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# Losses in wheat varieties upon *Sitophilus oryzae* L. Infestation

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**Abstract**
*Sitophilus oryzae* is a major stored grain pest affecting many varieties of food grains, among which wheat infestation is common. It has been estimated that females of *Sitophilus oryzae* can destroy 50 g of stored grain in three generations. Quantitative loss was ascertained by infesting a fixed quantity of wheat grains with a fixed number of male and female pairs and taking the weight *Sitophilus oryzae* is a major stored grain pest affecting many varieties of food grains, among before and after the infestation. Qualitative changes concerning the loss of amino acid species upon infestation were ascertained by scanning the amino acid content in the pre- and post-infestation phases through double-dimensional chromatography. It has been found that the infestation caused weight loss in different varieties, but the losses were not uniform, such as while PB 1105 suffered the most (24.57%), Sonalika suffered moderate (19.96%), and Kalyan Sona suffered the least losses (11.96%). Similarly, the amino acid content also induced the attack. The chromatogram of the post-infested sample revealed that in the PB-1105 variety of wheat, six amino acids were lost, including two essential ones. Whereas, in Sonalika and Kalyan Sona, out of twelve amino acids, only three were lost, of which one was an essential amino acid.

**Keywords:** *Sitophilus oryzae*, quantitative and qualitative losses, paper chromatography, essential amino acids, wheat varieties

## Introduction
The infestation by *Sitophilus oryzae* causes immense loss to the infested grains. Poor management at post-harvest and storage levels causes a great problem with insect infestation (Zulaikha *et al*., 2021) [13]. The losses include quantitative losses caused by the insect directly feeding on the grain kernel. It has been estimated that one female of *Sitophilus oryzae* can destroy 50 g of stored grain completely in three generations (Cogburn, 1974) [3]. But the losses may not be quantified uniformly as they vary depending on the climatic condition, the type of container, and the duration of storage. In addition, varieties and grains also influence the losses caused (Campbell and Sinha, 1976) [2]. The infestation of grain by *S. oryzae* not only causes quantitative losses but also causes qualitative changes, such as loss of palatability due to the accumulation of excreta and decaying remains of the insect, resulting in bad odour. Furthermore, the infestation by stored grain pests also causes biochemical changes in the food grains, such as increased concentrations of fatty acids, an increase in non-reducing sugars, and the loss of important nutrients. (Saujanya *et al*., 2013). The objectives of this study were to ascertain (i) loss of weight and (ii) qualitative losses, with special references to the loss of particular amino acid species caused by the infestation of *S. oryzae* in the three selected varieties of wheat.

## Material and Methods
Three high-yielding varieties of wheat, *e*. Kalyan Sona, PB 1105, and Sonalika, were selected for studying weight loss when subjected to infestation by *S. oryzae*. The study was conducted in laboratory conditions at the University Department of Zoology, B.R. A. Bihar University, Muzaffarpur, Bihar, a northern state of India, during the 2023 rainy season.

### Assessment of weight loss
Lots of 100 grains of each variety were weighed out in petri dishes. Seven replicates were prepared for each variety. Eight females and four males of newly emerged weevils were introduced into each of these samples, except for the control. The sorting of sexes was done based on the length of the rostrum and dorsal punctures. The females were recognised by their rostrum being longer, narrower, more curved, and with fewer dorsal punctures than in males. In addition, external genitalia characteristics were studied by Halstead (1963) [4]. The petridishes were covered with muslin cloth so as to allow ventilation and, at the same time, prevent the escape of the insect. After two weeks of infestation, the insects were removed. The dead insects were accounted for and thrown away. Samples were left undisturbed under laboratory conditions at 26 ± 2 ºC and 65% relative humidity for another two weeks for the F1 generation to emerge. Thereafter, samples were inspected every other day for the emergence of F1 adults in *S. oryzae*. While counting, the containers were emptied onto white paper, and insects were counted with a camel hairbrush. After counting, all the contents were carefully replaced so that no portion of the sample was lost in transit. Each trial replicate was terminated when no emergence was recorded for three consecutive days. The contents of each Petri dish were again examined at the end of the experiment. The number of infested grains, as indicated by white chalky spots, linear streaks, and other damages, was ascertained. Each grain was dissected to remove developmental stages such as larvae and pupae. While doing so, abundant precautions were taken to prevent loss of any grain particle. Finally, loss of weight was ascertained by weighing the remains of the samples. Weights were taken again for each sample. The difference between the initial and final weights of the control, i.e., the weight loss suffered by the control, was deducted from the initial weight of the sample from which the final weight of the infested grains of each sample of each variety was deducted. The value represents the loss in weight caused by the feeding activity of the weevil.

Actual weight loss W = X – (CF) – Y
Where X is the initial weight of the uninfested sample, Y is the final weight of the infested sample, and CF is the weight

loss in control, assumed to be due to moisture loss.
Percentage weight loss = W/X * 100

**Assessment of loss of amino acids**

The biochemical changes with respect to the loss of amino acids upon infestation were also ascertained to find out the loss of amino acids due to infestation, and also to find out any possible correlation between the amino acid contents of the grain and the feeding preferences of the weevil *S. oryzae*. The determination of the amino acid content of food grains was done by double-dimensional paper chromatography in both infested and uninfested grains (defated) of each of the three varieties of wheat. Amino acids were characterized by calculating and comparing their Rf values.

Rf factors of amino acids Alanine, Arginine, Asparagine, Aspartic acid, Cysteine, Glutamine, Glutamic acid, Glycine, Histidine, Isoleucine, Leucine, Lysine, Methionine, Phenylalanine, Proline, Serine, Threonine, Tryptophan, Tyrosine, Valine are 0.38, 0.20, 0.5, 0.24, 0.4, 0.13, 0.30, 0.26, 0.11, 0.72, 0.73, 0.14, 0.55, 0.68, 0.43, 0.27, 0.35, 0.66, 0.45, 0.61 respectively.

**Result and Discussion**

Feeding losses by *S. oryzae,* while feeding upon different food grains studied during this research work, caused extensive damage to the grains, boring holes in the grains, and in extreme cases of infestation, the grain is hollowed with its interior converted into powdered mass. Thus, causing extensive losses due to feeding on grains. The losses are both quantitative and qualitative (Fig 1).

**Fig 1:** *Sitophilus oryzae* infesting wheat grains and an adult *Sitophilus oryzae*

**Weight loss**

The study carried out on the feeding losses suffered by wheat varieties, which were the subject of this study, revealed that out of the three varieties, Kalyan Sona suffered the least percentage weight loss at 11.96% (Table 1), whereas wheat variety PB 1105 suffered the most, with a percentage weight loss of 24.57% recorded in it. The Sonalika, which suffered moderate losses, recorded a percentage weight loss of 19.96%, which is more than Kalyan Sona but remarkably less than PB 1105.

**Table 1:** Showing percentage weight loss in wheat variety Kalyan Sona caused by infestation of *S. oryzae*

| Variety | Mean initial weight of control (gm) | Mean final weight of control (gm) | Mean difference in weight of control (gm) | Mean weight of uninfested sample (gm) | Mean weight of infested sample (gm) | Mean actual weight loss | Mean percentage weight loss |
|---|---|---|---|---|---|---|---|
| kalyan Sona | 3.088 | 3.024 | 0.064 | 3.089 | 2.655 | 0.370 | 11.967 |
| Sonalika | 4.217 | 4.129 | 0.088 | 4.314 | 3.365 | 0.861 | 19.965 |
| PB1105 | 4.057 | 3.983 | 0.074 | 4.063 | 2.990 | 0.998 | 24.578 |

Sudhakar and Pandey (1982) have reported a positive correlation between population buildup and weight loss in the case of *S. oryzae*, in line with the findings of this study. They also categorised Kalyan Sona as less susceptible to *S. oryzae* infestation. The findings of this study also concur with the findings of Singh *et al*. (1974). While studying the oviposition and development of *S. oryzae* on different high-yielding varieties of wheat, they found Sonalika to be the most preferred one, whereas Kalyan Sona came after it. Chakraborty and Mathew (1972) have, however, differed from the findings of this study and have ranked Sonalika ahead of Kalyan Sona as far as the resistance to the *S. oryzae* infestation is concerned. The damage that can be positively correlated with the emergence of more adults has also been confirmed by Singh *et al*. (1985) [11] and Khan *et al*. (2014) [7]. Tiwari (2016) [10], while evaluating the susceptibility of different wheat cultivars, *viz*., CSH-14 and CSH-16, has also come to this conclusion. Similar work has been carried out by Kumar (2018) [8]. They have concluded that PBW 343 is the most preferred, whereas WH 1105 and WH 1124 are the least preferred. Mehta and Kumar (2020) categorise different wheat varieties as high, moderate, and least susceptible to *S. oryzae*.

**Qualitative losses and changes**

When the post-infested sample of the PB-1105 variety of wheat was subjected to chromatographic studies after careful removal of various development stages of the insect, six spots were found to be missing from the chromatogram. These corresponded to Alanine, Aspartic Acid, Cysteine, Histidine, Isoleucine, and Tyrosine. Out of these missing six amino acid species two, the Isoleucine and Histidine were essential amino acids (Table 2). In the non-infested Sonalika variety of wheat, twelve identifiable spots were visible, corresponding to Arginine, Aspartic Acid, Cysteine, Glutamic Acid, Histidine, Leucine, Lysine, Methionine, Phenylalanine, Serine, Tryptophan, and Tyrosine. Out of these twelve amino acids present in Sonalika, six were essential amino acids. Upon infestation, three amino acids were lost from the wheat grains of the Sonalika variety *viz* histidine, serine, and tyrosine, of which histidine is the only essential amino acid. Analysis shows that twelve spots were also found on the chromatogram of uninfested Kalyan Sona. These were corresponding to Arginine, Aspartic Acid, Cysteine, Glutamic Acid, Histidine, Lysine, Leucine, Methionine, Phenylalanine, Serine, Tryptophan, and Tyrosine of these, the six Histidine, Lysine, Leucine, Methionine, Phenylalanine and Tryptophan were essential amino acids. Upon infestation, three amino acids were lost of which only one the Histidine was an essential amino acid. One essential amino acid (histidine) was lost among six essential amino acids (Fig2). The study carried out on the feeding losses suffered by wheat varieties, which were the subject of this study, revealed that out of the three varieties,

Kalyan Sona suffered the least percentage weight loss at 11.96% (Table 1), whereas wheat variety PB 1105 suffered the most, with a percentage weight loss of 24.57% recorded in it. The Sonalika, which suffered moderate losses, recorded a percentage weight loss of 19.96%, which is more than Kalyan Sona but remarkably less than PB 1105.

**Table 2:** Showing percentage weight loss in wheat variety Kalyan Sona caused by infestation of *S. oryzae*

| Variety | Mean initial weight of control (gm) | Mean final weight of control (gm) | Mean difference in weight of control (gm) | Mean weight of uninfested sample(gm) | Mean weight of infested sample(gm) | Mean actual weight loss | Mean percentage weight loss |
|---|---|---|---|---|---|---|---|
| kalyan Sona | 3.088 | 3.024 | 0.064 | 3.089 | 2.655 | 0.370 | 11.967 |
| Sonalika | 4.217 | 4.129 | 0.088 | 4.314 | 3.365 | 0.861 | 19.965 |
| PB1105 | 4.057 | 3.983 | 0.074 | 4.063 | 2.990 | 0.998 | 24.578 |

**Table 3:** Comparison between the amino acid composition of fresh and infested wheat grains upon infestation by *S. oryzae*

| Sl.No | Amino acids | PB 1105 | | | Sonalika | | | Kalyan sona | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | AA in fresh grains | AA in infested grains | Total no. of lost amino acids | AA in fresh grains | AA in infested grains | Total no. of lost amino acids | AA in fresh grains | AA in infested grains | Total no. of lost amino acids |
| 1 | Alanine | + | - | 6 amino acids (Alanine Aspaertic acid Cysteine Histidine Isoleucine & Tyrosine) | - | - | 3 Amino acids (Histidine Serine & Tyrosine) | - | - | 3 Amino acids (Aspartic acid Histidine & Tyrosine) |
| 2 | Arginine | + | + | | + | + | | + | + | |
| 3 | Aspaertic acid | + | - | | - | - | | + | - | |
| 4 | Cysteine | + | - | | + | + | | + | + | |
| 5 | Glutamic acid | + | + | | + | + | | + | + | |
| 6 | Glycine | - | - | | + | + | | - | - | |
| 7 | Histidine* | + | - | | + | - | | + | - | |
| 8 | Isolucine* | + | - | | - | + | | - | - | |
| 9 | Leucine* | + | + | | - | + | | + | + | |
| 10 | Lysine* | + | + | | + | + | | + | + | |
| 11 | Methionine* | + | + | | + | + | | + | + | |
| 12 | Proline | - | - | | + | + | | - | - | |
| 13 | Phenylalanine* | + | + | | + | + | | + | + | |
| 14 | Serine | + | + | | + | - | | + | + | |
| 15 | Tryptophan* | + | + | | + | + | | + | + | |
| 16 | Tyrosine | + | - | | + | - | | + | - | |

Here *Essential amino acid and AA – Amino acid

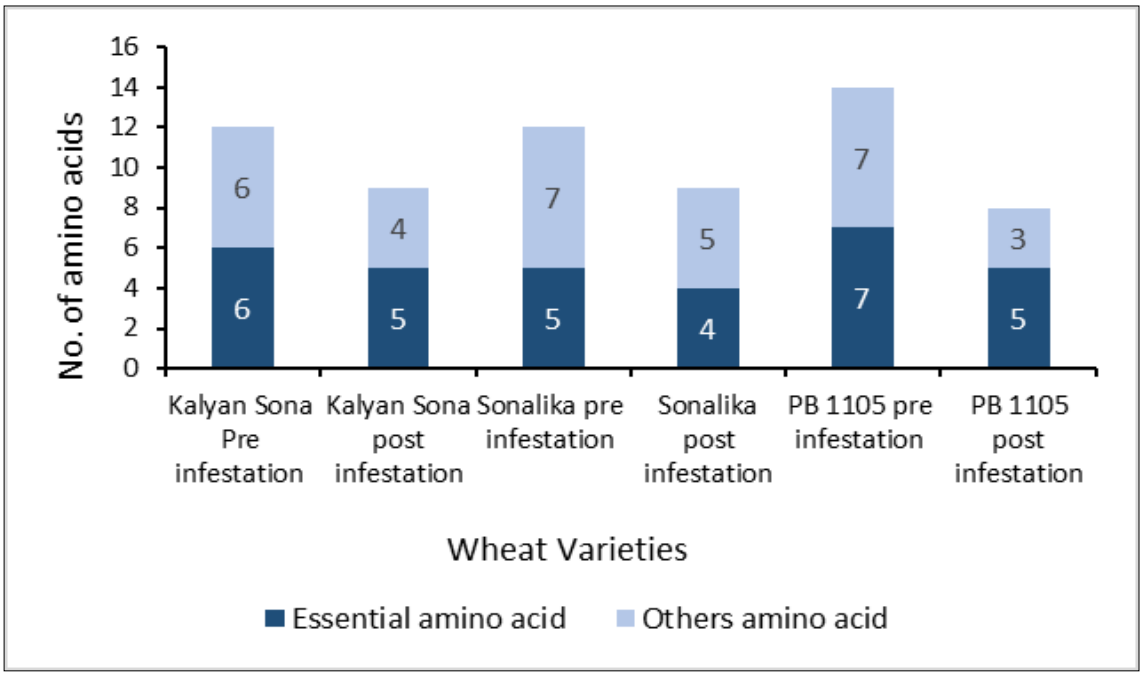


**Fig 2:** Comparison between the number of amino acids in fresh and *S. oryzae*-infested wheat varieties

Contrary to the findings of the instant study, many authors, such as Zulaikha and Yaakop (2021), have found no direct correlation between the chemical composition of grain and the preference of *S. oryzae*. This investigation, however, is based on amylose content rather than amino acid. Khan and Haldhar (2012) [6] and Akhtar (2017) have concluded that the preference factor is the largest of the grains. Islam (2007) [5] also conquered the view that the rate of infestation does not solely depend on the chemical makeup of the grain. Unlike the previous authors, the present study text considers

the amino acid species present in the grain, and the findings indicate that essential amino acids may be the attracting factor for the infestation. Essential amino acids, unlike the amylose content, may act as a limiting factor for infestation because of their essential nature in physiology.

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**Author contribution statement**
**BKPS:** designed and performed the experiment; FW: assisted BKPS in performing experiments; BKPS: manuscript writing; FW: reviewing, and editing the manuscript.

**Conflicts of interest**
No conflict of interest.

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