Source of resistance in pearl millet varieties against stem borers and the ear headminer

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Abstract
The most damaging pearl millet insect pests in Senegal include ear headminer (Heliocheilus albipunctella) and the complex of stem borers Sesamia calamistis, Coniesta ignefusalis, Eldana saccharina and Busseola fusca. To reduce damage and loss from these pests, resistant varieties would be most cost effective for smallholder farmers. Evaluation using Gawane, ISMI9507, Thialack2 varieties, IBV8004 as the resistant check and Souna3 as the susceptible check from the Senegalese of Agricultural Research Institute (ISRA) was conducted under natural infestation in the 2014 and 2015 cropping seasons. The results showed a low stem borer incidence and damage on Thialack2 and Gawane as yield the lowest. It is suggested, Gawane contains a source of resistance to stem borers i.e. S. calamistis while Thialack2 also exhibited tolerance to H. albipunctella for which ISMI9507 would be resistance as well. 

Keywords: pearl millet, H. albipunctella; stem borers; host resistance

Introduction
Pearl millet (Pennisetum glaucum (L.) R. Br.) is one of the main cereal crops in West Africa where it is grown by smallholder farmers for their own consumption. It is a hardy crop capable of growing on low soil fertility and in extreme dry conditions [1]. It has good nutritional value including proteins, fibres, calcium and phosphorous [2]. Despite this importance, pearl millet is subjected to many abiotic and biotic stresses which can cause significant crop losses. In Senegal, low yield levels ranging between 500 and 800kg/ha were recorded the last decade [3]. This is partly due to pests including the stem borers, Coniesta ignefusalis (Hampson), and the ear headminer, Heliocheilus albipunctella (De Joannis), which are the most damaging insect pests of pearl millet [1, 4-6]. Damage due to the ear headminer is characterized by larvae feeding on the floral structure or cutting and lifting the peduncles close to the rachis of the ear head and leaving spiral pattern behind on the ear heads. This can lead to yield loss up to 85% due to flower abortion or grain spilling [7-9].

For stem borers, after boring in the pearl millet stalks their feeding can kill whorl leaves i.e. deadhearts which is the most severe damage caused by the first generation of the pest. Later infestations by stem borers lead to retarded plant growth, reduced flowering and thus reduced grain production. Late infestations also cause flower abortion or chaffy grains and broken stalks due to larvae feeding as they destroy the apical tissues and vessels transporting the plant sap [4, 10]. Pearl millet stem borer damage vary between regions from 14% up to total crop failure for C. ignefusalis [1, 4, 6, 11]. The use of resistant or tolerant varieties as a first line of defence in integrated pest management for reducing damage due to insect pests is one of the cheapest and the most environmentally friendly alternative control method [1, 12-14].

The objective of our research is to assess the resistance of different pearl millet varieties screened against stem borers and headminer.

Materials and Methods
Location
This study was conducted at the Senegalese Agricultural Research Institute (ISRA) of Nioro du Rip (13°45’ N 15°48’W, altitude 28m) during the cropping season of 2014 and 2015. The experimental site is located in the Senegalese peanut basin, the main pearl millet production area with a high level of pest pressure.
Rainfall data was collected from the ISRA Nioro du Rip weather station.

**Varieties**
The varieties Gawane, ISMI 9507 and Thialack2 bred from the National Agricultural Research Centre of Bambey were evaluated with IBV8004 and Souna3 as resistant and susceptible checks respectively [15].

**Experimentation**
The experimental design randomized complete block (RCB) with three replicates was used. Each plot was of 69 m x 40.5 m including border rows and with a sampling subplot of 11.5 m x 11.5 m for each variety. Seeds were sown after the first significant rain using 6 grains per hill and plants were thinned to three plants two weeks later i.e. a density of 36,000 plants/ha. Top dressing was done using a complex fertilizer (NPK 15.15.15) one week after seedling germination at 150 kg ha⁻¹. Urea (50%N) at a rate of 50 kg ha⁻¹ was applied into two shares at two weeks after seedling germination and a two weeks later. Weeding was done manually as needed.

**Sampling**
Sampling of stalks and ear heads was conducted weekly from pearl millet jointing to maturity. To identify stem borers, stalks were cut and taken to the lab for incubation. For the headminer the ear heads were directly observed in the field. For each sampling date and from each treatment five hills were randomly selected and data collected on the number of infested stalks and ear heads, insect larvae, deadhearts, stalk holes, mines and their length as well as thousand grain weight and yield of the varieties.

**Estimate of variables**
The incidence (I) of headminer and stem borers was estimated using the following formula.

\[
I(\%) = \frac{\text{Number of infested plants}}{\text{Total number of sampled plants}} \times 100
\]

The damage and grain loss estimates due to the headminer were based on the number and length of the mines. The determination of those grain losses was done using the method of Jago [16] based on the number of mines per ear head classified in categories from 1 to 3 where 1= 1 mine; 2= 2-3 mines and 3= 4 mines and more. Based on these categories, the sampled ear heads from the field were grouped into three classes of grain losses where class1, class 2 and class 3 corresponded to 1 g of grain loss; 2.5 g of grain loss and 4 g of grain loss respectively.

The grain losses (GL) were estimated using the formula.

\[
GL(\%) = \frac{\text{Weight loss for each variety (Kg)}}{\text{Grain production for each variety (Kg)}} \times 100
\]

**Statistical analysis**
An ANOVA was conducted for the different variables using a General Linear Model (GLM) with Student Newman Keuls test for mean separation at a probability level \( \alpha=0.05 \). Prior to the analysis of variance and for eliminating heterocedasticity, data were transformed using \( \arcsin(\sqrt{\frac{l}{100}}) \) and \( \log_{10}(\sqrt{X+1}) \) where \( l \) = percentage of the incidence or deadhearts or grain loss and \( X \) = other considered count variables (number of larvae, number of mines, length of the mines, number of holes).

All analyses were performed using the software SAS 9.1 [17]. Except for rainfall and the stem borer composition, all results in the text are presented as mean ± standard error.

**Results**

**Rainfall pattern**
The rainfall was lower in 2014 with a dry spell noted during the last three weeks of June. The rainfall was more abundant and better distributed during the 2015 cropping season.

**Incidence of stem borers and headminer**
The incidence of the pearl millet headminer and stem borers in the tested varieties for 2014 and 2015 cropping seasons is shown in Figure 2. The headminer incidence was higher than the incidence of stem borers in both years. In 2014, the incidence of stem borers was low and showed no significant difference between tested varieties. A similar result was noted for the incidence of the headminer for the same time period.
In contrast, the stem borer incidence in 2015 was higher with a significant difference between varieties (P<0.05). The lowest incidence of 28±4% of infested plants was noted with Thialack2 compared to ISMI9507, Souna3 and Gawane for which the incidence ranged between 31±3% and 33±3%. The resistant check, IBV8004 was the least infested with 20±3% of the stalks affected in comparison to the tested varieties (Figure 2b). For the headminer, the incidence in 2015 was significantly lower for the ISMI9507 with 32±6% of the ear heads mined compared to IBV8004, Gawane and Souna3 for which the infestations ranged between 41±6% and 47±4%. The highest headminer incidence was recorded in Thialack2 with 56±7% of plants mined (Figure 2b).

**Stem borer larval density and damage**

The stem borer larvae density and damage in terms of holes and dead stalk or deadheart were variable in both two years and particularly low for 2014 compared to 2015 (Figure 3). For the cropping season 2014, the deadhearts were only noted on ISMI9507 and Gawane varieties with a low level of dead plants estimated at nearly 3% of the sampled hills. Nevertheless, no significant difference was noted between the varieties for the density of stem borer larvae and damage (Figure 3a). In contrast to 2015, the density of the stem borers larvae and damage were higher for all varieties, particularly Thialack2, Gawane and ISMI9507 which showed larval densities per hill varying from 8.40±1.46 to 12.07±1.84 as well as the holed stalks per hills ranging between 12.40±3.13 and 15.20±2.35. The larval density per hill and the number of holed stalks of 6.53±1.19 and 7.60±1.39 respectively were significantly lower for the resistant check IBV8004. Thialack2 recorded on average 9% of deadhearts per hill which was higher than the deadhearts exhibited by ISMI9507, Souna3 Gawane and IBV8004 (Figure 3b).
Stem borer composition
The pearl millet stem borer, *Cnopista ignefusalis*, the African pink borer, *Sesamia calamistis*, the African stalk borer, *Busseola fusca* and the African sugar cane borer, *Eldana saccharina* were the only stem borers recorded. The most frequently recovered species during the cropping seasons of 2014 and 2015 were *S. calamistis* and *C. ignefusalis* with 31-72% and 16-53% respectively (Figure 4).

**Fig 4**: Stem borers composition in the different pearl millet varieties in 2014 (A) and 2015 (B) cropping seasons. *Resistant check; ** Susceptible check.

Headminer larval density and damage
A more significant natural infestation of pearl millet by the headminer was noted in 2015 compared to 2014. The density of the headminer larvae and the number of mines as well as their length did not show a significant difference for the tested varieties in 2014 (Figure 5a). As for 2015, Thialack2 recorded more larvae, more mines and longer mines compared to the other tested varieties in average of $7.40\pm1.03$ larvae per hill, $13.27\pm1.87$ mines per hill and $5.75\pm1.14$ cm per mine. ISM9507 had the lowest larval density, number and length of mines with $3.20\pm1.32$ larvae per hill, $5.87\pm1.38$ mines per hill and $2.49\pm0.67$ cm per mine. However, this was comparable to the resistant check, IBV8004. Gawane and the susceptible check Souna3 were equally damaged (Figure 5b).

**Fig 5**: Mean (±SE) number of ear headminer larvae and mines and length of mines in the different pearl millet varieties in 2014 (A) and 2015 (B) cropping seasons. Means followed by the same letters are not significantly different. *Resistant check; ** Susceptible check.
Yield and grain yield loss

The grain yield of the different varieties tested was higher in 2015 but the 1000 grain weight was higher in 2014 (Table 1). The grain yield loss was low for both cropping seasons with a slightly better situation in 2014. Within a cropping season, no significant difference was noted between varieties for yield, yield loss and 1000 grain weight (Table1).

Table 1: Grain yield parameters and loss in the different pearl millet varieties in 2014 and 2015 cropping seasons. For each year means followed by the same letters are not significantly different. *Resistant check; ** Susceptible check.

<table>
<thead>
<tr>
<th>Cropping seasons</th>
<th>Varieties</th>
<th>Mean ± SE</th>
<th>1000 Grain yield (g)</th>
<th>Grain yield (t/ha)</th>
<th>Grain yield loss (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2014</td>
<td>Gawane</td>
<td>11.27±0.33a</td>
<td>1.50±0.14a</td>
<td>3.64±0.46a</td>
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<td></td>
<td>ISMI9507</td>
<td>10.96±0.26a</td>
<td>1.40±0.09a</td>
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<td></td>
<td>Thialack2</td>
<td>10.33±0.28a</td>
<td>1.83±0.31a</td>
<td>3.40±0.69a</td>
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<td></td>
<td>IBV8004**</td>
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<td>1.47±0.14a</td>
<td>3.45±0.29a</td>
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<td></td>
<td>Souna3**</td>
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<td>2015</td>
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<td>Souna3**</td>
<td>7.75±0.15a</td>
<td>2.31±0.24a</td>
<td>2.37±0.38a</td>
<td></td>
</tr>
</tbody>
</table>

Discussion

For all parameters recorded in 2014 cropping season, the behaviour of all varieties was similar with a slight variation in 2015. This might be related to the low population of the pests and the erratic rainfall in 2014 compared to 2015. Other factors affecting damage by insects could be population density, year or time of occurrence, unstable cultivated habitats and the production cycle of host varieties [18, 19].

During the two years of investigation, the incidence of stem borers was lower than the incidence of the headminer. This is generally noted on pearl millet as far as these two pests are concerned [6].

Considering overall behaviour, Thialack2 and Gawane exhibited resistance to stem borers. The incidence of stem borer larvae and holed stalks was very low compared to the susceptible check Souna3. For Thialack2, earliness may have played an important role in the mechanism of resistance. Previous research has shown that for stem borers early maturing varieties are less damaged than late maturing varieties [1, 20]. For Gawane leaf trichomes could be preventing easy egg laying for the stem borers as well as normal growth and development for the young larvae i.e. physico-chemical resistance to insects [1, 10, 21, 22, 23]. For all the varieties tested, similar to the response in sorghum when infested by *B. fusca* [32]. Additionally, the deadheart and plant height exhibits direct positive effects on yield if the new stalks are not stressed by water and nutrient deficiency [10]. For ISMI9507 the incidence of the headminer, its larval density and lower damage in 2015 seem to indicate a source of resistance to this panicle pest due to the morphology of the ear or the variety earliness creating unfavourable establishment conditions for the young larvae [14, 33]. Nevertheless, other stress factors such as bristle beetle damage among others [18] may have contributed to a reduction in yield of the tested varieties compared to their potential of 2 to 3 metric tons per hectare (t ha⁻¹). In 2014, the rainfall distribution within months, the water stress which occurred after the sowing and the late infestation by stem borers may have led to low 1000 grain weight as the panicle abortion increased with the increase in holed stalks [34].

Conclusion

All parameters monitored including yield of the different varieties compared to the checks in overall, suggested Gawane and Thialack2 possess a source of resistance to stem borers. Thialack2 exhibited additional tolerance to the headminer leading to a reduction in crop loss. ISMI9507 also showed good resistance to the headminer even though the actual yield was relatively low compared to the potential yield due to its susceptibility to others limiting factors.

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