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Study of new techniques of variability among teliospores of brown and yellow rust of wheat

Abha Mishra

Abstract
In the present study new technique for cutting sections of telia of brown and yellow rust of wheat was developed. This helped in studying the variability among teliospores of above rust fungi. Cut sections of telio sori of these rusts were also macerated on the slide with a drop of water. Above macerated material was spread on the slide. The bigger parts of macerated material were removed; a thin film of water was made and examined under the microscope. Teliospores were searched, located and allowed to dry. A drop of lacto phenol and cover slip was carefully placed over it. Slides prepared in this way were used for detailed studies of teliospored of both rust fungi. Bicelled and three celled teliospores along with hilum and apical digitations were found in *Puccinia recondite f. sp. Tritici* whereas in case of *P. striiformis* single celled and bicelled teliospores were observed with elongated rostrum at the apex, apical and lateral digitations and a hilum.

Keywords: Variability, teliospores, rusts, fungi

Introduction
The details of variability of teliospores were studied under microscope. Comprehensive morphological study of teliospores as made using camera lucida which helped in making suitable drawings for describing the actual size, shape and other variations. It is reported that yellow rust caused by *Puccinia striiformis* is not of common occurrence in the plains of India. However, it occurs in Gwalior (Madhya Pradesh) every year. Although with introduction of newly high yielding dwarf and tall varieties of wheat it was observed once in 1965-1966 at Jabalpur (M.P.). In brown and yellow rust of wheat the telio sori remain embedded deeply underneath the host epidermis which makes it difficult to cut the cross section of telia for hostopathological studies. In order to overcome this difficulty the authors attempted to cut transverse sections of these two rust species using variety of piths and fresh, dried and preserved specimens for the demonstrations to the graduate and post graduate students. The authors finally developed new techniques for cutting the transverse section, cutting and maceration or grinding and freeing of teliospores and preparation of good mounts of fine cross sections and loose teliospores out of rigidly conglomerate nonerumpent telia of these two rust species. These techniques are being described here as “New Techniques”.

This enabled the transverse section of the smuts, pith material included was dry pith of “Home light match Box” as it was quite soft. Narrow stripes cut off from dry leaf and infected leaf-sheaths of respective brown and yellow rust were used. A narrow slit at the top of the match stick was made by a sharp razor blade and then narrow and elongated stripes of telio sori of individual rust specimens (1 cm long) were inserted in between the two halves of the dry pith. Cross sections were cut by a new razor blade held in right hand after rotating the same just parallel to the top of the pith where specimen got inserted. The sections along with cut pieces of dry pith were water mounted on the slide and observed under compound microscope. Pith material, macro sections and other unwanted fragments were removed and fine good sections were further examined. On drying of thin film of water, lacto-phenol mounts with cover slip were prepared.

To study the morphological details of teliospores, slides of loose and scattered teliospores were prepared by cutting the sections, sorting out macro sections and other unwanted fragments on water mount on the slide was adopted. Remaining sections were macerated briskly by the help of ground edge of another glass slide and these razor blade and again other fine host parts were finally removed. As soon as thin film of water dried lacto-phenol mounts with cover slip were prepared.

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Observations and Findings
Various slides were prepared out of cross sections and loose teliospores of both the rusts. Prepared slides were observed for detailed morphological variations among the teliospores of *P. recondita f. sp. tritici* and *P. striiformis* by the help of a compound microscope under high power magnification. Observations recorded for teliospores of both the rusts included single celled teliospores and bicelled teliospores with and without rostrum at the apex, with and without apex with and without apex digitations, lateral digitations and with and without hilum region. Measurements of such morphological variability’s including teliospores were taken in microns. Appropriate camera Lucida drawings were also made wherever required. Finally the percentages of occurrence of aforesaid characteristics of teliospores were recorded separately for both rust organisms. Characteristics of teliospores along with morphological characters of different types of teliospores of these rust fungi are depicted in Table-1.

**Table 1:** Per cent occurrence of different teliospores and other morphological variants of *P. recondita f. sp. tritici* and *P. striiformis.

<table>
<thead>
<tr>
<th>S. No</th>
<th>Different king of teliospores</th>
<th>Percentage of occurrence of teliospores with variable morphological characters</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td><em>P. recondita f. sp. tritici</em></td>
</tr>
<tr>
<td>1.</td>
<td>Single celled teliospores</td>
<td>0.0</td>
</tr>
<tr>
<td>2.</td>
<td>Bicelled teliospores</td>
<td>98.5</td>
</tr>
<tr>
<td>3.</td>
<td>Three celled teliospores</td>
<td>1.5</td>
</tr>
<tr>
<td>4.</td>
<td>Teliospores with apparently thickened hilum region</td>
<td>85.0</td>
</tr>
<tr>
<td>5.</td>
<td>Teliospores without apparently thickened hilum region</td>
<td>15.0</td>
</tr>
<tr>
<td>6.</td>
<td>Teliospores with rostrum at apex</td>
<td>0.0</td>
</tr>
<tr>
<td>7.</td>
<td>Teliospores without rostrum at apex</td>
<td>100.00</td>
</tr>
<tr>
<td>8.</td>
<td>Teliospores with apical digitations</td>
<td>8.0</td>
</tr>
<tr>
<td>9.</td>
<td>Teliospores without apical digitations</td>
<td>92.0</td>
</tr>
<tr>
<td>10.</td>
<td>Teliospores with lateral digitations or folds</td>
<td>0.0</td>
</tr>
<tr>
<td>11.</td>
<td>Teliospores without lateral digitations or folds</td>
<td>0.0</td>
</tr>
</tbody>
</table>

Different symptoms of teliosori and morphological details of teliospores of the rusts under study are described as below:

**A. Puccinia Recondita f. sp. tritici Rob. Ex Desm**

telia are oval to linear, dull black, non-erumpent and chiefly formed on adaxial (lower) leaf surface as scattered and in cluster form. On leaf sheath telia are more linear and confluent also formed scantily on abaxial (upper) leaf surface. Teliosori are divided in to number of compartments by the partition wall of brown single celled incurved paraphyses. These compartments are filled with closely packed mass of dark brown teliospores which are variable in shape and size. Chiefly oblong clavate sometimes oblong to cylindrical or ellipsoid to broadly ellipsoid or even obvoid, very few spores tending to be diorchidoide. Bicelled teliospores (98.5%) and three celled one (1.5%) are brown to chestnut brown smooth walled slightly constricted at the septum (38-) 42-62.4 (-79) x (12) 15.4-19.2 (-23) µm, apex brown to cinnamon brown (1.9-) 2.9-3.8 (-5.8 µm thick. Excluding digitations. Sometimes bicelled teliospores are sparingly cororate (8%) with 0-2 digitations, 0.8-1.9 µm thick. Stalk (Pedicle) is short dull brown collapsing (3.88-) 7.6-19.2 (-21.2) µm long with an apical thickening adjacent to the hilum or the hilum region below the lower cell of teliospores is more apparently visible deep brown in colour and thickened to the extent of 3.8-11.5 µm. Eighty five percent teliospores possessed deep brown hilum and 15% were without hilum.

**B. Puccinia striiformis Westend**

telia are small oval shaped dull black, non-erumpent, arranged in linear rows or stripes formed chiefly on adaxial leaf surface, leaf sheath, stalks, spikelets, glumes and awns. Teliosori are divided in to compartments by single celled brown colourued incurved paraphyses. These compartments are filled with limited number of teliospores which are deep golden brown in mass teliospores are single to bicelled constricted at the septum and of variable shape mainly oblong to oblong clavate or ellipsoid to fusiform ellipsopoid) golden brown, smooth walled (44.5-) 62.76.8 (-86.8) x (13.5-) 19.2-23 (-26.8) µm, (Av. 66.5 x 20.2 µm), apex thick flat to pointed or obvoid deep golden brown to yellowish brown (2.3-) 5.7-8 (-9.6) µm (Av. 3.4 µm) thick, sometimes the apex of five percent teliospores was found with an elongated rostrum faintly yellowish brown below and dull yellowish or hyaline at the upper tapering end 7.5-25 µm long (Av. 13.4 µm). Other 95% teliospores did not possess rostrum. Rarely limited number of spores (3.5%) were found to have 0-3 digitations at the apex which were shorter in length or just like tuberciles, 1.9-4.8 µm long (Av. 3.4 µm) but 96.5 percent teliospores were without digitations at apex. The percentages of teliospores with lateral digitations were 7.5 percent and spores devoid of digitations were 92.5 per cent. Such digitations are 0-2 in number dull yellowish to hyaline (1.9-5.8 µm) thick. Stalk is short hyaline collapsing (5.8-) 7.8-11.5 (-25) µm (Av. 8.6 µm), long with an apical thickening of hilum region below the lower cell of teliospore golden brown or dull yellowish brown. Per cent occurrence of such typical spores with apparently conspicuous thickened hilum region was 40% whereas other 60 percent spores were without hilum. Thickening of hilum region measured 3.8-5.8 µm (Av. 4.6 µm). In the present investigation four per cent single celled teliospores were observed along with 96% bicelled teliospores they measured 52x13.4µm, with 5.6 µm thickened at the apex and 1.5-2 µm thick at sides, dull yellowish brown to yellowish in colour oblong ellipsoid to broadly ellipsoid in shape, stalk is short yellowish or hyaline and collapsing 5.8-9.6 µm long.

**Discussion**

Earlier Mishra and Khare (1985) [5] studied the variability within the teliospores of *Puccinia graminis f. sp. tritici* but under the present investigation it was found that in case of *P. recondita f. sp. tritici* too, the occurrence of three celled teliospores has not been mentioned and described by Butler (1918) [1] and Mehrotra (1980) except one unlabelled figure of three celled teliospores was shown in the diagram in the
above reference. Likewise, other scientists also neither mentioned the occurrence of three celled teliospores nor depicted figure in the text (Dickson, 1956; Mundkur, 1959; Rangaswami, 1975) [3, 6, 7]. Singh (1990) [8] mentioned about the occurrence of three celled teliospores in the text description of *P. recondita* f. sp. *Tritici.* Cummins (1971) [2] has treated *P. recondita* as “species complex” due to the occurrence of variable teliospores but no diagram of three celled teliospores was given in his monograph. Cummin’s monographic description of *P. Recondita* did not mention the oblong to cylindrical, ellipsoid to broadly ellipsoid and diorchidioid shape of teliospores and thickened browning of hilum region. However, Cummins (1971) [2] has described such type of brown thickening adjacent to the hilum in case of *P. crinitae* on *Dichelachne crinita.* Thus, above mentioned morphological characters of *P. recondite* f. sp. *tritici* are being described here for the first time.

Conspicuously thickened and coloured hilum region, elongated rostrum at the apex, lateral digitations and single celled condition of teliospores of *P. striiformis* have not been described in any of the text books on plant pathology (Dickson, 1956; Mundkur, 1959; Rangaswami, 1975; Mehrotra, 1980; Singh, 1990) [3, 6, 7, 9]. However, occurrence of single celled teliospores has been reported by Cummins (1971) [2]. The present studies have highlighted the methods of cutting fine sections and freeing loose teliospores by maceration which enabled to observe clearly entire contents of compartments of teliosori and teliospores of brown and yellow rusts of wheat. The new method of cutting sections will be helpful to study in detail the morphology of various contents of histopathology.

**Reference**