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# Gossypol as a Natural Insecticide in Cotton Plants against Cotton Thrips and Pink Bollworm

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ABSTRACT

Gossypol level plays an important role in self-protection as antibiosis of cotton plants (*Gossypium* spp.). Thus, it was found necessary to evaluated the level of gossypol in the different parts during the stages of growth of cotton plant for three cotton cultivars viz., Giza 86, Giza 80, and Giza 45. Their performance against cotton thrips, *Thrips tabaci* and pink bollworm, *Pectinophora gossypiella* under field conditions was studied. Statistical analysis revealed that there was a positive correlation between the gossypol content in the seed pulp of different varieties of cotton and the amount of gossypol detected in the different parts of the produced plants. The level of gossypol in seeds were significantly higher on Giza 86, and Giza 45 having 1.90% and 1.52%, respectively, whereas the level of gossypol was limited in Giza 80 (1.45%). Therefore, cotton variety, Giza 86 showed higher levels of gossypol in all parts of plant. In field tests during 2019, the results indicated that the higher content of gossypol had a negative effect on population size both *T. tabaci* (adult and nymph) and *P. gossypiella* larvae that infested cotton bolls. Cotton thrips and pink bollworm population exhibited negative response to gossypol content. According to the results, the cotton varieties could be arranged in the following descending order concerning their susceptibility to infestation with these pests: Giza 80, Giza 45 and Giza 86.

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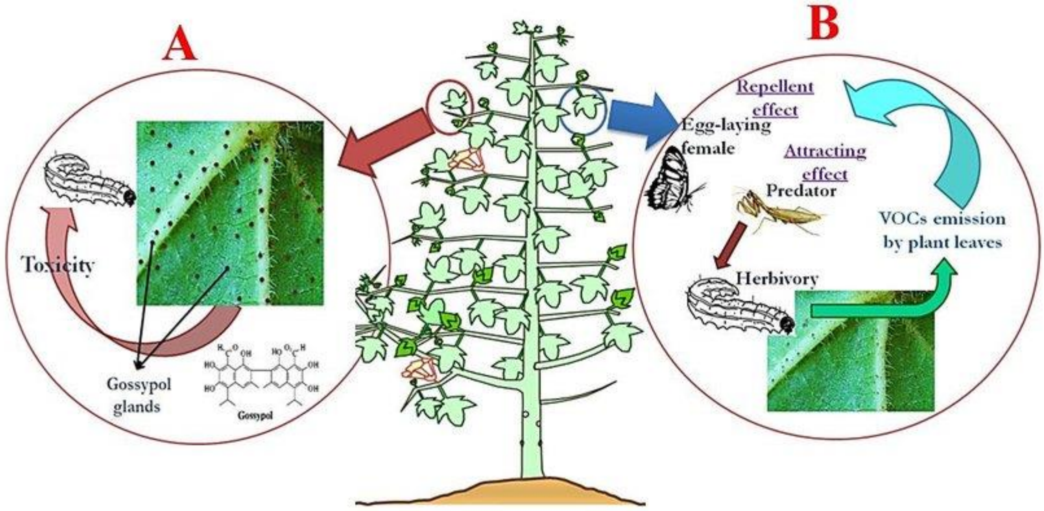
**GRAPHICAL ABSTRACT**




## 1. Introduction

Cotton plants is known as "white gold". It is the main fiber and cash crop of Egypt that grows in both tropical and sub-tropical regions throughout the world, in about 111 countries. One major challenge faced by cotton farmers is the high pest incidence which occurs at virtually all stages of cotton growth, limiting its productivity and quality [1]. There are 1326 species of insect pests on cotton, though most cause little or no economic damage. Pests that attack reproductive structures cause about 80% of the damage to cotton [2]. Among the cotton pests, the cotton thrips *Thrips tabaci* (Lindeman) (Thysanoptera: Thripidae) and pink bollworm, *Pectinophora gossypiella*, (Saunders) (Lepidoptera: Gelechidae) remain the principal pests in Egypt. Cotton thrips is the most vital early-season sucking insect pest on cotton that infests at the beginning of the crop season, delaying early plant development; Thrips cause great damage to the cotton seedlings where a maximum population of cotton thrips was recorded in the second fortnight of September. Thrips harmed seedlings occasionally show an explosion of monopodial branches. Also, pink bollworm is the most injurious insect damaging the bolls completely as well as the growing buds and flowers [3].

Plants protect themselves from herbivores and pathogens by synthesizing structurally diversified secondary (specialized) metabolites; many exert defense function by their cytotoxicity [4]. Cotton plants from the genus *Gossypium* are one of the most important economic crops and the major source of natural fiber for textile industry. Today 95% of all cotton are derived from *Gossypium hirsutum* family of Malvaceae [5]. They accumulate a large amount of sesquiterpenoids as phytoalexins, of which the most known is gossypol. Gossypol and related sesquiterpene aldehydes are the major phytoalexins against the parasites, pathogens and pests [6]. It is naturally produced in lysigeneous glands which are small black spots distributed throughout the cotton plant, but their greatest concentration is in the seeds. These lysigeneous glands are of size 50–400 µm. The amount of gossypol in different parts of cotton plant varies from 0.02 to 6.64% [7]. It plays the role of natural insecticide in tree cotton and acts as its defense mechanism, thereby defending the plant against subsequent insect attacks through anti-insect activity [8,9]. Gossypol levels can vary significantly by variety and within a variety grown under different environmental conditions [10,11]. Generally, resistance of cotton plants based on gossypol levels is promising within the pest management approach of cotton pests. therefore, this investigation was undertaken to determine the gossypol content in the different parts during the stages of growth of cotton plants of three cotton varieties including Giza 45, Giza 80, and Giza 86 and determine the optimal cotton variety through the extent of its sensitivity to infection with adult and nymph of cotton thrips, *Thrips tabaci* and larvae of pink bollworm, *Pectinophora gossypiella* under field conditions.

## 2. Experimental phase

### *2.1. Experimental Design*

The field experiments were conducted during the cotton-growing summer season in El-Beheira Governorate at Kafr El-Dawar district, Egypt. Before setting up the experiment, we prepared the field for seed sowing. Seeds were sowed on 4th April 2019 for three different cotton varieties listed in **Table1**. The experiments were laid out in Randomized Complete Block Design (RCBD) four replications. Three tested cottonseed varieties were designed in the total experimental area of 2160 $m^2$. This area was divided into plots (replications) size of each sub-plot was 175 $m^2$. The plots received all good recommended agricultural practices without any insecticidal treatments applied during the season.

### *2.2. Laboratory examination*

#### *2.2.1. Gossypol content determination in the different parts of cotton plants during the stages of growth*

Three complete seedlings were picked at random representing each plot 30 days after planting, to prevent any damage to the root system. These samples were taken to demonstrate the difference between the seeding grown under the field conditions regard to their gossypol content according to method [12]. Investigating the total gossypol content in the different parts of cotton plants during the stages of growth, we took samples of roots, true leaves, stems, sympodiums, leaves of sympodiums and flower buds 90 days after planting. This period represents the stage of growth of cotton plants two weeks before blooming. Also, samples of flowers were partitioned to receptacle, epicalyx, calyx, and pericarp at blooming day (104 days after planting). Marked bolls were divided to the receptacle, epicalyx, calyx, pericarp, and seeds 10, 20, 30 and 40 days after fruit setting. Samples of mature seeds representing each variety were taken from open bolls 60 days after fruit setting.

**Table 1.** Comparative classification of the different varieties of the three Egyptian cotton

| | *Cotton varieties* | | |
|---|---|---|---|
| *Properties* | Giza 45 | Giza 80 | Giza 86 |
| *Pedigree crossing between* | Giza 7 x Giza 28 | Giza 66 x Giza 73 | Giza 75 x Giza 81 |
| *Class– growing areas* | East of Delta | Middle of Egypt | Middle and North of Delta |
| *Country of origin* | Egypt | Egypt | Egypt |
| *Advantages* | Extra-long staple (over 13/8-inch fiber length) | long staple (1 1/4 -1 3/8-inch fiber length) | long staple (1 1/4 -1 3/8-inch fiber length) |

These data were obtained by Cotton Research Institute, F.C.R.I., A.R.C., Giza, Egypt.

Hulls were discarded, and the kernels were kept for free, bound, and total gossypol determinations [13]. All samples were dried and kept at 10 °C.

*2.2.2. Susceptibility to infestation during the early stage of growth*

Three weeks after planting, adult thrips (*Thrips tabaci*) started to appear on the cotyledons of the seedlings. Estimation of infestation was carried out by counting the adult, and nymphal thrips on 20 seedlings selected randomly from the four replicates of each variety. Counting was achieved every week. The samples were kept in nylon bags and taken immediately to the laboratory where they were carefully inspected. Using binocular infestation percentages, we calculated them according to the formula of [14].

*2.2.3. Susceptibility to infestation with bollworms*

To assess the infestation with pink bollworm, *Pectinophora gossypiella* (Saund.), we picked one hundred green bolls at random to represent each plot (400 bolls per each cotton variety). First samples were taken on August 3, 2019. Then, successive samples were taken at weekly intervals. Percentage of infestation by pink bollworm was determined in the laboratory by dissection of bolls and checking the bolls externally and internally. Any sign of infestation, i.e. a hole in the pericarp, the presence of any number of pink bollworm larvae inside the boll was recorded; the boll was infested. The percent of pink bollworm infestation and the population density and the population size of the pink bollworm larvae throughout the test period (5 weeks) was calculated according to [14].

*2.3. Statistical Analysis*

All data were analyzed using the SPSS 20.0 Software Package (SPSS Inc., Chicago, IL, USA). One-way ANOVA followed by the Tukey's HSD test in JMP 11.1.1. (SAS Institute Inc. 2013, Cary, NC, USA) was employed to analyze differences between developmental stages.

## 3. Results

*Gossypol content in tested cotton seedling before planting*

Determining the total gossypol content in the cottonseed pulp of the different varieties before planting revealed that Giza 86 and Giza 80 contained respectively the highest and the lowest amount of gossypol by 1.90% and 1.45%, respectively, while Giza 45, contained 1.52% gossypol.

*Gossypol content in tested cotton seedling after planting*

The amounts of total gossypol detected in the different parts of the cotton seedlings 30 days after planting. The parts of seedlings could be arranged in the following descending order concerning the values of gossypol estimated: roots, cotyledons, true leaves, and hypocotyls (**Table 2**).

**Table 2.** Total gossypol content in the different parts of cotton seedlings, 30 days after planting

| *Cotton varieties* | *Cotyledons (%)* | | *Roots (%)* | | *Hypocotyls (%)* | | *True leaves (%)* | |
|---|---|---|---|---|---|---|---|---|
| | F* | D* | F | D | F | D | F | D |
| *Giza 45* | $0.109^{c}\pm0.14$ | $1.006^{a}\pm0.20$ | $0.160^{b}\pm0.12$ | $1.162^{b}\pm0.15$ | $0.017^{a}\pm0.12$ | $0.111^{b}\pm0.22$ | $0.093^{b}\pm0.20$ | $0.606^{b}\pm0.23$ |
| *Giza 86* | $0.081^{b}\pm0.71$ | $0.787^{b}\pm0.61$ | $0.151^{a}\pm0.15$ | $1.252^{b}\pm0.23$ | $0.016^{a}\pm0.26$ | $0.102^{a}\pm0.28$ | $0.095^{b}\pm0.26$ | $0.637^{c}\pm0.21$ |
| *Giza 80* | $0.072^{a}\pm0.24$ | $0.801^{b}\pm0.58$ | $0.151^{a}\pm0.12$ | $1.036^{a}\pm0.20$ | $0.028^{b}\pm0.18$ | $0.193^{c}\pm0.23$ | $0.071^{a}\pm0.17$ | $0.422^{a}\pm0.13$ |

F*: Fresh weight D*: Dry weight
The data in the table are the means ± SE. Different letters in the same column indicate significant differences at P<0.05.

*Gossypol content in tested cotton plants varieties 90 days after planting*

There is considerable natural variation in contents of gossypol for developmental stages of cotton plants in most cultivars as shown in **Table 3.** The data for root samples indicated that Giza 86 was the most contained gossypol (1.248%) followed by Giza 80 (1.184%) compared with Giza 45 contained (0.974%). The samples of flower buds were almost the same as those detected in the root samples. Along the same lines, the highest amounts of gossypol (0.395, 0.315 and 0.116%) were recorded in the samples taken from true leaves, leaves of sympodiums and main stems respectively for Giza 86. Also**,** the values recorded were 0.066, 0.061 and 0.053% with the sympodiums (without leaves) of Giza 45, Giza 86 and Giza 80. Generally, it was shown that the different parts of cotton plants could be the arranged in the following descending order concerning the amounts of gossypol detected: roots, flower buds, true leaves, leaves of sympodiums, main stems and sympodiums.

*Gossypol content in the flowers*

Flowers samples were taken on a blooming day. The plants were 104 days old for analyzed total gossypol content. The data showed that the receptacle samples contained the lowest amounts of gossypol. The values recorded in descending order were as follows: 0.163, 0.102 and 0.077% where the samples were taken from Giza 86, Giza 45 and Giza 80 plants, respectively. Analyzing the epicalyx samples, we found the amounts of gossypol higher than those detected in the receptacle samples. As for the calyx samples, the values of gossypol recorded were higher than those recorded for the epicalyx samples in all the varieties tested. The highest amounts (0.313%) were found in the samples of Giza 86 and the lowest (0.187%) samples of Giza 80. The respective amounts detected were 0.412 and 0.156%, showing that the pericarp samples of these two varieties (Giza 86 and Giza 80 respectively) contained amounts of gossypol lower than those estimated in their calyx samples. The highest level of gossypol (0.611%) was recorded for samples Giza 45 as shown in **Table 4**.

*Gossypol content in the bolls*

Marked bolls were picked up 10 days after blooming, and the successive samples were taken at 10 days' intervals. The results concerning the amount of gossypol detected in the different parts during the development of bolls are summarized in **Table 5**.

**Table 3.** Total gossypol content in the different parts of cotton plants, 90 days after planting

| *Cotton varieties* | *Roots (%)* | | *True leaves (%)* | | *Main stems (%)* | | *Leaves of sympodium (%)* | | *Sympodium (%)* | | *Flower Buds (%)* | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | F* | D* | F | D | F | D | F | D | F | D | F | D |
| *Giza 45* | $0.161^{a}$ ±0.01 | $0.974^{a}$ ±0.18 | $0.53^{b}$± 0.08 | $0.323^{b}$ ±0.12 | $0.007^{a}$ ±0.01 | $0.047^{a}$ ±0.04 | $0.049^{a}$ ±0.33 | $0.250^{b}$ ±0.20 | $0.012^{a}$ ±0.17 | $0.066^{a}$ ±0.15 | $0.098^{a}$ ±0.25 | $0.551^{a}$ ±0.23 |
| *Giza 86* | $0.211^{b}$ ±0.18 | $1.248^{c}$ ±0.32 | $0.091^{a}$ ±0.21 | $0.395^{c}$ ±0.19 | $0.030^{b}$ ±0.06 | $0.116^{b}$ ±0.14 | $0.072^{b}$ ±0.32 | $0.315^{c}$ ±0.34 | $0.011^{a}$ ±0.23 | $0.061^{a}$ ±0.21 | $0.117^{b}$ ±0.01 | $0.665^{c}$ ±0.17 |
| *Giza 80* | $0.225^{c}$ ±0.12 | $1.184^{b}$ ±0.20 | $0.051^{a}$ ±0.17 | $0.255^{a}$ ±0.15 | $0.030^{b}$ ±0.03 | $0.134^{c}$ ±0.15 | $0.053^{a}$ ±0.11 | $0.206^{a}$ ±0.23 | $0.014^{a}$ ±0.21 | $0.053^{a}$ ±0.20 | $0.126^{c}$ ±0.03 | $0.638^{b}$ ±0.13 |

F*: Fresh weight D*: Dry weight
The data in the table are the means ± SE. Different letters in the same column indicate significant differences at $P<0.05$.

**Table 4.** Total gossypol content (%) in the different parts of flowers of the cotton plants

| *Gossypol content in Parts of flowers (%)* | *Cotton varieties* | | | | | |
|---|---|---|---|---|---|---|
| | Giza 45 | | Giza 86 | | Giza 80 | |
| | F* | D* | F | D | F | D |
| *Receptacle* | $0.028^{a}$±0.13 | $0.102^{b}$±0.01 | $0.042^{a}$±0.22 | $0.163^{c}$±0.15 | $0.023^{a}$±0.18 | $0.077^{a}$±0.23 |
| *Epicalyx* | $0.034^{a}$±0.21 | $0.174^{b}$±0.18 | $0.053^{a}$±0.12 | $0.233^{b}$±0.14 | $0.075^{a}$±0.17 | $0.169^{b}$±0.15 |
| *Calyx* | $0.091^{a}$±0.08 | $0.268^{c}$±0.20 | $0.095^{a}$±0.08 | $0.313^{c}$±0.12 | $0.058^{a}$±0.20 | $0.187^{b}$±0.11 |
| *Pericarp* | $0.163^{c}$±0.20 | $0.611^{e}$±0.15 | $0.130^{b}$±0.21 | $0.412^{d}$±0.18 | $0.041^{a}$±0.11 | $0.156^{c}$±0.24 |
| *Seed pulp* | ------ | ----- | ------ | ------ | ------ | ------ |

F*: Fresh weight D*: Dry weight
The data in the table are the means ± SE. Different letters in the same row indicate significant differences at P<0.05.

The percentage content of gossypol estimated in the receptacle samples taken from Giza 86 and Giza 80 tended to decrease gradually after the 30 days after blooming. As for the variety of Giza 45, the gossypol level was almost constant during this period. While epicalyx samples, the amounts of gossypol increased slightly 10 days after blooming and then kept on gradually. Calyx samples the percentage content of gossypol tended to increase gradually. It reached the maximum twenty days after blooming in Giza 45 and Giza 86 samples, 10 days after blooming in the sample of Giza 80. Following this period, gossypol levels decreased gradually. The pericarp samples of this part of the boll contained the highest level of the gossypol when compared with the previously mentioned parts. In the meantime, the highest amounts were recorded 10 days after blooming. Ten days later, the amounts dropped considerably. Following this period, it decreased gradually, and varieties could be arranged in following decreased order: Giza 45, Giza 86 and Giza 80.

*Gossypol content in mature seeds*

Samples were taken from the crop, picked up 60 days after blooming. The hulls were removed, and the total gossypol contents were determined in a portion of the kernels. The data showed the same results previously recorded for the different varieties tested before planting. It confirmed that Giza 86 kernels contained the highest level (2.03%). Those of the other varieties, Giza 45 and Giza 80, contained 1.55, and 1.37%, respectively (**Table 6**). Another portion of the kernels was subjected to the techniques for the free and the bound gossypol determinations. The results showed that the amounts of the free gossypol were as much as 3 to 4 times higher than those of the bound gossypol. The values of free gossypol detected in the kernels of Giza 86, Giza 45 and Giza 80 were 1.881, 1.461 and 1.363%, respectively. The respective amounts of bound gossypol were recorded by 0.433, 0.291 and 0.251%. Summing up the two values, free and bound gossypol gave the total amount of gossypol, which was about the same as the one determined chemically (**Table 7**).

**Table 5.** Total gossypol content in the different parts of bolls of the cotton plants

| *Cotton varieties* | *Days after blooming* | *Receptacle (%)* | | *Epicalyx (%)* | | *Calyx (%)* | | *Pericarp (%)* | | *Seed pulp (%)* | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | F* | D* | F | D | F | D | F | D | F | D |
| *Giza 45* | 10 | $0.023^{a}\pm 0.18$ | $0.099^{a}\pm 0.22$ | $0.047^{b}\pm 0.14$ | $0.227^{b}\pm 0.24$ | $0.119^{b}\pm 0.25$ | $0.381^{b}\pm 0.01$ | $0.102^{b}\pm 0.23$ | $0.693^{d}\pm 0.12$ | $0.007^{a}\pm 0.01$ | $0.028^{a}\pm 0.11$ |
| | 20 | $0.019^{a}\pm 0.20$ | $0.095^{a}\pm 0.18$ | $0.038^{a}\pm 0.03$ | $0.202^{a}\pm 0.12$ | $0.136^{c}\pm 0.15$ | $0.408^{c}\pm 0.23$ | $0.036^{a}\pm 0.12$ | $0.261^{c}\pm 0.24$ | $0.003^{a}\pm 0.15$ | $0.017^{a}\pm 0.10$ |
| | 30 | $0.023^{a}\pm 0.12$ | $0.92^{b}\pm 0.14$ | $0.039^{a}\pm 0.23$ | $0.201^{a}\pm 0.11$ | $0.107^{a}\pm 0.18$ | $0.282^{a}\pm 0.11$ | $0.031^{a}\pm 0.20$ | $0.246^{b}\pm 0.12$ | $0.322^{b}\pm 0.18$ | $1.077^{b}\pm 0.15$ |
| | 40 | ----- | ----- | ------ | ------ | ------ | ------ | $0.030^{a}\pm 0.22$ | $0.191^{a}\pm 0.15$ | $0.543^{b}\pm 0.12$ | $1.125^{c}\pm 0.13$ |
| *Giza 86* | 10 | $0.027^{a}\pm 0.03$ | $0.125^{c}\pm 0.18$ | $0.084^{a}\pm 0.01$ | $0.377^{c}\pm 0.19$ | $0.143^{b}\pm 0.14$ | $0.451^{b}\pm 0.18$ | $0.089^{b}\pm 0.03$ | $0.585^{c}\pm 0.24$ | $0.008^{a}\pm 0.01$ | $0.027^{a}\pm 0.02$ |
| | 20 | $0.033^{a}\pm 0.01$ | $0.118^{b}\pm 0.98$ | $0.065^{a}\pm 0.20$ | $0.323^{b}\pm 0.22$ | $0.155^{c}\pm 0.20$ | $0.496^{c}\pm 0.20$ | $0.044^{a}\pm 0.02$ | $0.262^{b}\pm 0.18$ | $0.009^{a}\pm 0.02$ | $0.027^{a}\pm 0.03$ |
| | 30 | $0.026^{a}\pm 0.02$ | $0.101^{a}\pm 0.98$ | $0.061^{a}\pm 0.04$ | $0.300^{a}\pm 0.12$ | $0.132^{a}\pm 0.12$ | $0.388^{a}\pm 0.14$ | $0.037^{a}\pm 0.03$ | $0.257^{b}\pm 0.23$ | $0.197^{b}\pm 0.18$ | $0.661^{b}\pm 0.20$ |
| | 40 | ------ | ------- | ------- | ------- | ------- | ------- | $0.026^{a}\pm 0.01$ | $0.208^{a}\pm 0.20$ | $0.582^{c}\pm 0.22$ | $1.242^{c}\pm 0.19$ |
| *Giza 80* | 10 | $0.020^{a}\pm 0.01$ | $0.118^{b}\pm 0.12$ | $0.036^{a}\pm 0.01$ | $0.233^{c}\pm 0.22$ | $0.081^{a}\pm 0.02$ | $0.364^{b}\pm 0.20$ | $0.047^{b}\pm 0.11$ | $0.282^{c}\pm 0.23$ | $0.007^{a}\pm 0.01$ | $0.035^{a}\pm 0.03$ |
| | 20 | $0.019^{a}\pm 0.02$ | $0.077^{a}\pm 0.03$ | $0.046^{a}\pm 0.11$ | $0.213^{b}\pm 0.18$ | $0.098^{b}\pm 0.01$ | $0.355^{b}\pm 0.19$ | $0.030^{a}\pm 0.01$ | $0.226^{b}\pm 0.18$ | $0.014^{b}\pm 0.03$ | $0.048^{b}\pm 0.02$ |
| | 30 | $0.011^{a}\pm 0.01$ | $0.071^{a}\pm 0.02$ | $0.042^{a}\pm 0.03$ | $0.201^{a}\pm 0.12$ | $0.096^{b}\pm 0.03$ | $0.296^{a}\pm 0.21$ | $0.022^{a}\pm 0.03$ | $0.139^{a}\pm 0.20$ | $0.188^{c}\pm 0.18$ | $0.777^{c}\pm 0.23$ |
| | 40 | ------ | ------- | ------- | ------- | ------- | ------- | $0.021^{a}\pm 0.02$ | $0.130^{a}\pm 0.15$ | $0.555^{d}\pm 0.14$ | $1.050^{d}\pm 0.20$ |

F*: Fresh weight D*: Dry weight
The data in the table are the means ± SE. Different letters in the same column indicate significant differences at P<0.05.

**Table 6.** Total gossypol content in mature cotton-seed kernels

| *Cotton varieties* | *Air dried samples (%)* | *On dry weight basis (%)* |
|---|---|---|
| *Giza 45* | 1.46$^{b}$ ± 0.18 | 1.55$^{b}$ ± 0.17 |
| *Giza 86* | 1.94$^{c}$ ± 0.24 | 2.03$^{c}$ ± 0.20 |
| *Giza 80* | 1.29$^{a}$ ± 0.11 | 1.37$^{a}$ ± 0.12 |

The data in the table are the means ± SE. Different letters in the same column indicate significant differences at P<0.05.

**Table 7.** The percentage content of free and bound gossypol in mature cottonseed kernels

| *Cotton varieties* | *Gossypol content (%)* | | |
|---|---|---|---|
| | *Free* | *Bound* | *Total* |
| *Giza 45* | 1.170$^{b}$ ± 0.21 | 0.291$^{b}$ ± 0.19 | 1.461$^{b}$ ± 0.24 |
| *Giza 86* | 1.448$^{c}$ ± 0.23 | 0.433$^{c}$ ± 0.22 | 1.881$^{c}$ ± 0.20 |
| *Giza 80* | 1.112$^{a}$ ± 0.21 | 0.251$^{a}$ ± 0.12 | 1.363$^{a}$ ± 0.18 |

The data in the table are the means ± SE. Different letters in the same column indicate significant differences at P<0.05.

*Rate of infestation of cotton thrips*

Cotton seedling inspected led to the finding that infestation with cotton thrips, *Thrips tabaci*, started three weeks after planting (April 19th). The highest rates of infestation with the thrips were recorded where the seedlings were inspected by the 4th week. A total number of cotton thrips (adults and nymphs) in the data are presented in **Table 8**. Samples taken 3 weeks after planting showed that the higher numbers of thrips were found on Giza 80 (131) and Giza 45 (113) seedlings, respectively. The lower numbers, 42, were found on Giza 86 seedlings. The highest rate of infestation was recorded on the seedlings 4 weeks after planting. The recorded numbers of insects on 20 seedlings were 498, 369 and 284 for Giza 80, Giza 45 and Giza 86, respectively. Inspecting cotton seedlings 5 weeks after planting showed the number of cotton thrips dropped to 53, 38 and 24 on the Giza 80, Giza 45 and Giza 86 seedlings, respectively. One week later the respective counts were 23, 21 and 12 on the Giza 45, Giza 80 and Giza 86 seedlings, respectively. By the seventh week after planting, all the seedlings were free from any pest. Statistical analysis of the data obtained throughout the infestation period (4 weeks) revealed that there were no significant differences between the different varieties of cotton seedlings concerning the numbers of cotton thrips infesting the seedlings during this early stage of growth. Further, the statistical analysis showed that there were significant differences between varieties of seedlings, save for between Giza 86. The different varieties of cotton could be arranged in the following descending order concerning their susceptibility to infestation: Giza 80, Giza 45 and Giza 86.

*Rate of infestation of pink bollworm*

The data presented in **Table 9** showed that the percentage rate of infestation with pink bollworm, *Pectinophora gossypiella* ranged from 14 to 24% when the bolls were first inspected on the 3rd of August. The lowest percentage was that of Giza 86 and the higher was that of Giza 80. Successive samples taken through the following 4 weeks showed about the same trend and the same rate of infestation. Following this period, by the 5th week, the rate of infestation increased suddenly in all the varieties which could be arranged in the following descending order: Giza 80 (81%), Giza 45 (71%) and Giza 86 (61%). Statistical analysis of the data obtained revealed that there were no significant differences between the different varieties of cotton seedlings concerning the rate of infestation.

**Table 8.** Number of thrips on 20 seedlings of certain cotton varieties of indicated weeks after planting

| *Cotton varieties* | *Number of thrips* | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 3rd week (19 April) | | | 4th week (26 April) | | | 5th week (3 May) | | | 6th week (11 May) | | |
| | N* | A* | Total | N | A | Total | N | A | Total | N | A | Total |
| *Giza 45* | 9$^{a}$ | 104$^{b}$ | 113$^{b}$ | 365$^{b}$ | 4$^{a}$ | 369$^{b}$ | 3$^{a}$ | 21$^{a}$ | 24$^{a}$ | 7$^{a}$ | 16$^{b}$ | 23$^{b}$ |
| *Giza 86* | 8$^{a}$ | 36$^{a}$ | 44$^{a}$ | 277$^{a}$ | 7$^{a}$ | 284$^{a}$ | 8$^{a}$ | 30$^{b}$ | 38$^{b}$ | 5$^{a}$ | 7$^{a}$ | 12$^{a}$ |
| *Giza 80* | 17$^{b}$ | 114$^{c}$ | 131$^{c}$ | 494$^{c}$ | 4$^{a}$ | 498$^{c}$ | 17$^{b}$ | 36$^{b}$ | 53$^{c}$ | 12$^{b}$ | 9$^{a}$ | 21$^{b}$ |

N*: Nymphs A*: Adults

Different letters in the same column indicate significant differences at P<0.05.

**Table 9.** Number of infested bolls in tested cotton plants

| *Cotton varieties* | *Infested bolls at indicated dates (%)* | | | | | |
|---|---|---|---|---|---|---|
| | 3 August | 10 August | 17 August | 24 August | 31 August | 7 September |
| *Giza 45* | 24$^{b}$ | 14$^{b}$ | 21$^{c}$ | 24$^{b}$ | 26$^{b}$ | 71$^{b}$ |
| *Giza 86* | 14$^{a}$ | 11$^{b}$ | 9$^{a}$ | 19$^{a}$ | 19$^{a}$ | 61$^{a}$ |
| *Giza 80* | 16$^{a}$ | 4$^{a}$ | 14$^{b}$ | 19$^{a}$ | 16$^{a}$ | 81$^{c}$ |

Different letters in the same column indicate significant differences at P<0.05.

*Rate of infestation of pink bollworm larvae*

The results showed lower numbers of larvae found inside the infested bolls (**Table 10**). The higher number 26, 13.5 and 11 in 100 infested bolls were recorded by September 7th. They were found in the bolls Giza 86, Giza 80 and Giza 45, respectively. The population density and the population size of the pink bollworms infesting different varieties of cotton plants during the season were calculated, and the whole results were tabulated in **Table 11**. It revealed that the population sizes ranged from 3.01 to 3.63. Additionally, statistical analysis showed that there were no significant differences between the population densities of this pest infesting the bolls of the different cotton plant varieties.

## Discussion

Our study demonstrated that high level gossypol was primarily found in the seeds of cotton plants, while other organs had a rather weaker level of gossypol. However, there was a positive correlation between the high content of gossypol in cottonseeds and the amount of gossypol detected in the other parts of the cotton plants. These results are in agreement with the previous findings [15-17], finding that gossypol is a polyphenolic aldehyde which constitutes 20–40% of the pigment glands weight and accounts for 0.4–1.7% of the whole cottonseed kernel. All the cotton varieties showed significant differences in the level of gossypol; Giza 86 contained highest level of gossypol followed by Giza 45 whereas Giza 80 had lowest. Our results are in conformity with those obtained by previous studies [18,19]. They reported that cotton plant especially the seeds, are a rich source of the gossypol. They found gossypol proportion ranges from 33.8 to 47.0% in the seeds of upland variety (*Gossypium hirsutum*) and from 24.9 to 68.9% of the seeds of *Gossypium barbadense*.

**Table 10.** Number of pink bollworm larvae per 100 infested green bolls

| *Cotton varieties* | *Number of pink bollworms at indicated dates* | | | | | |
|---|---|---|---|---|---|---|
| | 3 August | 10 August | 17 August | 24 August | 31 August | 7 September |
| *Giza 45* | 25 | 7.5 | 5 | 2.5 | 2.5 | 10 |
| *Giza 86* | 10 | 7.5 | 7.5 | 2.5 | 0.0 | 25 |
| *Giza 80* | 7.5 | 5 | 7.5 | 0.0 | 0.0 | 10 |

**Table 11.** Population density of pink bollworm larvae infesting cotton plants

| *Cotton varieties* | *Population density at indicated dates* | | | | | | *Population size* |
|---|---|---|---|---|---|---|---|
| | 3 August | 10 August | 17 August | 24 August | 31 August | 7 September | |
| *Giza 45* | 1.38[c] | 0.98[c] | 0.63[b] | 0.42[c] | 0.49[b] | 0.85[a] | 3.63[c] |
| *Giza 86* | 0.60[a] | 0.66[a] | 0.53[a] | 0.35[b] | 0.66[c] | 1.67[c] | 3.30[b] |
| *Giza 80* | 0.76[b] | 0.86[b] | 0.61[b] | 0.26[a] | 0.36[a] | 1.07[b] | 3.01[a] |

Different letters in the same column indicate significant differences at $P<0.05$.

In field tests during the season 2019, high gossypol level correlated with lessened insect damage, probably due to toxicity of gossypol to *Thrips tabaci* (adult and nymph) and *Pectinophora gossypiella*, larvae that infested cotton bolls and other cotton insects, suggesting a resistance of cotton plants based on gossypol levels that is promising against these pests. The different varieties of cotton could be arranged in the following descending order concerning their susceptibility to infestation: Giza 80, Giza 45 and Giza 86. The results reported that the differences in the amount of gossypol between varieties experimental of cotton plants where Giza 86 contained the highest value while Giza 80 contained the lowest value. Several studies have been conducted to evaluate the cotton plant variety of *Gossypium* spp. against many insect species [20,21], reporting that *Gossypium* spp., resistant to pink bollworm [22,23], certain leaf-feeding Lepidoptera [24,25] and insect sucking pests of cotton [26,27].

## Conclusion

Cotton thrips and pink bollworm are the most destructive pests on cotton and numerous agriculture crops. The control of these pests mainly relies on the application of chemical insecticides in Egypt. However, our results demonstrate that gossypol has potential for controlling these pests. These results are helpful for cultivating cotton varieties with natural defenses like gossypol that are the promising varieties which can be used to develop more environmentally friendly control strategies, such as plant resistance.

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