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like elevations, that are found on the outer part of the lower side of the inverted ventral plate.

The Groningen erratics, containing remains of Leperditia baltica, are limestones, varying between yellowish-grey and yellowish-brown. In these stones, or in others of exactly the same nature, are also found remains of Enerinurus punctatus Wahlenb. sp., Proetus concinnus Dalm. sp, var Osiliensis Fr. v. Schm., Calymene tuberculata Brünn., Cyphaspis elegantula Lov. sp., Bumastes barriensis Murch., Beyrichia spinigera Boll, Primitia seminulum Jones, Primitia mundula Jones, Strophomena rhomboidalis Wilk. sp., Strophomena imbrex Vern. (non Pander), Atrypa reticularis L. sp., Zaphrentis conulus Lindstr., Halysites sp. and Tentaculites sp. This proves sufficiently, that these erratics are of the same age with the Lower Ocsel Zone. The comparing-material at my disposal does not enable me to come to a positive conclusion with regard to their origin.

# Botanics. — "Contributions to the knowledge of some undescribed or imperfectly known Fungi" (1<sup>st</sup> Part). By Prof. C. A. J. A. OUDEMANS.

On entering upon the task which I have undertaken, I wish to express my kind thanks to Mess<sup>15</sup>. C. J. J. VAN HALL, Candidate in Botanics and Zoology at the Amsterdam University and Assistant to Professor Dr. J. RITZEMA BOS; Mr. C. J. KONING, Chemist at Bussum, one of my former disciples and author of an essay published by VAN HETEREN (Amsterdam) and ENGELMANN (Leipzig), and entitled: "Der Tabak. Studien über seine Kultur und Biologie", dedicated to Prof. Dr. J. FORSTER, Straatsburg, and to Mr. C. A. G. BEINS, private person at Nunspeet, who in different ways have helped me to facilitate that task, as well by the collecting and sending of objects, and the yielding of their observations there about, as, and this regards Mess<sup>15</sup>. VAN HALL and KONING by their putting at my disposal their drawing-pen and pencil, where I wanted these to elucidate here and there the text of my contribution. I highly value that help and am fully confident that in future it will not be denied me.

# I. ASCOMYCETAE.

### PYRENOMYCETAE.

#### Sphaeriaceae.

a. Phaeodidymae.

#### DIDYMOSPHAERIA Fuckel.

1. DIDYMOSPHAERIA RHODODENDRI Oud. n. sp. — On branches of a cultivated exotic *Rhododendron*; Wassenaar, 1894.

Perithecia fere destructa. Asci perfecte cylindracei, subsessiles,  $116 \times 7 \mu$ , paraphysibus quam plurimis filiformibus obvallati. Sporidia oblique monosticha, umbrina (Sacc. Chromotaxia, n<sup>0</sup>. 9), bilocularia, cylindrica, ad polos rotundata, vix constricta,  $14 \times 4 - 5 \mu$ .

### b. Phaeophragmiae.

#### LEPTOSPHAERIA Cesati et de Notaris.

2. LEPTOSPHAERIA GENISTAE Oud n. sp. On the pods of Genista anglica. — Nunspeet, 2 Jan. 1899; Mr. BEINS.

Perithecia innato-erumpentia, in maculis pallescentibus vulgo aggregata, nigra,  $1/_5$  mill. in diam., vertice p.m. depresso perforato; asci cylindracei, breve pedicellati, 8-spori; sporae distichae, amoene fuscae, 2-septatae (3-loculares), ad polos rotundatae, absque appendiculis,  $14-18^2/_3 \times 4^2/_3 \mu$ , loculo intermedio leniter incrassato.

Sporulis 2-septatis a pluribus affinibus descissit.

3. LEPTOSPHAERIA PHLOGIS Oud. n. sp. — On the leaves of *Phlox decussata*, cultivated at Dedemsvaart, 10 Nov. 1898. — Sent by Prof. Dr. RITZEMA Bos.

Perithecia parva, sparsa. Asci cylindracco-clavati, curvulı, sessiles, 46  $\times$  9<sup>1</sup>/<sub>3</sub>  $\mu$ . Sporidia disticha, cylindracea, curvula, ad polos obtusa, 3-septata, loculo penultimo antico ceteris ampliore, fuscidula, 23-25  $\times$  4-5  $\mu$ . (Pl. IV fig. 1).

4. LEPTOSPHAERIA VAGABUNDA Sacc. Fgi Ven. Ser. II, 318; Sacc. Mycol. Ven. p. 97 et tab. IX f. 37-46, sub titulo erroneo "Sphaeria fuscella"; Sacc. Syll. II, 31; Fabre Ann. Sc. nat. 6, IX, 89; Berlese Icones Fung. I, Fasc. II, tab. XLV f. 1; Penzig, Funghi Agrumicoli p. 30 et tab 1144 B; Winter Kr. Fl. II, 465; Oud. Ned. Kr. Arch. 2, V, 482 et 2, VI, 33; Oud. Rév. II, 288. (Pl. I fig. 1).

Ramicola. Perithecia corticola, sparsa vel aggregata, nigra, 1/5-1/2 mill. in diam., depresso-sphaeroidea, ostiolo parum vel nequa-10\* quam prominulo, primitus peridermate tecta, postremo exposita. Asci cylindraceo-clavati (d), breve stipitati, vertice rotundati, paraphysibus filiformibus copiosis obvallati, 8-spori, 132—154×22  $\mu$ . Sporidia disticha, primitus continua, hyalina, fusiformia, 4-guttulata; denique aeque hyalina, 2-locularia, ex partibus dimidiis conoideis conformata, singulis biguttulatis, basi sua sibi invicem arcte applicatis et coarctatis, infra apicem obtusum paullo collapsis; postremo cylindraceofusiformia, fuscescentia, quadrilocularia, ad septa constricta, recta vel curvula, nunc eguttulata, tunc vero loculo uno alterove guttula praedito, ad polos obtusata; sporidia hyalina 13<sup>1</sup>/<sub>2</sub>, colorata 22<sup>1</sup>/<sub>2</sub> $\mu$ longa, ultima praeterea 4<sup>1</sup>/<sub>2</sub> $\mu$  lata; utriusque generis in iisdem ascis mixta, quum varia evolutionis stadia representent. Sporidia immatura mire simulant ea plurium specierum Diaporthes.

On branches of *Tilia*. Bussum and clsewhere in "het Gooi". March, 1900. Mr. C. J. KONING.

Though this fungus has long been known already, we have yet reserved a place to L. vagabunda in this essay, 1<sup>st</sup> because we have to give some particularities from the life of the fungus itself; 2<sup>nd</sup> as we wished to sketch the changes which its presence brings about in the more profound tissues of *Tilia*, and 3<sup>nd</sup> because we wanted to draw attention to the result of some experiments performed by Mr. KONING about the nature and virulence of the poison secreted by the mycelium of the fungus.

The infection of branches of limetrees by the spores of Leptosphaeria vagabunda manifests itself by small black spots on the surface of the green, or brown-red, glossy young branches, of which the youngest internodes are first attacked. They are shorter- or longer-oval, a half to one and a half centim. long, and some millim. wide, and in the middle they always show one or two white dots. By and by the black colour changes into a dark brown and the spots take the appearance of solid, brittle scales, which after shorter or longer time isolate themselves from the surrounding parts to resemble little isles which are separated from the rest by a circleshaped groove, and finally also let loose the tissue underneath and fall off. Microscopic examination points out that they consist of flat table-shaped, brown air-bearing cells, and that their colour is due as well to a change of the cell-walls, as to a modification of their contents which is condensated to a shrivelled mass. The white dots are lacunac, filled with colourless, loosely contiguous globose cells, i.e. lenticels which, as in many other trees and shrubs, take the place originally occupied by a stoma.

The result of this research, combined with the appearance of the

black spots, can lead to no other view but that the stomata or lenticels are the localities where the spores of a former generation came down and germinated, and that the germinal tubes secrete a poisonous substance which caused the above described changes.

It was obvious that these germinal tubes and the thence proceeding mycelium-filaments ought to be found out. On the very first prepared transverse sections of the black spots and the tissue underneath, it seemed, however, that this end could not be gained. Very rarely a mycelium filament came into sight, so that the impression arose that a destruction, as figured on our plate, was not in the least proportioned to the number of germinal tubes or myceliumbranches wanted to bring about so much mischief. Meanwhile, however, after the knife had been introduced in other directions, and in particular in a tangential one through the spots, and more inwardly, more and more filaments were discovered, so that the proportion between the damage occasioned, and the cause of it, appeared in a quite different light from what had been supposed at the beginning of the research.

Before coming to this result, rather much time had however got lost, apparently uselessly, and that in consequence of the trouble which it gives to recognise the mycelium filaments. They go creeping in the intercellular canals, but are so extremely thin and quite colourless, so that they are not to be distinguished from the healthy cell-walls between which they make their way. Only after having got acquainted with the finely granulous contents of the mycelium filaments, by the use of stronger lenses, the task becomes lighter, and when, finally, the cell-walls of the surrounding bark tissue have begun to change colour under the action of the poison, it may be said that the research affords no more difficulty.

Here attention should be drawn to an accidental particularity which, previous to the examination of the diseased Tilia-branches, might well have disappointed our expectations. This, namely, concerned an investigation of diseased branches of *Negundo fraxinifolia* — an ash-tree frequent in gardens — which, by the thickness of the mycelium filaments, the brown tint of their walls, as well as by the presence of transverse partitions, combined with the accompanying nodated appearance in some places, had much more quickly carried us to our end. Under the impression of these observations our research of *Tilia* had begun, and so it was not to be wondered at that at first we thought it much more troublesome to get on, than later it proved in reality to be.

There can be no doubt but the changes, which are observed as

well in the tissues situated nearer to as in those farther from the mycelium-filaments, and to which belong 1st the decoloration of the bark- and bast-parenchyma-cells, of the phloem-layers, of the medullary rays, and of the wood-parenchyma, and 2nd the killing or liquefaction of those tissues, are caused by the more and more inwardly penetrating mycelium-branches and this in such a sense that by them a substance is secreted — an enzyme — which, as a poison for living plant cells, exerts a deadly influence upon them. The original contents of the cell grow unrecognisable, and are replaced by a brown-red shapeless precipitate, which proves indifferent to number of reagents (alcohol, ether, kalum chromate, ferrichlorid, caustic kalı, ammonia, nıtric- and sulphuric acid) and can but be decoloured by a few oxidising substances, as a mixture of kalium bichromate and sulphuric acid, or chromic acid. The poison leaves the bast fibres uncoloured and, in as much as can be ascertained by microscopic observation, unchanged.

In order experimentally to demonstrate the presence of a poisonous substance, Mr. KONING proceeded as follows:

He cut out some hundreds of black spots from young Tilia-bark, crushed them fine under addition of  $20 \text{ cM}^3$ . of sterilised water and filtered the viscous liquid through a Chamberland-Pasteur-candle. The filtrate amounted to  $7 \text{ cM}^3$ . and was of a light yellow colour.

With it branches and sections of branches of a healthy *Tilia* were treated; the former by longitudinal incisions with a flamed knife plunged in the liquid, or by injections, the latter by submerging with the liquid in a watch-glass or an experiment tube.

Experiments in both directions with sterilised water and back sap of healthy Tilia-branches served for control. The result of these proceedings was after 8 days for the incised and injected branches and of  $2 \times 24$  hours for the sections:

"that what had been treated with sterilised water or with healthy bark sap, hid remained uncoloured, but that the wound edges of the incised or injected branches on one hand, and the flat sides of the sections on the other, had suffered a brown, albeit light, colouring".

Another, later performed experiment, quite corresponded with the above described. It conceined some healthy Tilia-branches, cut off with the necessary precautions and of which some were placed in the filtrate of healthy, some in that of diseased Tilia-bark. After  $3 \times 24$  hours the poison proved, as might be expected, to have most positively exerted its influence, as the original colour of the branches placed in pure liquid, had remained unchanged, whilst that of the

branches which had been plunged into the infected sap had changed from light brown-red into dark brown.

The perithecia of *Leptosphaeria vagabunda* develop in the bark parenchyma, but gradually they make their way to the surface of the branches where, accordingly, they are then found, like the pycnidia.

The former are much more numerous than the latter and appear either at the surface of the scales, or at the wound edges, or in grooves and cavities. They have solid, black walls and a small ostium, with or without papilla, and contain numerous narrow clubshaped 8 spored asci. Their width or diameter averages from  $1/_5$ to  $1/_2$  mill. The spores are 132 to 154  $\mu$  long and 22  $\mu$  wide and show so much difference in appearance, according to their age, that one might often be inclined to believe in the existence of two different Pyrenomycetes. By this characteristic Leptosphaeria vagabunda is easily recognised among the numerous species of the genus.

In the very youngest period of development the spores are spindleshaped, colourless and one-celled; somewhat later there appears in their middle a transverse partition; still later the two halves take the form of a very obtuse cone, but which, at a third of its height, appears constricted and then consists of a pulvinate under- and a knob-shaped upperportion; moreover in each of the two portions appear two superposed drops of oil.

Between each two drops now a new partition makes its appearance and accordingly 4, instead of 2 loculaments, are observed. Then the spores again assume their original form, the drops disappear, the deeper constrictions are replaced by superficial ones and the spindle shape shows itself again. Now, however, the spores have become 4 celled and have got a light olive tint, which both characteristics secure the Fungus its place among the species of *Leptosphaeria* (ibid. g).

Notwithstanding the virulence of the poison produced by the mycelium of *Leptosphaeria vagabunda*, which brings about the destruction of an infected branch, nursery-men are not afraid of this parasite, because, according to their experience, the diseased parts are pushed off, and, as they say, the tree outgrows the evil. The justness of this observation is supported by the fact that *Tilia* belongs to the trees which regularly, first in the depth of the bark tissue, afterwards in that of the bast, produce cork-layers which exclude all beyond from the supply of water and thus abandon it to dessication. The thus killed tissue, in which the fungus had nestled, is sooner or later pushed off, or at least rendered harmless, and the absence of stomata and lenticels at the surface of the now exposed parts, deprives the spores of every opportunity again to infect the branches.

Besides a few perithicia cut through our figure shows at the outside of the section a pycnidium cut through, i.e. a spore-fruit, in which only free spores are to be seen but none enclosed in asci. This fruit has all the properties of the genus *Phoma*, but was not hitherto distinguished as a species. I call it therefore *Phoma Tiliae* and assign to it the following properties:

PHOMA TILIAE n. sp. Perithecia primo peridermate velata, denique hoc rupto semilibera, subsphaerica, nigra, tandem vertice perforata,  $154-225 \ \mu$  in diam.; sporulae ellipticae, continuae, hyalinae, ad polos rotundatae,  $4.5 \times 2.7 \ \mu$ . Differt a Ph. velata Sacc. et Phoma communi Rob. sporulis enucleatis et multo minoribus ( $4.5 \times 2.7 \ \mu$ contra  $10-12 \times 2.5 \ \mu$  et  $6-7 \times 1.5$ ).

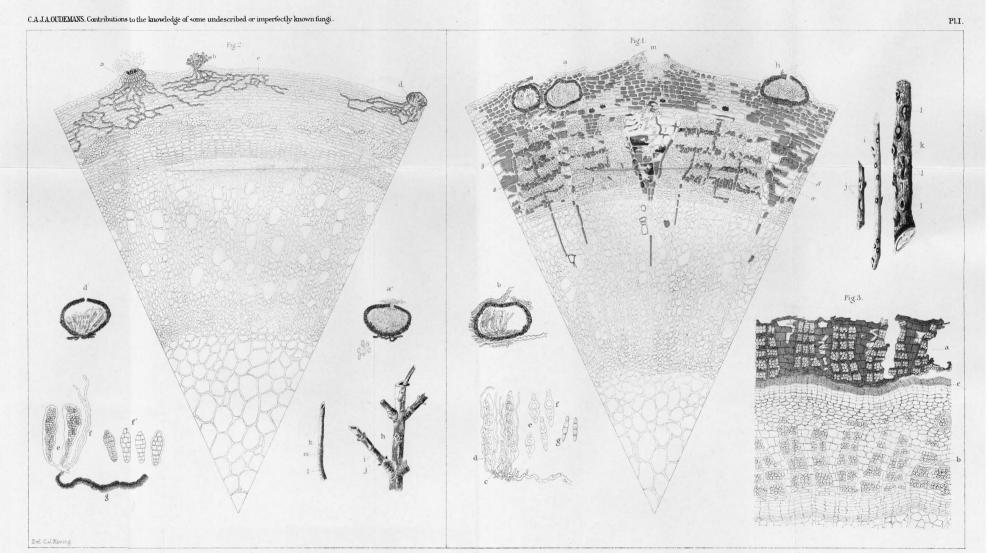
\* This *Phoma* belongs most probably to the cyclus of development of *Leptosphaeria vagabunda*, i.e. is most probably produced by the same mycelium filaments as the ascus-bearing form, but earlier. The proof for this supposition would be procured if from one and the same mycelium both forms of spore-fruits were seen to come forth; or, if from the spores of either form the second was seen to originate, or, lastly, if none of either forms were ever met with separately (unless by high exception), but constantly in company of the other. Hitherto these phenomena could not be stated, so, the last word is not yet said about the relation between the two mentioned forms.

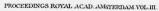
In the course of this paper we have already inferred that the nursery-man does not care for the infection of his lime-trees by *Leptosphaeria vagabunda*, as the diseased parts are thrown off and are not replaced by new ones. Meanwhile it remains advisable to render the infected branches harmless as the ripe spores of both the perithecia and the pycnidia, might afterwards again show their destructive power, and as it cannot be determined beforehand whether the infectious matter might not spread further, than has been observed till now. As to myself, among the thin branchlets, I sometimes met with much thicker ones, which had not a little suffered of the Leptosphaeria-disease.

#### EXPLANATION OF THE FIGURES.

Plate I. Fig. 1. – Part of transverse section of a one year's lime tree branch, infected by Leptosphaeria vagabunda Sacc.

The colourless spaces  $\alpha$  represent the bast bundles;  $\beta$  brown, in radial





direction extending medullary rays, of which some upwards fan-shaped elated;  $\gamma$  a fan-shaped elated portion of a medullary ray with a colourless mycelium filament and destroyed tissue.

a. Two perithecia with asci and paraphyses.

b. A perithecium with asci and paraphyses separately.

c. Young ascus.

d. Ripe asci with spores and paraphyses.

e. f. g. Spores of different ages; e. and f. 2-celled, uncoloured; g. 4-celled, coloured.

h. Pycnidium of Phoma Tiliae Oud.

- *i.* Branchlet at the beginning of the disease. The black spots with white dot in the middle are distinctly seen.
- j. Branchlet in a later period of the disease. Instead of the black spots a scale is seen (the highest) and a wound after the scale is fallen off.
  k. Older, knotty branch, upon which some closed perithecia (l).
- *m*. Lenticel, out of which a mycelium filament has penetrated more deeply and has occasioned decolouring of the bark-parenchyma cells and destruction of tissue.

Fig. 3. Piece of lime-tree bark cut transversely to show the detaching of a diseased part (a) from the still healthy, more profoundly situated (b) portion. At c the cork layer is seen which has brought about the separation between diseased and healthy tissue.

## c. Hyalophragmiae.

5. METASPHAERIA TAXI Oud. n. sp. On the leaves of Taxus baccata. — Nunspeet, 18 Sept. 1898; Mr. BEINS. — Perithecia epigena, valde numerosa, gregaria,  $\frac{1}{8} - \frac{1}{6}$  mill. in diam., continuo sub epidermide abscondita, tandem prominentia, vertice perforata, nigra, carbonisata, applanato-globulosa; asci claviformes, saepe curvati,  $65 - 70 \times 9 - 10 \mu$ , paraphysibus filiformibus obvallati; sporae 8, distichae, colore destitutae, lanceolatae vel obovato-lanceolatae, 3-septatae, ad septa non constrictae,  $18 - 23 \times 4^{2}/_{3} - 5^{1}/_{2} \mu$ .

## d. Dictyosporae.

6. PLEOSPORA NEGUNDINIS Oud. — On the one- to three years' branches of Negundo fraxinifolia and californica, often in company of Phoma Negundinis Oud. — March, 1900. Bussum, and elsewhere in "het Gooi". — Mr. C. J. KONING.

Ramicola. Perithecia gregaria, primo epidermate vel peridermate tumidulo velata, postea papilla apicali, postremo toto corpore exposita, globoso-depressa,  $\frac{1}{3}-\frac{1}{2}$  mill. in diam., papillata, nigra, glabra, contextu parenchymatico, fuligineo. Asci cylindracei vel cylindraceoclavati, subsessiles, vertice rotundati, 120-176×22-23  $\mu$ , paraphysibus paullo longioribus obvallati, octospori. Sporidia disticha, oblonga, medio leniter constricta, utrimque rotundata, 25 - 35 × 12-16  $\mu$ , primo hyalina, 1-septata, mox flavescentia, 3- et 5-, postremo mellea, 7-septata, atque, loculamentis interseptalibus fere omnibus septis 1 vel 2 longitudinaliter denuo divisis, muriformia. A *Pleospora Gilletiana* Sace. (Fgi ital. del. tab. 330 et Berlese Icon. Fung. vol. II, fasc. 1, tab. XX f. 2) differt ascis latioribus (23  $\mu$  contra 14-15  $\mu$ ), sporidiis distichis neque monostichis, rectis neque curvatis; absentia hypharum basilarium expositarum.

The above described fungus causes much damage to the plants which it attacks and destroys, and loss to the cultivator. According to informations obligingly afforded to Mr. KONING by Mr. JAC. SMITS, nursery-man at Naarden, the phenomena of the disease manifested themselves for the first time in 1898, on plants, cultivated on soil, deprived of sand. On argillaceous and sandy soils they were not observed as yet. In most cases, the variegated specimens of *Negundo* have much to suffer, though, as an exception to the rule, it is worth mentioning that in the nursery of Mr. VERSTEGEN at Naarden, p.m. 500 M. distant from that of Mr. SMITS, specimens of *Negundo californica*, with purely green leaves, were attacked by our *Pleospora*. On the var. *Kosteriana* of *N. fraxinifolia*, and on the aurated variety of the latter, the disease was not seen hitherto.

On branches, older than three years, the *Pleospora* does not occur. The nusery-man is usually not aware of the disease before St. John (21<sup>st</sup> June), which does not prevent, however, that in September ensuing many may be dead already.

Pleospora Negundinis Oud. and Phoma Negundinis Oud. seem genetically to belong to each other. Usually they are found on the same branch (Pl. I, Fig. 2 d and a and Fig. k, l and m) and in each other's vicinity, in which case the perithecia of the former are recognised by their larger dimensions and looser dispersion, those of the latter by their smaller dimensions and more compact crowding.

The perithecia of *Pleospora Negundinis* are concealed in the barkparenchyma, but with dried twigs, by the shrinking of the softer parts, they seem to repose on the bast-bundles. At their foot, hidden in the parenchyma, are seen numerous brownish mycelium filaments which spread around and ramificate (d). In our figures is seen under the same circumstances a *Phoma*-pycnidium (a) and a bundle of brown, upright filaments (b) of a black mould, all supported by hidden mycelium-hyphae, which cannot be distinguished from those of the *Pleospora*-perithecia. These filaments, like those of *Leptosphaeria*, secrete a poison, and cause a modification, though apparently not equally important, of the contents of the parenchymacells of the bark and of those, situated in the direction of the medullary rays. The diseased spots (h) grow, at the surface of the branches, from green to red-brown, over larger or smaller extents, in accordance with the bark-parenchyma, situated under the epidermis which, however, though likewise red-brown, keeps a lighter tint. If a branchlet, spotted by the disease, dies (h), the red-brown colour turns of a gray one, though the tint of the mycelium continues unchanged, and on the sharply marked spots the perithecia, often preceded by pycnidia, are seen to appear. On the drawing, the red-brown tint is not represented, for the very reason that it was borrowed from a dead branchlet. Some cells — those containing chlorophyll have till now escaped the influence of the poison.

Tissue-elements of an abnormal colour frequently appear in *Negundo* at places where no myceliumfilaments are found. Bastand phloem-bundles seem to resist the influence of the poison.

The largest *Pleopospora*-perithecia are found at the oldest internodes, so that it seems not doubtful but these organisms require much time for their complete development In accordance with this is the fact, that the larger perithecia may quite have thrown off the periderma above them, while with the smaller and younger oncs this protecting layer is still extant and is only perforated by the papilla perithecii.

Of destructions in the shape of resorption of tissues and the appearance of caverns, quite differently from Leptosphacria vagabunda on Tilia, nothing is observed. Notwithstanding this the poisonous power of the Negundo-fungus is much more vigorous than that of the Tilia-fungus, as is proved by the fact, that, according to the experience of nursery-men, the once infected Negundines are vowed to death, while the Tilias, as they say, overgrow the evil and persist.

The disease of the Negundo-branches is, as in *Tilia*, announced by local decolorations of the cork-tissue, upon which first redbrown, and later paled, black-encircled dots become visible. By-and-by the said dots begin to wrinkle and to detach from the lower bark parenchyma. Meanwhile tiny, black, corpusculae appear through the peridermis and gradually increase so much in height as to attain this membrane. By the pression which they exert on it the surface of the branchlet grows somewhat uneven, until the papillae of the perithecia perforate the periderma. In this state the asci and spores, which had been introduced into the inside of the perithecia, have not yet attained their full growth. Only when their diameter is increased to about  $\frac{1}{2}$  mill. these organs are not sought in vain, so that then a commencement can be made with their description and measurement. As we formerly inferred already, the ripe spores sometimes are oblong-elliptical, sometimes club-shaped, and have a yellow or yellow-brown colour. In well-developed specimens are found 7 partitions and a superficial constriction on a third of their height. Usually the foremost half, i.e. the one turned to the summit of the ascus, is a little wider than the backpart. Each loculament is divided by one or two longitudinal partitions, into smaller ones, so that the whole bears some resemblance to a brick wall, whence the expressions: "Sporae muriformes", "Spores muriformes", "Spores murées", "muriform spores". There are spores whose longitudinal partitions lie in each other's direction and together form a straight line, but there are others where 1 and 2 partitions alternate in the successive loculaments.

The infection in Negundo does not occur through interference of stomata, but probably through that of wounds, found near the foot of the leaf-buds, and without doubt caused by tensions during the growth. There at least are commonly found the first abnormally coloured spots. Other places are not excluded, probably, however, wounds will there, too, have given access to the spores.

If we survey the results to which have led the investigation of the *Tilia*- and *Negundo*-diseases, we find that they agree in so far as:

- 1<sup>st</sup>. they are caused by Pyrenomycetes: the *Tilia*-disease by Leptosphaeria vagabunda, the Negundo-disease by Pleospora Negundinis;
- 2<sup>nd</sup>. in both often lower fruit-forms, such as one or more kinds of pycnidia and *Dematicae*, precede the perithecia;
- 3<sup>rd</sup>. in both, not the fruits (perithecia or pycnidia), but the mycelium-filaments are the producers of the evil;
- 4<sup>th</sup>. in both by these filaments a poison is secreted, which in *Tilia* — and most probably also in *Negundo* — persists in its action, even after filtering through a Chamberland-Pasteurcandle, so, deprived of all solid components, and that consequently in both cases the nearest cause of the disease of the plantcells must be ascribed to the action of an enzyme;
- 5<sup>th</sup>. in both cases the same portion of the bark (the parenchyma) is affected, and the phloem-fasciculae seem beyond the noxious influence of the mycelium-filaments.

Both diseases, on the other hand, differ from each other, in as much as:

### (151)

- 1st. the mycelium-filaments of the Tilia-disease are colourless, devoid of partitions, very thin and delicate and so not easily perceived, while those of the Negundo-disease unite a brownish tint with the possession of partitions and a great solidity, and accordingly attract more the attention;
- 2<sup>nd</sup>. the volume of all the mycelium filaments jointly, when compared to the space in which they are spread, is much smaller for *Tilia* than for *Negundo*;
- 3<sup>rd</sup>. the enzyme of the *Tilia*-disease acts more locally, that of Negundo also at a distance; the former can give rise to liquefaction of tissue, the latter not.

#### EXPLANATION OF THE FIGURES.

Plate I, fig. 2. - Portion of a transverse section of a one year's branch of Negundo fraxmifolia (Acer Negundo), attacked by Pleospora Negundinis Oud.

- a. Pycnidium with Phoma Negundinis Oud.
- a'. 'The same, separately.
- b. A bundle of unripe hyphae of a Dematiea.
- c. Coloured mycelium.
- d. Perithecium.
- d'. The same, magnified.
- e. Ripe asci.
- f. Paraphyses.
- g. Mycelium.f'. Spores.
- h. A dying more-years' branchlet.
- i. Dying brown fragment of back.
- j. Dead colourless fragment of bark.
- k. Dead branchlet.
- *l*. Perithecia with asci.
- m. Pycnidia of Phoma.

#### SCLEROPLEA n. g.

Genus Pyrenomycetum e familia Sphaeriacearum et e sectione Dictyosporarum, generi "Plcospora" proximum, tamen ab eo distinctum perithecio duplici: uno nempe interiore (spurio) tenuiore, incompleto (i.e. sursum hiante), e cellulis rotundatis composito, ascos sporigeros et paraphyses fovente; altero exteriore (vero) crassiore, magis resistente, nigro, carbonaceo, strato parenchymatoso hyalino, satis voluminoso, a priore distincto.

7. SCLEROPLEA CLIVIAE n. sp. — Perithecia innato-erumpentia, subgregaria, sphaerico-depressa, calva, 0.5 mill. in diam., summo apice/tantum supra epidermidem prominentia et coriaceo-carbonacea; asci cylindraceo-subclavati, in pedicellum brevem et crassum abrupte

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desinentia, octospora,  $100-140 \times 15-35 \mu$ , paraphysibus longioribus articulatis obvallati; sporae distichae, fulvo-flavescentes, ellipticoobovatae, utrimque obtusissime rotundatae, muriformes, 7-, rarius 6-septatae, loculis interseptalibus omnibus, ultimis tantum vulgo exceptis, septis 1 vel 2 longitudinaliter divisae, ad septum medium constrictae, parte dimidia anteriore parum tumidiore,  $35 \times 10 - 12 \mu$ ; paraphyses ascis paullo longiores, articulatae.

On the leaves of cultivated specimens of *Clivia nobilis*. December 1899. Hees near Nijmegen.

In the Meeting of the section of 28 November 1896, a paper was presented by me, entitled: "Notice sur quelques "Champignons nouveaux", which was published the 9<sup>th</sup> December of the same year in the Proceedings of the Meeting. To the still unknown Fungi, discussed in this paper, belonged also *Chaetostroma Cliviae*, of which on pag. 226 of the said Proceedings an accurate description is found.

The plant, attacked by *Chaetostroma* was the well known *Clivia nobilis*, and the diseased specimen, sent me by Professor Dr. J. RITSEMA Bos, grown in the garden of Mr. GERRITSEN, at Hees, near Nijmegen.

Having after the 28<sup>th</sup> of November 1896 heard nothing more of diseased *Clivia*-leaves, I received, on the 27<sup>th</sup> December 1899, accompanied by a letter from Mr. C. J. J. VAN HALL, Candidate in Botanics and Zoology and assistant to Prof. RITSEMA Bos, in Amsterdam, some *Clivia*-leaves from the same garden, again diseased by *Chaetostroma Cliviae*, but moreover provided with perithecia with ripe asci and spores, which latter, by reason of their well-known characteristic structure, were recognized by Mr. VAN HALL as *Pleospora*-spores.

The diseased leaves looked like fig. 4 on Plate II. At their upper face, markedly separated by a dark line, could continually be distinguished two portions, of which only the smaller foremost one was beset with groups of perithecia, the larger back portion, on the contrary, was wholly devoid of these black points. The purely green colour of before was no more to be observed at the back portion, but it was replaced by a yellow or yellowish dingy one, whose equality had come forth from the confluence of primitively smaller, later more increased spots. At the foremost, perithecium-bearing portion, the change of green into yellow had never been observed, but instead a decrease in freshness of the green, combined with the appearance of a brown tint, and corresponding with that a drying and withering, so that it might be said that quite ripe perithecia were observed exclusively at the withered summits of the leaves. As afore mentioned, the *Cheatostroma*-pustulae occurred indeed on the yellow portions of the leaf, but then the withered

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portions were wanting there and the dark line proved to be the demarcation, not between gray-brown, withered perithecium-bearing, and green or stained unfertile parts, but between yellow, peritheciumbearing and green unfertile parts.

A vertical section of *Chaetostroma Cliviae* (Pl. II fig. 2) gives an idea of the structure of this fungus of which in our former paper only a conidium, supported by its basidium, was figured (Pl. II, fig. 3). The intact fungus, 25 times magnified, is seen on Pl. II, fig. 1.

The fungus allied to the genus Pleospora as regards its spores is, 25 times magnified, represented on Pl. III, fig. 2. In the leaf., parenchyma, arrived at its full growth, it tries to perforate the epidermis of the leaf, in which it succeeds at last. As is seen, this protective layer is hereby pushed asunder, sometimes into 3 lipshaped slips.

A vertical section of the perithecium and its environment is figured on Pl. II, fig. 5. Here the remarkable fact presents itself that the true perithecium (a) does not directly surround the so-called nucleus (asci and paraphyses) but is separated from it by a wide layer of parenchyma-tissue (c), and besides, by a pseudo-perithecium wall. The question whether this special condition of which no other instance was known to us among the singular dictyosporic Pyrenomycetes, had perhaps been observed by other mycologues, called forth a nearer investigation, and so we found that already SACCARDO had in some way alluded to it on p. 277 of Vol. II of his Sylloge, where the  $3^{1d}$  sub-division of the genus *Pleospora*, named "Scleroplea", is being discussed.

SACCARDO, namely, divided the numerous species of *Pleospora* into three categories:

I. Eu-Pleospora.

II. Catharinia.

III. Scleroplea.

Of these the 1<sup>st</sup> (p. 241) embraces the species with thin-walled, membranous perithecia and coloured spores;

the 2<sup>nd</sup> (p. 275) the species with thin-walled (membranous) perithecia and colourless spores, and finally;

the 3<sup>1d</sup> (p. 277) the species with thick-walled perithecia, and coloured spores.

Eu-Pleospora embraces the most numerous, and long since known, species, always indicated by the simple name of *Pleospora*; whilst to *Catharinia* are assigned no more than eight, and to *Scleroplea* only/ two species. Nevertheless *Catharinia* was in 1896 by SACCARDO in Vol. XI of his Sylloge, and in 1897, independently of SACCARDO, by myself in Vol. II of my "Révision des Champignons des Pays-Bas", raised from a sub-genus to an independent one.

With a view to the above diagnoses there was no doubt but the *Pleospora* produced by *Clivia* must belong to *Scleroplea*, as both characteristics: a thick, as carbonised wall, and coloured spores, were present.

The question now presented itself whether, now that Catharinia had been raised to an independent genus, the same measure should not be applied to Scleroplea too. It appeared to us that no motivated doubt could thereabout exist for the following reason: not only because a thick, as carbonised, opaque perithecium-wall might be called as great an exception among the Pleospora-species as the colourless spores of Catharinia, but also as the structure of the perithecia, as we have seen, is of a much more complicated nature than that found in the species of Eu-Pleospora and Catharinia.

Cause of astonishment might be found in SACCARDO's not recording among the characteristics of his sub-genus the by us observed special structure of the perithecia of *Scleroplea*, yet the reason of this lies at hand. The two species of *Pleospora* which are noted in the Sylloge under the afore-mentioned sub-genus, having selected for their dwelling-place the organs of exotic plants, flourishing in New-Zealand and Argentinia, were not, or only in dried state examined by him, so that he had to depend on the descriptions of others, who had, perhaps, likewise, worked under unfavorable circumstances, or whose attention had not by preference been directed to the internal structure of the perithecia.

Only for *Pleospora nuda* (Sac. Syll. II, 277 = Pyrenophora nuda Cooke, Grevillea VIII, 68) — called by COOKE "nuda", because he was of opinion that in this fungus no perithecium had come to development, SACCARDO alludes to the phenomenon observed by us, using the following expressions: "Secundum COOKE stratum perithecii exterius brunneo-cellulosum a perithecio vero interiore separabile est."

Meanwhile it should be borne in mind that COOKE who wrote in English, did not use the terms attributed to him by SACCARDO, but simply asserted: "These are no true perithecia. The cells surrounding the perithecial cavity are brown, globose and readily separable". SACCARDO has consequently wrongly understood COOKE's words, but still, at least as regards *Scleroplea Cliviae*, has come very near the truth. How much, however, the views of both mycologues diverge, is obvious to any-one who pays attention to the fact that COOKE disowns the existence of a true perithecium, whilst SACCARDO, on the con-

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trary, infers the presense of two perithecia, and that the English mycologue does not recognise our layer (d) as a perithecium-wall, whilst the Italian savant applies to it the name of "perithecium interius".

But let us leave to both authors the responsibility for their respective judgments and exclusively consider the results of our microscopic investigation most successfully reproduced by Mr. VAN HALL in Fig. 5 on Plate II, we then come to the conclusion that the perithecia of *Scleroplea Cliviae* are built up of:

- 1st a thick, solid, as carbonised black layer of cells (a) (the "stratum perithecii externum" of SACCARDO; probably the continuation downward and inward of the "cuticula nigrefacta" of COOKE;
- 2<sup>nd</sup> a many cells thick layer of thin-walled parenchyma built up of polyhedral elements (c);
- 3<sup>rd</sup> an imperfect, i.e. at the upperside not closed layer of light brown globose cells (d) surrounding the nucleus perithecii, that is to say the asci and paraphyses ("the brown and easily separable cells, surrounding the perithecial cavity" of COOKE; "perithecium interius" of SACCARDO);

and that the raising of the sub-genus *Scleroplea*, to a genus chiefly reposes with us on the presence of an external and internal perithecium, separated from each other by a rather vigorously developed interpushed parenchyma which, at a later period, probably passes into decay and in doing so breaks up the connection between the said two layers.

If we admit, — which however ought to be determined by a nearer research, -- that *Pleospora nuda* SACC. (= Pyrenophora nuda COOKE) and *Pleospora sclerotioides* correspond with *Scleroplea Cliviae* in the structure of their perithecium, then the latter genus, to be interposed between *Pleospora* and *Pyrenophora*, would for the present moment consist of 3 species, all bound to exotic plants.

By the addition of iodium the ascus-wall of Scleroplea Cliviae assumes a red-brown (presence of glycogene), the spore-wall a bluegreen, and the contents of the spores a blue colour.

The germination of the spores, tried by Mr. VAN HALL in a decoction of Clivia-leaves, was satisfactory. After the separate spores were strongly swollen, many began to send out germinal tubes. (Plate III, fig. 5.)

The question whether *Chaetostroma Cliviae* and *Scleroplea Cliviae* are to be considered either as parasites or as saphrophytes, should, we think, be answered in the latter sense, because in the tissue of

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the faded yellow leaves or parts of leaves, where no black spots appear, no trace of mycelium filaments is to be found <sup>1</sup>). The decoloration, the cause of which remained unknown to us, precedes accordingly the appearance of the fungus.

An effort was made by Mr. VAN HALL to infect a dying Clivialeaf with ascospores of Scleroplea in state of germination. The result of the experiment was not favorable. Small black points developed indeed in the environment of the inflicted wound, but they soon ceased growing, so that it remains undecided whether they belonged or not to *Chaetostroma*.

#### EXPLANATION OF THE FIGURES.

Plate II, fig. 4. A leaf infected by *Scleroplea Cliviae*. The foremost brown portion bears small groups of perithecia, of which the summits are only visible. The hindmost yellow portion bears neither of the two fungi and contains no mycelium filaments.

Fig. 2. Vertical section of a pustula of *Chaetostroma Cliviae* Oud., described more in details on pag. 226 of the Proceedings of the Meeting of 28 Nov. 1896.

Fig. 1. Some *Chaetostroma* pustulae 25 times magnified. The chaetae to which the genus owes its name, can be distinctly seen.

Fig. 3. A conidium of *Chaetostroma Cliviae*, reposing on its basidium, 500 times magnified.

Fig. 5. Vertical section of a perithecium of *Scleroplea Cliviae* (216 times magnified).

a. exterior or primary perithecium.

d. interior or secundary perithecium.

c. parenchyma tissue between the primary and secundary perithecium.b. asci.

Plate III, fig. 1. Older and younger perithecia of *Scleroplea Cliviae* (8 times magnified).

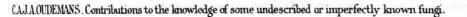
Fig. 2. Some perithecia of the same, which have torn as under the epidermis (25 times magnified).

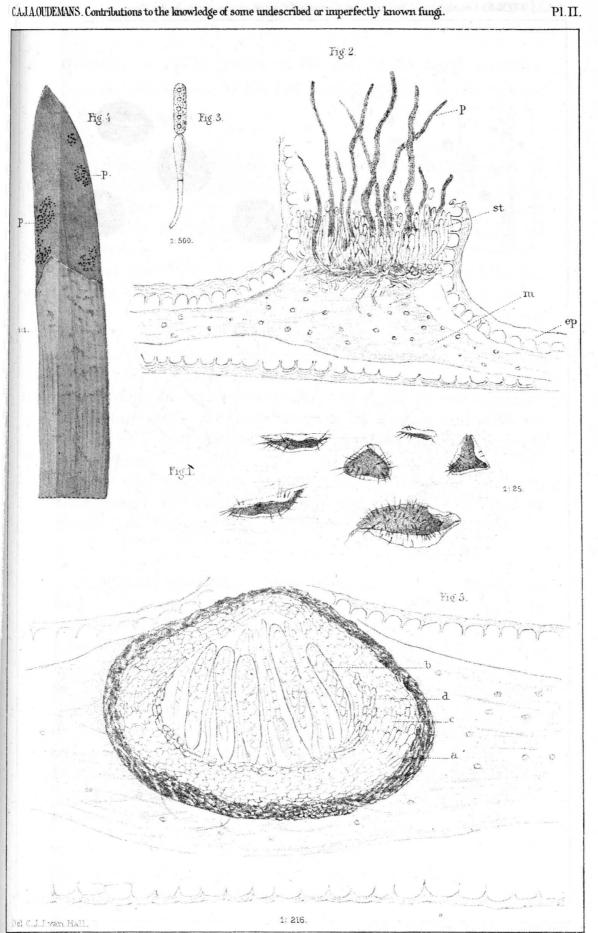
Fig. 5. Spore of the same in state of germination.

Fig. 3. Spore bearing ascus with its paraphyses (800 times magnified).

Eig. 4. Four spores of different ages and sizes of the same (800 times magnified); a. a. a. moistened spores, b. spores soaked in water.

1) At our request Prof. P. MAGNUS in Berlin had the kindness to test the accuracy of our research with some leaves sent to him; he fully confirmed it.







J.Bijtel, hth.P.J.Mulder, impr. Leiden.

