereupon. In fact, this confirmation has, to my delight, been made by botanists of the first rank. So, the development of *Cryptococcus* and *Arthroccoccus* from *Micrococcus*, by the chemical change of the matrix, is easily proved, and has been confirmed by different individuals.

It follows that the *Micrococcus* retains its own specific nature, so that any one can now get only that fungus from it by means of the sprouts which begot it, as clearly appears from the above mentioned investigations. The most difficult point is the motion of the swarming *Micrococcus*. Here are needed the best microscopes of the best manufacture, the highest powers and the best illuminating apparatus. But all these suffice only when one brings into use all the artificial helps which the laws of optics suggest. Now not all microscopists are in such favorable circumstances, possessing instruments of the first class and from the various workshops, and thus there is a dispute in relation to the motions of

swarming *Micrococcus*. PACINI, KLOB, THOME and some English observers very accurately attribute to one portion of the small cells (*Micrococcus*) of Cholera discharges self infusorial-like motion, in contradistinction to molecular movement. Less expert observers, provided with an ordinary instrument, have reproached a certain person for his statements; while here, nevertheless, the use of an acid would at once decide, by the immediate suspension of all self-motion, while the molecular movement would be seen at first to be very energetic.

With a powerful system of a good microscope one will with sufficient clearness perceive the motive organs of the small swimmers.

With the system G of Zeiss, even with the system F of the same optician, by the help of a weak eye-piece, as with the immersion system No. 11 Hartnack, and with an immersion system 1-18 of Merz, it can be seen that in the *Micrococcus* (not indeed in every *Micrococcus*) of some fungi, especially of the *Mucor*-kind, the swarming-cell has a longer or shorter tail-like prolongation, as I have often exhibited in my designs. These locomotive organs are not shown so plainly by the previous systems as by an immersion system 1-18, recently procured of Merz in Munich.

\* \* \* \* \*

With this system, by the help of ocular No. 4, I can perceive the tail, under the favorable western light of the morning or eastern light of the afternoon sun reflected from a still cloud to every *Micrococcus*, which, by its oscillation, produces its movement. That such is the fact (after very close focusing, arrangement of the diaphragm and mirror, the protection of the eyes against direct light by means of a screen, and that of the object from light from above by means of a small black shutter, and other delicate manipulations which are essential, as the skilled microscopist knows), would need



no confirmation, had not a false view been recently diffused, viz., that *only two sound, not experienced eyes*, were necessary; that to such everything must be plain, and that what such persons cannot through lack of practice find, they even deny the existence of, while it is readily seen by experienced observers. Had we not good microscopists, such observers would occasion more confusion than exists in the subject itself.

I will here give some examples of swarming Micrococcus which I had an opportunity to study lately. Moritz Willkomm, as already stated, had heretofore, and more recently noticed anew, the development of Micrococcus. He gives a description thereof from Corticium amorphum Fr. in his brilliant investigations upon the diseased bark (cancer) of the Larch, which he had the kindness to send me, together with material for investigation, so that I am in a position to compare his statements with my own research. I can corroborate that the spores of Corticium not arriving at maturity, and not capable of sprouting, develop Micrococcus in a swarming form. It appears, that the mycelium-cells bring forth Micrococcus, yet they are less easily and certainly to be identified. Still the sprouting of the ripe spores on the object-bearer I found closely as Willkomm had described and figured. I succeeded not only to develop from the double spores in a drop of glycerine on the object-bearer, the two end sprouts, as seen at T. 2, Fig. 19, *a*, but also with thinner sections through the Corticium to show the sprouts broken out even from the Asci, as seen in T. 2, Fig. 19, *b c*. This circumstance was, to me, of great importance in judging of the mould-form raised from spores by Willkomm.

It was Willkomm who succeeded in bringing up from the germs a pencil-fungus which he classed with Penicillium. As now the Penicillium already on the second or third day fructifies, so he succeeded here to follow the fungus of the



fruit-pencil backward to the sprouted spore. Willkomm sent with his preparations a drawing, which proved this, as I succeeded in doing, to follow back the same pencil-mould not only to the sprout spore, but even to the spores still covered in the Asci. But not more evidently could be proved the consanguinity of two fungi, and I was gratified to be able to ratify the brilliant observation of Willkomm.

Concerning the *Penicillium*, it is difficult to say, whether it should be regarded as *Penicillium* or *Cladosporium*. The fructification of both is so undecided and variable, that we can seldom make out for either a sure definition. The change of Generation is here the main thing. The *Penicillium* alluded to, has a brownish color, often it is dark-brown. The ramification is incipiently irregular, the spores or joints are long, spindle-shaped, sometimes divided or having septa, towards the end of the chain nearly globular, but generally with two very blunt-pointed ends. In the fluid the specimens soon became pale, and now resemble in some measure the fungus *Penicillium crustaceum* Fr., but in the form and arrangement of the spores there is always some difference, and the whole habit is different.

After a week's continued duration of the culture on starch paste with phosphate of ammonia, the fungus entered within the matrix; but here the joints become deep-brown, independent and fallen to pieces by division into a globular brand-spore. This brand-fungus, this anaërophytic form of *Corticium*, has entirely the figure and spore-form of an *Ustilago*. It strongly resembles the *Ustilago carbo* Tul., but it is somewhat distinguished by smaller and paler spores. Thus the consanguinity of three fungi-forms, which formerly were regarded as belonging to different species, viz.: *Ascomycetes*, *Corticium amorphum* Fr., with its already known change of generation, the anaërophytic brand-form; *Usti-*



lago corticii, and the appertaining acrospore-form; *Penicillium*, or *Cladosporium corticii*, is here again proved. \* \* \*

The swarming *Micrococcus* of the *Corticium* spores (T. 2, Fig. 21) does not belong directly to the larger forms, but there plainly appeared, already, with the system of Zeiss, a tail-formed prolongation of the cell. With the immersion system of Merz, these tails appeared as plainly, as is shown in T. 2, Fig. 21. One sees, that these swing here and there, but yet most have a revolving and spiral motion also. Upon longer and closer observation we come to the conclusion, that the *Micrococcus* cell is contractile. In motion they change form considerably.

More clearly are these changes of form seen in the *Micrococcus* cells of *Penicillium crust. Fr.*; for example, in blood and the stools of typhus (T. 2, Fig. 20). One can here distinguish the cell from the tail. The motions of the tail are closely like those of *Corticium*. But the cell is sometimes globular, sometimes pyriform, sometimes stretched out, and one sees these changes interruptedly take place, or by degrees. In both cases here described, the *Micrococcus* is perfectly colorless. Not so with *Rhizopus*. This is naturally unimportant, whether one observes the *Micrococcus* from the spores of *Rhizopus* or from typhus-stool. The swarmers appear here under the immersion system and with ocular No. 4 as large and plainly as is shown in T. 2, Fig. 22. One sees a large yellow-brown cell wall, which, as the parent cell, surrounds the true swarm-cell. This is found free, central, or at the side in the parent-cell, through whose wall the long tail proceeds. Its length amounted to five or six times the diameter of the cell. It swings, in motion, here and there, and is ordinarily found before the cell, this dragging behind. Sometimes, in particular when meeting with obstruction, it shoved the cell forwards.

Much more distinctly is the tail seen in the smaller but



quickly moving Micrococcus cell of *Mucor mucosa* Fr. of the measles blood, as also in the Micrococcus from the spores of the genuine *Mucor mucedo* Fr. (T. 2, Fig. 23). I have figured some of these as I saw them under an immersion system of Merz. They are nearly colorless, very light golden-greenish. They move here and there very quickly, sometimes forward, sometimes backward, sometimes revolving. I mentioned before that purely vegetable cells are able also to develop Micrococcus, as Willkomm declares. Rarely do they seem to divide easily, whether the Micrococcus is produced from the spores or from the cells of Mycelium also. A good example of this is *Oidium*, which the mould-fungus puts forth. Here we often have joints, only, without a proper fructification. Thus T. 2, Fig. 29, shows some joints of the *Oidium* of *Rhizopus*, as if they were partly filled with nuclei, and between the joints the Micrococcus multiplying as coming from the liberated nuclei.

The older elaborators of the yeast-theory came to their false conclusions and dogmas by the false system of their investigations, partly through the erroneous explanation of what they had seen, and partly through their entire ignorance of the later labors of the chemist and physiologist in the field of the doctrine of Fermentation.

With nothing else than the isolating apparatus, and that imperfect, one cannot bring out the culture which I have discussed in my writings upon fermentation. But a lack of knowledge upon fermentation in general necessarily occasions mistakes in the examination of the yeast. I might here bring forward an example. Everybody knows that the representatives of the older doctrine upon fermentation were acquainted with only one kind of fermentation, the spirituous. Thence the childish criterion: if at the decomposition carbonic acid is developed, or when, more rudely expressed, gases rise, so the acting organism was yeast and nothing else.



All the other formation precedents are disregarded by such dogmatic definition; and while now the spirituous fermentation is by no means so plain an event, as we must believe after the just mentioned theory, so must errors arising be very many and evident, and the discovery of a developmental history formed upon a distorted apprehension, be an impossibility. This impossibility of discovering the yeast development upon the elder theory, is far more evident when the false method of inquiry and necessarily false explanation of the observations is taken into account. To wit, when one employs only a closed apparatus (isolating apparatus) for culture, and this first opened at the close of the culture;—thus it is impossible, *a posteriori*, to obtain a clear idea of the development of the active organisms taking place.

One obtains only the final product of the experiment, and it is a pure sport of fancy when one any way judges of and interprets the earlier condition of the organism exciting fermentation, which is permitted to take place only step by step. And now the limitation of spirituous-fermentation! Here lies, first of all, the danger of gross errors, viz., the confounding of cell-sprouting and spore-sprouting with actual yeast-cells, which are similar. The intermediate forms between genuine yeast and mycelium are naturally many, and such has too often been regarded as yeast itself until my investigations. Such sprouting spores, as have been very closely observed by J. Sander, come on the surface of substances undergoing spirituous fermentation, when the air is admitted, as can be no otherwise in the isolating apparatus earlier made use of.

I have, for example, some sprouting vegetable cells of Favus-fungus (Oidium of Penicillium crust. Fr.) as T. 2, Fig. 30 shows. A certain resemblance between such a formation and cryptococcus is incontestable. But I have already

earlier shown, in my treatise on "Fermentation," that such intermediate forms, viz., the transition of yeast-cells into mould-forming (for example, *Hormiscium*), or inversed transition of mould-form into the formation of yeast, only happens on the surface, never within the fermenting liquid. It needs only the examination of the bottom yeast to be convinced of this; but one gets it not in using the former apparatus.

Where would it lead if all cell-sprouting was to be regarded as yeast? Those conidia and spores appearing acrogenous, always sprout closely in the same manner from the stem-spot like the young *cryptococcus* cell.

If one compares only the spore-forming of *Aspergillus*, *Penicillium* and other fungi with the spore-forming of *cryptococcus*, he will be convinced that the whole process is the same. But it is nothing else than one of the two main forms of the cell multiplying of the fungus. T. 2, Fig. 31 shows two stem-cells of *Aspergillus* with spores; compare T. 2, Fig. 3, *s p*. At *x* is seen a very young sprout-like spore, at *s p* a similar one ripe. Fig. 32 shows the same in *Penicillium*. In Fig. 33 is figured a sprouting brand-fungus. Everywhere among the thread-joints are seen sprouting conidia breaking forth. Should all these structures be regarded as yeast-forming because they pass through a similar histiological development, as *cryptococcus*, or shall it not rather be asked, is this sprout-forming really yeast, and the *cryptococcus* the only yeast-form, or is there still another kind of yeast generation present?

I have long since answered this question, and as my yeast doctrine has enjoyed universal recognition and ratification, I have put far away the renewal of any conflict with the fancy forming of the old yeast doctrine. But I regarded it necessary to show to those ignorant of botany, that in the three above mentioned points a large source of error



and false conclusion lies hid, which must be known to be avoided.

When one undertakes actual cultures which are free from the above censured errors, he will no longer retain those old dogmas.

I have in T. 2, Fig. 35, furnished an example, as the conidia of the before mentioned brand-fungus (Fig. 33), in the liquid medium its *Micrococcus* developes (T. 2, Fig. 34, *a-d*) as also the parent-cell subdivides (T. 2, Fig. 34, *b c, f g*), and at last, when the wall-membrane was dissolved the *Micrococcus* came out of its original confinement (Fig. 34, *e-g*). On dry nitrogenous soil *Micrococcus* also forms, but in an entirely different arrangement and is therefore wholly mistaken. I have already made observations on dry meat in dry air, and found that the mould-spores fallen from the air on the meat did not sprout, but formed *Micrococcus*. If the air is very dry, the *Micrococcus* is in chain-form. The chains (*Mycothrix*) anastomose on the side and throw out exceedingly soft fructifying threads, mutually ramified by means of fusions, and forming a proper fur. I have already shown that the *Mycothrix* fungus bears a close analogy to the *Sclerotium* forms.

Later I have found real *Sclerotium*, which was formed by the *Micrococcus* coming from common mould.

The form of *Sclerotium* is uniform. It appears everywhere when the fungus cell of similar form and origin developes in such large numbers that they cannot form sprout-threads on dry soil, for want of room. So, for example, appears the (*Mutterkorn*) ergot. In the sweet sap of the flowers of the cereals and grasses, we find, at first, some single yeast-cells or conidia of *Sphacelia segetum* Lev. These sprout and form fructifying threads of *Sphacelia*, the fallen conidia of which now come in such masses in the before mentioned fluid, that they absorb it and have no

more room to develop Mycelium. They extend any way, and are augmented still by means of subdivision. Thus they form the large, spongy and at last solid body called Mutterkorn.

Closely do they resemble the Micrococcus of Penicillium and Aspergillus in their peculiar appearance, and they have been observed lately in the hair made use of by hair-dressers.

Dr. Beigel, of London, had the kindness to send me such hair. It showed, here and there, knots (T. 2, Fig. 24, *a b c*) which embraced the hair wholly or in part, and already with low magnifying powers showed very tender cell-structure. The smallest and youngest knots consist of Micrococcus, which, in small heaps, covered the respective spots. In the larger knots are much larger cells (T. 2, Fig. 25), which have so flattened one another, that they form a false mycelium. It can now be plainly seen, that each possesses a large nucleus, and that it is about to be divided into two or four parts. Therefore the formation remotely reminds one of the quartering in the lower Algae (Merismopædia, Pleurococcus and others), but there can be no doubt, at first sight, that here a fungus is represented. To this its optical and chemical condition witnesses.

The cells in the compact knot exhibit, in their multiplying, great resemblance with those which I have called colony-yeast (Sarcina-form), and which, in fact, belong there, in so far as their origin is concerned. Only the aggregation is here different. By the close crowding together of a very large number of fungus cells, which are still in continual division, there is formed mycelium closely like ergot, i. e. a Sclerotium. Certainly it is proper henceforth to designate this knot "Sclerotium Beigelianum," when it is recollected, that Sclerotium is no especial fungus-genus, but a morphism appearing with many fungi. To find out to what fungus the



*Sclerotium Beigelianum* belongs, I cultivated the same, in air saturated with moisture. The *Sclerotium*, by and by, fell apart into its cells, whereby the parent cell was dissolved (T. 2, Fig. 26).

Each cell sprouted (Fig. 26, *k*) and brought forth, after a few days, pencils of *Penicillium* (Fig. 26, *p*). Among some few *Sclerotium* there formed only the sprouts of *Aspergillus*, so that we must grant a part in the knot-form to both fungi.

Therefore the whole process consists in this:—that the *Micrococcus* of *Penicillium* crust. Fr. and *Aspergillus glaucus* Lk., develope on the hair; that the *Micrococcus* cells swelled to sporoids, which did not come to sprouting, but, being crowded together in great numbers, were limited by subdivision, in one or two directions. Thus they formed *Sclerotium*.

It occurred to me to prove this explanation by an artificial nursery of this *Sclerotium* from the spores of *Penicillium* and *Aspergillus*.

I sowed the spores of *Aspergillus* on a well disinfected cork in the culture apparatus. On the cork I put some of my light hair. The spores which fell on the hair formed *Micrococcus*. These lay in very small heaps on the hair (T. 2, Fig. 27). The *Micrococcus* quickly multiplied by division, and formed large knots on the hair. In these the cells swelled to sporoids (Fig. 28), and now began the subdivision of the nuclei. This, thus formed *Sclerotium*, fell to pieces in a moist room, as is the case with *Sclerotium Beigelianum*; the single cells sprouted and brought forth on the hair bright *Aspergillus* pencils. (T. 2, Fig. 27, *sp.*)

## XI.

## APPARATUS.

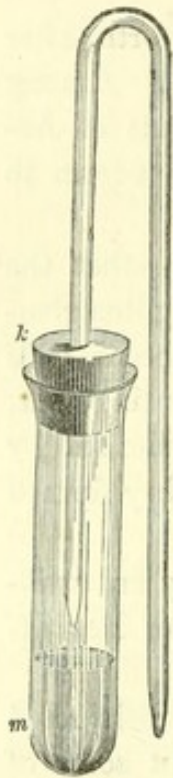


FIG. 1.

\* \* \* The most simple apparatus used by Professor Hallier, is represented at Fig. 1, as copied from his work on "Fermentation," and there described. It consists of a glass test-tube, first boiled out and cleansed by alcohol. It is then provided with the previously heated matrix (*m*) and hermetically closed by the cork (*k*), through which the inverted U-shaped tube (*r*) was passed, which can be easily removed as may be desired, only taking care that it shall not touch the matrix. I now use this apparatus so that, before putting the cork in its place, the matrix is boiled in the tube for two minutes. For negative experiments (tests), the cork with the tube can be put in while the boiling is going on. Only in this case we must proceed very cautiously. I cleanse, before use, in the following manner. If the vapor, at 100° C., is allowed to escape through the drawn end of the tube (*r*), it may be regarded as clean; but, for the sake of precaution, the tube is filled with pure alcohol, and allowed so to remain about an hour, generally until the matrix in the test-tube is heated and the substance to be cultivated introduced, which is accomplished as follows:—

The tube, filled with alcohol, is quickly emptied and as quickly dipped into the matrix with the same end which shall come into the vessel (or test-tube). Meanwhile the lamp beneath is blown out and the cork inserted as soon as the boiling has subsided. Where the substance to be examined is liquid, a certain quantity adheres to the entrance



of the tube if the outward opening is small enough. If the substance is solid, as, for instance, the scales and scurf of human exanthems, they adhere to the outer and inner part of the tube. This, at first, is pushed only so far down that it is found with the lower part still high above the matrix. The air ascending only from without through the bent tube towards the test-tube, by degrees a portion of the substance to be examined is pressed downwards; when the matrix is sufficiently cooled, through tilting and shaking, the adhering substances are brought in connection with the substratum.

I have made this apparatus of various calibre and form, and used it constantly with good results. With the greater number of substrata, I took a flask instead of the test-tube. PASTEUR says, positively and clearly, that a similar open apparatus gave as distinct and satisfactory results as the more complicated, and I have never neglected, when a substance was to be examined, to make controlling experiments.

\* \* \* \* \*

If one would have pure and clean air, he must use a pressure or suction apparatus. I have used both kinds, but to the latter (the air-pump) I give the preference.

I employ compression by means of gasometers such as the chemists use. The air is passed by suitable pressure. In use the upper part of the gasometer is connected by means of an air-tight tube, which conducts the passing air through boiled cotton and sulphuric acid. The cotton is put into a larger tube which it fills for some centimetres. The sulphuric acid is put into the well-known sulphuretted hydrogen apparatus. The culture apparatus, a flask in this case, bears a U-formed tube, and the air escapes as I press more in.

Naturally we can pump in the air more directly through clean tubes into the apparatus; however, the above described method has the preference on account of a more equal pres-

sure. I have used lately and for greater convenience, half the suction apparatus. For this purpose an aspirator, in general, is used; still it has many inconveniences. I make use of the air-pump.

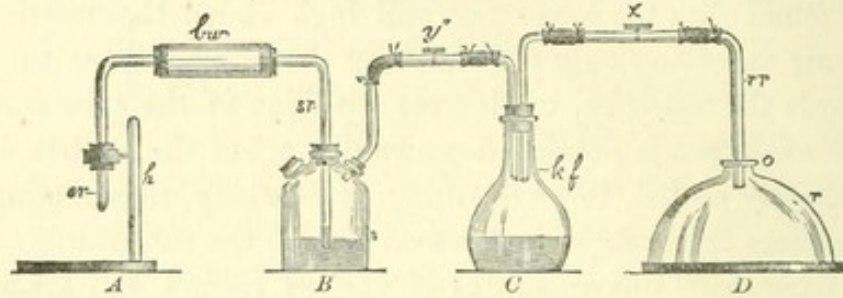


FIG. 2.

Figure 2 shows such an apparatus ready for use. *C* is the flask which contains the substance to be examined. It stands on the tripod of a Berzelius lamp. Its cork is doubly perforated for the bent tube. The matrix is heated in it, corked, for two minutes. Thereupon the substance to be examined is gently placed; afterward the glass tubes *x* and *y* are put in their places; these are capable of being closed by means of a stop-cock *x* and *y*. After the deposit the previously opened tubes are allowed to remain so. The tube *x* stands connected with the bell-glass of an air-pump whose plate is represented at *D*. A cork is adapted to the opening *o*, through which the tube *rr*, which is in connection with *x*, is passed. The tube *y* is in connection with the sulphuric acid apparatus *s*. The tube *sr*, partially filled with cotton, is introduced through the corked middle opening and terminates below the sulphuric acid, through which the air is admitted as the air-pump is worked. The sulphuric acid apparatus stands on the table *B*, as also does the holder *h*, which supports the tube containing the cotton, on table *A*. As a proof of the air-tight closure, appears the rising bubbles in the sulphuric acid, while the air-pump is slowly worked.



As soon as the substance to be examined is put into the flask the air is slowly pumped out of the whole apparatus, which is to be repeated once or oftener every day.

As is readily perceived, this apparatus admits of many modifications. Which to choose may be found explained hereafter. Still, I will here remark, that with aspirators or expulsors, it is better that the air should be passed through alcohol rather than through sulphuric acid, even in using the air-pump. In moderately concentrated sulphuric acid it happens that many fungi-elements are not killed, but actively vegetate, and it is questionable whether the acid in the highest state of concentration can kill them. The whole process must be regarded as a cleansing. At all events, filtration by means of cotton or other fibrous substances is indispensable; and then, even, the entrance of fungi-elements, by means of the air in the cleansing liquid, is not impossible.

\* \* \* \* For culture, to follow the vegetation step by step, we need a wholly open vessel, not a narrow-necked flask.

Very frequently we want to convey the organisms into the medium on the object-glass, sometimes under the glass cover. Here, then, is required a particular culture-apparatus, hence the following remarks. \* \* \* \*

In all the kinds of culture alluded to in my book on "Parasites," I have used the above specified apparatus, modified in a variety of ways; and now, for many years, I have from 20 to 30 of such apparatus in constant use in my various cultures, and have had occasion to use no other. The principle of the apparatus, in all its variations, is very simple. A flat dish (*s*, Fig. 3), is filled with water. In the water a small inverted dish with an even bottom is placed (*fs*, Fig. 3). On this, another small dish, *o*, is put, which contains the matrix and the substance to be examined. Over the object, the inverted bell-glass, *g*, is now

set in the water, so that the sowing is kept air-tight. Sometimes I use an open-mouthed bell-glass with the mouth

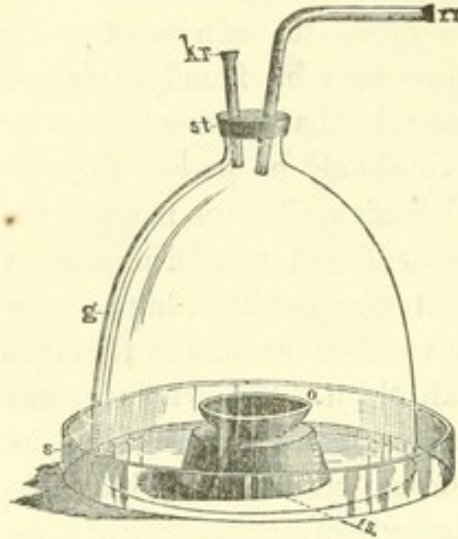


FIG. 3.

closed by a doubly perforated cork (*st*, Fig. 3), which permits the air to be removed without taking off the bell-glass. One of the openings of the cork carries a tube (*rr*, Fig. 3), which connects with the recipient of the air-pump. Another tube (*kr*) is placed in the other aperture of the cork to connect the cleansing apparatus. For ordinary purposes both tubes through the cork are closed.

In this modification the apparatus has the advantage, that the air can be renewed at different times before opening the bell-glass. Which of the above described apparatus will come into use, and which will be preferred, must depend on each determined case and the taste of the investigator. \* \* Immediately before culture all the pieces of the apparatus, &c., should be well washed in alcohol and rinsed in distilled water; and the matrix, previously heated for two or three minutes and until moderately cool, set under a clean bell-glass.



## XII.

## RÉSUMÉ.

*General Outline of the Views entertained by Professor Hallier.*

[Translated from an Essay of M. Baudouin, published in Professors Coze and Feltz's work upon "Infectious Diseases."]

"For a long time it had been believed that fungi, even the most inferior (moulds), might be divided into well characterized species. In 1851, M. Tulasne first discovered that the same species might bear very different kinds of fructification, and, that thus, many forms, which had been regarded as distinct species, were only modifications of one and the same species. A little later, in 1857, Bail pretended to demonstrate that ferments were no other thing than the most simple forms of certain moulds long known, as for example *Penicillium*. These ideas were adopted by Berkely and Hoffman. Hallier extended them farther. He thinks that he has proved that ordinary moulds reproduce diverse forms, and that these changes in the aspect of the plant depend on many causes: to wit, the free admission or exclusion of the air during their vegetation; its temperature, and the moisture or dryness of the matrix upon which the plant vegetates; the chemical nature of the elements upon which it feeds (hydro-carbons, sugar or azotized matter), &c. &c.

"I proceed rapidly to review the different forms admitted by Professor Hallier:—

"All moulds, even the highest forms, as Agarics, can act as ferment: he calls *Ferment* the forms or morphes which develope in the interior of a substance capable of undergoing fermentation (and consequently organic), of being nourished at the expense of this substance, and of producing

multiplied generations, which rapidly succeed each other, and always remaining one-celled. These ferments may come from the spores (organs of reproduction) of moulds, or, indeed, from the mycelium (that is to say, the vegetative organs). The moulds can present themselves under three different aspects :—

"A. That of *true ferment*. In this case, the globules remain separate, the one from the other, and are always suspended in the liquid.

"B. The *ferments in chains*. This second form, which M. Hallier calls *imperfect ferment*, is composed of globules agglutinated in filaments ; it appears when the air is imperfectly admitted.

"C. *Colony-ferment*—that when the first cell, instead of producing buds, quadruplicates itself (for example, *Merismopædia* or *Sarcina*) ; or, indeed, when it is composed of cells confusedly conglomerated.

"I proceed to examine what are the varieties met with in each of these divisions :—

"A. Of the true ferments there are three forms : that of *Micrococcus*, that of *Cryptococcus*, and that of *Arthrocooccus*.

"1. *Micrococcus* (*Microzyma* of Bechamp). This form is that where the spore-membrane of a mould, or indeed, that of a joint of mycelium, breaks and allows the escape of the plasma. If this protoplasm meets with organic substances capable of putrefaction, the granules of the *Micrococcus* organize, and take the form of a small sphere very often provided with a pedicle or tail, which enables it to move (*Swarmers*), when they multiply abundantly. The *Micrococcus* may be regarded as the promoter of putrefaction.

"2. *Cryptococcus*. If, instead of coming in contact with a substance capable of putrefaction, the protoplasm of the spore, or the *Micrococcus* already developed, is in contact with a substance slightly azotized or apt to undergo



alcoholic fermentation, the form changes. The *Cryptococcus* develops very rapidly, not by division, but by budding.

"3. *Arthroccoccus*. In fine, when the same organs already cited are put in a medium which has a tendency to take on an acid fermentation, they assume an elliptic, elongated form, and are called *Arthroccoccus*.

We may hence conclude, that Professor Hallier's theory is just the opposite of M. Pasteur's. He believes, that for every fermentation, a special ferment is needed; for example, that the *Micrococcus* of milk is requisite to produce lactic-acid fermentation, while M. Hallier, on the contrary, regards the form of the ferment as depending on the particular kind of fermentation: thus, if we put *Micrococcus* in a solution of sugar, we shall obtain *Cryptococcus*; if we put it into an alcoholic liquid in contact with the air, we get the acetic ferment, that is to say, the *Arthroccoccus*.

"B. One has only to suppose the diverse forms, enumerated in the first section, adhering together endwise, to have those of the second section.

"1. *Mycothrix*. These are no other than the filaments composed of *Micrococcus*, united together in a bead-form. This takes place when the *Micrococcus* is on the surface of the liquid, and consequently a little more exposed to the air, or when floating in an oxygenated liquid. These forms were formerly known as *Bacteria* or *Leptothrix*; but as the name *Leptothrix* had been previously appropriated to an Alga, Hallier prefers the nomenclature of Itzigsohn, which recalls the mycologic origin of this form.

"2. *Torula*. When the *Cryptococcus* are in the same condition as the *Micrococcus*, they produce like chains, only composed of larger joints, which have received from Mycologists the name of *Torula*.

"3. Mycologists have likewise given the name of *Torula* to

chains composed of the joints of *Arthrococeus*, and which appear under the same conditions as the two preceding.

"When the three kinds of chains exist at the surface of the liquid, they adhere together and form a kind of net-work, appearing to the naked eye as a membrane: this is the *Mycoderm* of Pasteur.

"C. Under this last division we have only the forms which extend to the surface, as *Sarcina*, or indeed which are divided irregularly in every direction, and form agglomerations.

"I pass to the more developed forms, i. e. to the **MOULDS**.

"*Moulds*. M. Hallier believes that the absence or presence of the air modifies their degree of development. Thus he divides the moulds into anaërophytic or aërophytic moulds.

"A. *Anaërophytic Moulds*. These are intermediate forms, a kind of transition between the last varieties of ferments and the moulds most developed. They appear only when the access of the air is hindered; for example, when they are plunged into the liquid or develop in the interior of a plant or animal: most of the varieties arranged in this category are true parasites.

"1. *Oidiate-form*. In this section we may place *Achorion Schænlinii* (*Oidium albicans*, &c.). These forms develop in the living tissues filled with liquid, or indeed in the interior of pasty or mucilaginous substances. The mycelium then ordinarily remains little developed and produces at the extremities of the numerous joints what is known as a chain of spores. We meet with them in *Favus*.

"2. *Corruption-forms*. These occur in very thick substances.

"a. The form of *Ustilago* develops in thick starch, or in the interior of the culms of certain grasses very rich in sugar. It produces ramifications which terminate in a chain



of brown, spherical spores. This form belongs especially to *Aspergillus*.

"*b. Tilletia-form.* This appears under the same influences as the preceding form; but instead of terminating in spores with a continuous membrane, it terminates in spores with a reticulated membrane; for example, *Penicillium*.

"3. *Schizosporangiolen-form.* Here are found the Moulds whose filaments penetrate the atmosphere to terminate in conceptacles enclosing confined spores.

"B. *Aërophytic Moulds.* These are the true moulds. They grow on the surface of the substances which nourish them and in contact with the air. They produce a developed mycelium and ascending filaments, which bear the reproductive organs. In this section we find again three different forms.

"1. *Acrospores.* These plants bear free spores at the extremity of the ramifications. It is in this section that *Penicillium* and *Aspergillus* are found. They are produced when the nitrogenous soil is moist. One can, therefore, according to Hallier, raise, at pleasure, *Penicillium* and *Mucor*, or *Aspergillus* and *Stemphyllium*, as we sow, indifferently, the spores of one of these forms on a matrix\_dry or moist.

"2. *Thecaspores* or *Mucor-forms.* In this category is found the *Mucor* belonging to the genus *Penicillium*, and *Stemphyllium*, which is only a form of *Aspergillus*. The *Thecaspores* spring up only on very nitrogenous and dry soil. The organs of fructification of these species are in the form of a large vesicle containing many spores. Hence from *theca*, the name given to the envelope, *Thecaspores*.

"3. *Sexual-form.* The highest development of the mould family presents, in fine, sexual organs, some of which are called *Oogonia*, corresponding to pistils, and *Antheridia*,

corresponding to the stamens of phanerogamous plants. The contents of the Oogonia are fecundated by the Antheridia, and produce durable and winter spores. I will cite, as an example of this last form, *Eurotium herbariorum*, which developes upon the dried leaves of our herbaria, and which belongs to the species *Aspergillus*."



## APPENDIX.

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### CULTURES AND RESULTS.

BEFORE entering upon the details of the following cultures, which were intended to be like those of Prof. Hallier, it should be stated, that his work\* in which the apparatus used by him was described, was not to be procured in this country, and we were left to devise our own; which consisted of glass tubes from four to five inches in length and somewhat more than half an inch in diameter, closed at both ends by a perforated cork, for the admission of smaller glass tubes bent downwards at right angles and which were stuffed with clean cotton. Also open-mouthed phials of about one and a half fluid ounces capacity, whose apertures were closed by a cork doubly perforated, for the insertion of small glass tubes stuffed with clean cotton, for the extraction and renewal of the air, by suction, and for straining out any spores that might be floating about. Care was taken, if possible, to prevent the occurrence of anything that would have a tendency to vitiate the results of the cultures, although it must be confessed that had we known of the apparatus Prof. Hallier used, we should have endeavored to follow him more closely.

FIRST CULTURE. On the 21st of April, 1871, sowed vaccine lymph on a paste made with a boiled solution of phosphate of ammonia and starch, in a clean, open-mouthed phial whose aperture was closed by a clean doubly perforated cork through which passed two glass tubes, stuffed with cotton moistened with alcohol.

On the 27th of April, examined a bit of the paste with B.5 (350 lin.). Saw a few small dark-colored points in

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\* "Gährungserscheinungen."

the liquid around the starch granules, none within them. It did not occur to test with an acid, so that it must be left doubtful whether this motion was molecular (Brownian) or not. Within the starch, as seen under C.10 (1200 lin.), were very many minute dark *still* points; many of the starch granules were, as usual, cracked and broken, while some were entire.

On May 5th, put a piece of kine-pock scab on the same paste, pressing it partially into the paste. These phials were allowed to remain unopened for more than a month, but no fungus whatever was to be seen. *Quere*—Were the fungi-spores from the outside and within (had any been admitted and included) destroyed by the vapor of the alcohol? Hallier says, "Alcohol is a disinfectant, destructive of spores."

SECOND CULTURE. May 5th, 1871, planted a piece of kine-pock scab on lemon. On the 9th found a fruitage, which was pronounced to be a bastard between *Penicillium* and *Aspergillus* (see Figs. 6, 8). In this culture the pellets of cotton had not been moistened with alcohol.

THIRD CULTURE. May 10th, 1871, placed pieces of kine-pock scab on boiled white of egg in two of the glass tubes previously described. In the first tube (1) the cotton was moistened with alcohol, in the second (2) it was not. On the 12th, examined with A.C.10 (425-1200 lin.), and found in tube 1 many moving points (probably molecular movement). In tube 2, the white of the egg exhibits signs of commencing decomposition. Saw specimens of *Mucor mucedo* (T. 3, Fig. 4), with burst sporangia and scattered spores. The *Mucor* is a morphism of *Penicillium*. May 16th, in the same tube (2) *Mucor* was found with interstitial sporangia (Fig. 7, T. 3). Eleven days had now passed, but no fungus was to be seen in tube 1, i. e. where the cotton had been moistened with alcohol. This state of things, in tube 1, remained the same for a week or more, while the white of egg in tube 2 was covered with fungi.

FOURTH CULTURE. May 24th, 1871, put a piece of kine-pock scab on starch moistened with a cold solution of phosphate of ammonia, into one end of the clean tube A, while at the other end of the same tube was placed a piece



of the same scab on paste made with a solution of phosphate of ammonia and then boiled. At the same time and place, a mixture of a cold and also a boiled solution of the above salt and starch, but without any kine-pock matter, was put into another similar clean tube, B. The small tubes, bent at right angles and plugged with clean cotton not moistened with alcohol, were passed through a perforated cork which closed each end of the tubes. I then sucked through the small tubes twenty times to strain out and prevent (if this would do it) the presence of any spores.

May 26th, 1871, saw a faintly red spot, which on the 27th became very distinctly of a pinkish color on the unboiled starch pellet in tube A. A similarly colored spot appeared on the bit of boiled paste at the other end of the tube (A). These colored spots were at a small distance from the pieces of scab.

On the 25th of May, I vaccinated three children and two infants from fragments of the same scab which I had used in the above culture. In three of the cases normal vaccine vesicles formed and matured on the 8th and 9th days; in one, the constitutional symptoms were very strongly marked. The vaccination failed in the infants, probably from too superficial puncture.

The *Micrococcus* of kine-pock, as well as those from *Torula rufescens* (the fungus which Hallier believes to be the source of kine-pock *Micrococcus*), are both reddish; the former, he says, is of a wine-red color, the latter of a darker red. The color before me on the unboiled pellet of paste in tube A, is of a pinkish color; that on the boiled pellet is a little more of a wine-red color.

To the above remarks upon the color, it may be objected—that fresh and active kine-pock lymph is colorless. So is newly crystallized biniodide of mercury; but if we touch one of these crystals with the point of a needle, the whole crystal becomes instantly of a beautiful red color, while it still remains biniodide of mercury, the arrangement of its particles only having changed; and in this manner the difference in color between vaccine-lymph and vaccine-scab, may be explained, as well as by the multiplicity of the *Micrococcus*.



May 29th, examined a bit of the reddish part on the unboiled paste, with C.10 (1200 lin.). Found Mycelium-threads and some very small spores, which proved to be *Penicillium* crust. with *Macroconidia*; the same fungus was found on the unboiled paste in tube A, not colored red. None of the characteristic cells of *Palmella* (which was suspected) could be found.

June 1st, again examined portions taken from the reddish colored spots, tube A, with C.10 (1200 lin.); find in that from the unboiled pellet large numbers of moving *Micrococcus* and starch granules, nothing else; and as the starch is white, the reddish color would seem to come from the color of the *Micrococcus*.

In a portion taken from a part not colored, sprouts of *Aspergillus* and *Penicillium* were found. In a part of the boiled paste from tube A, where the color was wine-red, find a great number of very active *Micrococcus*, and but very few starch granules. Examined, the same evening, parts of the contents of tube B (where there was no kine-pock matter); found very fine mycelium and many spores, apparently of *Penicillium*, but no bastard forms; some resting or still *Micrococcus*.

FIFTH CULTURE. On June 2d, 1871, after carefully cleaning two open-mouthed phials as well as the tubes, sowed on boiled milk in A, kine-pock, by means of freshly charged quills, as also by vaccine scab; while in phial B, no kine-pock was placed, but only boiled milk.

June 5th, examined with C.10 (1200 lin.), what was on the cork of phial A, and found from the under surface of the cork very beautiful specimens of *Aspergillus glaucus* (Fig. 9, T. 3). Examined also the cork of phial B, and found only a few spores.

June 8th, examined some of the surface of the milk in phial A (where the kine-pock matter was present); find *Arthrococcus lactis*, also some whirling and rapidly moving bodies, probably vibrios. Examined also the under surface of the cork of phial B (where there was no vaccine virus); find only a few spores, supposed to be from *Penicillium*, but no *Aspergillus*: from near the surface of the milk were multitudes of fat-globules, some moving *Micrococcus*, but no *Arthrococcus*.



SIXTH CULTURE. June 14th, 1871. Repeated the fourth culture and with similar results. In two hours after the sowing I saw on the boiled paste, a reddish spot, which gradually faded away.

June 17, evening, examined with A.10 and C.10 (425. 1200 lin.) the contents of the tube, where bastard forms between *Aspergillus* and *Penicillium*, and what appears to be *Oidium* (Fig. 10, T. 3), were found.

SEVENTH CULTURE. June 17th, 1871, a cork was well washed in alcohol and a fresh surface given it. It was applied, with some *Micrococcus* of kine-pock on its under surface, to an open-mouthed phial, A, previously cleansed by washing in alcohol, and then with clean water, and set away. On the 18th, the same was done with the phial B, except that no vaccine matter was placed on the cork, and this phial placed alongside the other.

June 21st, evening, examined with C.10 (1200 lin.), the fungus on the under side of cork in phial A, and found much *Penicillium* (T. 3, Fig. 3) and some bastard forms between *Penicillium* and *Aspergillus*, but no perfect *Aspergillus*. On the corresponding part in phial B, no fungi were found.

June 23d, 1871. Having noticed, for a few days, that the unboiled paste in the tube where the kine-pock matter was placed on the 14th of June, was becoming of a reddish-brown color, it was examined with C.10 (1200 lin.), and we found that this color was owing to the conidia, which had fallen in great numbers from the sterigma. *Oidium* (Fig. 14, T. 3), and moving *Micrococcus* are also seen.

Professor Hallier says, "An *Oidium* first precedes *Mucor*, which could be but with difficulty distinguished from *Torula rufescens*." \* \* \* "I have said that the *Torula* was an *Oidium*, that *Oidium* belongs to *Mucor*, which shows at the commencement, small, pale, reddish-brown conidia, closely like *Torula rufescens*."

June 24th, saw a pink spot on the white of egg (repetition of Culture 3), where on the 19th were placed pieces of kine-pock scab in tube A, and none in tube B. Strong mycelium was also seen shooting forth at a distance from the scab. Nothing in tube B.

June 25th, examined by reflected sun-light a portion of



the pale rose-red heap on the white of egg, which to the naked eye appears to be a *Tubercularia*. A bit of this commingled with water and placed on the glass-slide, resolves itself into a small cloudy spot, which under C.15 (1800 lin.), appears to be cells. See Fig. 11. Examined also the fungus, which proved to be *Penicillium*. In corresponding tube B, saw nothing.

June 27th, noticed in tubes A and B, some dark-colored spots on the boiled white of egg, where on the 30th was found under A, C.10 (425-1200 lin.), *Oidium*-like sprouts closely resembling Figs. 11, 12, of Professor Hallier's plate 1, and also *Cryptococcus*.

**EIGHTH CULTURE.** Rubbed up some kine-poek scab with pure and fresh pump-water and put a bit on the under side of a clean and freshly cut cork, which closed a phial in which was left a little of the water. On the 30th examined the above, which exhibited specimens of *Penicillium*.

On July 4th, 1871, I took vaccine-lymph from a young lad, some in capillary tubes, and some on a piece of clean glass or points.

July 5th, examined some of the above lymph, mingled with a minute drop of pump-water under C.10 (1200 lin.), and saw moving *Micrococcus*, on one of which it was thought could be seen what Hallier considers as the moving organ (the tail); this cilium appeared to be swinging in the direction of an obtuse angle. Some of the lymph blown out from the capillary tube was next examined, and although some small bodies were found, none were in motion.

**NINTH CULTURE.** July 5th, boiled a filtered solution of the phosphate of ammonia and added starch; boiled the starch with a small tube in the cork, so as to allow for the escape of steam and air; then, having put a portion of the vaccine lymph upon the drawn out end of the small tube, brought it just into contact with the paste; then slightly withdrew it. July 8th, a portion of the above was examined, but nothing found.

On the same day, under a fine light reflected from a house painted white (almost equal to the sunlight reflected from a cloud), examined a portion of the vaccine lymph from a capillary tube with C.10 (1200 lin.), and saw many bright bodies, some in motion, some quiet.



With C.15 (1800 lin.), saw *Micrococcus*, found in a liquid, where spores of *Penicillium* were put, about two months since: saw the cilia (or tails), a part of the time in motion, a part of the time at rest. This statement is made only to show the optical quality of the microscope used in the above examinations, and for comparison of the small bodies found in vaccine lymph and their instrument for motion.

July 10th, 10 A.M. Under bright light from the reflected sun-light, examined with A.10, the liquid paste (ninth Culture), and find *Penicillium* and bastards between it and *Aspergillus*. Above the surface of the liquid, the form is that of *Penicillium* (T. 3, Fig. 3), below the surface of the liquid, *Oidium*.

On the 13th of July, examined the under surface of the cork (Culture nine), and found *Aspergillus*.

July 14th, took some vaccine lymph in a capillary glass tube, from a young child vaccinated on the 5th instant, which was immediately examined (i. e., as soon as I could reach my office). With C.15 (1800 lin.), found many moving corpuscles, some of which strikingly resembled the two right hand forms figured in Dr. Beale's "Disease Germs, Their Real Nature," Fig. 39. Upon examining another drop of the same lymph, saw more cells, some resembling the spores of *Aspergillus*, others of about the same size, appearing like empty cysts, resembling those figured by Hallier, and presenting a trembling motion (T. 1, Fig. 5).

July 18th, 1871, examined with C.15 (1800 lin.), vaccine lymph taken (within half an hour) from the vesicle on the arm of an infant vaccinated ten days since; the vesicle was umbilicated, well-filled, and surrounded with the areola; many moving corpuscles, mostly round but smaller at one end, were seen. The motion resembles that of the swimmers of *Algæ*.

TENTH CULTURE. August 9th, 1871, planted vaccine matter on boiled white of egg, and placed in the tube; then sucked through the tube forty times.

Aug. 22d, the egg had become quite dry and covered with a fungus. Examined with A.10, on a dry glass slide, and found *Torula* chains, some free, some still standing on the branched hyphens (Fig. 15, T. 3).

In conclusion, while I would not presume to pass judgment on Professor Hallier's hypothesis upon the causes of zymotic diseases, I desire to state, that I have attempted a plain and simple statement of facts in the cultures made, and in the results obtained; but must confess that it was gratifying to find, under like circumstances, the same vegetable forms: viz. *Penicillium*, *Aspergillus*, the bastard forms between these two fungi, *Oidium*, *Mucor* and *Torula*, and to see the moving corpuscles in the fresh lymph of the vaccine vesicle: while it may be admitted, with Drs. Billings and Curtis, that "the laws of development are not sufficiently known to enable one to draw decisive inferences from such results."



## EXPLANATION OF THE PLATES.

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### PLATE I.

- FIG. 1. Zeiss system F. Ocular 2. Swarming Micrococcus of small-pox.
- FIG. 2. Zeiss F.2. Cryptococcus forming in twenty-four hours from the Micrococcus of small-pox raised on glycerine, under an hermetically sealed bell-glass.
- FIG. 3. Z. F.2. Mycothrix chains from the small-pox. In the links of the chain is seen a dark nucleus.
- FIG. 4. Z. F.2. Swarming Micrococcus from sheep-pock, here and there Mycothrix chains, with a nucleus in each joint (*k*).
- FIG. 5. Z. F.2. Quiet Micrococcus from the Munich kine-pock fluid.
- FIG. 6. Z. F.2. Micrococcus from the sheep-pock, cultivated on sugared water. The Micrococcus cells are for the greater part grown to short Leptothrix or Mycothrix chains, Bacteria (auct).
- FIG. 7. Z. F.2. The same culture. At the edge of the liquid and at the bottom of the vessel, the Micrococcus cells slowly swelling to large clear sporoids, ready to sprout, with one or more nuclei, after culture of fourteen days.
- FIG. 8. Z. F.2. The same culture. The large round cells (Sporoids), which swim in the liquid, discharge their nuclei, which were formed from a plasm investing the wall.
- FIG. 9. A sprout of Sporoid (Fig. 7), bearing in every branching thread a chain of at first oval, at last round spores, which become pale in the fluid and discharge their contents (Fig. 8).
- FIG. 10. End of some sprouts at the brim of the vessel. At every termination is a chain of brown spores. There lay thrown out spores partly roundish, partly cladospore-like Septate (*cl*), partly very large and pale bodies (*m*), scattered in the fluid.
- FIG. 11. Sprouts from the Micrococcus of Sporoids, germinating on the white of an egg. The spores were drawn out in an irregularly branched pencil.
- FIG. 12. Micrococcus of sheep-pock, slowly swelling to cells able to sprout (Sporoids), which already, here and there, have put forth long shining Oidium-like sprouts.

- FIG. 13. Spore-bearing thread of the sprouts from sporoids within the white of egg.
- FIG. 14. Similar thrown off Tilletia-spores, getting pale in the liquid, forming Micrococcus.
- FIG. 15. Sprouts from sporoids with Sporidesmium fruit at the brim of the vessel.
- FIG. 16. Similar fruit with vigorous or swelling, divided spores.
- FIG. 17. Another fruit branch from the same culture.
- FIG. 18. Culture on paste and tartrate of ammonia. Micrococcus forming. The Micrococcus cells, in some places swollen to Sporoids (*sp*), multiplied largely within the soil. Single Mycothrix chains with nuclei were to be seen (*m*).
- FIG. 19. Micrococcus in round, and changing to rod-shaped Arthrocooccus, raised from the Micrococcus of the sheep-pock lymph on pear.
- FIG. 20. Full-grown Arthrocooccus from the same fruit, forming a close Mycoderm.
- FIG. 21. Thread with six Sporidesmium fruits, from the white of egg culture.
- FIG. 22. Culture from paste with the tartrate of ammonia. Fragments of a vegetative Mycelium thread.
- FIG. 23. Pale red thread from the same culture, at the brim of the vessel beginning to fructify.
- FIG. 24. Farther stage of fructification.
- FIG. 25. Spore formation from the same culture.
- FIG. 26. Sprouting Mycothrix-fungus, with tender Cladospore-fruit, from the same culture.
- FIG. 27. Culture on lemon, Cladospore thread with Macrospore ; (*m*), Monilia cinerea Bon.
- FIG. 28. Rhizopus nigricans. Lemon culture, seen by an instrument of Beniche. System 4, Oc. 1.
- FIG. 29. Penicillium grande from the same culture, produced from the sprouting thecasore of Rhizopus. Shown by the same optical power as Fig. 28.
- FIG. 30. Fruit-bearing branch of the same Penicillium, as seen with Zeiss F.2.
- FIG. 31. Pleospore from Lolium perenne.
- FIG. 32. Monilia cinerea Bon., obtained from Pleospore on pear. (*p*), a Puccinia-like conidia ; (*m*), a Macroconidia.
- FIG. 33. Oidium-lactis, appertaining to Monilia-Rhizopus-Botrytis.
- FIG. 33 *a*. Monilia seen in a succulent pear, sprouted and forming a Penicillium thread ; (*k*), Normal sprouts at the end.
- FIG. 34. Sprout of Rhizopus nigricans Ehr., from an apple.
- FIG. 35. Arthrocooccus from the interior of a spore of Rhizopus borne on pear. Transition stage of Micrococcus to Arthrocooccus.
- FIG. 36. Spore-bearer (Hyphen) of Botrytis elegans Corda. From the Monilia raised on pear.



- FIG. 37. *Rhizopus nigricans* Ehr. (as seen with a good lens), covered up by *Penicillium*.
- FIG. 38. A single theca somewhat magnified, with *Rhizopus* twining up the bearer.
- FIG. 39. Spore of *Botrytis* sprouting to a *Rhizopus* thread.
- FIG. 40. *Rhizopus nigricans* Ehr., hanging downward from an apple peel, as seen by a good lens.
- FIG. 41. Young sporangium of *Rhizopus* at the commencement of spore-forming.
- FIG. 42. Sporangium of *Rhizopus*, the spores partly discharged through the cracked wall of the sporangium. From its bearer ascends a club-shaped mass of plasma.
- FIG. 43. Wholly empty sporangium of *Rhizopus*. The wall is torn away and a shred (*w*) only is visible. The club-shaped mass of plasma is furnished with a new wall, with a distinctly two-fold outline (*c*).
- FIG. 44. Amylum nuclei from the culture of kine-pock lymph on paste and phosphate of ammonia. The starch nucleus tears from the centre (*a*). In these rents, after a short time, the Micrococcus settle and largely multiply. After twenty hours, is seen the centre of the nucleus and often the layers filled with Micrococcus. Frequently the layers break into small fragments.
- FIG. 45. Amylium nucleus from the same culture, some days after the sowing. The Micrococcus cells are mostly swollen to Sporoids, which, in part, are already sprouting, bursting and lacerating the nucleus until it is wholly dissolved.
- FIG. 46. Example of *Mucor mucedo* Fr., as seen by a good lens, appearing on the cork in the same culture.
- FIG. 47. Macroconidia of this *Mucor* raised from *Aspergillus*.
- FIG. 48. Filled sporangium of the same *Mucor*, in the under part of the bearer Macroconidia, come to development.
- FIG. 49. Discharged sporangium of the same *Mucor*; the round stalk-cell (Columella) is seen.
- FIG. 50. Culture of kine-pock on lemon, fourth day. Formation of sporoids from the Micrococcus, at *k* a sprouting sporoid.
- FIG. 51. Macroconidia from the same culture.
- FIG. 52. Culture of kine-pock fungus on the white of egg. Spores of *Cladosporium* (*Oidium albicans* auct.).
- FIG. 53. Sprouting sporoids at *k*, from the same culture.
- FIG. 54. Bastard between *Aspergillus* and *Penicillium* on the cork of the culture of kine-pock lymph in milk.
- FIG. 55. *Arthrocooccus lactis*, originating from the Micrococcus of the same culture.

## PLATE II.

- FIG. 1. *Mucor* from the culture of kine-pock on paste and phosphate of ammonia placed on a cork. *p l* are the swellings of the tender *Oidium* threads. At *v* is seen the Macroconidia of some chains united. *k* is the empty *Mucor* capsule with a round basal cell, and some attached spores.
- FIG. 2. *Cladosporium* (*Oidium albicans* auct.) from the culture of vaccine-lymph on white of egg. *u*, *Ustilago* spore-chains not yet arrived at maturity, because they immediately shoot forth cladospores (*c l*), whose young spores (*o*) closely exhibit the nature of *Aphthous*-fungi.
- FIG. 3. *Aspergillus glaucus* Lk., from the sowing of the vaccine-lymph on cork, at *s* the mostly hollow bearer of the pencil. *s p*, spores; *l s p*, these in air; *a s p*, the same in alcohol.
- FIG. 4. *Arthrococcus lactis*, brought forth from the small-pox matter on boiled milk.
- FIG. 5. A bastard between *Aspergillus* and *Penicillium* from the same culture.
- FIG. 6. A fur of *Cladosporium* (*Oidium albicans*), appearing on lemon from the sporoids of *Micrococcus* of small-pox, under a good lens.
- FIG. 7. The same fungus in fragments with Z. F.2. It showed *Cladosporium* chains (*c l*); *Ustilago* spores (*u*), in part very large and often septate (*u u*), large terminating round cells (*p*), partly with a mitre-formed end cell (*p m*); in part developed *Septosporium-Stemphyllium* fruit (*s p h*), partly becoming *Pycniden* (*p p*), with sporoids (*s p*).
- FIG. 8. *Sclerotium* appearing on the cork of the sowing of small-pox on sugared water with phosphate of ammonia.
- FIG. 9. Crypto-crystalline formation in Typhus-stool from Munich.
- FIG. 10. Vegetable organisms appearing in the monkey's stool. (*a*), thread-sprout; (*b*), fungus cells; (*c*), *Mucor* spores; (*d*), *Micrococcus*-balls from the nearly dissolved spore-wall, still holding together.
- FIG. 11. From the same, twenty-four hours after feeding with the spores of *Rhizopus nigricans* Ehr.; (*a*), *Mycothrix* chain; (*b c*), *Micrococcus* formation within the parent cell in two different stages; (*d*), conidia, one of which is about sprouting.
- FIG. 12. Yeast and thread formation in human excrement, after the inhalation of *Rhizopus*.
- FIG. 13. Sporoid-formation from the mucous membrane of the throat.
- FIG. 14. *Micrococcus* and therefrom produced *Mycothrix*-chains on epithelial cells.
- FIG. 15. Fungus-formation in the monkey's intestine, on the fourth day after the feeding with *Rhizopus*.
- FIG. 16. Fungus-formation in human faeces, sixteen hours after the feeding with *Rhizopus*.



- FIG. 17. Yeast-formation in the monkey's stool, on the third day after feeding with *Tilletia caries* Tul.
- FIG. 18. Fecundation-ball of *Ustilago carbo*, surrounded spirally with a thread.
- FIG. 19. (a) Double-spore (Thecasporium) from an Ascus of *Corticium amorphum*: every half spore has pushed out a sprouting tube. (b c) Ascus of *Corticium* cultivated in glycerine on the object glass; all the spores have sprouted; the Ascus is broken through.
- FIG. 20. Swarming Micrococcus of *Penicillium crustaceum* Fr., as shown by the immersion system 1-18 Merz, Oc. 4.
- FIG. 21. Swarming Micrococcus cells of *Corticium*, drawn with 1-18, Oc. 4.
- FIG. 22. Swarming Micrococcus (from Munich typhus stool), belonging to *Rhizopus nigr.* Ehr., drawn under the same system.
- FIG. 23. Swarming Micrococcus of *Mucor mucedo*.
- FIG. 24. Brown hair with *Sclerotium Beigelianum*. Z. system D. Oc. 2.
- FIG. 25. A small fragment of such, under Z. system F. Oc. 2.
- FIG. 26. Sprouting cells of a *Sclerotium* bursting in moist air; at (k) a longer sprout; at (b) a fructifying pencil of *Penicillium*, Syst. F, Oc. 2.
- FIG. 27. Artificially raised *Sclerotium* from *Aspergillus*, near by a fruit pencil (*as p*) from light hair. Z. syst. D. 2.
- FIG. 28. Some cells of such *Sclerotium*, partly sprouting. Z. F. Oc. 2.
- FIG. 29. Oidium of *Rhizopus*; the cells have partly discharged their nuclei, which, outside the parent cell, form nuclei and Mycothrix chains.
- FIG. 30. Sproutings on the joints of *Favus*-fungus like cryptococcus.
- FIG. 31. Development of spores of *Aspergillus* from the end twigs of *Sterigma*, at x.
- FIG. 32. The same from *Penicillium*.
- FIG. 33. Origin of Conidia from side-sprouts of the thread-joints of a Pleospore.
- FIG. 34. Development of Micrococcus from the Conidia, and joints of the same fungus.











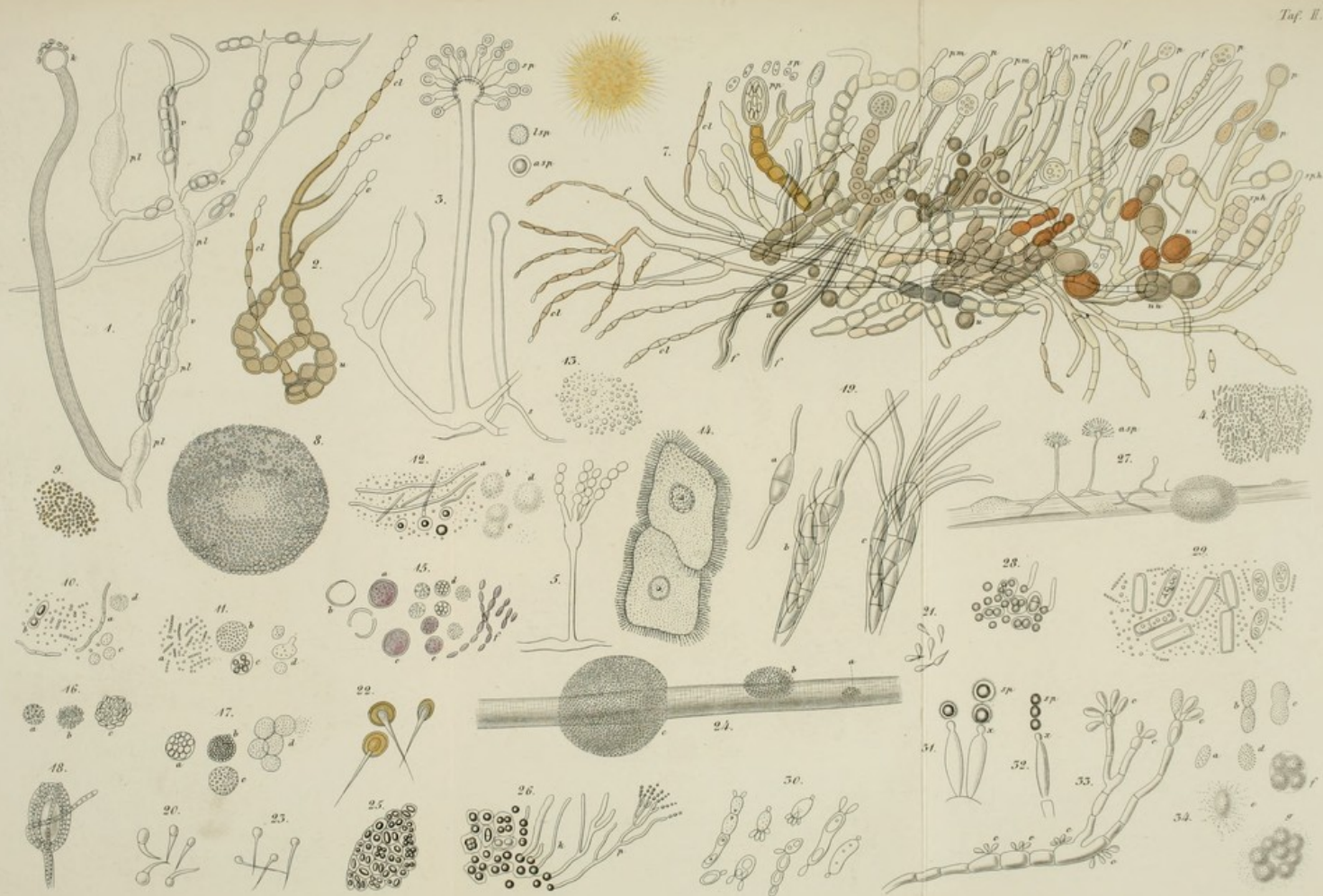
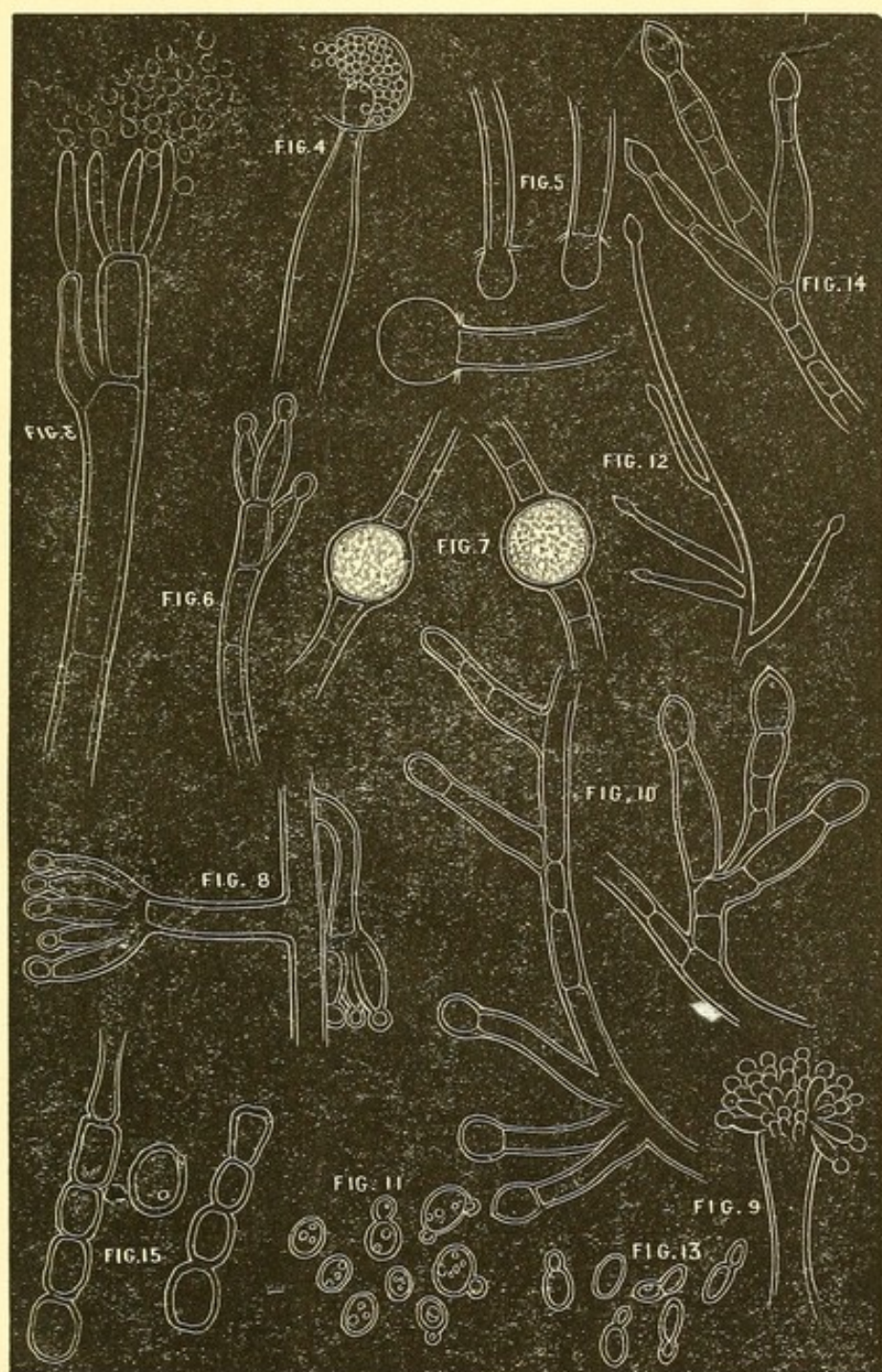






TABLE III.



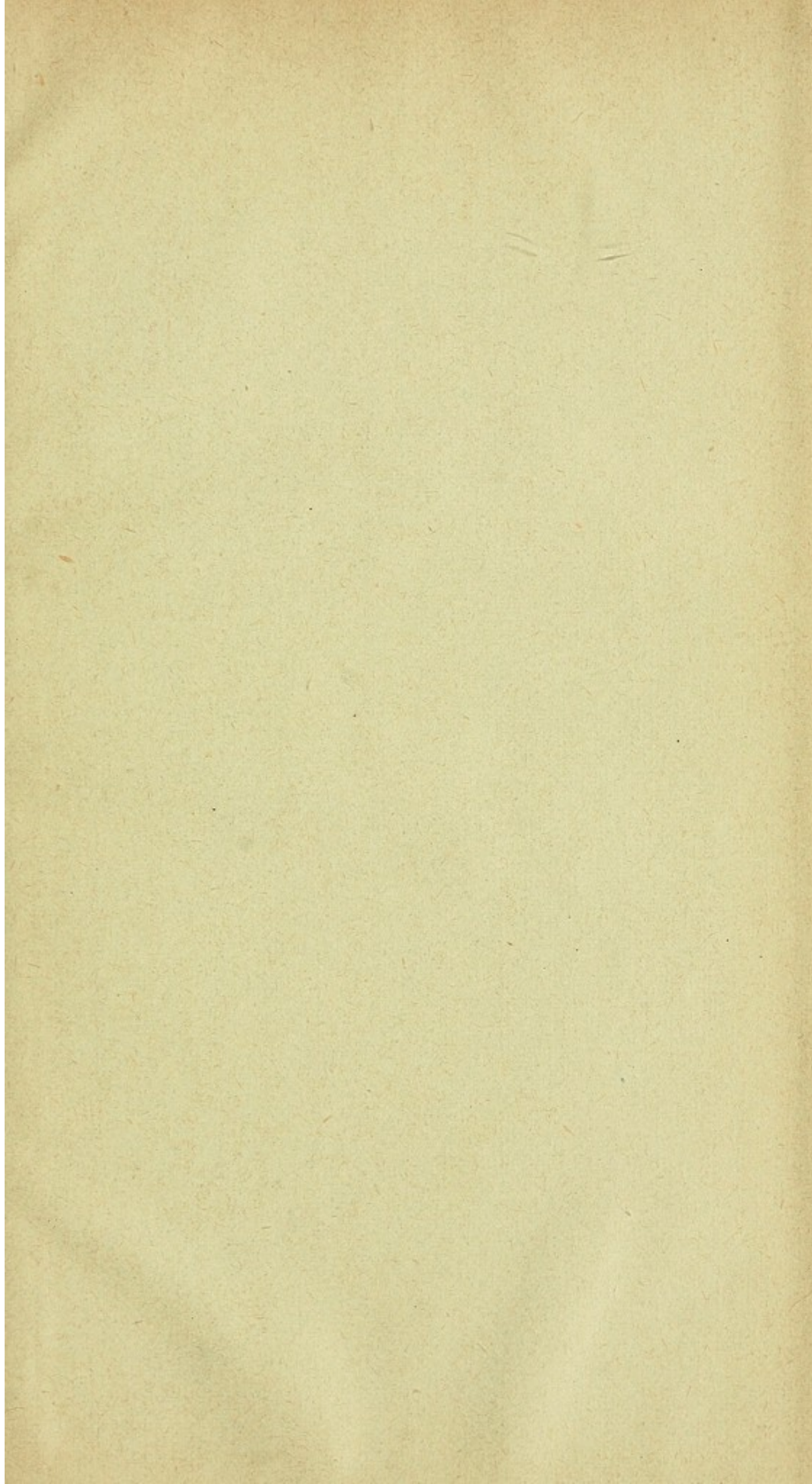


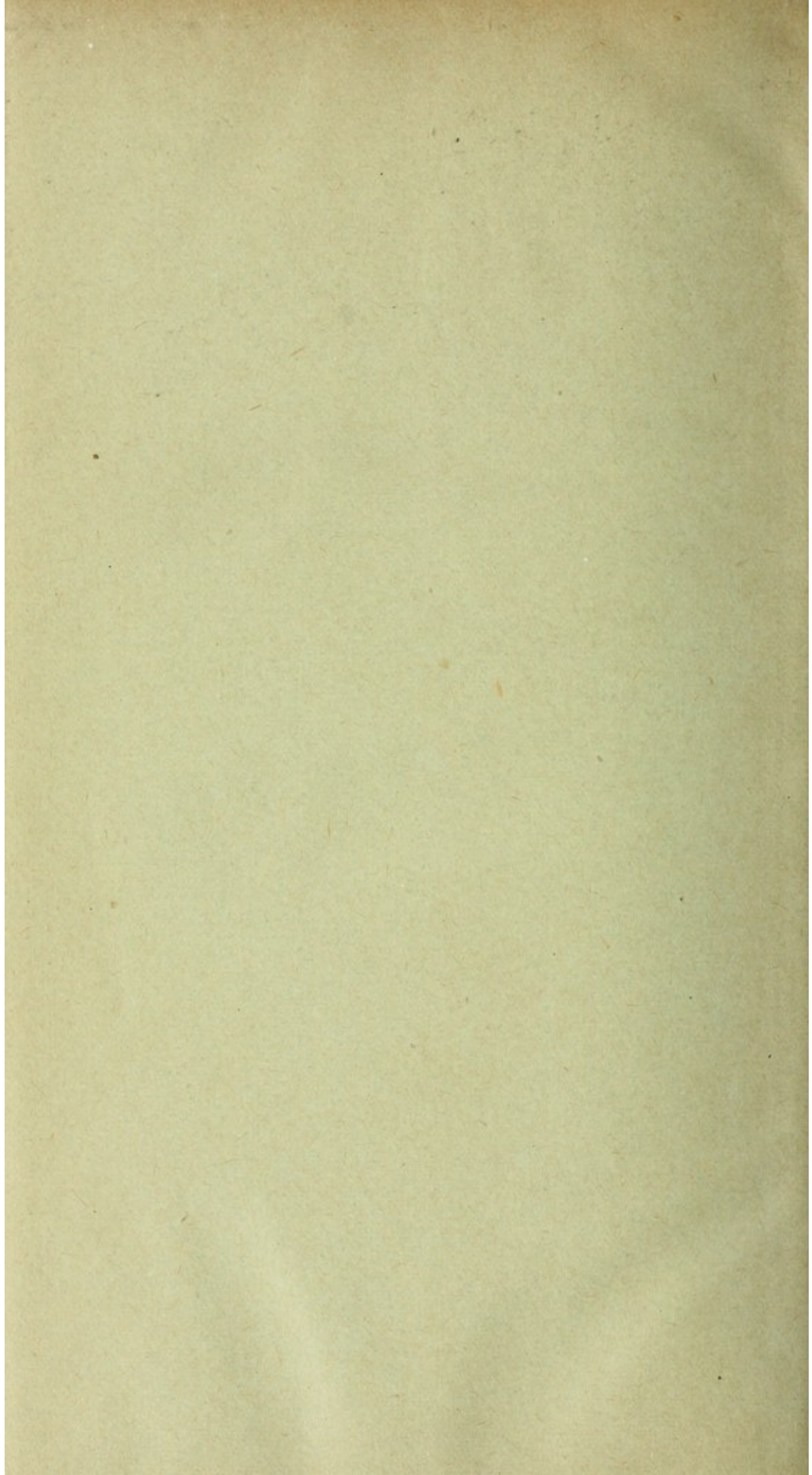














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