NEW MARINE PYRENOMYCETES.

PLATE III.

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INTRODUCTION.

To our knowledge of the marine Pyrenomycetes little has been added since the discovery of their existence nearly a century ago. Isolated species, collected incidentally, have been described at long intervals with the result that little is known as to their numbers, distribution or development. Consequently, no inclusive nor coherent account is available. In 1907, Cotton, while describing Mycosphaerella Ascophylli Cotton and the pycnidial stage of Leptosphaeria Chondri, took the opportunity to survey briefly the position up to that date. He included all the known marine Pyrenomycetes in a list of nine species. Of these, only two, collected by himself, are recorded for Britain.

During the past year the writer has collected and obtained algal material from various parts of the coast of Britain, in which a number of new species and even new genera have been found. The shores of Orkney yielded the richest harvest, but that may be due to the more careful search and the excellent and varied facilities for prosecuting it. Four of these occurring in Pelvetia alone, have been described recently.* To these may now be added the three treated briefly in the present account. Several others are under observation and examination, thus more than doubling the number hitherto noted and investigated.

OPHIOBOLUS LAMINARIAE, nov. sp.—(Pl. III., fig. I., 1-4).†

Perithecia densely crowded, remaining embedded in tissue with tip of long cylindrical beak merely breaking through, irregularly spherical and flattened, large, 350-450 μ in diameter.

- * Sutherland.—" New Marine Fungi on Pelvetia," New Phytologist, Vol. 14, Feb. and Mar. 1915.
- † Perithecia dense conferta, continue immersa, ostiolo elongato cylindraceo, apice solo erumpente, subsphaeroidea vel complanata, magna, 350-450μ diam., contextu coriaceo, atro; asci cylindracei, curvati, octospori, 300-360μ × 20-22'5μ, aparaphysati; sporidia filiformia, ascos subaequantia, hyalina, 50-75-septata, tandem in articulis cylindraceis (5-6μ × 5'5-6'5μ) truncata.

Hab. In thallo putrescente Laminariæ digitatae, Orkney.

Perithecial wall thin, leathery, black. Asci cylindrical, curved, 8-spored, $300-360\mu \times 20-22^{\circ}5\mu$. Paraphyses absent. Spores filamentous with numerous (50-75) cross-walls, hyaline, $300-350\mu \times 5^{\circ}5-6^{\circ}5\mu$, breaking up later into cylindrical portions $5-6\mu$ long by $5^{\circ}5-6^{\circ}5\mu$ broad.

On stalks of Laminaria digitata.

The specimens examined were collected from various parts of the north and east coasts of Scotland, where the fungus occurs abundantly on detached plants lying in rock pools. Sections of occasional blackened spots on attached, actively-growing Laminaria revealed the presence of corresponding mycelium, accompanied, however, invariably by bacteria and sometimes by one of the Dematiaceae. The presence of these two, growing luxuriantly on the outer cortical layers, prevents any definite conclusion being drawn as to the power of this fungus to gain entrance by itself.

The mycelium penetrates to the mucus-passages, whose anastomosing tubes, full of mucilage, form ideal centres for rapid development and extension. Long, black, septate hyphae, with an average diameter of 10-15 μ , although 20 μ is not uncommon, run along these ducts sending numerous branches into the outer layers towards which nearly all bend. Their passage through the cells presents the appearance of irregular nodulose lines, owing to the alternation of hyphal dilations in the cells with constricted portions passing through the walls. These branches, varying from 4-7.5 μ in diameter, give rise to still finer hyphae, 2-3 μ , which form a dense network in the cortex. There seems to be little or no attempt to attack the zone beneath the ducts with the result that a sharp separation of these two tissues occurs later.

The perithecia are formed at the tips of special branches about $300\text{-}350\mu$ below the epidermal layer, and are densely crowded all over the infected area. The mature perithecium is a relatively large body easily visible to the naked eye; some of the largest approach 0.5 mm. in diameter. In outline it presents a flattened irregular spherical appearance with a long cylindrical beak $175\text{-}275\mu\times65\text{-}70\mu$. The latter breaks through the outer layers but without projecting. The perithecial wall is thin, black and leathery. Neither periphyses nor paraphyses are present.

The long cylindrical thin walled asci are strongly curved owing to their great length. They become free at an early stage and are coiled inside the perithecium. The long filamentous septate spores lie parallel and completely fill the ascus whose length they practically equal. At first they are unseptate, but at maturity numerous cross walls are formed, and finally each spore breaks up into short cylindrical portions which pass out through the ostiole.

The systematic position of the fungus is open to question. and it has been placed in the genus Ophiobolus only after considerable hesitation. The delicately walled asci, with their filamentous spores breaking up later, are characteristic of Ophiobolus. The absence of paraphyses, however, would tend to refer it to the family Mycosphaerellaceae, in which there is no genus with similar spores. A disclination to create a new genus on such a character alone has prevented me from describing it as such, although there is abundant precedent, as for example in the separation of Stigmatea from Mycosphaerella. The inclusion in the genus Ophiobolus of one or two forms wanting paraphyses, as O. maritimus would seem to justify this naming, while at the same time it bears eloquent testimony to the unsatisfactory condition of classifications of the Pyrenomycetes. The long cylindrical beak differs also from the type generally met with in Ophiobolus. The simplest way out of the difficulty would undoubtedly be the creation of a new genus, but that is undesirable, in the writer's opinion, until the life history is fully investigated.

TRAILIA, nov. gen.*

Stroma wanting. Perithecia deeply sunk in substratum, with tip of long narrow straight or curved cylindrical beak or ostiole breaking through surface, but not projecting. Wall of perithecium soft and white. Asci cylindrical and curved with thin membrane, 8-spored. Paraphyses absent. Spores hyaline, filamentous, tapering, septate, bent double and coiled in ascus.

TRAILIA ASCOPHYLLI, nov. sp.—(Pl. III., fig. II., 1-3).†

Mycelium localised, hyaline, septate. Perithecia grouped irregularly in blackened regions of thallus, spherical or clubshaped, $50-60\mu$ in diameter with long narrow straight or bent cylindrical beak $300-450\mu \times 7.5-10.5\mu$. Perithecial wall thin, soft, and white. Asci cylindrical and curved, thin-walled, 8-spored, $45-50\mu \times 9-10\mu$. Paraphyses absent. Ascospores filamentous, tapering from $3-3.5\mu$ at broad end to 1μ at narrow

- * Stroma nulla; perithecia simplicia, thallo alte immersa, mollia, alba vel pallida, ostiolo longissimo, angusto, recto vel curvato, cylindraceo; asci cylindracei, octospori, aparaphysati; sporidia hyalina, filiformia, fastigata, septata, in asco duplicata contortaque.
- † Mycelium in quibusdam locis, hyalinum; perithecia conferta vel sparsa, thalli regionibus inaequaliter atris factis immersa, subglobosa vel clavata, 50-60μ diam., mollia, alba, ostiolo longissimo, angusto, recto vel curvato, 300-450μ longo, 7.5-10.5μ lato; asci cylindracei, contorti, octospori, 45-50μ×9μ, aparaphysati; sporidia hyalina, filiformia in exilitatem fastigata, septata, altero tanto asco longiora, et igitur duplicata contortaque. Hab. In thallo Ascophylli nodosi, Orkney.

end, hyaline, septate, twice the length of ascus and, consequently, bent double and coiled.

Parasitic on thallus of Ascophyllum nodosum.

This fungus, observed for the first time in Orkney during the summer and early autumn of 1914, was found occurring in great abundance on the thallus of Ascophyllum nodosum just to the outside of the Pelvetia zone on the flat rocks. It was also obtained on free-floating masses in the North Sea.

At first circular patches of brown appear, giving place later to irregular dark blotches which stand in striking contrast to the pale olive of the healthy thallus. The spread of the disease is rapid, and frequently it renders portions of the thallus so brittle as to be easily broken off by the action of the waves. Once the fungus has broken the epidermal layer, the host is exposed to the action of various bacteria. These latter working in consort with the parasitic fungus kill and break down the outer tissue destroying protoplasm and chloroplasts. The result is that frequently the thallus, in section under the microscope, is seen to be surrounded by a hyaline envelope of disorganised tissue, permeated by branching hyphae and bacterial masses.

The hyaline mycelium, consisting of fine septate strands with a diameter varying from $1.5-2\mu$ in the finer branches to 3μ and occasionally more in the main ones, forms a closely woven network throughout the tissue of the infected areas. The finer hyphae are difficult to distinguish, at first glance, from the mycelium of Mycosphaerella Ascophylli Cotton with which I have invariably found them interwoven. The slightly greater thickness of the main branches and the habit of both makes it easy to differentiate them. The parasitic species, in addition to occupying the intercellular spaces, coils branches tightly round the protoplasmic contents into which slightly dilated haustoria are sent, or through which absorptive branches pass. Azo-blue is quite effective in singling these out.

The perithecia are produced deep within the tissue, sometimes being found in the very centre of the thallus. As a rule they are scattered irregularly, but frequently three or four are grouped together, generally at different levels. They are minute bodies rarely exceeding 50μ in diameter. Owing to their depth within the host they require long ostioles. Thus in the case of one young perithecium the developing beak had reached a length of 245μ with about another 300μ to travel to the surface. This characteristic, long, attenuated neck seems out of all proportion to the size of the body. Usually it grows straight outwards in the form of a cylindrical tube of about 8 or 9μ in diameter, consisting of loosely woven hyphae, but, whenever the perithecium is bent over, it curves gradually upwards. It is quite a common occurrence for perithecia em-

bedded in the tissue of the air bladders to grow towards the enclosed cavities, and in that case the ostioles usually project into these in a curiously bent worm-like fashion with slightly swollen tips. Normally, however, they penetrate the cortex and reach the surface beyond which they never project.

The perithecial walls remain soft and white. This character is maintained even in the superficial layers when the epidermis and outer cortex have been destroyed as already described.

The asci are few in number, and bunched together. When young, they are tapering and relatively thick-walled, later becoming thin-walled and cylindrical, with a slight constriction at the curve above the thickened lower end. The spores are most distinctive. They are hyaline and twice the length of the ascus. This necessitates their being doubled and coiled in the manner characteristic of the genus. The filamentous and tapering spores, at first unseptate, ultimately develop 1-3 septa in the wide end. None have been observed so far in the narrow end. When released from the ascus they retain a characteristic coiled attitude which gives them a distinctive snake-like appearance. This may assist in their passage through the long, narrow ostiole.

The formation of a new genus to include this fungus is clearly necessary. Not only does the character of the perithecium separate it from all other genera of the Hyponectrieae among which it must be placed; the type of spore is also distinct.

ORCADIA, nov. gen.*

Stroma absent. Perithecia entirely immersed in thallus of host with long tapering beak. Perithecial wall soft, white never becoming black. Asci cylindrical or clavate, 8-spored. Paraphyses present. Spores septate.

ORCADIA ASCOPHYLLI, nov. sp.—(Pl. III., fig. III., 1-3).†

Mycelium localised, hyaline, forming network on which fruiting bodies are borne. Perithecia entirely sunk in thallus. spherical or arched, $160-200\mu$ in diameter, with long broad cylindrical beak $80-125\mu \times 35\mu$. Asci clavate, thin-walled, 8-

* Stroma nulla; perithecia simplicia, thallo immersa, contextu distincte molli pallidoque (nunquam carbonaceo), ostiolo elongato, fastigato; asci cylindracei vel clavati, octospori, paraphysati; sporidia hyalina, septata.
† Mycelium in quibusdam locis, hyalinum; perithecia thallo omnino immersa,

Hab. In thallo vivo Ascophylli nodosi, Orkney.

[†] Mycelium in quibusdam locis, hyalinum; perithecia thallo omnino immersa, globosa vel arcuata, mollia, pallida, 160-200μ diam., ostiolo elongato, lato, cylindraceo, 80-125μ longo, 35μ lato, ad superficiem pertinente; asci clavati, octospori, 65-75μ x 17'5-18μ; periphyses numerosae; paraphyses paucae, filiformes, simplices; sporidia tempestiva flavescentia, cylindracea, parum curvata, 3-septata.

spored, $65-75\mu \times 17\cdot5-18\mu$. Periphyses abundant, paraphyses few, filamentous, unbranched, with slightly dilated tips. Ascospores light yellow when mature, cylindrical, slightly curved, 3-septate.

Parasitic on thallus of Ascophyllum.

This fungus was also found in great abundance on Asco-phyllum nodosum along the shores of the Orkney islands during the summer of 1914. In external appearance the disease presents a strong resemblance to the damage caused by Trailia The blackened patches, at first circular, become irregular in outline and run into one another so that finally whole areas of the thallus show infection. The fungus aided again by bacterial action, causes a similar breaking down of the epidermal and outer cortical layers, and, in these, various bacteria and fungifind a footing.

This pale coloured mass of disorganised cell walls, stains distinctly with azo-blue, but fails to show any reaction with iodine, as in the case of a similar layer in Stigmatea on Pelvetia.

The mycelium, consisting of numerous, slender, muchbranched, curved hyphae 2-3.5 μ in diameter, is strongly developed. It forms a very dense somewhat cylindrical network round the algal cells, which are strangled by close coils or penetrated by haustoria. This habit enables us to distinguish it from the endophytic non-parasitic mycelium of Mycosphaerella, but gives no aid in separating it from that of Trailia, with which it is sometimes associated. When the three occur together the thallus becomes simply riddled with hyphae.

The perithecia, produced irregularly in the cortex, are spherical or arched with long, cylindrical, slightly tapering beaks, level with the surface except where, owing to the decay of the upper layers, they may project for a very short distance, rarely more than 5-10 μ . The perithecial walls consist of closely woven, fine, hyaline hyphae remaining soft, fleshy and white. They never become black even when the ostiole projects slightly. Numerous asci are present, mixed with sparse short paraphyses slightly dilated at the tips, and surrounded by numerous periphyses. The 3-septate spores tend to break easily into sections.

Arising from similiar mycelium, which penetrates the disorganised outer tissue, are short unbranched conidiophores cutting off elliptical hyaline conidia $6-8\mu \times 3\mu$. While the mycelium bearing them is similar to, and appears to spring from that bearing the perithecia, it is inadvisable to designate this the conidial form of Orcadia without further investigation

The septate spores correspond to those of Leptosphaeria, but the structure of the perithecial wall taken in connection with the elongated soft beak, may be regarded as sufficient reason for considering it distinct. The occurrence of hard, black perithecia of the Sphaeriales type on Pelvetia, Laminaria, etc., prevents our regarding this as a Leptosphaeria whose walls remain soft and whitish owing to the different conditions met with in algal hosts. Its natural position in our present artificial classification is undoubtedly among the Hypocreales near Trailia to which it bears a resemblance both in the soft whitish walls and its total immersions in the host tissue. Taking these points into consideration, it seems desirable to make this generic separation.

The foregoing, added to the other four new Pyrenomycetes recently described by the writer, make seven new species, two of which are also new genera. The investigation so far has revealed some interesting characters and adaptations, which it is hoped to link into a coherent whole when the present

examination of species is complete.

Appended are diagnostic characters and brief preliminary notes on two other new species still under observation. Special interest attaches to the former of these, Hypoderma, as, so far as the writer is aware, this is the first record of the occurrence of any of the Hysteriineae on marine algae.

HYPODERMA LAMINARIAE, nov. sp.

Perithecia in areis rotundis gregaria, immersa, longoelliptica, apicibus aliquid praeacutis, 150-220 μ longa, circa 130 μ lata, scissuris longis aperientia, parte superiori atra, carbonacea, basi submembranacea, pallide brunnea; asci clavati, apice attenuato, 69-90 μ × 12.5-14 μ , octospori; paraphyses filiformes, extremibus leviter inflexis vel dilatis; sporidia disticha, fusiformia vel ovoidea, hyalina, 1-septata, leviter constricta, flavescentia, 20-25 μ × 5-7.5 μ .

Hab. In thallo vivo Laminariae saccharinae, Orkney.

During the summer and autumn of 1914 this fungus was found in great abundance along the shores of the Orkneys, where its occurrence on living plants, both attached and newly

cast up after storms, fully bears out its parasitic habit.

The first appearance of the disease is indicated by small brown discoloured patches on the stalk; these gradually extend and darken, forming circular or somewhat elongated areas, which ultimately merge into one another. On examination they prove to contain immense numbers of densely crowded, oblong, irregular, or even branched perithecia with the habit and mode of opening characteristic of the Hysteriineae.

The dark-coloured, thick-walled, irregular mycelium penetrates in wedge fashion between the mucilaginous cell walls of the outer cortical region. Haustoria enter the cells and absorb their contents. In this way the outer layers are ultimately

broken down, exposing the inner to the action of numerous saprophytes. The perithecia are formed in the cortex immediately underlying the epidermis, along which are laid down the dark streaks which split later to form the thick-lipped openings. These are carbonaceous, while the lower portions of the perithecia are merely membranous. Lining the wall are several layers of pale-coloured mycelium from which the asci and paraphyses spring. The former, rising from the somewhat flattened base, show great variation in outline and in the arrangement of the ascospores within them. The paraphyses are filamentous and bent towards the tips. The uniseptate thick-walled spores are hyaline when young but become slightly coloured at maturity.

DOTHIDELLA PELVETIAE, nov. sp.

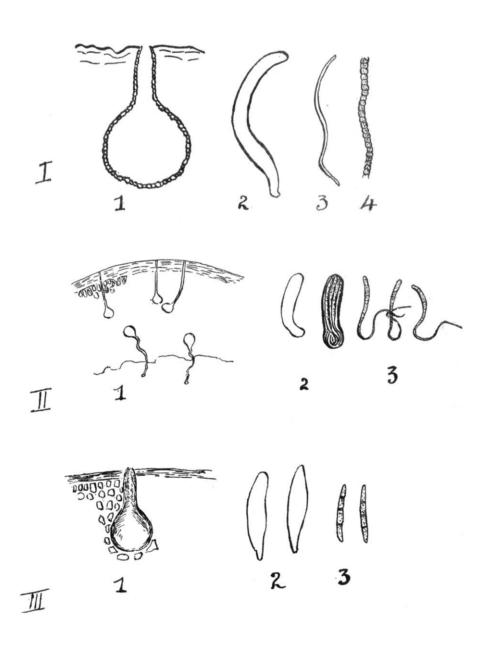
Stromata sparsa, parva, $210-650\mu$ diam., convexa, initio immersa tandem in summa parte, atra; loculi immersi, subglobosi vel complanati, $100-120\mu$ lati, $80-90\mu$ alti; asci cylindracei vel clavati, octospori, $40-45\mu \times 12-15\mu$, paraphysati; sporidia disticha, ovoidea vel oblongata, hyalina, 1-septata, vix constricta, tegumento denso, $12\cdot5-15\mu \times 5-6\mu$.

Hab. In thallo Pelvetiae canaliculatae, Orkney.

This fungus was found on cast-up and dried thalli of Pelvetia, and consequently it is uncertain yet whether its habit is parasitic or saprophytic. An extensive examination of attached and living plants in that particular locality has so far failed to show any trace of the fungus. As the infected thalli may have been sea-borne, this absence does not prove conclusively that the fungus does not occur elsewhere as a parasite.

The stromata, produced in the epidermis and the layers immediately underlying it, ultimately become superficial through the breaking down of these layers by various organisms. They are relatively small bodies, rarely exceeding 0.5mm. in diameter, and containing frequently only one loculus. Seldom, even in the largest stromata, are more than 3 to 5 found. The loculi are small and generally flattened, flask-shaped bodies with numerous asci and paraphyses borne on the basal portions. The former are cylindrical or clavate, with two membranes, the outer of which usually splits transversely. They contain eight, oval or oblong, two-celled, biseriate spores, which are much smaller than those of *Dothidella Laminaria* Rostr., found on the stipes of *Laminaria longicruris* in Greenland.

University College, Southampton, February 1915.



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I. Ophiobolus Laminariae. II. Trailia Ascophylli. III. Orcadia Ascophylli.

EXPLANATION OF FIGURES. PLATE III.

- I. I. Perithecium of Ophiobolus Laminariae.
 - 2. Ascus
 - 3. Immature ascospore
 - 4. Portion showing cross walls.
- 1. Section of air bladder wall of Ascophyllum showing perithecia of Trailia.
 - 2. Asci.
 - 3. Ascospores.
- I. Perithecium of Orcadia Ascophylli.
 - 2. Asci.
 - 3. Ascospores.

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