A NOVEL TAXONOMIC STUDY OF THE HELMINTHOSPORIUM LINK EX FRIES STATUS AND CLOSED RELATED GENERA

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ABSTRACT: This study isolated and examined many isolates of Drechslera spp., Bipolaris spp. and Exserohilum from rice cultivars and treated the most changes that occurred in the taxonomy and nomenclature of the genus Helminthosporium according to the ICBN rules. This genus was considered as complex of species and deeply entrenched in the taxonomic literature. 100-668 species were recorded in the phytopathological literature and database, but in this study only 49 species were fixed as correct names in Helminthosporium, and split it into four genera, also the common species of the genus Helminthosporium were distributed in the four genera: Drechslera (28 spp.), Bipolaris (10 spp.), Exserohilum (7 spp.) and Marielliottia (3 spp.). Most of the species of Helminthosporium become synonym for species of the genera Drechslera, Bipolaris, Exserohilum and Marielliottia and the names of Helminthosporium still as basyname.

Key words: Classification, Drechslera, Bipolaris, Exserohilum, Marielliottia taxonomical treatment, Helminthosporium status.

INTRODUCTION

The fungus Helminthosporium a customs to the dematiaceous of Deuteromycetae, it was treated as a form of a genus (form genus) according to the rules of scientific naming (ICBN). This fungus behaviour as facultative saprophytic fungus and causing brown spot disease on cereals (Bakonyi et al., 1997) and has widely hosteda range (Rodriguez-Decuadro et al., 2014). Helminthosporium was established for the first time by the Link ex Fries in 1809 and the type species of this genus was H. velutinum Linke ex Ficinus & Schubert. There are inconsistencies in the number of its species, Hesseltine et al. (1971) were stated that genus includes 175 species, while the number of species that bears the name genus Helminthosporium were 686 species based on fungus MycoBank database (www.mycobank).

Since the fifties of the last century and tell time, many taxonomic changes were conductedon the nomenclature for those species. Some species been belonging to other genera far down the phenotypes such as Cladosporium and Curvularia and other genera (Shoemaker, 1959).

The Helminthosporium was considered heterogeneous fungus, it includes a species differ in good morph characters, so the Helminthosporium was subdivided into two subgenera: subgenus: Cylindro-Helminthosporium and subgenus: Eu-Helminthosporium (Raemaekers, 1988), depending on the most important taxonomic characteristics like spores germination and shape of spores and the shape of the hilum, septate characters, the species of Helminthosporium were renamed again.

The species belonging to the subgenus Eu-Helminthosporium were placed into the genus Bipolaris and the species within the subgenus Cylindro-Helminthosporium were placed into the genus Drechslera and based on the germination mode, where the germination occurred at the poles only considera diagnostic character to the Bipolaris, while if the germination occurred in most the phragmospore considered as a diagnostic character to the genus Drechslera (Drechsler, 1923; Shoemaker, 1959; Raemaekers, 1988).

Based on the helium characters like prominent or non-prominent been designated the new genus Exserohilum Leonard & Suggs. This genus included 8 species were previously registered in the genus Helminthosporium (Leonard and Suggs, 1974). The septa number was considered as a diagnostic character for new genus Marielliottia Shoemaker (1999), which includes three species was part of Drechslera; they basically were among Helminthosporium (Aslani et al., 2006). Recently using analysis based on ITS sequences to determine evolutionary relationships and taxonomic status of Bipolaris oryzae and other Cochliobolus species (Dela Paz et al., 2013).
names became Illegitimate names or Ambiguous names according to the rules of the ICBN. On the other hand, the aim of the present study is to clarify the taxonomic status of the **Helminthosporium** spp and remove the confusion surrounding the status of this genus, and re-taxonomic treatment of the species belonging to it. Also to compare between the taxonomic characteristics of the species under study, which addresses this research associated problems in terms of the diagnostic characters and taxonomic status.

**MATERIALS AND METHODS**

**Sample collection**

A total of 50 leaf samples of rice (*Oryza sativa*) bearing symptoms of leaf spotting, at various stages was collected and kept in sealed plastic bags. The samples were examined for the possible presence of pathogen structures. *Bipolaris, Drechslera* and *Exserohilum* as a dematiaceous fungi were regularly associated with the necrotic tissues was inoculated into plates containing potato dextrose agar (PDA) as described by Imran and Al.Rubaiy (2015). Rice leaf samples were collected from October to December 2015-2016, in the middle regions of Iraq. Daly temperatures ranged were approximately around 5°C in January, 56°C in August and 37°C in October.

**Culturing and isolating of Bipolaris, Drechslera and Exserohilum**

The pure cultures of *Bipolaris, Drechslera* and *Exserohilum* were isolated by the serial dilution technique using Potato Dextrose Agar (PDA) medium. The isolates of *Bipolaris, Drechslera* and *Exserohilum* were sub cultured on the PDA medium in separate triplicate plates for each one of the fungi and incubated at 25°C for 7 days (Imran and Al-Shukry, 2014; Imran and Al Asadi, 2014). Microscopic examination was performed using mounted tiny portion from colony margins each fungus. This was done by using adhesive transparent tape placed on a slide with a drop of lacto-phenol cotton blue stain. *Bipolaris, Drechslera* and *Exserohilum* isolates were identified phenotypically using the taxonomic key created by Alcorn (1988). They were maintained on PDA slants at 25°C for four days and were kept in refrigerator at 4°C until use; they were sub cultured every two weeks. The frequency of a fungus is denoted by the number of samplings in which it is recorded against the total: Frequency (%) = No. of observation in which colony appears / total number of observations recorded × 100 (Imran and Al-Rubaiy, 2015).

**Taxonomic treatment method**

In this study, it has been the most species recorded

<table>
<thead>
<tr>
<th>Table 1: List of Helminthosporium spp with their authorities.</th>
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</table>
statistic in genus *Helminthosporium* with all of the articles, year of publication and published since 1809 until now, have been identified and diagnostic characteristics and document species names and authority of species in each genus and scheduling those species in the tables, each table included synonyms and telomorph for each species if present. Illustrated the most important diagnostic characteristics of the species under interest by modern images for slides from fresh cultures performed in this study. The main diagnostic characters included conidiophores shapes, conidiogenisis and conidial appearance of fungal species.

Approved the diagnosis each of the genera: *Bipolaris*, *Drechlera*, *Exserohilum* and *Marieliottia* also these diagnostic characters for separation of *Bipolaris*, *Drechlera*, *Exserohilum* and *Marieliottia* and demonstrated the taxonomic value of conidium apical characteristics: round or truncate, number of septa, constituent double barriers of conidium, triple or multiple septa, present of false septa, characters of Central cells, presence or absence and numbers, Helium being prominent privileged or slightly raised or low-profile perfectly, scars characters in terms of being a small, narrow or wide dark-colored, lobes or bulges that appear on the body of conidia, germination mode of conidia and the location of germination from one pole of the or the two poles, as well as some cultural characters and review their pathogenicity for plant species and humans.

**RESULTS AND DISCUSSION**

**The species belonging to the *Helminthosporium***

There is high similarity in the conidial characters between *Helminthosporium*, *Drechlera*, *Bipolaris* and *Exserohilum*. *Helminthosporium* included 48 species.

### Table 1 continued...

41 *Helminthosporium spiciferum* (Bainier) Nicot.
42 *Helminthosporium stenacrum* Drechsler (1923)
43 *Helminthosporium siccans* Drechsler (1923)
44 *Helminthosporium sorghicola* Lefebvre & Sherwin (1949)
45 *Helminthosporium setariae* Lind (1919)
46 *Helminthosporium tritici-repentis* Diedicke (1903)
47 *Helminthosporium turcicum* Pass. (1876)
48 *Helminthosporium vagans* Drechsler (1944)

**RESULTS AND DISCUSSION**

**The diagnostic characteristics of the *Bipolaris***

Fig. 1: Conidal characters: Hilum shapes; septet number in: A: *Drechlera*, B: *Exserohilum*, C: *Bipolaris* and D: *Marieliottia*.

The genus *Bipolaris* was diagnosed by Shoemaker in 1959 as a genus differently because it includes many telomorphic species belong to *Trichometasphaeria* other species located in the telomorph: *Cochliobolus* has been removed this disparity valuable introduction of a new genus: *Exserohilum*. This genus which is characterized by special hilum which prominently seamlessly termed exerted helium (Safarimotlagh and Kaviani, 2008).

The *Bipolaris* differ from *Drechlera* by the germination mode: the first one germination at polar ends while the second germinated randomly (Alcorn, 1983), while the number of septa about 2-3 considered as taxonomic character of *Marieliottia* (Aslani et al, 2006) (Fig. 1).

It was found that the species involved under the *Helminthosporium* have conidia characterized by important changes from the taxonomic feature, those differences in microscopic characteristics led to the split
and transfer Helminthosporium species to other four genera: Bipolaris, Drechslera, Exserohilum and Marielliottia. The former three ones have telomorphic phases Cochliobolus (Sivanesan, 1987; Fetch and Steffenson, 1994), Pyrenophora and Setosphaeria respectively (El-Nashaar and Stack, 1989) as shown in the Tables 2-5. All species of Helminthosporium transformed to the four genera and been as synonyms (Hesseltine et al, 1971) as shown in Tables 2-5.

### Bipolaris Shoemaker

The main taxonomic characters of Bipolaris spores are being with parallel walls usually with erected conidiophores and upright character feature that spores germinate at the poles (Duveillerand Garcia Altamirano, 2000; Kumar et al, 2001) some characters of spores shown in Fig. 2.

### Drechslera Ito

The Drechslera colonies dense, hairy and sometimes velvety, grey or brown or dark brown to black, mycelia immersed. Most of this genus are forming stroma and sclerotia in some cultures. Conidiophores are specialized, mononemative or in clusters, straight or zigzag shape, non-branched and are rarely branched in some species, brown smooth in most species. Single spores were organized in some species, simple, straight or curved, clavate or cylindrical in shape, curved at the ends or be elliptical or spindle clavate inverted shape. Brown to dark brown to a pale olive to brown. The cells are irregular in color. Terminal cells are paler than those that exist in the centre of spore, mostly smooth and rarely identical, with false septa (Fig. 3). Our finding agrees to report of Zeiders (1980), Dehne and Oerke (1985).

The taxonomic treatment of 28 species belonged to Drechslera were shown in Table 3.

### Exserohilum Leonard & Suggs

This genus includes seven species, these species characterized by good taxonomic character its prominent hilum of conidia as in Fig. 4. Five species of this genus contain the telomorphs: Setosphaeria: S. rostratum, S. turcica, S. halodes, S. pedicellatum and S. holmii as the recording is a new species: Exserohilum mcginnisii Padhye and Ajello, sp. Nov. 1986 (Chang and Fan, 1986) (Table 4).

Seven species of Exerohilium belonged to Drechslera were shown in Table 4.

### Marielliottia Shoemaker

This a new genus of dematiaceous fungi, it includes

### Table 2: Bipolaris spp list and common synonyms and their telomorphes if present.

<table>
<thead>
<tr>
<th>No.</th>
<th>Anamorphe</th>
<th>Telomorphe</th>
<th>Synonym</th>
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<tbody>
<tr>
<td>1</td>
<td>Bipolaris cynodontis (Marignoni) Shoemaker 1959</td>
<td>Cochliobolus cynodontis Nelson 1964</td>
<td>Drechslera cynodontis (Marignoni) Subram. &amp; Jain 1966</td>
</tr>
<tr>
<td>2</td>
<td>Bipolaris hawaiiensis (Ellis)Uchida &amp; Aragaki 1979</td>
<td>Cochliobolus hawaiiensis Alcorn 1978</td>
<td>Drechslera hawaiiensis (Bugnic) Subram. &amp; Jain 1966; Drechslera hawaiiensis Ellis 1971</td>
</tr>
<tr>
<td>3</td>
<td>Bipolaris maydis (Nisik. &amp; Miyake) Shoemaker 1959</td>
<td>Cochliobolus heterostrophus (Dreschler) Dreschler</td>
<td></td>
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<tr>
<td>5</td>
<td>Bipolaris sorokiniana (Sacc.) Shoemaker 1959</td>
<td>Cochliobolus sativum (Ito &amp; Kurib.) Drechsler ex Dastur 1942</td>
<td>Helminthosporium sativum Pammel, King &amp;Bakke 1910</td>
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<td>7</td>
<td>Bipolaris cactivora (Petr.) Alcorn 1983</td>
<td>Unknown</td>
<td>Drechslera cactivora (Petr.) M.B. Ellis (1971) Helminthosporium cactivorum Petr. 1931</td>
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<tr>
<td>8</td>
<td>Bipolaris australiensis (M.B.Ellis) Tsuda et Ueyama 1981</td>
<td>Unknown</td>
<td>Drechslera australiensis (Bugnicourt) Subram. &amp; Jain ex M.B.Ellis; 1971 Subram. &amp; Jain Helminthosporium australiensis Bugnicourt</td>
</tr>
<tr>
<td>9</td>
<td>Diporalis sacchari (E.J. Butler) Shoemaker, (1959)</td>
<td>Unknown</td>
<td>Helminthosporium sacchari(E.J. Butler,) 1913</td>
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<td>10</td>
<td>Bipolaris sorghicola (Lefebvre &amp; Sherwin) Alcorn 1983</td>
<td>Unknown</td>
<td>Drechslera sorghicola (Lefebvre &amp; Sherwin) M. J. Richardson &amp; E. M. Fraser 1968 Helminthosporium sorghicola Lefebvre &amp; Sherwin 1949</td>
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<td>11</td>
<td>Bipolaris spicifer (Bainire) Subram 1971</td>
<td>Unknown</td>
<td>Brachycladium spiciferum Bainier</td>
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</table>
Novel taxonomic study of the *H. sporium*

**Fig. 2:** Conidial arrangement on the conidiophores of *Bipolaris.*

**Fig. 3:** Conidial arrangement on the conidiophores of *Drechslera.*

**Fig. 4:** Conidiogenesis and conidia arrangement on the conidiophores of *Exerohillium.*
<table>
<thead>
<tr>
<th>No.</th>
<th>Anamorphe</th>
<th>Telomorph</th>
<th>Synonym</th>
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<tr>
<td>1</td>
<td>Drechslera avenae (Eidam) Scharif 1963</td>
<td>Pyrenophora avenae S. Ito &amp; Kurib. 1930</td>
<td>Helminthosporium avenae Eidam 1891</td>
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<td>4</td>
<td>Drechslera bromi (Died.) Shoemaker 1962</td>
<td>Pyrenophora bromi (Died.) Drecshler 1923</td>
<td>Helminthosporium bromi (Died.) Died 1903</td>
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<td>5</td>
<td>Drechslera campanulata (Lév.) B. Sutton, 1976</td>
<td>Pyrenophora semeniperda (Brittleb. &amp; D.B. Adam) Shoemaker 1966</td>
<td>Bipolaris cyclops (Drechsler) R. Sprague 1962; Chaetostigme horridula (Syd.) Clem. &amp; Shear; Helminthosporium cyclops Drechsler 1923; Pleosphaeria semeniperda Brittleb. &amp; D.B. Adam 1924; Pyrenophora horrida Syd. 1924</td>
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<td>6</td>
<td>Drechslera joicis (Nisikado) Subram. &amp; Jain 1966</td>
<td>Unknown</td>
<td>Helminthosporium joicis Nisikado Curvularia joicis Castellani</td>
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<td>7</td>
<td>Drechslera dematioidea (Bubák &amp; Wróblewski) Subram. &amp; Jain 1966</td>
<td>Unknown</td>
<td>Drechslera dematioidea (Bubák &amp; Wróbl.) Scharif¡ (1963); Helminthosporium dematioideum Bubák &amp; Wróbl.¡ (1916); Marielliottia dematioidea (Bubák &amp; Wróbl.) Shoemaker ¡ (1999)</td>
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<td>8</td>
<td>Drechslera. dictyoides (Drechsler) Shoemaker 1978</td>
<td>Pyrenophora dictyoides Paul &amp; Parberry</td>
<td>Helminthosporium dictyoides Drechsler</td>
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<td>9</td>
<td>Drechslera erythrostipa (Drechsler) Shoemaker 1959</td>
<td>Unknown</td>
<td>Helminthosporium erythrostipa Drechsler 1935</td>
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<tr>
<td>10</td>
<td>Drechsleraeuphorbiae (Hansford) M.B.Ellis comb. nov.1971</td>
<td>Unknown</td>
<td>Helminthosporium euphorbiae Hansford 1943</td>
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<td>11</td>
<td>Drechslerafugax (Wallr.) Shoemaker 1958</td>
<td>Unknown</td>
<td>Helminthosporium fugax Wallr.H.stenacr um Drechsler</td>
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<td>12</td>
<td>Drechslera graminea (Rabenh. ex Schlidl.) S. Ito 1930</td>
<td>Pyrenophora graminea S. Ito &amp; Kurib. 1930</td>
<td>Helminthosporium graminea Rabenh. ex Schlecht1888</td>
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<td>Drechslera iridis (Oud.) M.B.Ellis 1971</td>
<td>Unknown</td>
<td>Clasterosporium iridis Oudem. 1898; Bipolaris iridis (Oudem.) C.H. Dickinson 1966</td>
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<td>14</td>
<td>Drechslera miyakei (Y. Nisik.) Subram. &amp; B.L. Jain 1966</td>
<td>Unknown</td>
<td>Bipolaris miyakei (Y. Nisik.) Shoemaker 1959; Helminthosporium miyakei Y. Nisik. 1929</td>
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<td>15</td>
<td>Drechslera nodulosa (Berk. &amp; M.A. Curtis ex Sacc.) Subram. &amp; B.L. Jain 1966</td>
<td>Cochliobolus nodulosus Luttrell 1957</td>
<td>Helminthosporium nodulosum Berk. &amp; M.A. Curtis ex Sacc. 1886</td>
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<td>16</td>
<td>Drechslera oryzae (Breda de Haan) Subram. &amp; Jain 1966</td>
<td>Cochliobolus miyabeanus (Ito &amp; Kuribayashi) Drechsler ex Dastur</td>
<td>Helminthosporium oryzae Breda de Haan1900</td>
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<td>17</td>
<td>Drechslera papendorfii (Van der Aa) M.B. Ellis</td>
<td>Unknown</td>
<td>Curvularie papendorfii Van der Aa</td>
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<td>18</td>
<td>Drechslera phlei (Graham) Shoemaker 1959</td>
<td>Unknown</td>
<td>Helminthosporium dictyoides Drechslervar.phlei</td>
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<td>19</td>
<td>Drechslera poae (Baudys) Shoemaker 1962</td>
<td>Unknown</td>
<td>Helminthosporium poae BaudysH.vagans Drechsler</td>
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<td>21</td>
<td>Drechslerasacchari (Butler) Subram. &amp; Jain 1966</td>
<td>Unknown</td>
<td>Helminthosporium sacchari Butler</td>
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<td>22</td>
<td>Drechslera sativus</td>
<td>Cochliobolus sativus (Ito &amp; King &amp; Bakke</td>
<td>Helminthosporium sativus Pammel</td>
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<th>Telomorphe</th>
<th>Synonym</th>
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<td>23</td>
<td><em>Drechslera setariae</em> (Sawada) Subram. &amp; B.L. Jain 1966</td>
<td><em>Cochliobolus setariae</em> (S. Ito &amp; Kurib.) Drechsler ex Dastur 1942</td>
<td><em>Bipolaris setariae</em> (Sawada) Shoemaker 1959; <em>Helminthosporium setariae</em> Lind 1919; <em>Helminthosporium setariae</em> Sawada 1919; <em>Ophiobolus setariae</em> S. Ito &amp; Kurib. 1930</td>
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<td>24</td>
<td><em>Drechslera siccans</em> (Drechsler) Shoemaker</td>
<td>Unknown</td>
<td><em>Helminthosporium siccans</em> Drechsler</td>
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<td>25</td>
<td><em>Drechslera teres</em> (Sacc.) Shoemaker 1959</td>
<td>1923 <em>Pyrenophora teres</em> Drechsler</td>
<td><em>Helminthosporium hordie</em> Eidam <em>Helminthosporium teres</em> Pleospora teres Diedick: 1903</td>
</tr>
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<td>26</td>
<td><em>Drechslera tritici-repentis</em> (Died.) Shoemaker 1962</td>
<td><em>Pyrenophora tritici-repentis</em> (Died.) Drechsler 1923</td>
<td><em>Helminthosporium tritici-repentis</em> Diedicke: 1903</td>
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<td>27</td>
<td><em>Drechslera victoriae</em> (Meehan &amp; Murphy) Subram. &amp; Jain 1966</td>
<td><em>Cochliobolus victoriae</em> Nelson</td>
<td><em>Biplores victoriae</em> (Meehan &amp; Murphy) Subram 1959; <em>Helminthosporium victoriae</em> Meehan &amp; Murphy 1946</td>
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<td>28</td>
<td><em>Drechslera heterostrophus</em></td>
<td><em>Cochliobolus heterostrophus</em> (Drechsler) Drechsler</td>
<td><em>Helminthosporium maydis</em> NisikadoD. (Nisikado) Subram. &amp; Jain</td>
</tr>
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Table 4: *Exerohillum* spp. list and common synonyms and their telomorphes, if present.

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<th>Telomorphe</th>
<th>Synonym</th>
</tr>
</thead>
<tbody>
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<td><em>Exerohillum frumentacei</em> (Mitra) M. B. Ellis, 1971</td>
<td>Unknown</td>
<td><em>Helminthosporium frumentaceum</em> Mitra</td>
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<tr>
<td>7</td>
<td><em>Exerohillum mcginnisii</em> Padhye and Ajello, sp. Nov 1986</td>
<td><em>Setosphaeria monoceras</em> Alcorn 1978</td>
<td></td>
</tr>
</tbody>
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Novel taxonomic study of the *H. sporium* 933
3 species, these species have been described within the *Drechslera*: *D. biseptata*, *D. dematioidea* and *D. triseptata* (Table 5). It was originally registered under the *Helminthosporium* spp (Table 1), the three species differ from the species of *Drechslera* based on conidia that contain mostly three septa and be oval shapes upside down to the oval (Fig. 1). The germination of *Marielliottia* from the basal cell or sometimes from the apical, no central cells in the conidial form. This fungus parasitic on weeds (Aslani et al, 2006).

Due to the truth that *Helminthosporium* is a source for many of species, there are seeking to reduce the number of their species. Alcorn (1988) mentioned to the need to reduce and re-identify the species of this genus with 20 species. The number of this genus listed in Table 1 is actually not really species at the present time, it has become synonyms in other species, but the one which nor been synonyms are not listed in Tables 2–5 are in a neglected species that counter because they are not supported at the present time and what enhances this opinion that sources have indicated to the neglect of all neglected species that counter because they are not non been synonyms are not listed in Tables 2–5 are in a neglected species that counter because they are not supported at the present time and what enhances this opinion that sources have indicated to the neglect of all neglected species that counter because they are not non been synonyms are not listed in Tables 2–5 are in a neglected species that counter because they are not supported at the present time and what enhances this opinion that sources have indicated to the neglect of all neglected species that counter because they are not.

Our conclusion in the current study, the four genera were shown high variation and the diagnostic characters based on conidial evidence, not sufficient to classify the four genera, because there are high ranges of variation of spore characters: shapes, heliomer and locations of germination of spores, so we observed a significant change in these species sites (Tables 2–5). Any future study of these genera should be investigated taxonomic criteria based on genetic criteria.

**REFERENCES**


