

# The Cereal Rusts.

## I. The Development of their *Uredo* mycelia. -

BY

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With Plates **XL-XLIII.**

### INTRODUCTION.

**I**N recent years very considerable attention has been attracted to a group of species of *Puccinia*, the more important members of which are parasites of our chief cereals. All these species were at one time included under the three names *P. graminis*, Pers., *P. rubigo-vera*, DC., and *P. coronata*, Corda.

Recently they have been subdivided into thirteen species, each of which is readily recognized by well-defined morphological and physiological characters. These have further been subdivided into no less than twenty-four forms, distinguishable from each other by apparently physiological characters, that of adaptation to particular hosts.

Further, the well-known Swedish mycologist Eriksson in endeavouring to account for special cases of infection was led to formulate a view since known as the 'Mycoplasm' hypothesis, according to which the mycelium perennates in the form of a naked intercellular plasma.

Unfortunately our knowledge of the cytology of these fungi, notwithstanding the interesting work which had been done by De Bary (4), Poirault and Raciborski, Sapin-Trouffy (31), Marshall Ward (33) and Blackman (6), is still in a very unsatisfactory condition.

It was obviously desirable that someone should undertake the detailed investigation of the histology of these forms.

It was under such circumstances that the late Professor Marshall Ward very kindly suggested that I should attack, partly in conjunction with himself, but mainly working alone under his advice, some of these problems.

The work was begun in November, 1903, and was carried on for two years in the Cambridge Botany School, and has since been continued in my own Laboratory at Pretoria.

As would be expected, a vast amount of information relating to the

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histology, physiology, and ecology of these Fungi has now been obtained, in addition to many interesting facts connected with the relative immunity of the various hosts.

The development of the *Uredo* mycelium of the Fungi (to be described) has been followed in detail from the time of spore-germination to spore-production. In such a course of development we have to recognize three distinct stages:—

1. The attack by the parasite on its host, or the first phenomenon of occupation.

2. The course taken after occupation by the further growth of the parasite.

3. The reaction on the host after occupation and the subsequent reciprocal action of host and parasite.

It is mainly to the attack by the parasite on its host, or the first phenomenon of occupation, that we shall confine our attention in this paper.

Up to the present the complete series of these rusts has not been obtained; but as I have examined all the species which are known to occur on Cereals, it is thought advisable to publish them without further delay, and leave several species which occur on the more uncommon grasses for a future investigation, should they be obtained.

A detailed histological study has also been made of the so-called 'physiological species,' 'biologic species,' 'sister species,' or 'specialized forms' of rusts, and points of great interest have come to light. These I hope to publish at an early date.

#### METHODS.

The methods adopted for studying the histology are as follows:—

1. Selection of host plants. For infection purposes, seedlings of plants known to be susceptible to the particular kind of rust which it was desired to study were raised. In most cases, directly the first foliage leaf was fully expanded, a series of inoculations were made by laying the spores on the inner face of the leaves with a flat platinum needle. Sowings of spores were also made at the same time in watch glasses of distilled water as control experiments to observe the germination capacity of the spores.

2. After inoculation damp bell-jars were placed over the plants for forty-eight hours.

3. The inoculated portions of the leaves were then removed and fixed daily until the crop of *uredo*-pustules made their appearance. As a rule this generally took place about the tenth day, but was liable to considerable variation, according to the different rusts under investigation.

In this way the life-history of the Fungus was studied from the period of germination of the spore up to the time that it had successfully reproduced itself on its particular host.

### THE FIXING SOLUTIONS.

1. Chrom-acetic solutions. On the whole the best results were obtained with this solution, in strengths varying from 1 per cent. to one-tenth per cent. solutions. Whenever possible the fixing was done under the air-pump, and the fixed portions were allowed to stay in the solution from twelve to forty-eight hours.

2. Flemming's Chrom-osmium acetic solution. The weaker formula was most frequently used, and sometimes it was diluted down to one-half its volume with water. The chief objections to these solutions are:—

- (a) The blackening due to the osmium.
- (b) Their slow penetrating power.
- (c) They cause the protoplasm of the hyphae to become vacuolated.

3. Picro-formol solution:—

Commercial formol	.	.	.	30 c.c.
Water	.	.	.	20 c.c.
Acetic acid	.	.	.	5 c.c.

saturated with picric acid.

The advantages of this solution over the two previous ones are:—

- (a) Its quick penetrating power.
- (b) Its not causing the material to become brittle on cutting.

After washing and hardening in the usual way, the material was taken into paraffin by means of Xylol or cedar-wood oil. The sections were cut from 5 to 10  $\mu$  thick, and all the staining done on the slide.

The chief stains used were:—

- 1. Diamant Fuchsin and Lichtgrün.
- 2. Safranin, gentian-violet and orange G.
- 3. Iron-alum-haematoxylin with safranin, eosin or orange G.

### GENERAL.

The Uredineae, like most other true parasites, possess two methods of penetrating their host.

In the one case the germ-tube emitted from the spore penetrates the firm membrane or cuticle. This usually takes place with the sporidia developed from the teleutospores.

In the other case, the germ-tubes penetrate their host by means of a natural opening or orifice, the stoma. This is always the method adopted in the case of uredospores and aecidiospores.

The method of infection by means of uredospores was known to de Bary, who has given a general description of the sequence of events involved. Several other investigators have from time to time given various descriptions of infection phenomena. But no one seems to have realized

that each set of phenomena is of a very definite nature for each species of *Uredo*, i. e. the infection phenomena presented by *Uredo graminis* on wheat is always of the same definite type and differs markedly from say *Uredo triticea* on wheat or any of the other *Uredo* forms to be described, which again in their turn are all constant. In uredospore infection it is usual to recognize two distinct steps: entry or inoculation and true infection.

In addition to the preliminary act of germination, entry or inoculation includes the swelling up of the tip of the germ-tube over the stoma, to form the appressorium. From the appressorium a slender branch passes through the stomatal slit and at once swells out into a vesicular body, the substomatal vesicle, into which the whole contents of the spore is poured. The entry of the fungus is now assured.

The next step in the development is the putting forth from the substomatal vesicle of one or more infecting hyphae, which closely apply themselves to the host-cells and send in their haustoria, thus bringing about the act of infection.

*Puccinia graminis Tritici*, Erikss. and Henn.

The Black Rust of Cereals, which is heteroecious on Barberry, has been broken up by Eriksson (12) into six specialized forms:—

1. *Puccinia graminis Secalis*, found on *Secale cereale*, *Hordeum vulgare*, *Agropyrum repens*, *Elymus arenarius*, and *Hordeum comosum*, but not on *Triticum vulgare* or *Avena sativa*.

2. *Puccinia graminis Tritici* on *Triticum vulgare*. He regards this form as 'not sharply fixed', because he says *Uredo* infection from Wheat feebly infects Barley, Rye very slightly, and in the case of Oats he regards the infection as doubtful.

3. *Puccinia graminis Avenae* on *Avena sativa*, *Alopecurus pratensis*, *Dactylis glomerata*, but not on *Triticum vulgare*, *Hordeum vulgare*, or *Secale cereale*.

4. *Puccinia graminis Airae* on *Aira caespitosa* only.

5. *Puccinia graminis Agrostis* on *Agrostis canina* and *A. stolonifera* only.

6. *Puccinia graminis Poae* on *Poa* only.

In this paper we shall concern ourselves solely with the form *Uredo graminis Tritici*, Erikss. and Henn., found on Wheat.

This rust, formerly so common in England, now appears to have been replaced by the Yellow Rust of Wheat, *Uredo glumarum*, which is particularly abundant in the Fen District.

*Uredo graminis* is the rust responsible for the damage wrought in the Cereal crops of the United States of America, Australia, and Tasmania.

In India, according to Barclay (3), it is comparatively unimportant.

In South Africa, especially the Transvaal, *Uredo graminis* prevents the successful raising of summer cereal crops.

*Uredo graminis*, the Black Rust, is so called from the dark colour produced on the straw by the teleutospore patches. In the field the Uredo stage is recognized by its rusty orange to brown ochre coloured pustules, which are from 12–15 mm. long or even longer. Under the microscope the spores are broadly elliptic, dirty yellow, spinulose,  $17-40 \times 14-22 \mu$ .

#### THE HISTOLOGY OF UREDO GRAMINIS.

Although *Uredo graminis* has probably received more attention at the hands of mycologists all the world over than any other parasitic Fungus, I think it will be clear that the complete sequence of infection as represented in my figures has never heretofore been observed or represented. Apart from de Bary (5), Marshall Ward (32), and more recently Bolley (7), have figured the germination of and infection by means of uredospores of *Uredo graminis*.

De Bary has figured a slight swelling of the germ-tube over the stoma, but nothing more. Bolley has depicted the germ-tube passing straight through the stoma to the mesophyll-cells below. This I find never occurs.

Further, Bolley (7), p. 630, says that the germ-tube from these uredospores 'may bore its way through the skin of a wheat plant and thus start another point of infection'; in the case of uredospores of '*Puccinia rubigo-vera*,' he illustrates some of the filaments passing directly through the leaf. I have now examined some hundreds of thousands of uredospore infections on all the cereal crops and various other Gramineae, and have never met with a single infection which was not brought about by way of a stoma.

The germination of the uredospore on the epidermis is usually effected within the first twenty-four hours, and by the third day infection is well established.

When the spore germinates, two germ-tubes frequently appear at first, but one quickly gets ahead of the other and arrests its growth. The tip of the germ-tube, as soon as it reaches a stoma, instead of entering straight away, swells up and forms the appressorium (Figs. 1 and 2). From the appressorium a narrow branch passes through the stomatal slit, always situated somewhat nearer to one of the corners. As soon as the neck has got through the aperture, it enlarges to form the sub-stomatal vesicle, which applies itself closely to the inner face of the stoma (Figs. 2 and 3). Into this vesicle the whole contents of the spore are poured, and the entry of the fungus is completed. The germ-tube and appressorium now quickly wither and are soon lost sight of.

A point to be noted in connexion with the substomatal vesicle, both in

these species and those to be described below, is that it always occupies a definite position relative to the stoma, i.e. in *Uredo graminis* and most other species its long axis is always parallel to the stomatal slit. From one end of the substomatal vesicle the infecting hypha is given off, which at once penetrates the host cells by means of its haustoria (Fig. 3), and thus a successful infection of the host is accomplished.

A very characteristic feature connected with the substomatal vesicle of this species is that the end from which the infecting hypha springs always clings very closely to the head of the guard cell, or the epidermal cell abutting on it.

From the position of the organs depicted in Figs. 2 and 3 it will be clear that the complete sequence of infection, as has usually been figured, cannot be attained by transverse section; for the relative position of these organs is such as to make this almost impossible with anything like moderately thin sections. In Fig. 4 is shown such a transverse section, which happens to pass through the point of union of appressorium and substomatal vesicle.

In diameter the appressorium is  $9\mu$ , while in length it is roughly  $27\mu$ , the substomatal vesicle being approximately of the same dimensions. The hyphae are of uniform size, and measure  $3.5\mu$  in thickness.

The haustoria are very varied in shape. They attain their greatest development in the cells bordering on the vascular bundles, where they often become very branched and contorted. A very young haustorium, which has just penetrated one of the mesophyll cells, is seen in Fig. 5, while in Fig. 6 forms commonly found in the epidermal cells are shown.

#### *Puccinia Phlei-pratensis*, Erikss. and Henn.

The Timothy rust was set apart from *P. graminis* in 1894 by Eriksson and Henning (16) as a distinct species, because out of ninety-two infections with good germinating material on *Berberis*, only one feeble infection resulted. From this they concluded that this form does not probably form its aecidium on *Berberis*.

Further experiments on *Berberis* by Eriksson (10) (14) have simply served to confirm this result. He has also shown that it can be transferred in the Uredo stage from *Phleum pratense* to *Festuca elatior* and vice versa (Eriksson) (9); and in addition that it will feebly infect *Phleum Michelii*, *Secale cereale*, and *Avena sativa* (Eriksson) (14), but does not infect *Triticum vulgare*, *Hordeum vulgare*, and *Poa pratensis*.

Consequently Eriksson regards *P. Phlei-pratensis* as 'nicht scharf fixiert'.

*P. Phlei-pratensis* is distinguished in the Uredo stage by the dark yellowish-brown pustules which are from 1–2 mm. long; these may frequently run together in a long line as much as 10 mm. long or more on

the leaf edges and sheaths. The spores are oblong, somewhat pear-shaped, spinous, and of a dirty yellow colour. They measure from  $18-27 \times 15-19 \mu$ . It is interesting to note that according to Eriksson and Henning (16) the *Uredo* mycelium is able to survive the winter. For during 1891-2 fresh pustules were produced on the 28th of December, and they continued even up to as late as the 28th of March, and during the winter 1892-3 the latest and first appearance of fresh pustules was on the 30th of November and the 27th of March.

Also, according to them, the germinating capacity of the uredospores falls with the increase in severity of the winter.

#### GERMINATION OF THE SPORE.

From the spore on germination a fine delicate germ-tube runs along the surface of the epidermis, and when the tip reaches a stoma, it swells up a little into a thin and delicate vesicle, the appressorium. Directly the contents have passed from the appressorium into the sub-stomatal vesicle, the delicate and now empty appressorium shrivels up and breaks away at its junction with the firmer walled sub-stomatal vesicle, and so is rarely seen after the formation of the latter.

The sub-stomatal vesicle is a very definitely shaped body (Fig. 7), closely resembling that of *Uredo graminis*, but narrower. Like that of *Uredo graminis*, it gives rise at one end to one infecting hypha only, from which the others subsequently arise. In diameter it measures from  $4-6 \mu$ , and in length from  $30-35 \mu$ . It differs from *Uredo graminis* chiefly in the fact that the end from which the hypha springs does not cling to the head of the guard-cell.

The resulting hyphae, and especially their haustoria, bear a very striking resemblance to those of *P. Symphyti-Bromorum*. As a general rule the hyphae are thinner and more thread-like than even those of *P. Symphyti-Bromorum* or *P. simplex*.

The first formation of septa in the hyphae occurs about the third day, and always takes place in connexion with the development of haustoria.

In addition to these septa, others appear at longer or shorter intervals in the branched hyphae up to about the tenth day, when, with the preparation for spore production, the hyphae become drained of their protoplasm and uniform septation occurs in all the older hyphae. So that at this stage the only hyphae which are full of contents, and for the most part are unseptate, are those which run in the margins of the pustules, the so-called 'Protomycelium.'

## NUCLEI.

The nuclei closely resemble those of *P. Symphyti-Bromorum*, *simplex*, and *coronifera*, and are usually of an oval shape, and in the vegetative hyphae they all divide by the direct method of division. Two such nuclei are shown in Fig. 12.

## HAUSTORIA.

The haustoria of this form, both in the earliest stages of development and when they reach maturity, bear very striking resemblances to those of *P. Symphyti-Bromorum* as figured by Marshall Ward (33). This is especially noteworthy, because Eriksson ('04) (15) in his last paper on 'The vegetative life of the Cereal Rust Fungi' expressly states that 'In no single case has he found such young stages of haustoria as those figured by Marshall Ward' (33).

Consequently I have thought it advisable to show that such stages of young haustoria are to be found in this form (*P. Phlei-pratensis*). See Figs. 8, 9, and 10.

Hauatoria are produced from the apices of the hyphae.

Before a haustorium is formed the hypha becomes distinctly swollen to a distance of about 10  $\mu$  from the extreme tip. This swollen portion becomes cut off by a transverse wall, and contains a protoplasm which is more vacuolar than that of the rest of the hyphae.

In it two or more nuclei of much smaller size than those in the hyphae are seen.

Simultaneously with the transverse wall which cuts off the haustorium, a tube-like structure begins to grow out and pierce the host cell-wall. After penetrating the wall, the little tube then begins to swell at its distal end, and very soon a brightly staining granule is seen in its interior, and this generally lies near the aperture of the tube.

As the body of the haustorium increases in size, its proximal swollen end becomes quite empty, and the small round nuclei which were present in it are now seen to have passed over into the distal end of the haustorium (Fig. 11). The large haustorium embracing the host cells nucleus in Fig. 11 is the form commonly seen in the chlorophyll containing cells, while the small hammer-headed forms are rather common in the epidermis.

*Puccinia Glumarum* (Schmidt), Erikss. and Henn.

This is the Yellow rust of wheat, by far the most prevalent of rusts on the wheat fields around Cambridge. In South Africa as yet I have not found this rust, and doubt whether it exists here.

It was originally included under *P. rubigo-vera*. DC. (*P. striaeformis*, Westd., and *P. straminis*, Fuck.).



It was first distinguished as a single species, with very distinct morphological and biological characteristics by Eriksson and Henning (16), and a few years later Eriksson divided the old *P. rubigo-vera*, DC., into seven additional forms, so that to-day under *P. rubigo-vera*, DC., he includes:—

1. *P. glumarum* (Schm.), Erikss. and Henn.  
The Yellow rust. Aecidium unknown.
2. *P. dispersa*, Erikss.  
The Brown rust of the rye. Aecidium on *Anchusae*.
3. *P. triticina*, Erikss.  
The Brown rust of wheat. Aecidium unknown.
4. *Symphyti-Bromorum*, Müll.  
The Brown rust of Bromes. Aecidium on *Symphytum*.
5. *P. agropyrina*, Erikss.  
On *Triticum repens*. Aecidium unknown.
6. *P. holcina*, Erikss.  
On *Holcus lanatus*, *H. mollis*. Aecidium unknown.
7. *P. Triseti*, Erikss.  
On *Trisetum flavescens*. Aecidium unknown.
8. *P. simplex* (Kcke.), Erikss. and Henn.  
The dwarf rust on *Hordeum vulgare*.

Eriksson (9) divides *P. glumarum* into five specialized forms.

1. *P. glumarum Tritici*, Erikss.

In which uredospores from wheat will only infect wheat, but not barley or rye.

2. *P. glumarum Hordei*, Erikss.

Uredospores from barley will only infect barley, not rye or wheat.

3. *P. glumarum Secalis*, Erikss.

Uredospores from rye infect only rye, not barley or wheat, although Eriksson says he once got a feeble success from rye to wheat, and therefore is inclined to regard his form as 'nicht scharf fixiert'. But we need further experiments on this point before it can be regarded as such.

4. *P. glumarum Elymi*, Erikss.

Uredospores from *Elymus arenarius*, L., will not infect either wheat, barley, or rye.

5. *P. glumarum Agropyri*, Erikss.

Uredospores from *Agropyrum repens*, Beauv., will not infect rye, wheat, or barley.

*P. glumarum* is also found on the following Bromes:—*Bromus adoensis*, *asper*, *arvensis*, *mollis*, *secalinus*, *sterilis*, *tectorum*.

The Uredo stage of *P. glumarum* is readily distinguished in the field by its bright yellow colour, sulphur to pale cadmium. The pustules have a great tendency to run in long rows up and down the leaf, sometimes producing a disease spot as much as 70 mm. long, but this is by no means

always the case, for frequently the Fungus will invade a leaf from an infected area in the most methodical fashion, e. g. it will spread uniformly through the leaf blade both up and down the leaf from an infected area at the rate of 10–12 mm. per day, that is to say the total length of leaf infected per day usually amounts to 20–24 mm.

The pustules are always much more numerous on the inner face of the leaves than the outer.

They are from .5–1 mm. long and .3–.4 mm. broad.

The spores are round or shortly elliptical echinulate and yellow, measuring 25–30  $\mu$  in diameter.

The uredospores are easily distinguished under the microscope through their colourless membrane from the spores of the Brown rust with its brownish coloured membrane.

An aecidium of *P. glumarum* has so far not yet been discovered, and this is not by any means surprising, for very few trials appear to have been made in this direction.

Eriksson, it is true, has tried to infect with the teleutospores (which like those of *P. dispersa* germinate directly they are ripe) *Anchusa arvensis*, *officinalis*, *vulgaris*, *Cynoglossum officinale*, and *Pulmonaria officinalis*, but with no success.

Around Cambridge the Uredo pustules are to be found all the year round, both on the side tillers in the unploughed stubble fields and also on the fields of young winter corn. It should be mentioned that especially during the months of January and February it is those leaves of the winter corn which lie or come into contact with the damp soil that are very liable to show the uredo pustules and characteristic yellow flecks.

#### THE HISTOLOGY OF UREDO GLUMARUM.

The following works dealing more or less with the histology of *P. glumarum* have already appeared:—

Eriksson and Henning (17) gave a short description of the anatomy of the Yellow Rust with a few figures.

Klebahn (24) in a short article on 'The anatomy of the Yellow Rust' drew attention to the great thickness of the hyphae in this species when compared with those of the other cereal rusts. He also suggested that Eriksson had probably mistaken the haustoria of these rusts for his 'mycoplasma.'

In January, 1904, Eriksson and Tischler (18) published as the first of a series of articles 'Über das vegetative Leben der Getreiderostpilze' a paper on '*Puccinia glumarum* (Schm.), Eriks. and Henn., in der heranwachsenden Weizenpflanze'.

This paper deals with the mycoplasma hypothesis. Both normal and abnormal hyphae are figured, and Eriksson maintains that one is only a stage in the development of the other.

In May, 1904, Klebahn (25) in 'Bemerkungen über das Mycel des Gelbrostes und über die neueste Phase der Mycoplasma-Hypothese' tries to find an explanation for some of the phenomena represented in Eriksson and Tischler's paper.

In June, 1904, Eriksson (15) published a second paper on *P. glumarum*, entitled '*Puccinia glumarum* (Schm.), Eriks. and Henn., in der heranwachsenden Gerstenpflanze', the third of the series, 'Über das vegetative Leben der Getreiderostpilze.'

But as this paper is solely concerned with what Eriksson still calls the mycoplasma stage of the Fungus, it does not concern us here, for we shall confine our attention for the present to the normal histology.

In conjunction with Marshall Ward (34) I studied the histology of this rust in very great detail. It has been examined in a number of varieties of wheat, under various conditions of growth, normal as well as abnormal. Two varieties were used rather more extensively than the others for this purpose.

1. The one known as 'Red King', because it was one of the most susceptible of the many varieties grown by Biffen at the University Experimental Farm.

2. The other was Michigan Bronze, the same variety as that with which Eriksson worked. This was chosen partly because it also is a very susceptible wheat to Yellow rust, and partly because it had been specially recommended by Eriksson.

Histologically the most striking point of difference between the *Uredo* of *P. glumarum* and the other forms under discussion is that in *P. glumarum* the hyphae are extremely thick, frequently measuring from 10–19  $\mu$  across, and at an early stage are seen to be crammed with nuclei; whereas in the other forms the hyphae are only 3.5  $\mu$  thick, and have but few nuclei arranged usually in single file.

#### GERMINATION OF THE SPORE.

The spore on germinating puts out a simple and very delicate germ-tube, which runs along the surface of the epidermis until it comes to a stoma, where the tip of the germ tube swells slightly and forms a fragile vesicle, the sub-stomatal vesicle, which applies itself closely to the inner face of the stoma.

Into this large and thick-walled vesicle the whole contents of the spore are poured, and it is frequently seen crammed with nuclei only forty-eight hours after infection (Pl. XL, Figs. 13, 14, and 15).

This sub-stomatal vesicle is generally of a definite shape, being usually cylindrical with rounded ends.

In diameter it measures from 8 to 19  $\mu$ . Its position relative to the stoma is also very definite, for it lays itself with its long axis parallel to

the stomatal slit, so that in a transverse section of the stoma it is seen as a circular body blocking up the stoma (Fig. 16), and in longitudinal section as a long oval body fitting against the guard cells.

With the formation of the sub-stomatal vesicle we may regard the Fungus as having effected an entry into its host.

The next step in the development is the formation of the true infecting hypha.

This springs from one end of the sub-stomatal vesicle, and either strikes straight across the sub-stomatal intercellular space or runs between the mesophyll-cells and the epidermis.

The infecting hypha at once forms a haustorium in the first cell with which it establishes contact, and then we may say that infection has taken place. Frequently, in the same section, spore, germ-tube, appressorium, sub-stomatal vesicle, and the first infecting hypha with its haustorium, may be seen.

Before describing the hyphae and their general behaviour, it should be pointed out at once that they always possess a definite membrane which is clearly visible from their first formation to the production of spores.

Even in cases where the hypha fits itself into large and irregular intercellular spaces, the outer membrane is well developed. From the third day onwards to the seventh day the hyphae attain to the maximum development, and two distinct kinds of branches are seen.

1. The short branches, which are frequently septate and ramify in the intercellular spaces between the chlorophyll-containing cells.

2. The long branches or runners. These are very vacuolated, very rarely septate, and apparently do not form haustoria.

These run up and down the leaf, and are the hyphae which cause the rapid spread of the Uredo pustules (Fig. 22).

One of these hyphae running from the margin of a Uredo pustule will in twenty-four hours attain a length of 12 mm., it then branches repeatedly at its apex, forming a nest of hyphae, which in the next twenty-four hours form the beginnings of a new Uredo pustule.

#### SEPTATION OF THE HYPHAE.

There are three distinct formations of septa:—

1. Those formed in connexion with the haustoria. Directly a haustorium is formed from the tip of a hypha, a transverse septum makes its appearance and cuts off the tip. These septa may be formed as early as the third day.

2. In the same way septa are formed in the short branches of the hyphae, which become so abundant in the intercellular spaces from the sixth day onwards.

3. Uniform septation of the normal hyphae does not occur as a rule until the day before spore formation, so that it is brought about in a comparatively short space of time.

#### NUCLEI.

Each nucleus consists of a central body which stains as a deep homogeneous mass with fuchsin, gentian violet, and haematoxylin.

This central body lies in a clear space, the nuclear zone, and in the young and actively growing hyphae, no definite nuclear membrane can be made out surrounding it. But in the older hyphae, especially in the long runners where the protoplasm is more vacuolated, and the nuclei not quite so numerous, the typical resting nucleus is seen. Here it consists of a central body lying in a clear area surrounded by a definite nuclear membrane, and in some cases connecting strands can be seen running from the central body to the nuclear membrane.

In connexion with the nuclei, which must be in an active state of division in the young hyphae, distinct centrosomes can be made out. These appear as a single, deep-staining small body just outside the border of the nuclear zone. As a rule they are surrounded by a clearer space, but no definite membrane or radiations are to be seen (Fig. 15). The nuclei divide very rapidly up to the sixth and seventh day, when a change begins to take place. Some of them no longer stand out sharply, but they appear as dull red specks with no distinct membrane, and there is a great tendency for these small and degenerating nuclei to mass together in patches so that the hyphae often show a corroded appearance. The surviving nuclei are conspicuous by their large size and staining capacity.

#### HAUSTORIA.

The commonest form for *P. glumarum* is a small club-shaped body, which is very frequently seen in close connexion with the nucleus of the cell (Fig. 24), whereas the hammer-headed form so frequently met with in *P. Symphyti-Bromorum* is seldom seen.

The haustoria often contain as many as five nuclei, and attain their largest dimensions in the cells surrounding the vascular bundles, where they are usually very numerous and much branched. In no case have they been seen in the vascular elements themselves.

As the haustoria become older, they become very thick walled, and have an empty or very vacuolated appearance, while the wall stains very deeply and the nuclei can no longer be made out. Arborescent forms, such as are shown in Figs. 18 and 19, are frequently found.

#### PREPARATION FOR SPORE FORMATION.

Towards the eighth day the hyphae become very vacuolated, contain fewer nuclei, which are frequently of an oval shape and stain very deeply.

The hyphae from now onwards become extremely thin (Figs. 20 and 21), from  $3.5-6\mu$  in thickness, and always have a great tendency to make for the stomatal cavities. Here they branch very rapidly, forming a dense web, giving rise to the *Uredo sori*, which are seen bulging out the cells in the region of the stomata.

*Puccinia dispersa*, Erikss.

This is the Brown rust of the rye, originally included under the old *P. 'Rubigo-vera'* (DC.), as *Aecidium asperifolii*, Pers.

Eriksson (9) was the first to point out the characteristics of the 'Braunrost' (*P. dispersa*, Erikss. and Henn.), and very soon afterwards he split it up into the specialized forms (f. sp. *Secalis*, *Triticici*, *Bromi*, *Agropyri*).

Later on Eriksson (11) showed that the brown rust of the rye *P. dispersa*, Erikss. (not Erikss. and Henn.), is a distinct species from the other brown rusts, for its uredo and teleutospores are only found on *Secale cereale*, L., and *S. montanum*, Guss., not on *Triticum*, &c.

The aecidium is found only on *Anchusa arvensis* and *A. officinalis*. The teleutospores are capable of germination immediately after their formation, and as was shown by Eriksson and confirmed by Klebahn (22), when the teleutospores from *Secale cereale* are sown on *Anchusa arvensis* and *A. officinalis* the aecidia are always produced, whereas sowings of the teleutospores from *Triticum vulgare*, *Bromus mollis*, *Bromus macrostachys* failed to produce the aecidia on *Anchusa*. The aecidiospores from *Anchusa arvensis* and *A. officinalis* always successfully infected *Secale cereale*, but these aecidiospores when sown on *Triticum vulgare*, *Bromus arvensis*, *Hordeum vulgare*, *Avena sativa*, *Agropyrum repens*, *Holcus lanatus* produced no results.

Also uredospores from *Secale cereale* would only successfully infect *Secale cereale*, but failed when sown on *Triticum vulgare*, *Bromus mollis*, *Bromus arvensis*, *Agropyrum repens*, *Holcus lanatus*, *Trisetum flavescens*, to produce the *Uredo* pustule.

In the field the uredo stage is distinguished by the pustules being from 1 to 1.5 mm. long, nearly 1 mm. broad, and scattered irregularly over the leaf. As a rule the pustules are of a Terra Sienna colour (but too much reliance must not be placed on colour).

The spores are usually round, spinous, of a dirty yellow, and measure in diameter 19-20  $\mu$ .

THE HISTOLOGY OF UREDO DISPERSA, ERIKSS.

The general development of the mycelium of *P. dispersa* is very similar to that of *P. glumarum* and *P. Symphyti-Bromorum*, and the other species to be described.

Morphologically the mycelium of *P. dispersa* more closely resembles that of *P. Symphyti-Bromorum* than that of *P. glumarum*, but it is at once distinguished from either of these two species by its characteristically shaped sub-stomatal vesicle.

When the spore germinates a delicate germ-tube is formed, which may branch repeatedly until the tip reaches a stoma, where it swells up and forms the appressorium (as is seen in the oblique section of Fig. 25).

A short tube grows out from the appressorium, which passes through the stoma, and then immediately swells out into the large substomatal vesicle, which differs very strikingly in shape from that of *P. Symphyti-Bromorum*, which is more triangular and does not become transversely septate.

In diameter the substomatal vesicle is from 11–14  $\mu$ , and like that of *P. glumarum*, it lays itself with its long axis parallel to the stomatal slit (see Figs. 26 and 27), but it differs from *P. glumarum* in that it gives off at each end an infecting hypha, and it very soon becomes divided by a transverse septum, which never takes place in the substomatal vesicle of *P. glumarum*.

#### THE HYPHAE.

The hyphae in general appearance and structure are very like those of *P. Symphyti-Bromorum*, containing few nuclei, which are usually seen in pairs and arranged in single file in the hyphae. The hyphae measure from 3–5  $\mu$  across, and in the young condition are full of protoplasm and non-septate, but as they grow older from the sixth day onwards they lose their protoplasmic contents and become septate, so that at the time of pustule formation the hyphae become quite empty and closely septate, forming a dense pseudo-parenchyma.

#### NUCLEI.

The nuclei in the young and actively growing hyphae are of an oval shape, consisting of a single deeply staining body surrounded by a clear space, the nuclear zone. Within this clear space a distinct centrosome can usually be made out towards the polar end (Fig. 33). No definite nuclear membrane can be made out in connexion with the nuclei in this condition. But in the resting condition the typical nucleus, as has already been described under *P. glumarum*, is seen.

#### THE HAUSTORIA.

The haustoria of *P. dispersa* are much more uniform in shape than those found with *P. glumarum* or *P. Symphyti-Bromorum*.

The common forms met with are those shown in Fig. 34; branch forms are very rarely found.

*Puccinia triticina*, Erikss.

The Brown rust of wheat, now known as *P. triticina*, Erikss., is probably identical with the rust still known in the United States as the 'orange-leaf rust of wheat,' *P. rubigo-vera Tritici*, originally included under *P. rubigo-vera*, DC. (*P. straminis*, Fuck. = *P. striaeformis*, West). Eriksson and Henning (16) first sorted it out under the name *P. dispersa* forma specialis *Tritici*.

Five years later Eriksson (11) showed that the brown rust of wheat was a distinct species, which he named *P. triticina*, Erikss. For he found that unlike *P. dispersa* the teleutospores germinated only after the winter, they would not infect *Anchusa* through their sporidia, and no infection resulted on wheat with the aecidiospores from *Anchusa*.

In the field *Uredo triticina* is easily distinguished from *Uredo graminis* by its smaller pustules, which usually are found on the leaves only. The pustules measure from .3–.5 mm. in length and .1–.2 mm. in breadth, and are closely crowded together on the leaf surface. Large erumpent pustules so characteristic of *Uredo graminis* never occur. The spores are round to shortly elliptical, 19–20  $\mu$   $\times$  24–25  $\mu$ , smooth and dirty yellow in colour.

## THE HISTOLOGY OF UREDO TRITICINA.

When we come to compare the histology of *Uredo triticina* with that of *Uredo dispersa* we at once find a striking difference between the infection organs of these two parasites, as will be seen by comparing Figs. 25–27 with Figs. 28–30.

The substomatal vesicle of *Uredo triticina* is globular in shape, and gives origin to a single infecting hypha, from which other hyphae may eventually arise. A typical vesicle is shown in Fig. 28. In length this vesicle is 21  $\mu$ , breadth 12  $\mu$ , and diameter 15  $\mu$ .

In Fig. 30 is shown a transverse section through a stoma, in which the appressorium, neck, substomatal vesicle, and infecting hypha with the first haustorium is seen.

The resulting hyphae are very similar to the forms already described (with the exception of *P. glumarum*) and need little mention.

*Puccinia Symphyti-Bromorum*, F. Müll.

Originally the Brown rust of the Bromes was placed together with the Brown rust of Rye under the name *Puccinia rubigo-vera*, DC. Eriksson (9) was the first to separate it off as *Puccinia dispersa*, Erikss. et Henn., forma specialis *Bromi*.

Later on he regarded it as a distinct species, *Puccinia bromina*, Erikss. (11), from the fact that he was unable to infect plants of Rye with uredospores from the Bromes, and vice versa to infect Bromes with uredospores from the Rye.



Müller (26) (27) showed that the Aecidium of this species is formed on *Symphlytum officinale*, L. and *Pulmonaria montana*, Lej., and for this reason it has now been called *P. Symphyti-Bromorum* by Klebahn.

Not only is it to be distinguished by these physiological differences from *P. dispersa* on Rye, but as will be seen from my figures, the uredo mycelium of *P. Symphyti-Bromorum* is morphologically distinct from that of *P. dispersa*.

As the histology of *P. Symphyti-Bromorum* has been very fully described by Marshall Ward (33) there is no need to devote any further space to it here, except to say that my observations entirely coincide with those of Marshall Ward.

To avoid confusion in nomenclature it should be noted that this *P. dispersa* examined by Marshall Ward (33) is now known as *P. Symphyti-Bromorum*.

### *Puccinia simplex* (Kcke.), Erikss. and Henn.

This is the brown or dwarf rust of the barley. It is very common on the barley fields around Cambridge, and is to be found there in the uredo stage all the year round. Up to the present I have not found this rust in South Africa.

By the earliest observers it was distinguished under various synonyms, e.g.

(*P. stramineis* var. *simplex* Kornicke, 1865; *Uromyces Hordei* Nielsen, 1875; *P. anomala*, Rostrup, 1876).

However, Eriksson and Henning (16) have shown that it must be regarded as an independent species.

In the field it is characterized by its extremely small and very scattered pustules, which in length are from .3–.5 mm. and in breadth .1–.2 mm. The uredospores are spherical to oval, with a brownish coloured membrane, which is distinctly spiny. They measure from 20–30  $\mu$  by 17–22  $\mu$ .

The teleutospores according to Klebahn are mostly one-celled, and germinate only after the winter's rest.

The Aecidium is as yet unknown, Klebahn during 1900 and 1901 made sowings on some forty-three plants, but with no success.

Klebahn (24) has found *P. simplex* on *Hordeum vulgare*, L., *vulgare* 'cornutum,' *coeleste trifurcatum*, *distichum*, L., *distichum* 'nigricans,' *hexastichum*, L., *seocriton*, L.

### THE HISTOLOGY OF UREDO SIMPLEX (KCKE.), ERIKSS. AND HENN.

Klebahn (24) in two figures on the anatomy of this rust, shows the general appearance of the hyphae and some young haustoria. He

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says the mycelium shows nothing remarkable and gives the measurement of the hyphae at  $3\mu$ .

The general development of *P. simplex* agrees in every way with the species already described, and the general structure of its mycelium very closely resembles some of these species, but it can easily be distinguished from them by its characteristic and definitely shaped sub-stomatal vesicle.

The spore on germination forms a delicate germ tube, the tip of which swells up to form the appressorium and then follows the sub-stomatal vesicle, which is at first a long sausage-shaped body, which always lays itself with its long axis parallel to the stomatal slit. At first (in shape) it is very much like *P. glumarum*, but it soon begins to differ from it in the following important points.

1. Infecting hyphae simultaneously spring from both ends of the vesicle.

These hyphae only measure from  $3-3.5\mu$  across, and contain few nuclei, which are usually seen in pairs, whereas in *P. glumarum* the sub-stomatal vesicle only gives rise to a single infecting hypha from one end. From this hypha, which measures  $10-12\mu$  across, the other hyphae arise, and these are always crammed with nuclei.

2. Soon after two or more pairs of infecting hyphae have sprung from the vesicle, it becomes divided transversely into two equal halves. Close to this partition wall, and on each side of it, a hypha then usually springs, and the vesicle may then become further sub-divided by transverse walls. (Figs. 35-37.)

Thus the sub-stomatal vesicle in *Uredo simplex* becomes multi-septate.

#### THE HYPHAE.

The hyphae closely resemble those of *P. Symphyti-Bromorum*, as described by Marshall Ward (33), both with regard to size, vacuoles, and septation.

As the hyphae begin to get older, they lose their protoplasmic contents, become very vacuolated, with a diminution in size of their contained nuclei, which are seen in the last stages of degeneration as small, round, dull, staining dots. Directly the *Uredo* pustule-formation begins, the mycelium that gave rise to it, including also the sub-stomatal vesicle, is seen to be quite empty and septate, except in the immediate neighbourhood of the pustule, where the hyphae are full of protoplasm and contain large and sharply staining nuclei.

In *P. simplex*, the long runner-like hyphae so characteristic of *P. Symphyti-Bromorum* and *P. glumarum* are not anything like so numerous or well developed, but single ones are found here and there running through the tissue of leaf as a simple thread for some distance

before they show any signs of branching. This is naturally what we would expect, for the pustules are always few, and scattered at great distances on the leaf.

## NUCLEI.

The nuclei closely resemble those of *P. Symphyti-Bromorum*. They are oval in shape, and nearly always seen in pairs, lying just one behind the other in the thin hyphae. They divide very rapidly by the direct method. A large nucleus becomes drawn out into a long oval-shaped body and is then gradually constricted at the middle, the two halves finally separate, regain their normal size and shape, and remain for some time in close proximity to one another.

## HAUSTORIA.

The fully-developed haustoria all appear to be of the shape shown in Fig. 44, but frequently this form is seen coiled on itself, as is shown by Marshall Ward (33) for *P. Symphyti-Bromorum*. Here, again, the hammer-headed form so common in *P. Symphyti-Bromorum* is very rarely seen.

*Puccinia coronifera*, Kleb.

This is the 'crowned rust' of the Oats; for some time it was included under the Crown Rust proper, *P. coronata*, Corda, but for reasons given below it must now be regarded as a distinct species.

Nielsen (28) first showed that the aecidiospores from *Rhamnus cathartica* when placed on *Lolium perenne* produced the uredospores, and eventually teleutospores, whereas aecidiospores from *Rhamnus Frangula* failed to infect *Lolium perenne*.

Plowright (30) found that the teleutospores from *Dactylis glomerata* and *Festuca silvatica* readily produced the aecidium on *R. Frangula*, and so he says that he thinks two species are confounded under the name *P. coronata*. He also adds the following interesting and important fact, which I also have noted during the past two years, viz. the fungus which occurs on *Lolium perenne* is accompanied by a profuse development of uredospores, but only in the autumn, from September to November, after which the teleutospores follow; whereas the fungus on *Dactylis* is an early summer species with a much less free development of uredospores. Klebahn (20) in 1892 showed that the teleutospores from *Lolium perenne* when sown on *Rhamnus cathartica* readily produced the spermogonia and aecidia, whereas similar sowings on *R. Frangula* failed.

On this account Klebahn (20) in 1892 insisted on the splitting up of the old *P. coronata*, Corda, into two species.

I. *P. coronata*, Corda. With its aecidium on *Rhamnus Frangula* (*Aecidium Frangulae*, Schum.), uredo- and teleutospores on *Dactylis glomerata*, *Festuca silvatica*, and other grasses.

II. *P. coronifera*, Kleb. With its aecidium on *Rhamnus cathartica* (*Aecidium Rhamni*, Gmel.). Uredo- and teleutospores on *Avena sativa*, *Lolium perenne*, and *Festuca elatior*.

There are six specialized forms of *P. coronifera*:—

1. *P. coronifera Avenae*, Erikss. Uredospores from Oats will infect only oats, not other grasses.
2. *P. coronifera Lolii*, Erikss. Uredospores from *Lolium perenne* will only infect *Lolium perenne*.
3. *P. coronifera Festucae*, Erikss. Uredospores from *Festuca elatior*, but not *Avena sativa*, *Alopecurus pratensis* (Erikss.). *Holcus mollis* and *H. lanatus* (Kleb.).

Further experiments are needed in the case of its transference to *Lolium perenne*, for Eriksson entirely failed to infect this latter, whereas Klebahn got a very fair percentage of infections on it.

4. *P. coronifera Holci*, Kleb. Uredospores from *Holcus mollis* and *H. lanatus* infect only these plants.

5. *P. coronifera Alopecuri*, Erikss. Uredospores from *Alopecurus pratensis* infect only *Alopecurus pratensis*.

6. *P. coronifera Glyceriae*, Erikss. Uredospores from *Glyceria aquatica* infect only *Glyceria aquatica*.

*Uredo coronifera* is easily distinguished in the field from *Uredo graminis* on Oats, by its light yellow-coloured pustules. These are of smaller size than those of *U. graminis* and are usually only found on the leaf, rarely on the stem.

The spores are round, yellow, and measure from 20–22  $\mu$  in diameter.

I have evidence to show that the Crowned Rust of Oats does considerable damage to the Oat crops in the Transvaal. A point of interest and one worthy of note is that this rust appears later in the season than *Uredo graminis*. As far as I am aware the aecidium has not been definitely shown in this country, but it should be mentioned that MacOwan in 1877 collected an aecidium on *Rhamnus prinoides* in Cape Colony.

According to Carleton this rust is widely distributed in the States, and is heteroecious on *Rhamnus lanceolata*.

#### THE HISTOLOGY OF UREDO CORONIFERA AVENAE, ERIKSS.

The general development of the Fungus agrees in every way with the species already described, so that only the more important points will be mentioned here.

The appressorium (only the remains of which are seen in Figs. 38, 39) is very similar to that of *Uredo graminis*, but the attendant substomatal vesicle is of very different shape and gives rise at once to two infecting hyphae, one from each end of the vesicle. These in turn frequently branch dichotomously.

The haustoria, especially in the cells bordering on the vascular bundles, are of large size and cylindrical in shape. The pin-head type so common in the case of *Uredo graminis* is rarely met with.

*Puccinia Sorghi*, Schwein.

This rust is probably to be found wherever Maize is cultivated. In some parts of South Africa considerable damage has been wrought in the 'Mealie' crop through the agency of this parasite; but as a general rule little attention is paid to it.

Kellerman (19) has shown that the uredospores of *P. Sorghi* readily infect any and all of the 'agricultural species' of Maize, and that Teosinte (*Euchlaena luxurians*) was also a host for this species of rust, but that attempts to inoculate *Sorghum vulgare*, *Saccharum officinarum* and *Tripticum dactyloides* failed.

Recently Arthur (1) (2) has shown that this rust is heteroecious on *Oxalis cymosa*, successfully producing the aecidia on *Oxalis* from the teleutospores, and in turn infecting the Maize with the aecidiospores so obtained.

In this country MacOwan, as far back as 1879, found an *Aecidium* in Cape Colony on *Oxalis Bowei*, and quite recently I find that Burt-Davy has collected an *Aecidium* in Swaziland on *Oxalis setosa*. Whether these two act as hosts for *P. Sorghi* remains to be seen.

The pustules of *Uredo Sorghi* are found on both sides of the leaf. When they are closely crowded, they frequently run together to form a large dark brown erumpent pustule.

The uredospores are globose, elliptical or ovate, slightly verruculose, and measure from  $23-30 \times 22-25 \mu$ .

The sub-stomatal vesicle is somewhat triangular in shape when seen in longitudinal section (Figs. 40, 42), and gives rise to two or more infecting hyphae from the angles of the base.

In transverse section it is of the form shown in Fig. 43.

The hyphae are a little more robust than those of the other species described above, with the exception of *P. glumarum*, measuring as a rule from  $4.5-5 \mu$  in diameter.

The haustoria are frequently branched, as is shown in Fig. 41, and are constantly found in contact with, or carefully wrapping round, the nucleus of the host cells.

SUMMARY OF RESULTS.

As the accompanying table will show, these different species of *Puccinia*, in the early stages of development of their *Uredo* mycelium exhibit morphological characters (seen especially in connexion with the formation of the sub-stomatal vesicle) which serve at once to distinguish them from one another.

The sub-stomatal vesicle is of a definite shape for each species. It may be non-septate, septate, or even multi-septate, and may give rise to one, two, or more infecting hyphae, according to the species. In some species a well-defined appressorium is present, in others it is not so apparent.

The vegetative hyphae of the different species closely resemble each other in size and general behaviour, except in the case of *P. glumarum*, where they are much thicker than any of the other forms, and contain a much greater number of nuclei.

The haustoria of some of the species are very distinctive, e.g. the hammer-headed form is one most commonly met with in *P. Symphyti-Bromorum*, whereas in *P. glumarum*, especially in the cells surrounding the vascular bundles, they are frequently very branched, while in the chlorophyll-containing cells the typical form of haustoria is that of a small club-shaped body.

Whether such slight morphological differences as are exhibited by the mycelia of *P. graminis* on Wheat and *P. Phlei-pratensis* are sufficient to warrant them specific rank, may at first sight seem doubtful. Consequently, I will leave the discussion of these points, until I treat of the so-called 'physiological species' which will, I think, have an important bearing on the matter.

TABLE SHOWING HISTOLOGY OF UREDO MYCELIA.

Species.	Germ Tube.	Appressorium.	Substomatal Vesicle.	Hyphae.	Haustoria.	Nuclei.
<i>P. graminis</i> on Wheat.	Two present as a rule, one generally becomes branched.	Well defined.	Cylindrical, non-septate, one end applies itself closely to the head of the guard cell, and gives off the infecting hypha. 27 $\mu$ in length. 9 $\mu$ in diameter.	Narrow, 3.5 $\mu$ in thickness.	Branched and unbranched.	Not numerous.
<i>P. Phlei-pratensis</i> on <i>Phleum pratense</i> .	Two present as a rule, one soon develops more quickly than the other.	Not so well defined as in <i>P. graminis</i> .	Cylindrical, non-septate. Gives rise to a single infecting hypha. 5-7 $\mu$ in diameter.	Very narrow and thread-like. Hardly 3 $\mu$ in thickness.	Mostly unbranched.	Not numerous.
<i>P. Glumarum</i> on Wheat.	Unbranched as a general rule.	No very definite structure, germ tube just becomes slightly swollen.	Somewhat oval in shape. Non-septate, only gives rise to single infecting hypha. 18-19 $\mu$ in diameter.	Broad and crammed with nuclei. From 10-19 $\mu$ in thickness.	Both branched and unbranched.	Very numerous.
<i>P. dispersa</i> on Rye.	Branched.	Well defined.	Cylindrical in shape, divided by transverse septum. Gives rise to two infecting hyphae, one from each end of the cylinder. 11-14 $\mu$ in diameter.	From 3-5 $\mu$ in thickness, with nuclei arranged in single file.	Unbranched mostly, cylindrical in shape.	Not numerous.
<i>P. triticea</i> on Wheat.	Branched.	Well defined.	Spherical in shape, non-septate. Gives off single infecting hypha. 15 $\mu$ in diameter.	Narrow, 3.5 $\mu$ in thickness.	Branched and unbranched.	Not numerous.
<i>P. Symphyti-Bromorum</i> on <i>Bromus mollis</i> .	Generally branched.	Rather variable in shape.	Somewhat triangular in shape. Non-septate. Gives rise to one or two infecting hyphae.	Narrow and thread-like, about 3.5 $\mu$ in thickness.	Very variable in shape. Hammer-headed form very common.	Not numerous.
<i>P. simplex</i> on Barley.	Rarely branched.	Not well defined.	Cylindrical, multi-septate. May give rise to four or more infecting hyphae. 12-14 $\mu$ in diameter.	Narrow, 3.5 $\mu$ in thickness.	Unbranched, mostly of cylindrical shape.	Not numerous.
<i>P. coronifera</i> on Oat.	Branched or unbranched.	Not well defined.	Cylindrical, one septate, gives rise to two infecting hyphae, one from each end of the vesicle.	Narrow, 3.5 $\mu$ in thickness.	Unbranched, mostly of cylindrical shape.	Not numerous.
<i>P. Sorghi</i> on Maize.	Branched or unbranched.	Not well defined.	Triangular in shape, an oblique septum frequently present. Usually gives rise to two infecting hyphae from the angles at the base of the triangle.	Narrow, from 4.5-5 $\mu$ in thickness.	Branched and unbranched.	Not numerous.

## LITERATURE.

1. ARTHUR ('04): Bot. Gaz., xxxviii, 64, 1904.
2. ——— ('06): Jour. Mycol., xii, 1906.
3. BARCLAY: Journal of Bot., xxx, 45.
4. DE BARY ('68): Ann. des Sc. Nat., 4, xx.
5. ——— ('65): Monatsber. Akad. Berlin, p. 23.
6. BLACKMAN ('04): Ann. Bot., xviii, July.
7. BOLLEY ('06): Bull. 68 North Dakota, p. 627.
8. CARLETON ('89): Div. of Veg. Phys. and Path., Bull., xvi.
9. ERIKSSON ('94): Ber. Deutsch. Bot. Ges., xii, 310.
10. ——— ('96): Zeitsch. f. Pfl.-Krank., vi, 197.
11. ——— ('99): Ann. des Sc. Nat. 8, 9, 1899, p. 241.
12. ——— ('01): " " xiv, p. 101.
13. ——— ('02): " " Nos. 8 and 15.
14. ——— ('02): Ofv. Vet. Akad. Forh., p. 191.
15. ——— ('04): Kungl. Svenska. Vetenskaps. Akad. Handl., Bd. xxxviii, No. 3.
16. ERIKSSON and HENNING ('94): Zeitschr. f. Pfl.-Krank., iv, 141.
17. ——— ('96): Die Getreideroste.
18. ERIKSSON and TISCHLER ('04): Kungl. Svenska. Vetenskaps. Akad. Handl., Bd. xxxvii, No. 6.
19. KELLERMAN ('05): Jour. Mycol., January 11, 1905.
20. KLEBAHN ('92): Zeitsch. f. Pfl.-Krank., ii, 258.
21. ——— ('96): " " v, 332.
22. ——— ('97): " " vi, 36.
23. ——— ('98): " " viii, 338.
24. ——— ('00): " " x, 77.
25. ——— ('04): Ber. d. D. Bot. Ges., xxii, 255.
26. MÜLLER ('00): Vovl. Mitt. Bot. Centralbl., lxxiii, 1900.
27. ——— ('01): Bot. Centr., x, 181.
28. NIELSEN ('77): Bot. Tidsskr., 3 R. 2, p. 26.
29. PLOWRIGHT ('82): Gard. Chron., p. 234.
30. ——— ('88): Grevillea, xi, 52.
31. SAPIN-THOUFFY ('98): Le Botaniste, p. 29.
32. MARSHALL WARD ('88): Ann. Bot., ii, p. 217.
33. ——— ('08): Phil. Trans., cxcvi, B.
34. ——— ('05): Ann. Bot., xix, Jan., p. 35.

## EXPLANATION OF PLATES XL-XLIII.

Illustrating Mr. Pole Evans's paper on Cereal Rusts.

All the figures were drawn with the help of the camera lucida.

Figs. 1-6, 28-30, 38-43 were drawn with No. 2 eye-piece, and Zeiss Achromatic Apert. 1.30 Homog. Immers.

The remaining figures were drawn with Zeiss Apochromat. 2 mm. Apert. 1.40 Homog. Immers., using the No. 2 eye-piece for Figs. 13, 14, 16, 17, 19-24, the compensating ocular 8 for Figs. 7-11, 15, 18, 25-7, 31-7, 44, 45, and for Fig. 12 the compensating ocular 18.

## PLATE XL.

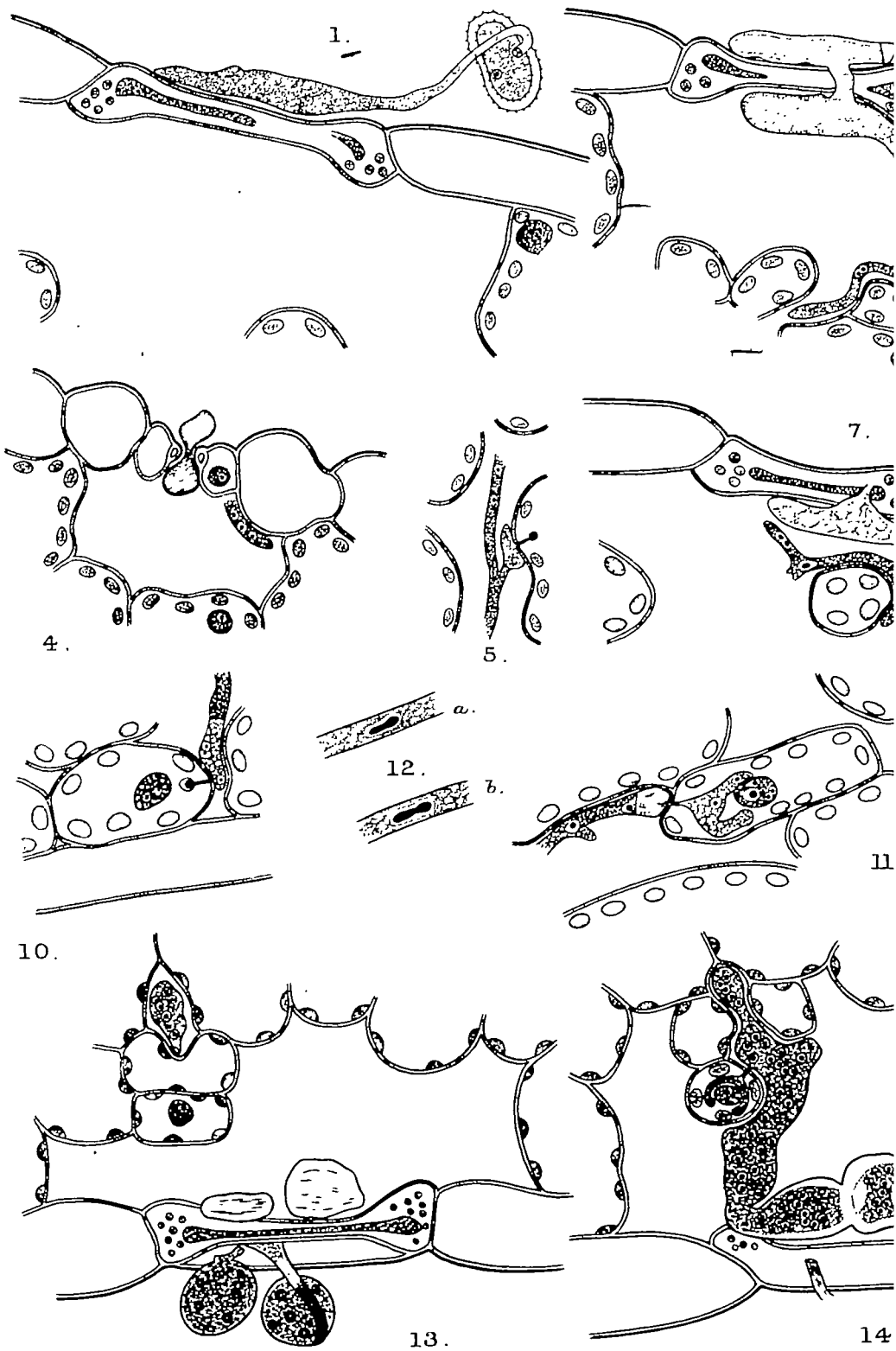
Figs. 1-6. Uredo infection of *P. graminis* on Wheat.

Figs. 7-12. Uredo infection of *P. Phlei-pratensis* on *Phleum pratense*.

Fig. 1. Germination of uredospore of *Puccinia graminis*, with formation of appressorium over the stoma of wheat leaf. From a culture twenty-four hours old.

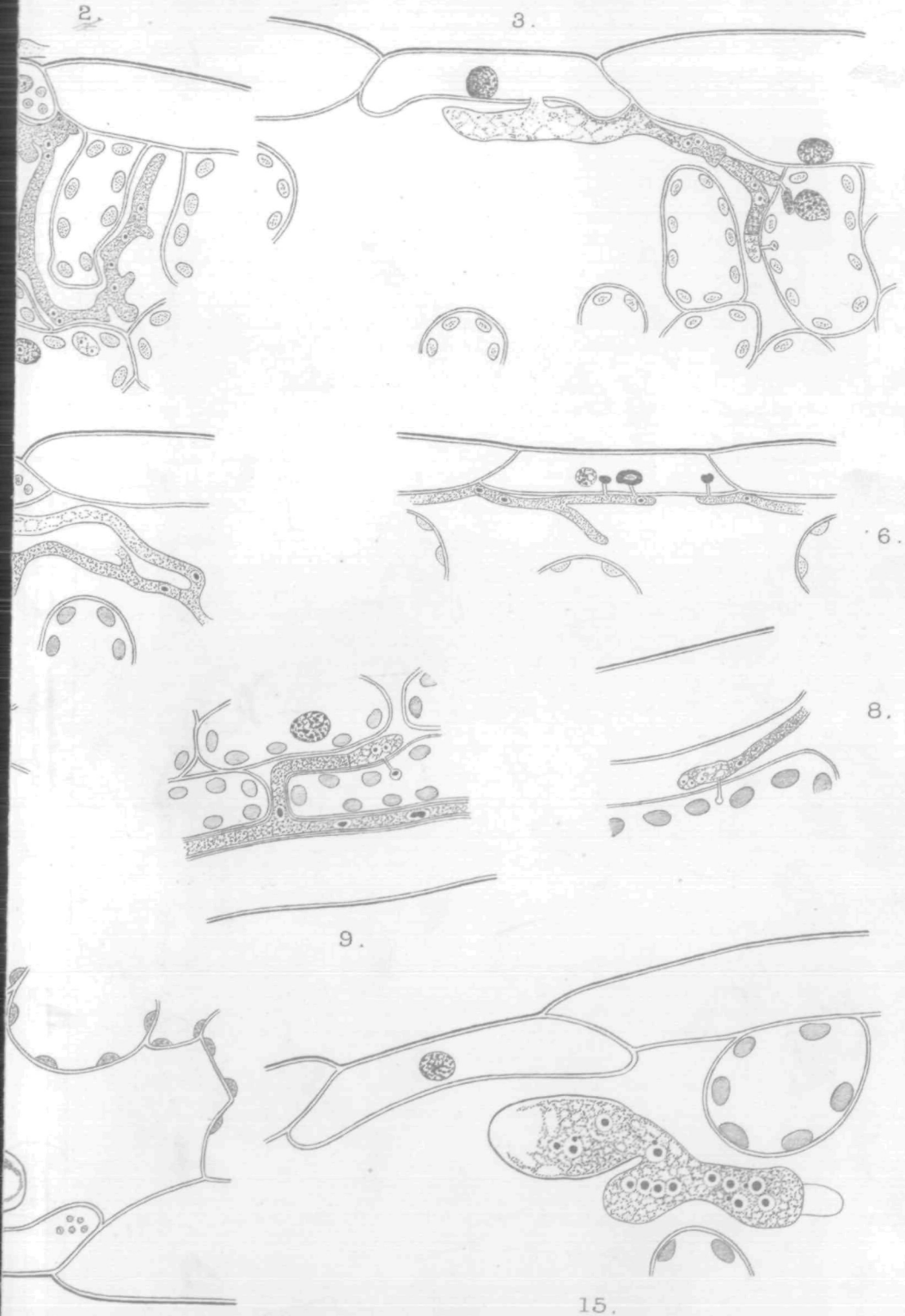






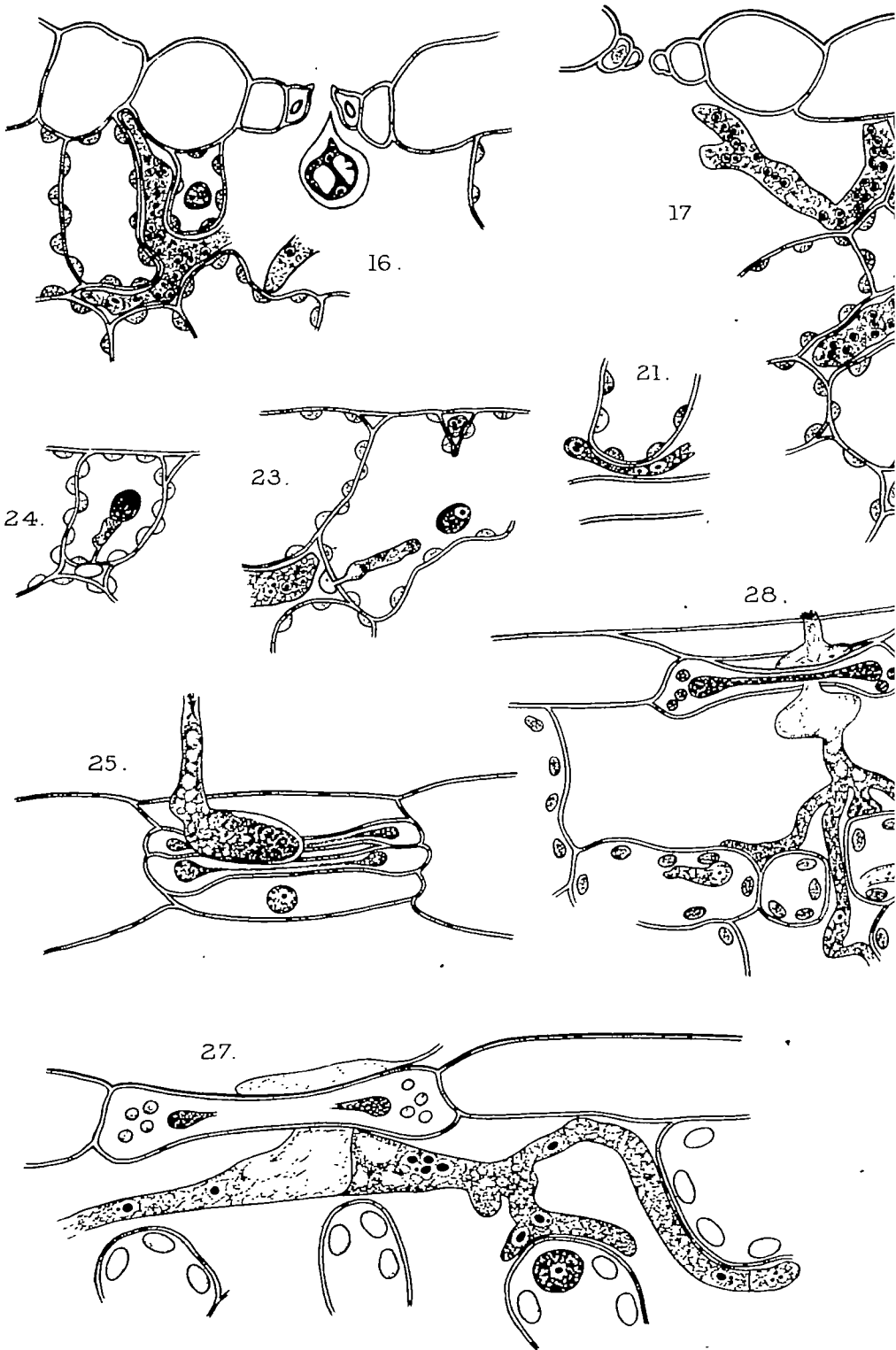
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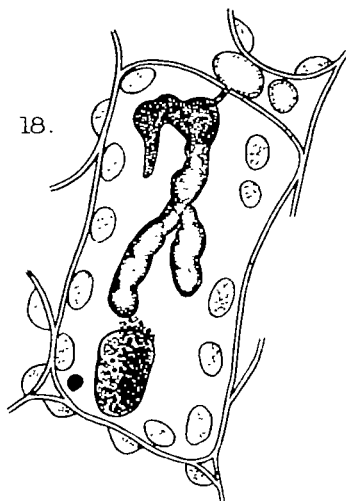




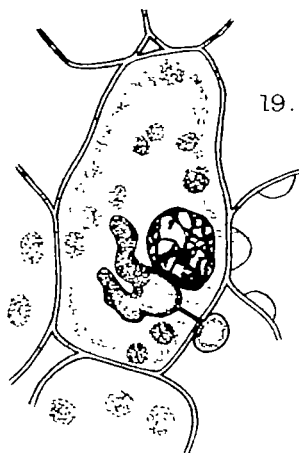


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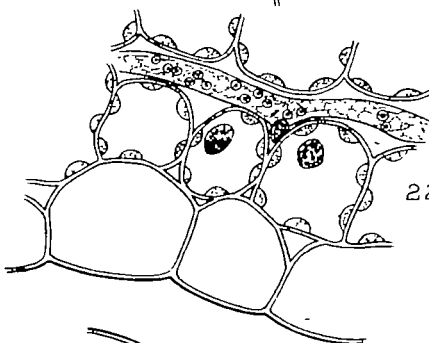
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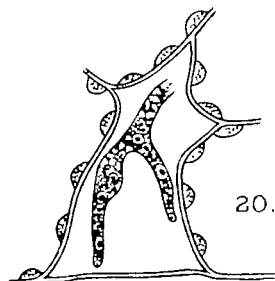
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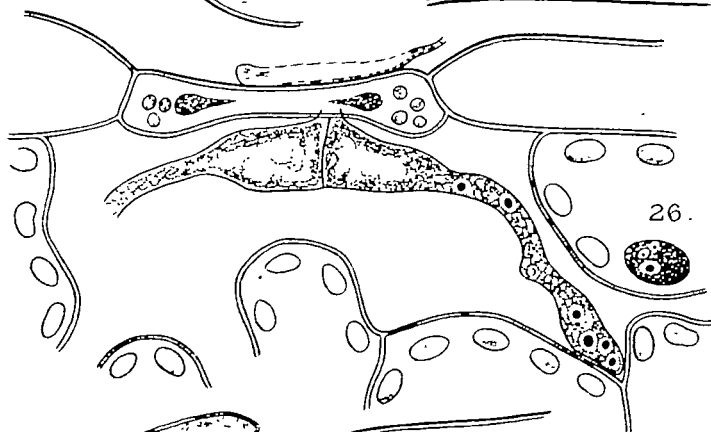
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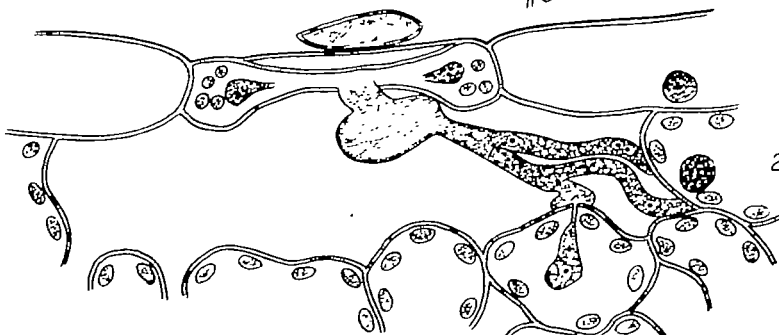
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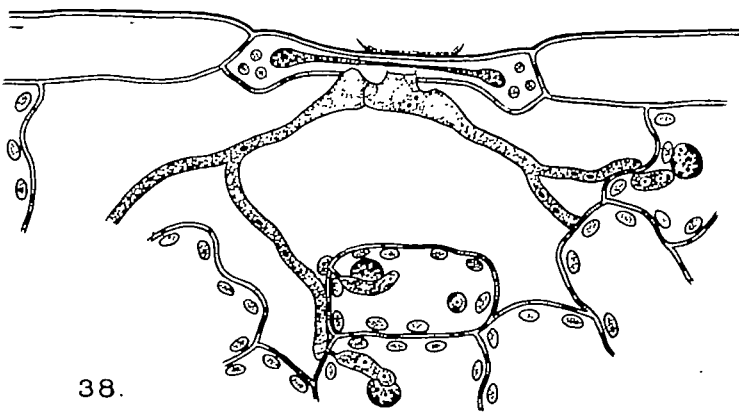
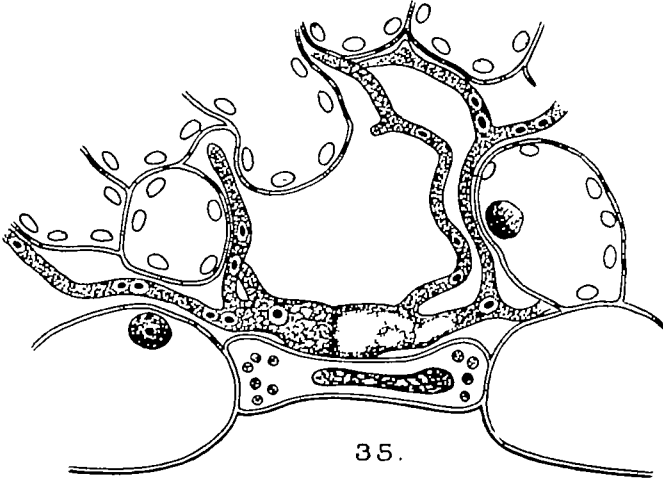
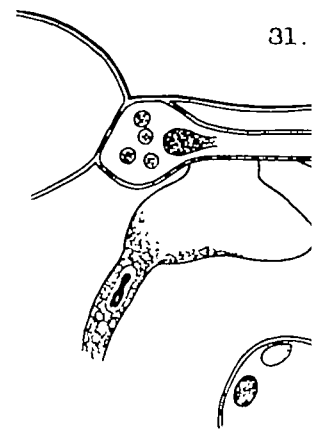
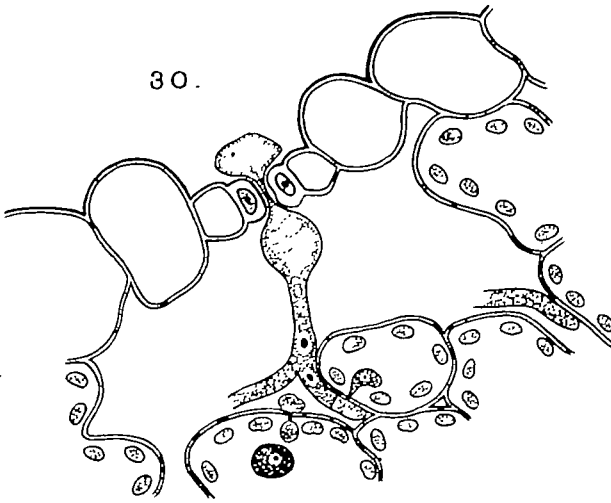
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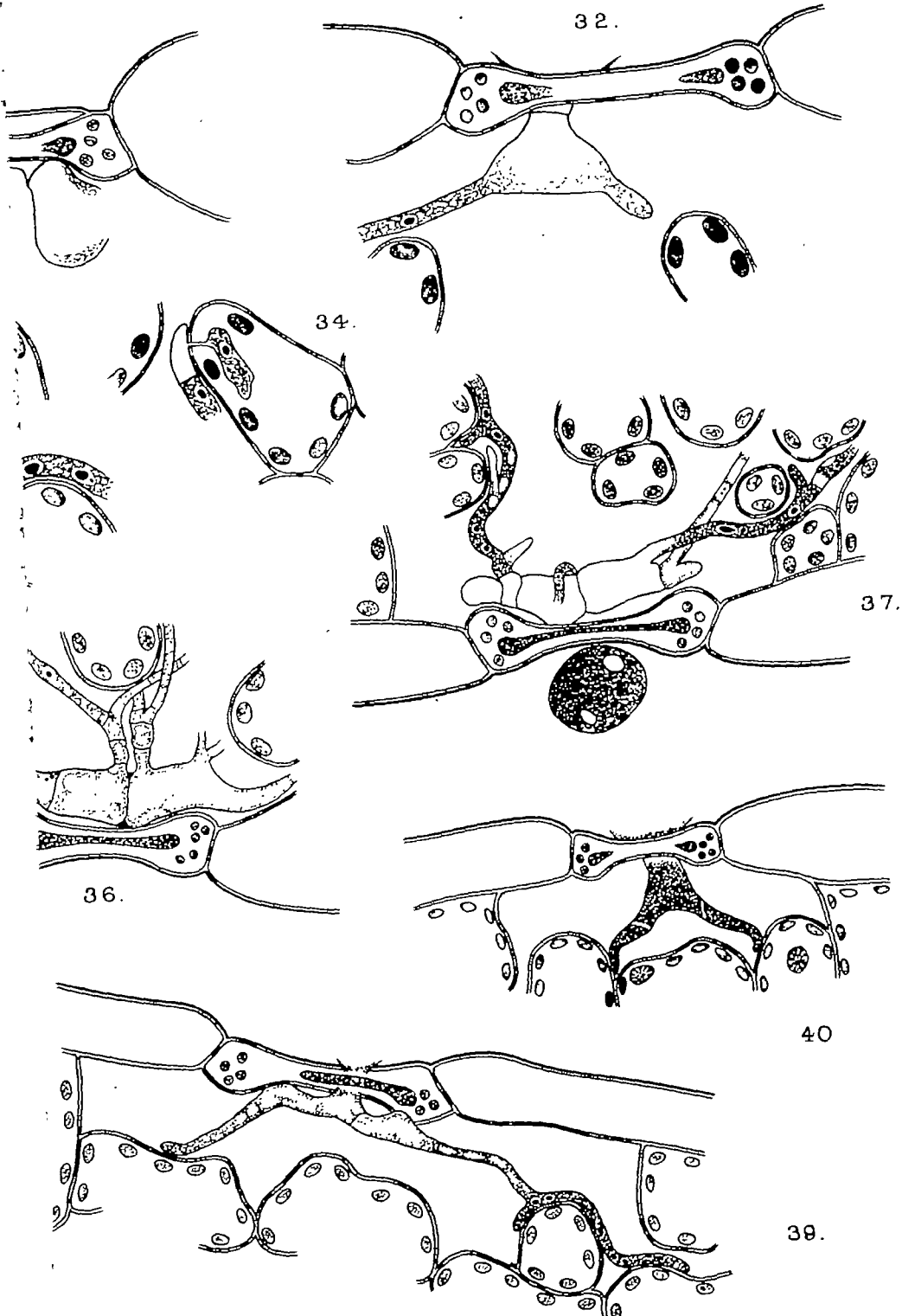




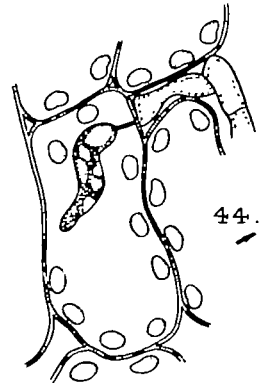
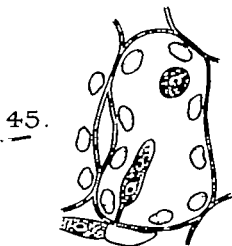
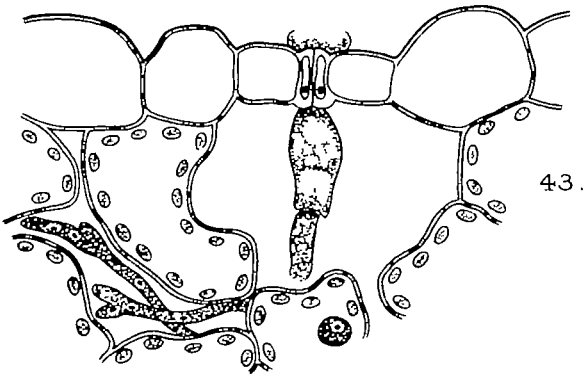
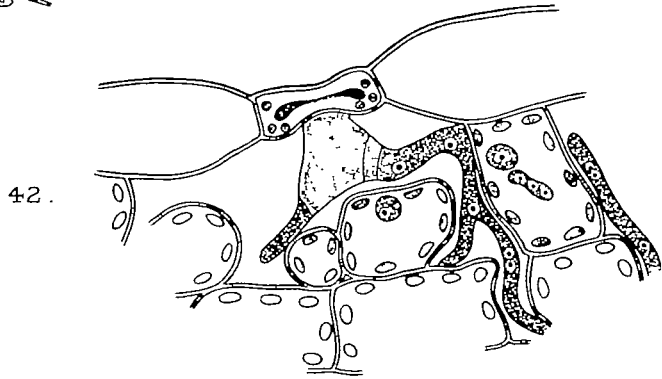
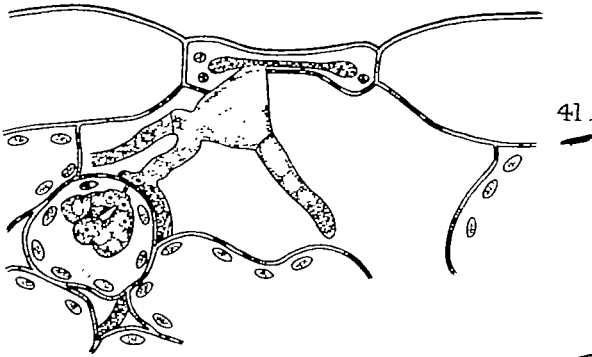












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Fig. 2. Longitudinal section through stoma of wheat leaf, showing appressorium, substomatal vesicle, and infecting hypha of *P. graminis*. Four-day culture.

Fig. 3. Similar section of wheat leaf, showing substomatal vesicle and infecting hypha with haustoria. Four-day culture.

Fig. 4. Transverse section of wheat leaf, passing through a stoma, in which the appressorium and substomatal vesicle are seen, with the tip of an infecting hypha in the stomatal cavity. Four-day culture.

Fig. 5. Portion of hyphal filament with young haustorium.

Fig. 6. Typical haustoria found in the epidermal cells. Four-day culture.

*P. Phlei-pratensis*.

Fig. 7. Longitudinal section through stoma of leaf of *Phleum pratense*, showing stomatal vesicle and infecting hypha. From an eight-day culture.

Figs. 8, 9, and 10. Typical young haustoria of *P. Phlei-pratensis*.

Fig. 11. Older haustorium, its behaviour towards the nucleus of the chlorophyll-containing cell is very characteristic.

Fig. 12. Two vegetative nuclei in the process of direct division.

All the figures of uredo infection of *P. glumarum* on Wheat.

Fig. 13. Longitudinal section of leaf. Two spores are seen germinating at one stoma. From one the germ tube and appressorium are seen, with just a graze of the substomatal vesicle.

Fig. 14. The next section to the above showing the substomatal vesicle with its first infecting hypha, which has formed a haustorium in the first cell with which it has come in contact. From a five-day culture.

Fig. 15. Longitudinal section showing the oval substomatal vesicle just under a stoma, with the first infecting hypha, which has become septate at the tip previous to the formation of a haustorium. In connexion with some of the nuclei of the hypha, centrosomes are clearly seen. Third-day culture.

#### PLATE XLI.

Fig. 16. Transverse section across a stoma, showing the substomatal vesicle in section. Eight-day culture.

Fig. 17. Typical hyphae with their numerous nuclei. Eight-day culture.

Fig. 18. Large branched haustorium. The host cell nucleus, where the haustorium is almost in contact with it, is just beginning to show signs of a fragmentation. Eleven-day culture.

Fig. 19. Branched haustorium affecting host cell nucleus. The chlorophyll corpuscles are seen to have become rounded and swollen, in the act of breaking down. Eleven-day culture.

Figs. 20 and 21 show how narrow the hyphae may become in the intercellular spaces before spore formation. At the tip of the hypha in Fig. 21 a nucleus is just dividing. Both from a ten-day culture.

Fig. 22. Portion of a runner, the protoplasm has become much less dense and is very vacuolated. The nuclei are in a resting condition, and have distinct nuclear membranes.

Figs. 23 and 24. Common forms of haustoria.

Figs. 25-7, 33 and 34. Uredo infection of *P. dispersa* on Rye.

Figs. 28-30. Uredo infection of *P. triticina* on Wheat.

Figs. 31 and 32. Uredo infection of *P. Symphyti-Bromorum* on Bromus.

Fig. 25. Oblique longitudinal section of a stoma of a Rye leaf, showing a portion of a germ tube with its appressorium of *P. dispersa* just over the stomatal slit.

Fig. 26. Longitudinal section of a five-day culture of *P. dispersa* on Rye, showing the remains of the appressorium, the substomatal vesicle with its two infecting hyphae.

Fig. 27. Similar preparation to the above. In both cases the sub-stomatal vesicle has become divided by a transverse septum. Small centrosomes are seen in connexion with some of the nuclei.

Figs. 28 and 29. Longitudinal sections of Wheat leaf showing infection phenomena in *P. triticina*.

#### PLATE XLII.

Fig. 30. Transverse section across a stoma showing appressorium, substomatal vesicle, and infecting hypha with its haustorium of *P. triticina*.

Figs. 31 and 32. Longitudinal sections of stomata, showing infections with characteristically shaped sub-stomatal vesicles of *P. Symphyti-Bromorum*.

Fig. 33. Small portion of a typical hypha of *P. dispersa* with its nuclei. The tip of the hypha has just become transversely septate, preparatory to the formation of a haustorium. From a five-day culture.

Fig. 34. Typical haustorium of *P. dispersa*.

Figs. 35-7. Uredo infection of *P. simplex* on Barley.

Figs. 38 and 39. Uredo infection of *P. coronifera* on Oats.

Figs. 40-3. Uredo infection of *P. Sorghi* on Maize.

Fig. 35. Four-day culture of *P. simplex* on Barley, showing sub-stomatal vesicle with its infecting hyphae. The first transverse wall in the vesicle is just beginning to be formed.

Figs. 36 and 37. Both from the margins of Uredo pustules of *P. simplex* on Barley, taken from an outbreak in the field (and not artificially inoculated).

The sub-stomatal vesicles have become empty and divided by transverse septa. The hyphae are beginning to lose their protoplasm and become septate.

Figs. 38 and 39. Longitudinal sections of Oat leaf showing infection phenomena of *P. coronifera*. A single transverse septum is formed in each sub-stomatal vesicle, which gives rise to an infecting hypha from each end. Both five-day cultures.

Fig. 40. Infection of Maize leaf by *P. Sorghi*. The sub-stomatal vesicle is full of protoplasm, and not yet fully developed.

#### PLATE XLIII.

Figs. 41 and 42. Older infections of Maize leaf by *P. Sorghi*. Four-day cultures showing characteristically shaped vesicles with their infecting hyphae and haustoria.

Fig. 43. Infection of Maize leaf by *P. Sorghi*, showing shape of sub-stomatal vesicle in transverse section.

Fig. 44. Typical haustorium of *P. simplex* on Barley.

Fig. 45. Haustorium of *P. dispersa* on Rye.