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New knowledge on the sorghum midge, *Stenodiplosis sorghicola* Coquillet 1899 (Diptera: Cecidomyiidae), in the south of France

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Abstract
Complete floret abortion on sorghum panicles due to sorghum midge (*Stenodiplosis sorghicola*) infestation was observed just after flowering onset in early August (2008) in fields near Montpellier (France). With the aim of controlling these infestations, mathematical models simulating the biological development of the insect as a function of temperature, which were developed in Texas (USA) and adapted to the climatic conditions in southern France, were used within the framework of an experimental research study on sorghum. The hypothesis put forward clearly accounts for the local dynamics of this insect. Entomophagous parasitoids (*Aprostocetus* spp.) of sorghum midges were also found to be present.

Keywords: sorghum, sorghum midge, *Aprostocetus*, mathematical model, sorghum insects

Résumé
Dans le sud de la France à Montpellier, en 2008, on a observé des avortements complets de panicules de sorgho à partir des dates de floraison du début août, dus à la cécidomyie du sorgho (*Stenodiplosis sorghicola*). Afin de maîtriser ces attaques, dans un contexte de recherche expérimentale sur le sorgho, des modèles mathématiques simulant le développement de la mouche en fonction de la température mis au point au Texas (USA), ont été adaptés aux conditions climatiques du sud de la France. Les modèles appliqués rendent bien compte de la dynamique locale de la cécidomyie. Les autres insectes présents sur les panicules de sorgho ont également été inventoriés, notamment les parasitoïdes de la cécidomyie, dont certains n’ont pas encore été cités en France, comme les entomophages du genre *Aprostocetus* spp.

Mots-clés : sorgho, cécidomyie, *Aprostocetus*, modèle mathématique, insectes du sorgho
Introduction

The sorghum midge, *Stenodiplosis sorghicola* (Dipt., Cecidomyiidae), or cecidomyia, is associated with the genus *Sorghum*. A native of Africa, it causes considerable damage to sorghum crops on all five continents (Young & Teetes 1977). For Europe, Coutin (1969) indicates that it was reported for the first time in Italy in 1963; Coutin himself observed it for the first time in 1969 in the south of France, in the region close to Nimes; however he provided no details concerning the practical conditions under which the observation was made, reporting only limited damage to a late-flowering variety. In 2008, trials conducted in Montpellier in the south of France, including a large number of sorghum cultivars (*Sorghum bicolor* ssp bicolor [L.] Moench) were infested by sorghum midges. The infestation spread rapidly from the beginning of August, in some cases leading to complete abortion of the spikelets of the panicles (Photo 1). The midge is not considered as a pest in France, and this is the first time that very high damage has been reported in the region.

To understand and control the rapid infestations of the midge, we adapted mathematical models developed in Texas (USA) that simulate the dynamics of the fly as a function of temperature (Baxendale et al., 1984a, 1984b) to climatic conditions in the south of France. The presence of other insects on the sorghum panicles was also recorded, especially parasitoids of the sorghum midge, some of which have not previously been reported in France.

I. Bio-ecology of the sorghum midge

The adult sorghum midge is a small fly measuring around 1.5-2.0 mm (around a quarter of the size of a mosquito). Its abdomen is red-orange (Photo 2). Its flight capacity is limited. The female, whose life span is 2-3 days, lays between 30 and 100 eggs on the ovaries and lower glumes of the flowers (Sharma et al. 1990; Magallanes-Cedeno & Teetes 1991; Chantereau & Nicou 1991) just before and during flowering (Photo 3) when the anthers are exposed. The larva feeds on the ovary and on the developing seed (Photo 4) preventing further growth. When the nymph emerges, the adult leaves a characteristic exuvium at the apex of the flowers (Photo 5). In Texas conditions, two generations can be observed on local wild sorghum (Johnson grass) and five generations on cultivated sorghum (Pendleton et al. 1994; Baxendale et al. 1984b).

To survive the cold season, the midges hibernate as cocooned larvae in aborted flowers of the panicles. Empty panicles are usually left in the field and buried in the soil during subsequent tillage operations. Diapausing insects are highly resistant. In Texas, 74.4% of sorghum midges that initiate their diapause during one crop season emerge the following spring, 23% emerge in the second year, and 2.6% in the third year (Baxendale et al. 1983). The damage threshold is one to two adult midges per panicle in susceptible genotypes, and five in more resistant genotypes (Castro et al. 2000).
New knowledge on the sorghum midge, *Stenodiplosis sorghicola* Coquillet 1899 (Diptera: Cecidomyiidae), in the south of France – B. Vercambre et al., 2010

Photo 1. Damage: aborted panicle after sorghum midge infestation (G. Trouche, CIRAD)

Photo 2. Adult sorghum midge (R. Coutin, INRA)

Photo 3. Sorghum midge eggs (R. Coutin, INRA)

Photo 4. Larvae in a sorghum flower (R. Coutin, INRA)

Photo 5. Exuvium at the top of the spikelet (R. Coutin, INRA)
II. Agronomic conditions

CIRAD (French Agricultural Research Centre for International Development) and GEVES (French group for the study and monitoring of varieties and seeds) conduct annual trials on sorghum at Domaine de Lavalette (experimental station) in Montpellier, south of France [43°35'N 3°52'E]. Research conducted by CIRAD focuses on genetic diversity and breeding of sorghum in tropical and subtropical regions including enhancement of biodiversity, and production of grain for food, silage, and as a source of energy. GEVES’ mandate is to approve new sorghum varieties at the French national level by conducting trials for “agronomic and technological value” (VAT) and trials for “distinctiveness, homogeneity and stability” (DHS). Trial plots for sorghum cover 10.5% of the total surface area of the Lavalette research station, (4.16 ha out of a total of 39.4 ha). The soil is silty and argillo calcareous, deep and pH 8; it is fully irrigable.

In 2008, 13 trials involving almost 500 sorghum cultivars showed high genetic and phenotypic diversity, especially for the sowing-heading/flowering period due to new interest in sorghum biomass. The three trials, established in the Pont de Fer Ouest and Est plots (table 1) in which sorghum midge damage was measured included 192 varieties planted in three different trials (fig. 1): hybrid trial for silage (P1), biomass trial for agronomic and technological value (VAT) and DHS trials. The trials were surrounded by wide borders planted with an early flowering commercial hybrid (Arlys).

Table 1. Plot characteristics of the three sorghum trials (Domaine de Lavalette, Montpellier, France, 2008).

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>P1 hybrid trial</th>
<th>VAT sorghum trial</th>
<th>DHS trial (GEVES)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>4/5 seeds/hill</td>
<td>pneumatic</td>
<td>pneumatic</td>
</tr>
<tr>
<td>Thinning</td>
<td>05/06/2008</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Plot dimensions</td>
<td>3 x 3 m²</td>
<td>5 x 2.56 m²</td>
<td>variable</td>
</tr>
<tr>
<td>Number of varieties</td>
<td>11 varieties (2 replicates)</td>
<td>14 hybrids (3 replicates)</td>
<td>167 cultivars</td>
</tr>
<tr>
<td>Lines/variety</td>
<td>3</td>
<td>4</td>
<td>-</td>
</tr>
</tbody>
</table>

Figure 1. Distribution of half-bloom dates for sorghum (sum of three trials, P1, VAT and DHS, Lavalette, Montpellier, France, 2008).
Climate data were provided by CEMAGREF (a French public research institute that targets results directly usable in land and water management). The CEMAGREF weather station is located 150 m from the trial plots. For the study of the emergence of the midges, soil temperatures were measured at a depth of -10 cm, while the length of the life cycle was based on mean air temperatures.

Field observations were made from 01/VII/2008 to 20/XI/2008. In all three trials and for each genotype, the 50% heading or half-bloom dates were recorded plot by plot. In the two trials (P1 and VAT) and in each plot, the percentage of seed loss per panicle due to midge damage was estimated, then the average of two replicates was calculated for the P1 trial and of three replicates for the VAT trial. Next, an average value of damage for genotypes with the same 50% flowering date was calculated for the two trials. Between September 16, and October 5, 2008, two panicles of several sorghum varieties (Autan, entries 9, 128, 315, 317, 428) were harvested and placed in plastic boxes (28x28x10 cm³) between 16/IX/2008 and 05/X/2008 at room temperature (20-25°C, with a natural photoperiod) to collect the emerging insects.

III. Observation of damage as a function of flowering date

Figure 2 shows the relationship between the half-bloom date and seed loss in the P1 and VAT trials. The midge infestations began on 07/VIII/2008 at a level that was already visible (20% of seed loss per panicle), then intensified rapidly between 9 and 16/VIII/2008 with 80% of seed loss, and culminating by up to 90-100% seed loss after August 20/VIII/2008. However, some genotypes did not fit this curve, for example hybrid V3 in the P1 trial displayed a high level of damage (60%) for an early flowering date (06/VIII/2008), while hybrid DSM 7-521 in the VAT trial was not affected at all, despite the fact that its half-bloom date (8/VIII/2008) was at the beginning of the infestation.

In the DHS trials, overall seed loss was sometimes high, but varied after heading, which occurred between August 2 and August 7, and was complete after 7/VIII/2008. Given that flowering occurs three to 7 seven days after heading, we can conclude that the dates on which damage appeared in the three trials were similar.
Figure 2. Transposition of the emergence curve for diapausing adult sorghum midges from Texas to Montpellier with application to following generations and relationship between seed loss (% of seeds aborted per panicle) due to the sorghum midge and half-bloom dates for sorghum (P1 and VAT trials, Montpellier, 2008).

IV. Application of a mathematical modelling of the dynamics of the sorghum midge infestations

We attempted to explain the infestation phenomenon by adapting two deterministic mathematical models developed at the University of College Station [30°34’N 96°21’W] in Texas (USA). These models take into account: i/ the onset and duration of adult sorghum midge emergence after the winter diapause as a function of soil temperature and rainfall (Baxendale et al. 1984a); and ii/ the length of the life cycles of the following generations of sorghum midges as a function of ambient (air) temperature (Baxendale et al. 1984b).

The climate at College Station in Texas, is warmer than in Montpellier (table 2). A comparison of mean temperatures revealed a time lag of two months in Montpellier for the period of emergence of the adult sorghum midge from the soil (difference of 0.4°C for sums of cumulative mean air temperatures from February to May in College Station [National and Local Weather forecast, 2009] and April to July in Montpellier [Météo France, 2009]).
Table 2. Comparison of mean monthly temperatures at College Station (Texas) and Montpellier (France) and mean monthly temperatures in Montpellier in 2008 (°C) – Illustration of two-month time lag in Montpellier of the period of adult emergence from diapause

<table>
<thead>
<tr>
<th>Month</th>
<th>Jan</th>
<th>Feb</th>
<th>Mar</th>
<th>Apr</th>
<th>May</th>
<th>Jun</th>
<th>Jul</th>
<th>Aug</th>
<th>Sep</th>
<th>Oct</th>
<th>Nov</th>
<th>Dec</th>
</tr>
</thead>
<tbody>
<tr>
<td>College Station (mean)</td>
<td>10</td>
<td>13</td>
<td>17</td>
<td>20</td>
<td>24</td>
<td>28</td>
<td>29</td>
<td>29</td>
<td>27</td>
<td>22</td>
<td>16</td>
<td>11</td>
</tr>
<tr>
<td>Montpellier (mean) 2008</td>
<td>7.9</td>
<td>8.0</td>
<td>9.9</td>
<td>12.6</td>
<td>16.7</td>
<td>21.1</td>
<td>22.8</td>
<td>22.8</td>
<td>18.0</td>
<td>14.4</td>
<td>9.1</td>
<td>5.9</td>
</tr>
</tbody>
</table>

Model of adult emergence after diapause
Emergence curves for adult midges were observed over a period of four years at College Station (1979-1982). Among these curves, the year 1980 was the most similar to the results obtained in Montpellier. Figure 2 shows the limits of the earliest and latest emergence curves obtained in College Station, where the curve recorded in 1980 is individualised. We moved back the 1980 curve two months to account for the difference in temperature at the two study sites, making it possible to determine the probable appearance of adults emerging from diapause in Montpellier (G0, fig. 2). In this scenario, midges from the overwintering generation would be expected to emerge in Montpellier from early July until early August, with the 50% adult emergence rate around 20/VII. In the American model, Baxendale et al. (1984a) consider that rainfall exceeding 12.7 mm delays the emergence of adults. In Montpellier, rain exceeding this amount fell on only one day (12/VIII/2008).

Model of the length of life cycles
The midge life cycle lasts between 14.6 days (at 30°C) and 28.5 days (at 20°C). Applying these data to the temperatures recorded in Montpellier in 2008, the potential emergences of adults are given by curves G1 to G3 in figure 2. In the model, there is an interruption between the end of generation and the beginning of the third generation as temperatures in Montpellier drop below 20°C and no data is available on the length of the life cycle at such temperatures. There was a very clear correlation between the gradual emergence of adults in curve G1 and the progression of the severity of damage, which is based on the half-bloom date, i.e. the stage at which the sorghum midge lays its eggs in the two days following its emergence from the nymph.

In the Montpellier trials, adults in egg-laying posture were observed until 29/IX/2008. In laboratory trials, adult emergences continued at a relatively high rate until the same date, then continued at a very limited rate until mid-October. In a fourth trial similar to the P1 trial, we also noted that the V492 entry, whose half-bloom date was 20/IX/2008, was not affected by sorghum midge infestation, indicating a considerable drop in the adult population at this date.
V. Observation of sorghum midge parasitoids and other insects on the sorghum panicle

During observations of sorghum panicles collected in the field, we also recorded the presence of other insects.

First, sorghum midge parasitoids, which were present from the start of rearing (16/IX/2008) in very limited numbers, appeared in large numbers from 29/IX until 19/X/2008, and were probably responsible for the significant reduction in the last generation of sorghum midges before diapause. Among these parasitoids, *Aprostocetus diplosidis* Crawford 1907 (Hymenoptera: Eulophidae) was the most abundant species and, to our knowledge, have never previously been described in France. This is a native of India that is also described in the United States, it was introduced in Italy for the purposes of biological pest control. A second, less abundant species of the same genus, *A. aff. phragmitinus* Erdös, 1954, was also observed, but this determination requires confirmation.

The following insects were also identified: *Sitobion avenae* Fabricius 1775 (Homoptera: Aphididae) or cereal aphid, a potential virus carrier (barley yellow dwarf virus or BYDV, Paliwal 1982); *Liorhyssus hyalinus* Fabricius 1794 (Hemiptera: Rhopalidae), is a bug which is widespread throughout the world, it is highly polyphagous, and causes damage to developing seeds and fruits (Cermeli et al. 2004); *Pinalitus conspurcatus* Reuter 1875 (Hemiptera: Miridae), a species observed for the first time in France, whose diet is unknown, but this genus is probably a pest (boring holes in seeds); *Orius* sp (Hemiptera: Anthocoridae), a genus which generally preys on acarids, aphids and thrips; *Euxoa tritici* L. 1761 (Lepidoptera: Noctuidae) or white-line dart, which feeds on flowers; and *Eupithecia* Curtis 1825 sp (Lepidoptera: Geometridae), which feeds on the flowers and ovaries of food plants.

Discussion

Limitations of the study

Our hypotheses do not account for a number of factors that may influence biological development. For example, we did not check whether there were differences in the biotype of the insects in the southern United States and those we observed in the south of France, but the estimation of the thermal threshold for the emergency of the first or last adult from diapausas is similar (14.8°C) that the one defined by Baxendale & Teetes (1983a) in Texas, based on a sum of Heat-Units. Likewise, sorghum midges have very reduced mobility and wind may play a role in the movement of the flies (Pendleton & Teetes 1994). Furthermore, the spatial distribution and density of the midge inocula emerging after diapause in the test plots depend on the density of panicles that harbour larvae in diapause during the two previous crop cycles, and may influence the dates and degree of infestation (Wolfenbarger 1972; Lampo & Mediaide 1994). In the case of the Domaine de Lavalette, sorghum had already been grown on the same plots in 2006, but few infestations were observed. Furthermore, adult sorghum midges have a very complex system of sensilla on their antenna (Slifer & Sekhon 2005) allowing them to perceive anthers: removal of the anthers would prevent egg laying (Sharma et al. 1990). Finally in the segmented experimentation design used (1 to 4 rows per variety), the interaction between the biological characteristics of the insect and the local environment could explain the attraction to genotypes in full bloom or, alternatively the groupings of midges whose determinants have not yet been studied in detail (Sharma & Franzmann 2001);
these characteristics could explain the early infestations of the V3 entry and the absence of damage to the DSM 7-521 entry. Specific trials are needed before we can conclude that V3 is a sensitive variety, or that DSM 7-521 shows particular resistance.

**Dynamics of sorghum midge infestations**

Despite these limitations, the use of the models developed in Texas by Baxendale et al. (1984a and b) and adapted to the climatic conditions in Montpellier enabled us to explain the dynamics of sorghum midge infestations in this region of southern France. All of the additional observations (variety with a late flowering date not infested, adults in egg-laying posture observed until 29/IX/2008, or adult emergence in the laboratory until mid-October) matches the results shown in figure 2 and with the onset of the sorghum midge diapause. In temperate regions, the winter diapause is often initiated by decreasing day length to reach 12 hours of daylight per day, with the equinox falling on 21/09. The gradual entry into diapause of sorghum midge larvae reduces the number of adults capable of egg laying, hence the lack of damage to late flowers; adult emergence ends very gradually, meaning some late emergences may occur until mid-October, with some nymphs being delayed by low temperatures. Our results therefore provide as a solid basis for future experimental confirmation.

It is generally agreed that a long flowering stage encourages infestation by the sorghum midge. This applies fully in our case, given that the varieties we studied had very different growth cycles, not including the role that could have been played by wild sorghum *Sorghum halepense* (L.) Pers., a common host of the sorghum midge, which is rigorously removed from plots in the Domaine de Lavalette. The practice of sowing borders of grain sorghum with early-flowering varieties around test plots (sometimes 50% of areas planted) is justifiable from an agronomic point of view (homogeneity of fertility, bird traps) but encourages the early development of the insect. In retrospect, it would have been wise to choose later-flowering genetic material.

Pursuing these scientific observations would have three advantages: first, it would make it possible to determine the latest cycle lengths of sorghum varieties that would be compatible with profitable production; this is particularly important to obtain varieties to be grown for silage. It would also provide a means of detecting resistance in certain varieties that suffer few infestations during the insect’s intense egg-laying period (Dakouo et al. 2000). Finally, it would make it possible to decide on specific dates for the insecticide treatments needed to control this insect.

Coutin (1969) thought that the climatic conditions in the Montpellier region (temperatures and rainfall regimes) would prevent severe infestations by the sorghum midge, considering the usual crop conditions (sowing in early May, the use of early to mid-early varieties). The hypothesis of a very rapid entry into diapause of the insect nevertheless needs to be clarified. Our observations indicate that the insect has a very long infestation period in one includes the latest individuals (egg-laying until mid-September?). For the time being, sorghum is cultivated in the Montpellier region to produce seed for early or mid-early varieties. But this production could be extended to include later-flowering sorghum varieties grown for biomass as a source of energy.

The value of modelling biological phenomena is very clear here: the results obtained at very distant locations (Texas and the south of France), which are different yet match those obtained in Montpellier, made it possible to better understand the dynamics of an insect pest.
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