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# MANAGEMENT OF PEANUT CERCOSPORA LEAF SPOT USING RESISTANT CULTIVARS AND INDUCER RESISTANCE CHEMICALS

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**ABSTRACT:** Six cultivars of peanut *i.e.* Giza 5, Giza 6, R92, Ismailia 1, Gregory and Virginia. were evaluated for their susceptibility to the natural infection by Cercospora leaf spot during two successive growing seasons of 2016 and 2017. Generally, Ismailia 1, followed by R92 were the most resistant tested cvs., for the disease and produced the highest pod yield in both seasons. However, Gregory, followed by Virginia were the most susceptible ones recording the highest percentages of disease incidence and severity in tested both seasons. Induction of disease resistance was carried out using salicylic acid, nicotinic acid, butyric acid, ascorbic acid and bion which were applied at three concentrations, *i.e.* 2, 4 and 8 mM. The obtained results proved that bion and salicylic acid at 8 mM followed by ascorbic acid at the same conc., were the most effective inducers for minimizing disease incidence as well as disease severity and consequently increased the produced pod yield during both investigated seasons . Four plant growth regulators namely indole butaric acid (IBA), naphthalene acetic acid (NAA), gibberellic acid (GA3) and baclobtrazole were applied at three concentrations, *i.e.* 50, 100 and 200 ppm proved that naphthalene acetic acid and indole butaric acid at 200 ppm were the most effective inducers for minimizing disease incidence as well as severity and consequently increased the prodced pod yield during both investigated seasons. Four sulfate mineral salts *i.e.*, copper sulphate ($CuSo_4$), zinc sulphate ($ZnSo_4$), magnesium sulphate ($MgSo_4$) and manganese sulphate ($MnSo_4$) at three concentrations, *i.e.* 1, 2 and 4mM, were tested. Copper sulphate followed by magnesium sulphate gave the lowest percentages of disease incidene and severity without significant differences in between and consequently increased the produced pod yield. The effect of five silicate salts, *i.e.* calcium silicate ($CaSio_3$), magnesium silicate ($MgSio_3$), potassium silicate ($K_2Sio_3$) aluminum silicate ($Al_2Sio_3$) and seal- matreax (commercial compound) at four concentrations, *i.e.* 200, 400, 800 and1600 ppm were evaluated. Generally, calcium silicate followed by potassium silicate gave the lowest percentage of disease incidence and severity and increased the produced pod yield.Thus, there is a correlation between induced resistance and some biochemical changes in peanut leaf tissues including phenol contents (free, conjugate and total phenols) and oxidative enzyme activities, *i.e.*, peroxidase and polyphenoloxidase.

**Key words:** Peanut, Cercospora leaf spot, cultivar reactions, induction, disease resistance, chemical inducers, plant growth regulators, sulfate salts, silicate salts, biochemical changes.

## INTRODUCTION

Peanut (*Arachis hypogaea* L.) is one of the major sources of protein and oil in the world. It is, cultivated on 24 million hectares in over than100 countries **(FAO, 2011)**. In Egypt peanut is one of the most crops for both exportation and locally consumption **(Hilal *et al*., 1994)**. Peanut cultivated area in Egypt was around 156044 fad., produced around 3243848 ardab as mentioned by the yearly book 2017 of Economics and Statistics of the Economic Affairs Sectors,

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Agriculture Ministry in Egypt. Leaf spots caused by the fungi *Cercospora arachidicola* S. Hori (telemorph = *Mycosphaerella arachidis* Deighton) and *Cercosporidium personatum* (Berk. and M.A. Curtis) Deighton (telemorph = *Mycosphaerella berkeleyi* Jenk.) are the most important foliar diseases of peanut worldwide (**Lijun *et al.,* 1999; Maninderpal, 2011**). Yield losses near 50% may result from failure to control early leaf spot (**Lijun *et al.,* 1999**). Yield losses in peanut cultivars are produced by diverse causes, mainly Cercospora leaf spots and the recognition of peanut genotypes being tolerant to them and simultaneously having higher production potentials should benefit growers and breeders to carry out the proper cultivar for sowing or for further breeding **(Gaikpa *et al.*, 2015)**. The cultivation of resistant and tolerant peanut cvs., does not only eliminate the crop losses caused by diseases, but also reduce costs related to fungicide spray and other control methods. The high expense associated with 8 to 10 fungicide sprayings during the crop cycle, is economically not feasible but serve as a challenge to develop resistance/tolerant cvs., against foliar diseases such as early leaf spot (*C. arachidicola*) and late leaf spot (*C. personatum*) **(Alderman and Nutter, 1994; Ambang *et al*., 2011).**

Wherever fungicidal applications cause hazards to human health and increase environmental pollution **(Garcia, 1993)** therefore, alternatives, eco-friendly approaches for control of plant diseases are needed including induced resistance (**Mandal *et al.,* 2009).** Induced disease resistance can be defined as the process of active resistance dependent on the host plants physical or chemical barriers activated by biotic or abiotic agents, **(Meena *et al.*, 2001; Walters *et al.,* 2007)**. These responses include phytoalexin accumulation, phenols, lignifications and activation of many enzymes such as peroxidase, polyphenoloxidase, catalase and chitinase (**Boller, 1991; Meena *et al.*, 2001; Mahmoud *et al.,* 2006; Hussein, 2011; Abdel Aal *et al.,* 2012; Ibrahim *et al*., 2013).**

The present investigation have been conducted to investigate the effectiveness of cultivar reactions and environmentally safe chemicals for management of peanut Cercospora leaf spot.

# MATERIALS AND METHODS

## Varietal Susceptibility to Cercospora Leaf Spots

This experiment was carried out under sprinkler irrigation system during the first week of May in a field at Ismailia Experimental Station (ARC), Ismailia Governorate Egypt . Six peanut cvs. *i.e*. Giza 5, Giza 6, R92, Ismailia 1, Gregory and Virginia were kindly obtained from the Oil Crops Research Department, Field Crops Research Institute, ARC. The six cvs were evaluated for their susceptibility to Cercospora leaf spot **(Abdel Aal *et al.,* 2019)** during two successive growing seasons (2016 and 2017).

## Disease Assessment

Percentages of disease incidence were calculated as follows:

Disease incidence (%)=

$$\frac{\text{Number of infected leaves in the sample}}{\text{Total number of leaves in the sample}} \times 100$$

Disease severity was assessed, three months after planting and before harvesting. One hundred leaves from each plot were randomizly sampled to determine disease severity using (0-8) scale adopted by **Ibrahim *et al.* (2013)**.

Pod yield was calculated as follows: Plants in individual plots were dug and inverted based on optimum maturity index. Pods were air-dried for three days then weighed as kg/plot (10.5m$^2$) at the end of the experimental periods and the expected pod weight (Ton/faddan), was calculated.

## Induction of Cercospora Leaf Spot By Different Inducer Resistance Chemicals (IRCS)

The effectiveness of different chemical materials and concentrations were applied for induction of disease resistance against natural heavily infected by Cercospora leaf spot. The investigation was conducted in field conditions under sprinkler irrigation system in Ismailia Experimental Station (ARC), Ismailia Governorate, Egypt during 2016 and 2017 growing seasons. Different concentrations of inducers for each experiment were used as a foliar spray at 20 and

40 days after sowing to evaluate their effectiveness in manging Cercosporal leaf spot of peanut. Disease assessment and pod yield were recorded as previously mentioned. Five chemical inducers *e.g.* salicylic acid, nicotinic acid, butyric acid, ascorbic acid and bion were applied at three concentrations, *i.e.* 2, 4 and 8 mM for each. Four plant growth regulators namely indole butaric acid (IBA), naphthalene acetic acid (NAA), gibberellic acid (GA3) and baclobtrazole each was applied at three concentrations, *i.e.* 50, 100 and 200 ppm and evaluated. Four sulfate mineral salts *i.e.*, copper sulphate ($CuSo_4$), zinc sulphate ($ZnSo_4$), magnesium sulphate ($MgSo_4$) and manganese sulphate ($MnSo_4$) each at three concentrations *i.e.* 1, 2 and 4mM, were tested. Five silicate mineral salts, *i.e.* calcium silicate ($CaSio_3$), magnesium silicate ($MgSio_3$), potassium silicate ($K_2Sio_3$) aluminum silicate ($Al_2Sio_3$) and Seal-Matreax (commercial compound) at four concentrations,for each *i.e.* 200, 400, 800 and1600 ppm were also evaluated.

Biochemical changes associated with induced resistance were evaluated to identify the probable mechanisms by which the tested chemical agents act as inducer disease resistance to Cercospora leaf spot. Thus, the activity of oxidative enzymes, *i.e.* peroxidase and polyphenol-oxidase as well as phenolic compounds were determined in the leaves of treated and untreated peanut plants with the indcer resistance chemicals.

Methods described by **Tuzum *et al.* (1989)** were followed for extraction of oxidative enzyme activities, *i.e* peroxidase and polyphenol oxidase. Peroxidase assay (based on oxidation of pyrogallol to purpurogallin in the presence of $H_2O_2$) was determined according to the method described by **Allam and Hollis (1972)**. The activity of polyphenoloxidase (PPO) was measured as mentioned by **Matta and Dimond (1963)**. Conjugated phenols, free and total ones using Folin and Ciocatalteus reagent were determined as described by **Snell and Snell (1953)**.

The obtained Results were statistically analyzed by analysis of variance (ANOVA) using **MSTAT-C (1991).** The least significant difference (LSD) test (0.05) was used to find out the significance of the means of various treatments **(Gomez and Gomez, 1984).**

# RESULTS AND DISCUSSION

## Varietal Susceptibility to Cercospora Leaf Spots

Six cvs. of peanut (Table 1) were evaluated for their susceptibility to Cercospora leaf spot during 2016 and 2017 seasons under natural infection conditions, and sprinkler irrigation at Ismailia Governorate. Results presented show that, Ismailia 1, was the most resistant cv. where significulty recorded the lowest value for each of disease incidence and severity (18.3 and 10.7%) and (15.0 and 8.9%) in both seasons, respectively. However Gregory, followed by Virginia were the most susceptible cvs., recording the highest percentage for each of disease incidence and severity in both seasons. Pod production of peanut cultivars (Table 1) showed that R92 yielded the highest in both seasons, being (4.63 and 4.75 Kg/plot) respectively and (1.852 ton/ fad., and 1.900 Ton/fad., respectively). However, cv. Giza5 produced the lowest pod yield in both growing seasons, being (2.85 and 3.12 Kg/plot) and (1.140 and 1.248 Ton/fad.) respectively.

The difference among the tested cvs., in their resistance and susceptibility to the disese, might be due to a given pathogen exhibited different reactions by the host due to their genome structures that master various biological and physiological behaviors **(Knauft and Gorbet, 1990).** The presence of genetic variability in crop plants have been described as essential in plant breeding where it encourage selection **(Izge *et al.*, 2005)**.

## Induction of Resistance to Cercospora Leaf Spot by Different Chemicals

The effectiveness of different materials used for induction of disease resistance to peanut Cercospora leaf spot was investigated under sprinkler irrigation system under field conditions at Ismailia Experimental Station (ARC), Ismailia Governorate, during 2016 and 2017, growing seasons.

### Induction by inducer resistnce chemicals (IRCS)

Five IRCS, *e.g.* salicylic acid, nicotinic acid, butyric acid, ascorbic acid and bion were applied at

**Table 1. Susceptibility of six peanut cvs. to Cercospora leaf spot and pod yield**

| Peanut cvs. | Disease incidence and severity (%) | | | | Peanut pod yield | | | |
|---|---|---|---|---|---|---|---|---|
| | 2016 | | 2017 | | Kg/plot (10.5m$^2$) | | Expected pod (ton/fad.) | |
| | DI (%) | DS (%) | DI (%) | DS (%) | 2016 | 2017 | 2016 | 2017 |
| **Giza 5** | 39.3 | 23.8 | 40.0 | 20.5 | 2.85 | 3.12 | 1.14 | 1.25 |
| **Giza 6** | 38.3 | 17.4 | 36.0 | 16.9 | 3.79 | 3.91 | 1.52 | 1.56 |
| **R92** | 23 | 12.6 | 20.0 | 10.2 | 4.63 | 4.75 | 1.85 | 1.90 |
| **Ismailia 1** | 18.3 | 10.7 | 15.0 | 8.9 | 4.03 | 4.11 | 1.61 | 1.64 |
| **Gregory** | 58.3 | 34.9 | 55.0 | 31.4 | 3.45 | 3.63 | 1.38 | 1.45 |
| **Virginia** | 52 | 29.1 | 49.0 | 26.8 | 3.65 | 3.97 | 1.46 | 1.59 |
| **LSD 5%** | 5.3 | 1.5 | 4.9 | 1.4 | 0.78 | 0.58 | 0.31 | 0.23 |

DI = Disease incidence DS = Disease severity

three concentrations, *i.e.* 2, 4 and 8 mM. Results presented in Tables 2 and 3 indicate that all tested chemical inducers and their concentrations significantly decreased incidence and severity of peanut Cerospora leaf spot and consequently increased the prodced pod yield during both investigated seasons (2016 and 2017) if compared with the control. The obtained results proved that bion and salicylic acid, both at 8 mM, followed by ascorbic acid at 8 mM were the most effective for minimizing incidence and, severity of the disease and, consequently increased the prodced pod yield(Kg/plot) during both investigated seasons as the obtained yields for both seasons were (4.05 and 3.91), (3.85 and 3.92) and (3.76 and 3.61) respectively. However, butyric acid as well as nicotinic acid at 2mM recorded the lowest reduction of both incidence and severity of the disease in both tested seasons.

The obtained results might be taken as a further support that such acids at special concentrations were clearly effective in enhancing yield production when applied as foliar spray. The improved performance of such acids might be one of the important agents implicated with the disease reaction leading to lower values of peanut Cercospora leaf spot. Induced disease resistance can be defined as the process of active resistance dependent on the host plants physical and/or chemical barriers activated by biotic or abiotic agents, **(Meena *et al.*, 2001; Walters *et al.*, 2007)**. Some compounds, *e.g.,* nicotinic acid, salicylic acid (SA), butyric acid have been shown to induce resistance in plants (**Mahmoud *et al.*, 2006; Mandal *et al.*, 2009; Hussein, 2011; Khalifa *et al.*, 2011; Abdel Aal *et al.*, 2012; Ibrahim *et al.*, 2013**). They added that, induction of systemic resistance might sensitizes the plant to respond rapidly after infection. These responses include one or more of phytoalexin accumulation, phenols, lignifications and activation of many enzymes such as peroxidase, polyphenoloxidase, catalase and chitinase. Thus application of such antioxidants proved suffiecient protection against peanut Cercospora leaf spot and most of such antioxidants increased pod yield.

There was significant effect of all examined IRCS at all their tested concentrations in reducing incidence and severity of the disease as well as consequently increased the total pod yield. Bion at the three concentrations studied followed by salicylic acid were the most effective ones during the two growing seasons in 2016 and 2017. Bion, activates various defense responses ranging from hypersensitive cell death (HR) of pathogen-attacked cells up to accumulation of reactive oxygen intermediates (ROI) like $H_2O_2$ and the expression of a number of pathogenesis-related genes (PR) genes, which together might control microbial pathogens **(Sauerborn *et al.*, 2001)**.

The effect of the different investigated IRCS on various biochemical changes, *i.e.* phenol contents and oxidative enzymes (peroxidase and polyphenol-oxidase) activities in peanut leaf plants, was studied.

Table 2. Effect of foliar spraying with some inducer resistance chemicals on incidence and severity of Cercospora leaf spot under field conditions during two successive seasons (2016 and 2017)

| IRCS | Conc. (mM) | Disease incidence and severity (%) | | | |
|---|---|---|---|---|---|
| | | 2016 | | 2017 | |
| | | DI | DS | DI | DS |
| **Salicylic acid** | **2** | 30.15 | 13.69 | 26.42 | 11.27 |
| | **4** | 26.27 | 11.31 | 23.19 | 10.09 |
| | **8** | 20.49 | 7.86 | 17.64 | 7.12 |
| **Nicotinic acid** | **2** | 38.76 | 20.13 | 37.68 | 21.45 |
| | **4** | 35.19 | 17.51 | 34.93 | 19.38 |
| | **8** | 26.63 | 13.75 | 29.88 | 16.81 |
| **Butyric acid** | **2** | 43.19 | 29.14 | 46.27 | 31.87 |
| | **4** | 39.42 | 24.68 | 41.90 | 29.15 |
| | **8** | 33.16 | 19.57 | 32.75 | 21.06 |
| **Ascorbic acid** | **2** | 28.71 | 16.52 | 27.34 | 15.75 |
| | **4** | 26.80 | 14.38 | 25.50 | 13.41 |
| | **8** | 20.13 | 9.22 | 21.76 | 10.73 |
| **Bion** | **2** | 25.84 | 10.13 | 26.92 | 12.95 |
| | **4** | 18.33 | 9.46 | 24.67 | 10.31 |
| | **8** | 16.57 | 6.97 | 19.31 | 7.59 |
| **Control** | **-** | 68.69 | 46.28 | 73.14 | 52.37 |
| **LSD 0.05% for** | | | | | |
| **Chemical inducers (A)** | | 0.74 | 0.46 | 0.94 | 0.60 |
| **Concentrations (B)** | | 0.52 | 0.33 | 0.67 | 0.43 |
| **Interactions (A) × ( B)** | | 1.27 | 0.80 | 1.63 | 1.05 |

DI= Disease incidence DS= Disease severity

Table 3. Effect of foliar spraying with some chemical inducers on peanut pod yield during two successive seasons (2016 and 2017)

| IRCS | Conc. (mM) | Peanut pod yield | | | |
|---|---|---|---|---|---|
| | | Kg/ plot (10.5m$^2$) | | Expected pod (Ton/fad.) | |
| | | 2016 | 2017 | 2016 | 2017 |
| **Salicylic acid** | **2** | 3.20 | 3.37 | 1.280 | 1.348 |
| | **4** | 3.46 | 3.53 | 1.384 | 1.412 |
| | **8** | 3.85 | 3.92 | 1.540 | 1.568 |
| **Nicotinic acid** | **2** | 2.91 | 2.74 | 1.164 | 1.096 |
| | **4** | 3.08 | 2.96 | 1.232 | 1.184 |
| | **8** | 3.17 | 3.07 | 1.268 | 1.228 |
| **Butyric acid** | **2** | 2.81 | 2.74 | 1.124 | 1.096 |
| | **4** | 2.97 | 2.87 | 1.188 | 1.148 |
| | **8** | 3.14 | 3.03 | 1.256 | 1.212 |
| **Ascorbic acid** | **2** | 3.25 | 3.38 | 1.300 | 1.352 |
| | **4** | 3.49 | 3.55 | 1.396 | 1.420 |
| | **8** | 3.76 | 3.61 | 1.504 | 1.444 |
| **Bion** | **2** | 3.81 | 3.72 | 1.524 | 1.488 |
| | **4** | 3.88 | 3.83 | 1.552 | 1.532 |
| | **8** | 4.05 | 3.91 | 1.620 | 1.564 |
| **Control** | **-** | 2.63 | 2.38 | 1.052 | 0.952 |
| **LSD 0.05% for** | | | | | |
| **Chemical inducers (A)** | | 0.34 | 0.04 | 0.02 | 0.03 |
| **Concentrations (B)** | | 0.24 | 0.03 | 0.01 | 0.02 |
| **Interactions (A) × ( B)** | | 0.59 | 0.07 | 0.04 | 0.05 |

Results presented in Fig. 1 indicate that phenol contents including the free, conjugated and total phenols were obviously higher in plants treated with any of IRCS than the untreated control during the two growing seasons. Peanut plants treated with bion and salicylic acid recorded the highest phenol contents. Meanwhile, butyric acid gave lower values compared to the other treatments. Increasing the concentration of IRCS led to an increase of phenol contents in peanut leaves during the two growing seasons.

All tested IRCS increased the activity of oxidative-reductive enzymes, *i.e*. peroxidase (PO) and polyphenoloxidase (PPO) in peanut leaves compared to untreated control during the two growing seasons (Fig. 2). The highest activity of PO was shown when salicylic acid was applied followed by bion. The same trend was recorded for PPO activity. Results also showed that increasing the concentration of any of the IRCS was accompanied by an increase in enzyme activities during the two successive seasons.

Salicylic acid treatment might led to a reduction of Cercospora leaf spot by increasing activities of many classes of PR-proteins **(Ata *et al.,* 2008; Nighat *et al.,* 2011).** Effect of salicylic acid (SA) in induced resistance of peanut leaf spot might be also due to the increase of oxidative-reductive enzymes activity and phenol compounds content. This is also in agreement with **Mahmoud *et al.* (2006)**, who stated that, there is asignificant increase in the total peroxidase activity after treatment with salicylic acid.They added that salicylic acid in generation of the oxidative burst in incompatible interactions by inducing a rapid transient generation of $O_2^{-}$ which is responsible for regulation of peroxidase activity. Effect of nicotinic acid and butyric acid as inducers might be also due to the increase of oxidative-reductive enzymes activity and content of phenol compounds that were clear in increasing of enzymes activity and phenol content and this was in agreement with **Meena *et al.* (2001), Khalifa *et al*. (2007), Khalifa *et al*. (2011), Abdel Aal *et al.* (2012) and Ibrahim *et al.* (2013)**.

**Mahmoud *et al.* (2006)** found that, the IRCS showed changes in the activity of oxidative-reductive enzymes and phenolic contents in primordial pods of peanut. This biochemical changes became a marker to induce resistance (**Cadena-Gomez and Nicholson, 1987; Edreva, 1989; Reuveni *et al.,* 1992**). Another possible role for peroxidase is the oxidative cross~ 1inking of pre-existing hydroxyproline-rich structural proteins in the cell wall, making the cell wall more resistant to degradation by microbial enzymes (**Bradley *et al.,* 1992**) as well as generation of hydrogen peroxide consider an antimicrobial agent (**Peng and Kuc, 1992**). While, phenol compounds play an important role in plant defense such phenols are essential for the biosynthesis of lignin, which consider an important structural component of plant cell walls (**Hahlbrock and Scheel, 1989**).

### Induction as affected by plant growth regulators

Four plant growth regulators were applied and evaluated at three concentrations for each, *i.e.* 50, 100 and 200 ppm. Resuls in Tables 4 and 5 prov that all tested plant growth regulators decreased disease parameters and consequently increased peanut pod yield during both seasons (2016 and 2017) comparing with the control. Both naphthalene acetic acid (NAA) and indole butaric acid (IBA) at 200 ppm were the most effective regulators for promising disease control and consequently increased peanut pod yield during both investigated seasons. However, baclobtrazole at 50 ppm revealed the lowest reduction for each of incidence and severity of the disease in both seasons.

Naphthalene acetic acid (NAA) is widely used in agriculture for various purposes. **Tomlin (2006)** has been shown that NAA greatly increased cellulose fiber formation in plants when paired with another phytohormone called gibberellic acid. Because it is in the auxin family it has also been understood to prevent premature dropping and thinning of fruits from stems. In order to obtain its desired effects it must be applied in concentrations ranging from 20–100 µg/ml **(Navalon *et al*., 1997).**

Phenol contents were obviously higher in plants treated with any of growth regulators as resistance inducers than the untreated control during the two growing seasons (Fig. 3). In this respect peanut plants treated with naphthalene acetic acid (NAA) and indole butaric acid (IBA)

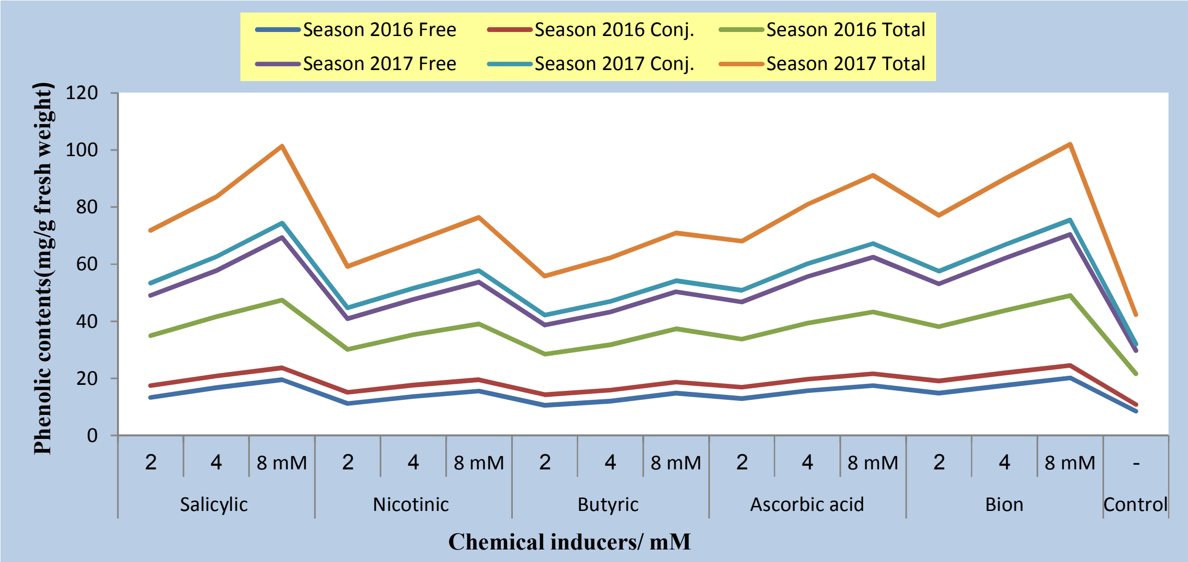


**Fig. 1. Effect of certain IRCS on phenolic contents (mg/g fresh weight) in peanut leaves**

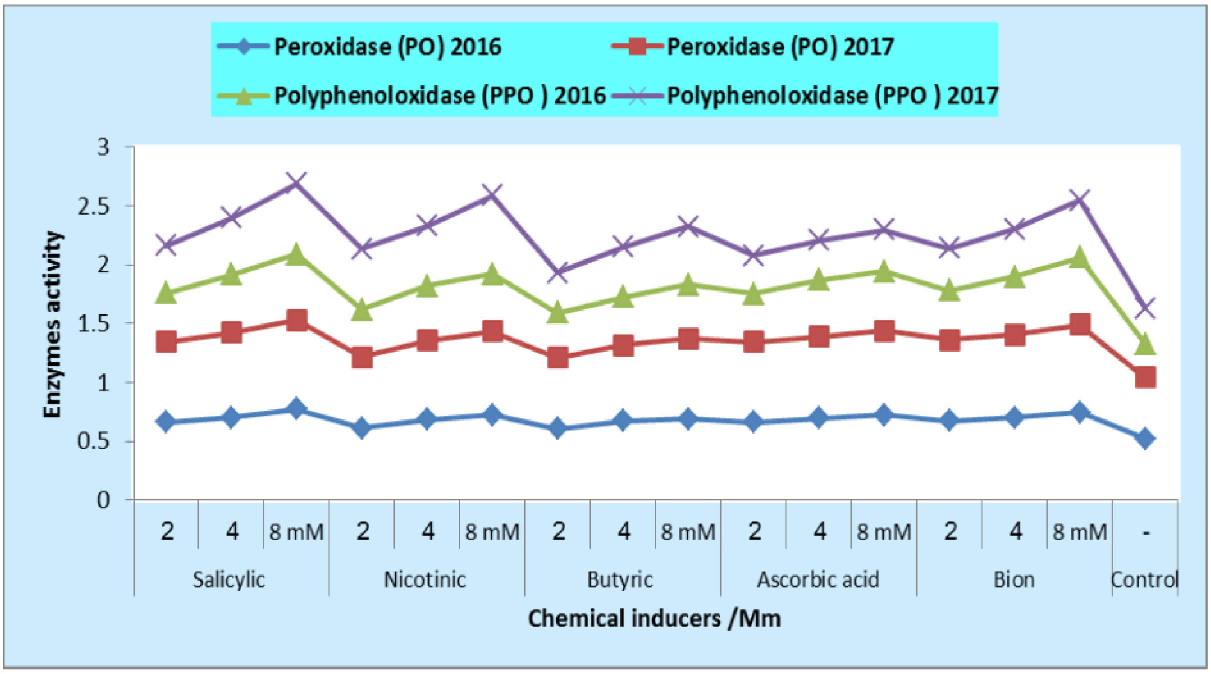


**Fig. 2. Effect of foliar spraying with some IRCS on peroxidase (PO) and polyphenoloxidase (PPO) activity in peanut leaves**

**Table 4. Effect of the foliar spraying with some growth regulators on incidence and severity of the disease during two successive seasons (2016 and 2017)**

| Growth regulator | Conc. (ppm) | Disease incidence and severity (%) | | | |
|---|---|---|---|---|---|
| | | 2016 | | 2017 | |
| | | DI | DS | DI | DS |
| **Indole butaric acid (IBA)** | **50** | 24.76 | 12.15 | 27.49 | 14.78 |
| | **100** | 18.29 | 9.32 | 22.34 | 11.20 |
| | **200** | 12.85 | 7.16 | 17.55 | 9.15 |
| **Naphthalene acetic acid (NAA)** | **50** | 20.63 | 10.24 | 23.67 | 11.53 |
| | **100** | 16.72 | 8.11 | 18.24 | 9.41 |
| | **200** | 10.38 | 6.23 | 12.73 | 7.94 |
| **Gibberellic acid (GA3)** | **50** | 22.66 | 11.82 | 26.44 | 13.07 |
| | **100** | 19.47 | 9.33 | 21.19 | 11.72 |
| | **200** | 13.54 | 6.98 | 15.08 | 8.33 |
| **Baclobtrazole** | **50** | 29.15 | 17.52 | 31.73 | 19.20 |
| | **100** | 24.36 | 13.26 | 26.11 | 16.83 |
| | **200** | 19.03 | 9.80 | 21.70 | 13.71 |
| **Control** | **-** | 56.17 | 38.46 | 62.42 | 41.94 |
| **LSD 0.05% for** | | | | | |
| **Growth regulators (A)** | | 0.44 | 0.40 | 0.35 | 0.41 |
| **Concentration (B)** | | 0.34 | 0.31 | 0.27 | 0.30 |
| **Interactions (A) × ( B)** | | 0.76 | 0.69 | 0.60 | 0.70 |

DI= Disease incidence DS= Disease severity

**Table 5. Effect of foliar spraying with some growth regulators on peanut pod yield during two successive seasons (2016 and 2017)**

| Growth regulator | Conc. (ppm) | Peanut pod yield | | | |
|---|---|---|---|---|---|
| | | Kg/plot (10.5 $m^2$) | | Expected pod (Ton/fad.) | |
| | | 2016 | 2017 | 2016 | 2017 |
| **Indole butaric acid (IBA)** | **50** | 2.79 | 2.65 | 1.116 | 1.060 |
| | **100** | 2.93 | 2.80 | 1.172 | 1.120 |
| | **200** | 3.16 | 3.08 | 1.264 | 1.232 |
| **Naphthalene acetic acid (NAA)** | **50** | 2.84 | 2.77 | 1.136 | 1.108 |
| | **100** | 2.99 | 2.84 | 1.196 | 1.136 |
| | **200** | 3.46 | 3.35 | 1.384 | 1.340 |
| **Gibberellic acid (GA3)** | **50** | 2.63 | 2.54 | 1.052 | 1.016 |
| | **100** | 2.94 | 2.69 | 1.176 | 1.076 |
| | **200** | 3.29 | 2.94 | 1.316 | 1.176 |
| **Baclobtrazole** | **50** | 2.55 | 2.42 | 1.020 | 0.968 |
| | **100** | 2.71 | 2.64 | 1.084 | 1.056 |
| | **200** | 3.06 | 2.84 | 1.224 | 1.136 |
| **Control** | **-** | 2.41 | 2.10 | 0.964 | 0.840 |
| **LSD 0.05% for** | | | | | |
| **Chemical inducers (A)** | | 0.18 | 0.26 | 0.23 | 0.03 |
| **Concentrations (B)** | | 0.14 | 0.20 | 0.18 | 0.02 |
| **Interactions (A) × ( B)** | | 0.31 | 0.45 | 0.40 | 0.05 |

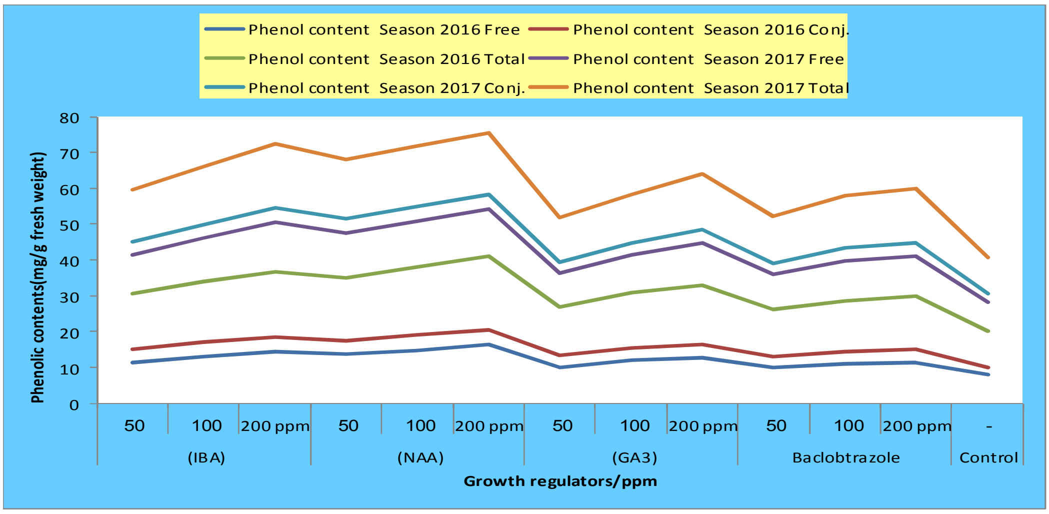


**Fig. 3. Effect of some growth regulators on phenolic contents (mg/g fresh weight) in peanut leaves**

recorded the highest phenol contents in both seasons. However, baclobtrazole and gibberellic acid (GA3) revealed the lowest phenol contents. Results also indicated that, increasing the concentration of plant growth regulators led to an increase in phenols of peanut leaves during the two growing seasons.

Results illustrated in Fig. 4 show that all tested growth regulators increased the activity of oxidative-reductive enzymes in peanut leaves compared to untreated control in both growing seasons. The highest PO and POP activity was shown when naphthalene acetic acid was evaluted followed by indole butyric acid (both at 200 ppm), while gibberellic acid and baclobtrazole at 50 ppm recorded the lowest PO and POP activity compared to other treatments in the two successive seasons.

Naphthalene acetic acid present in the environment undergoes oxidation reactions with hydroxyl radicals and sulphate radicals. In micro propagation of various plants, NAA is typically added to media containing nutrients essential to the plants survival. It is added to help induce root formation in various plant types. It can also be applied by spraying it onto plants and is typical in agricultural use **(Navalon *et al*., 1997).**

Several reports have been published on the use of growth regulators releasing the compound ethiphon for inductions of resistance in plants (**Abd-El-Kareem *et al.,* 2001; Hussein, 2011**). The effect of growth regulators in reducing the disease might be due to its effect on synthesis of pathogenesis-related proteins (PR-proteins), lignification, papilla formation and activity of oxidative enzymes (PO, PPO and CAT) which realized in this study by increasing the activity of oxidative-reductive enzymes (**Matsumoto and Asada, 1990; Abd-El-Kareem *et al.,* 2001**).

**Induction disease resistance by sulfate mineral salts**

All sulphate minerals at the different concentrations showed significant reduction of incidence and severity of the disease compared to control (Table 6). Copper sulphate ($CuSo_4$) revealed the lowest percentage of disease parameters, while manganese ($MnSo_4$) and Zinc sulphates ($ZnSo_4$) recorded the lowest ones in this respect. Results also showed that, there is a positive relationship between sulphate mineral concentrations and their effect on the infection by Cercospora leaf spot in the two successive season.

Regarding peanut pod yield, results in Table 7 indicate that, pod yield significantly varied among the tested sulphate minerals and their concentrations, in both successive seasons. Generally, copper sulphate ($CuSo_4$) at all concentrations tested gave the highest peanut pod yield in both seasons followed by zinc sulphate ($ZnSo_4$) .General positive correlations were obtained between sulphate mineral concentrations and their effect on peanut pod yield in the two successive seasons.

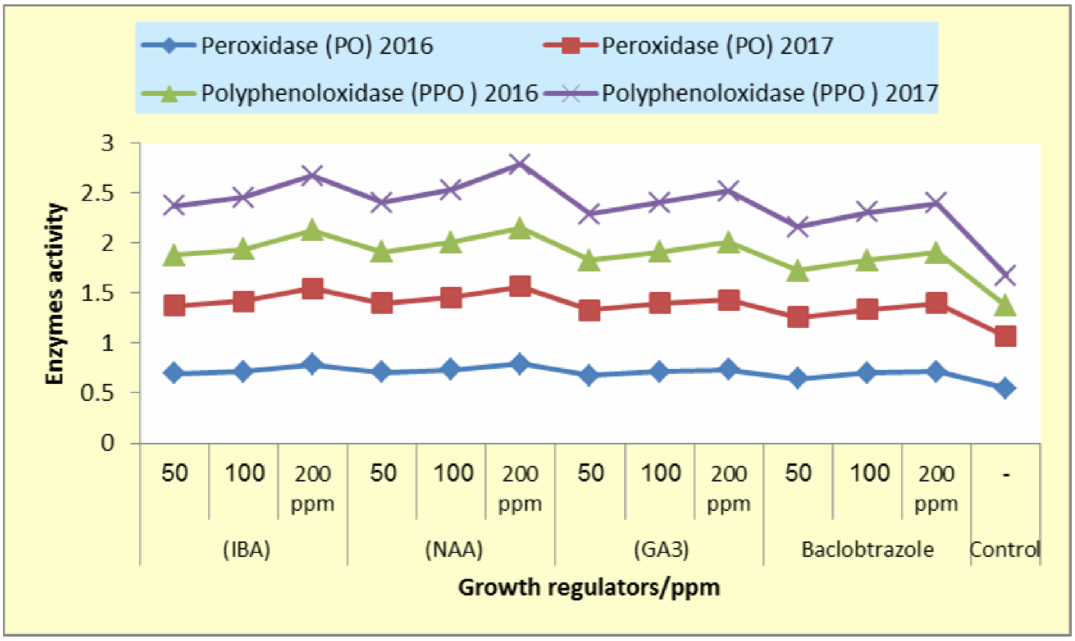


**Fig. 4. Effect of the foliar spraying with some growth regulators on peroxidase and polyphenoloxidase activity in peanut leaves**

**Table 6. Effect of the foliar spraying with some sulfate mineral salts on incidence and severity of the disease during two successive seasons (2016 and 2017)**

| Sulfate mineral salt | Conc. (mM) | Disease incidence and severity (%) | | | |
|---|---|---|---|---|---|
| | | 2016 | | 2017 | |
| | | DI | DS | DI | DS |
| **$CuSo_4$** | 1 | 44.75 | 11.18 | 47.50 | 13.43 |
| | 2 | 32.25 | 9.07 | 33.50 | 11.57 |
| | 4 | 26.50 | 8.90 | 28.25 | 9.50 |
| **$ZnSo_4$** | 1 | 42.25 | 12.37 | 43.50 | 14.87 |
| | 2 | 34.75 | 10.47 | 37.50 | 12.47 |
| | 4 | 31.50 | 9.59 | 32.75 | 10.59 |
| **$MgSo_4$** | 1 | 45.25 | 12.08 | 47.00 | 13.08 |
| | 2 | 43.75 | 10.07 | 45.00 | 11.32 |
| | 4 | 37.75 | 9.13 | 40.00 | 10.43 |
| **$MnSo_4$** | 1 | 50.50 | 13.12 | 51.75 | 15.01 |
| | 2 | 46.50 | 11.21 | 47.50 | 12.46 |
| | 4 | 41.25 | 10.62 | 43.75 | 11.37 |
| **Control** | - | 68.75 | 44.30 | 71.75 | 46.52 |
| **LSD 0.05% for** | | | | | |
| **Sulfate minerals (A)** | | 1.66 | 0.70 | 2.07 | 0.66 |
| **Concentration (B)** | | 1.29 | 0.54 | 1.61 | 0.51 |
| **Interactions (A) × ( B)** | | 2.88 | 1.21 | 3.59 | 1.14 |

DI= Disease incidence DS= Disease severity

**Table 7. Effect of foliar spraying with some sulfate mineral salts on peanut pod yield during two successive seasons (2016 and 2017)**

| Sulfate mineral salt | Conc. (mM) | Peanut pod yield | | | |
|---|---|---|---|---|---|
| | | Kg/ plot (10.5m$^2$) | | Expected pod (Ton/fad.) | |
| | | 2016 | 2017 | 2016 | 2017 |
| **$CuSo_4$** | **1** | 3.39 | 3.30 | 1.355 | 1.320 |
| | **2** | 4.40 | 4.26 | 1.760 | 1.704 |
| | **4** | 4.33 | 4.25 | 1.730 | 1.699 |
| **$ZnSo_4$** | **1** | 3.38 | 3.17 | 1.352 | 1.269 |
| | **2** | 3.58 | 3.43 | 1.430 | 1.373 |
| | **4** | 4.29 | 4.09 | 1.714 | 1.634 |
| **$MgSo_4$** | **1** | 3.16 | 3.09 | 1.262 | 1.235 |
| | **2** | 3.32 | 3.17 | 1.327 | 1.267 |
| | **4** | 3.75 | 3.65 | 1.499 | 1.460 |
| **$MnSo_4$** | **1** | 3.25 | 3.09 | 1.302 | 1.235 |
| | **2** | 3.34 | 3.29 | 1.337 | 1.317 |
| | **4** | 3.86 | 3.66 | 1.545 | 1.463 |
| **Control** | **-** | 2.59 | 2.56 | 1.038 | 1.025 |
| **LSD 0.05% for** | | | | | |
| **Sulfate minerals (A)** | | 0.06 | 0.09 | 0.005 | 0.002 |
| **Concentration (B)** | | 0.04 | 0.07 | 0.004 | 0.001 |
| **Interactions (A) X ( B)** | | 0.10 | 0.16 | 0.009 | 0.003 |

Results illustrated in Fig. 5 indicate that phenol contents were obviously higher in plants treated with all sulfate mineral salts as inducers than the untreated control during the two growing seasons (2016 and 2017) with visible increase when copper sulfate, was tested.

All tested sulfate mineral salts increased the activity of oxidative-reductive enzymes in peanut leaves compared to untreated control in both growing seasons (Fig. 6). Among all tested treatments, the highest value for each of (PO) and (POP) activity was produced when copper sulfate at 4mM was sprayed followed by magnesium sulfate at 4mM, while zinc and manganese sulfates at 1mM recorded the lowest value for each of (PO) and (POP) activities.

Microelements might play a positive role for stimulating natural defense mechanisms in peanut plants such as increasing the level of phenols and activities of the oxidative enzymes **(Meena *et al.*, 2001**). Moreover, microelements interacts with N metabolism and is intimately involved in carbohydrate synthesis, photosynthesis, coenzymes to many of plant enzymes and synthesis of other compounds associated with the defense of plant against pathogens like phytoalexins and lignin **(Engelhard, 1993)**.

Magnesium deficiency rarely limits plant growth, however, it is necessary for groundnut stems from its role as a carrier of phosphorus in oil formation, and its effect on seed viability. Magnesium supply may be omitted from the pod zone without adverse effects on pod development of some cultivars provided adequate Mg in the root zone **(Zharare *et al.,* 1993)**.

On the other hand, copper activate four distinct mitogen-activated protein kinases (MAPKs). Copper also present in three different forms in proteins: (a) blue proteins without oxidase activity (*e.g.,* plastocyanin); (b) non-blue proteins, which produce peroxidases and oxidize monophenols to diphenols; and (c) multicopper proteins, which act as oxidases and catalyze (**Jonak *et al.,* 2004)**.

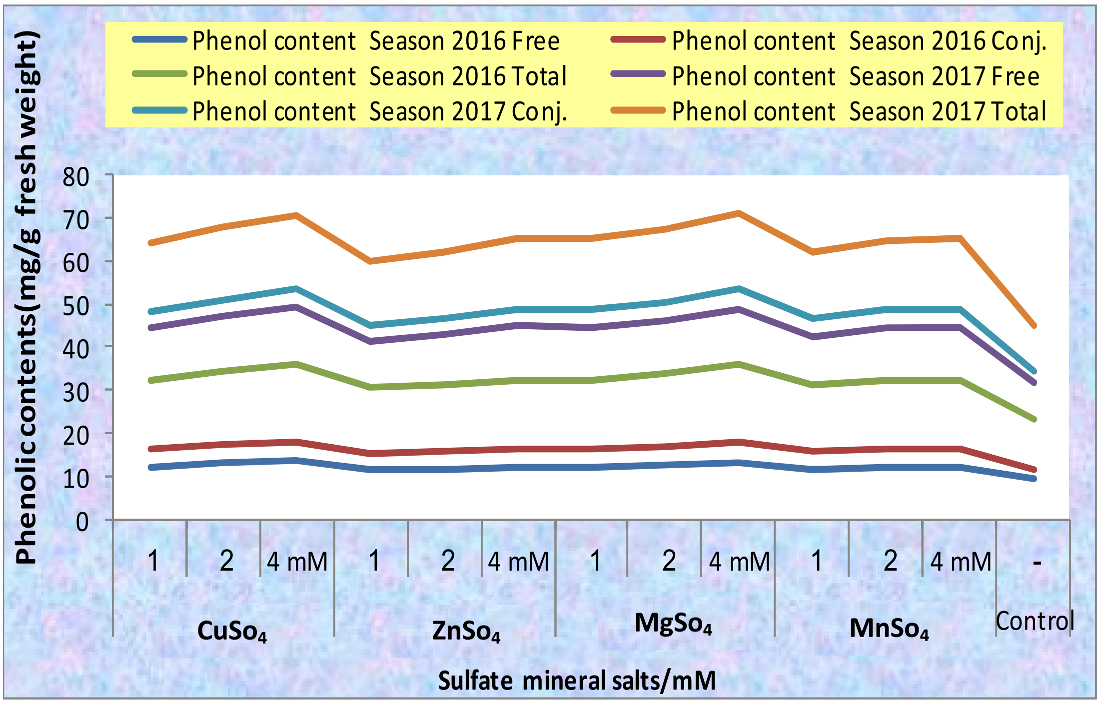


**Fig. 5. Effect of some sulfate salts on phenolic contents (mg/g fresh weight) in peanut leaves**

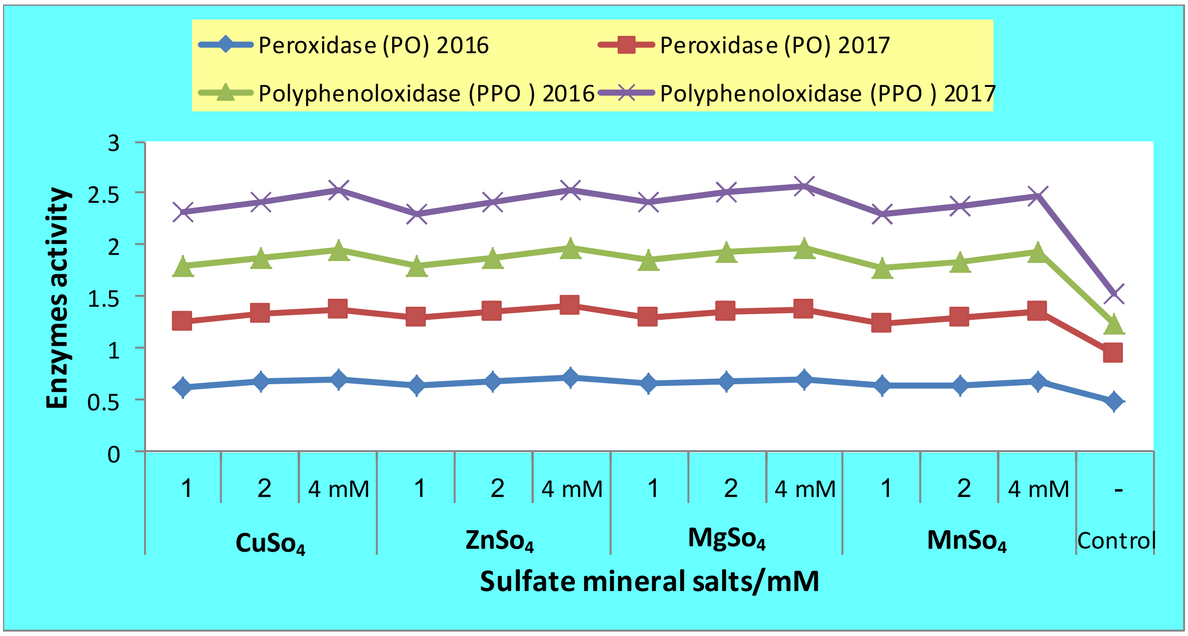


**Fig. 6. Effect of the foliar spraying with some sulfate salts on peroxidase (PO) and polyphenoloxidase (PPO) activity in peanut leaves**

## Induction disease resistance by silicate minerales

Results in Table 8 indicate that all silicate minerals at the different concentrations and the fungicide Seal-Matreax resulted in significant reduction the incidence and severity of the disease compared to non-treated control in the two successive seasons. Generally, calcium silicate ($CaSio_3$) followed by potassium silicate ($k_2Sio_3$) gave the lowest percentages without significant differences in between. Aluminum silicate ($Al_2Sio_3$) and Seal-Matreax recorded the lowest ones in this respect. Results also showed that, there is a positive relationship among silicate mineral concentrations and their effect on the infection by Cercospora leaf spot. An opposite way of both aluminum silicate and Seal-Matreax and a reduction in pod yield was recorded as their concentrations increased in the two successive season (2016 and 2017).

Regarding peanut pod yield results in Table 9 reveals that, peanut pod yield significantly varied among the tested silicate minerals at their different concentrations in the two successive seasons. Generally, calcium silicate ($CaSio_3$) at 1600 ppm reslted in the highest peanut pod yield in the two growing seasons followed by potassium silicate ($K_2Sio_3$) at the same concentration. Results also showed that, there is a positive relationship between silicate minerals concentrations and their effect on peanut pod yield where, aluminum silicate ($Al_2Sio_3$) and Seal-Matreax recorded the lowest pod yield in this respect.

Results presented in Fig. 7 indicate that phenol contents were obviously higher in plants treated with all silicate mineral salts as inducer resistance than the untreated control during both growing seasons. Peanut plants treated with calcium silicate ($CaSio_3$) recorded the highest phenol contents followed by potassium silicate. Meanwhile, aluminum silicate ($Al_2Sio_3$) and Seal-Matreax resulted in the lowest values. Results also indicated that, increasing the concentration of silicate mineral salts led to an increase in phenol contents in peanut leaves during both growing seasons.

Results also, show that all tested silicate mineral salts increased the activity of oxidative-reductive enzymes (Fig. 8). The highest (PO) and (PPO) activity was produced by both calcium silicate and potassium silicate at 1600 ppm. However, aluminum silicate and Seal-Matreax recorded the lowest (PO) and ((PPO) activity compared to the other treatments in the two successive seasons.

A possible effect of foliar application of silicon (Si) sources on disease control might be explained on the basis of the establishment of a physical barriers on the host tissue **(Samuels *et al*., 1991; Bowen *et al*., 1992)**. In case of bean anthracnose (*Colletotrichum lindemuthianum*), Si applied on foliage was effective even without establishing a physical barrier. Thus, increased plant resistance to diseases through Si treatment is associated with active and/or passive mechanisms **(Datnoff *et al*., 2007)**. Several modifications may occur in the plant surface after Ca or Si application, including pH, increase and changes in the osmotic potential and on the populations of microorganisms.

Calcium plays an important role in reducing many plant diseases (**Biggs, 2004; Sugimoto *et al.,* 2008**) as it might has a critical metabolic role in carbohydrates removal, cell wall deposition and formation of pectates in the middle lamella **(El-Neshawy *et al.,* 2004)**. Also, it forms strengthening bridges, especially in the pectate materials that form the middle lamella of plant cells. Calcium is, also, important in maintaining selectivity of cell plasmalemma, and in binding the plasmalemma to the cell wall. Calcium also binds strongly to oxalic acid, an important toxin produced by many pathogens, thus reduced host penetration by the pathogen and thus might as major limiting factor to groundnut production. The developing pods require adequate Ca in the surrounding soil for proper pod development and production of high quality seed **(Cox *et al.,* 1982; Gascho and Davis, 1994).**

**Table 8. Effect of the foliar spraying with some silicate mineral salts on disease incidence and severity during two successive seasons 2016 and 2017**

| Silicate mineral salt | Conc. (ppm) | Disease incidence and severity (%) | | | |
|---|---|---|---|---|---|
| | | 2016 | | 2017 | |
| | | DI | DS | DI | DS |
| **$Al_2Sio_3$** | 200 | 46.50 | 14.84 | 50.75 | 15.92 |
| | 400 | 50.50 | 16.65 | 51.50 | 17.57 |
| | 800 | 60.25 | 17.70 | 57.50 | 18.57 |
| | 1600 | 62.50 | 18.85 | 62.50 | 19.63 |
| **$MgSio_3$** | 200 | 40.50 | 13.90 | 42.50 | 14.68 |
| | 400 | 39.50 | 12.74 | 41.00 | 13.62 |
| | 800 | 36.50 | 11.91 | 38.25 | 12.96 |
| | 1600 | 33.25 | 11.31 | 36.50 | 12.53 |
| **$K_2Sio_3$** | 200 | 26.50 | 11.09 | 29.00 | 12.08 |
| | 400 | 21.75 | 10.19 | 22.50 | 11.17 |
| | 800 | 18.25 | 9.64 | 20.00 | 10.75 |
| | 1600 | 17.50 | 8.89 | 17.50 | 9.39 |
| **$CaSio_3$** | 200 | 14.75 | 10.32 | 18.25 | 11.21 |
| | 400 | 15.75 | 9.54 | 17.50 | 10.30 |
| | 800 | 10.50 | 7.39 | 13.25 | 8.17 |
| | 1600 | 10.25 | 7.65 | 11.75 | 8.08 |
| **Seal-Matreax** | 200 | 47.50 | 17.52 | 48.75 | 18.36 |
| | 400 | 48.25 | 18.31 | 50.75 | 19.32 |
| | 800 | 52.50 | 19.12 | 52.75 | 20.45 |
| | 1600 | 53.75 | 20.21 | 54.25 | 21.38 |
| **Control** | - | 71.75 | 42.72 | 74.25 | 44.25 |
| **LSD 0.05% for** | | | | | |
| **Silicate minerals (A)** | | 1.20 | 0.42 | 1.22 | 0.44 |
| **Concentration (B)** | | 1.21 | 0.40 | 1.20 | 0.41 |
| **Interactions (A) × ( B)** | | 2.39 | 0.84 | 2.44 | 0.87 |

DI=Disease incidence DS=Disease severity

Table 9. Effect of the foliar spraying with some silicate mineral salts on peanut pod yield during two successive seasons 2016 and 2017

| Silicate mineral salt | Conc. (ppm) | Peanut pod yield | | | |
|---|---|---|---|---|---|
| | | Kg/ plot ($10.5m^2$) | | Expected pod (Ton/fad.) | |
| | | 2016 | 2017 | 2016 | 2017 |
| $Al_2Sio_3$ | 200 | 3.96 | 3.93 | 1.585 | 1.573 |
| | 400 | 3.25 | 3.20 | 1.300 | 1.279 |
| | 800 | 3.22 | 3.16 | 1.288 | 1.265 |
| | 1600 | 2.91 | 2.88 | 1.164 | 1.152 |
| $MgSio_3$ | 200 | 3.99 | 3.90 | 1.595 | 1.558 |
| | 400 | 4.09 | 4.07 | 1.635 | 1.627 |
| | 800 | 4.31 | 4.18 | 1.723 | 1.673 |
| | 1600 | 4.91 | 4.73 | 1.965 | 1.890 |
| $CaSio_3$ | 200 | 4.30 | 4.11 | 1.720 | 1.558 |
| | 400 | 4.49 | 4.42 | 1.795 | 1.627 |
| | 800 | 4.84 | 4.81 | 1.935 | 1.673 |
| | 1600 | 5.42 | 5.13 | 2.169 | 1.890 |
| $K_2Sio_3$ | 200 | 3.56 | 3.17 | 1.423 | 1.268 |
| | 400 | 3.97 | 3.90 | 1.589 | 1.559 |
| | 800 | 4.46 | 4.25 | 1.786 | 1.698 |
| | 1600 | 4.95 | 4.86 | 1.982 | 1.945 |
| Seal-Matreax | 200 | 3.97 | 3.96 | 1.589 | 1.585 |
| | 400 | 3.41 | 3.26 | 1.365 | 1.305 |
| | 800 | 3.97 | 3.24 | 1.589 | 1.295 |
| | 1600 | 2.95 | 2.89 | 1.180 | 1.155 |
| Control | - | 2.66 | 2.56 | 1.065 | 1.025 |
| LSD 0.05% for | | | | | |
| Silicate minerals (A) | | 0.08 | 0.04 | 0.04 | 0.05 |
| Concentration (B) | | 0.07 | 0.03 | 0.03 | 0.04 |
| Interactions (A) × (B) | | 0.16 | 0.07 | 0.07 | 0.10 |

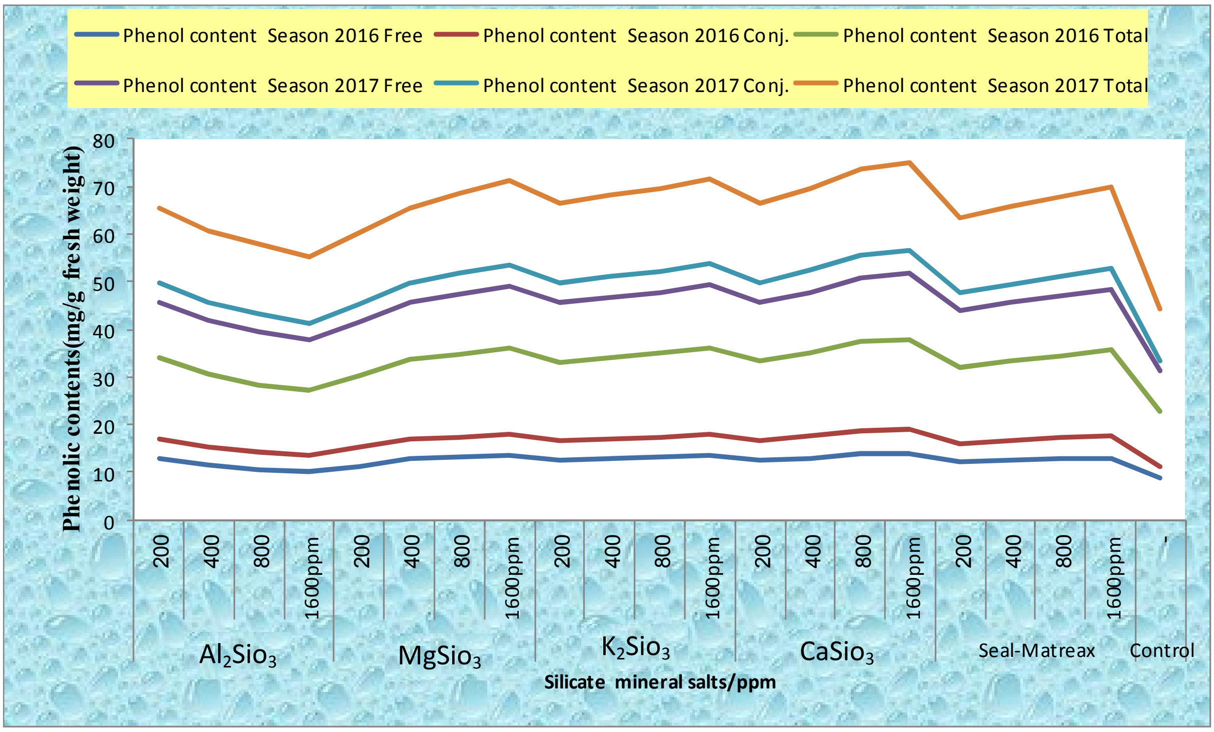


**Fig. 7. Effect of some silicate mineral salts on phenolic contents (mg/g fresh weight) in peanut leaves**

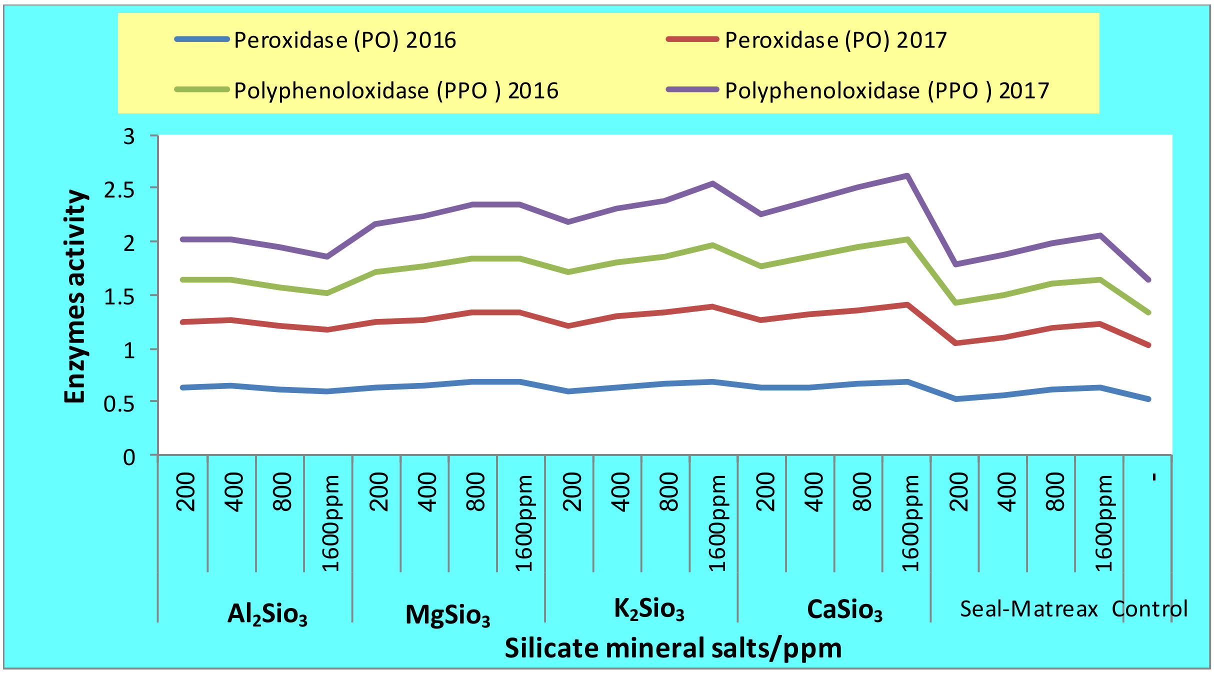


**Fig. 8. Effect of foliar spraying with some silicate mineral salts on peroxidase (PO) and polyphenoloxidase (PPO) activity in peanut leaves**

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# مكافحة مرض تبقع الأوراق السـركسـبوري في الفـول السـوداني باستخدام الأصناف المقاومة والكيماويات المستحثة للمقـاومــة

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تم إختبار رد فعل ستة أصناف من الفول السوداني للإصابة بمرض تبقع الأوراق السركسبوري وهى إسماعيلية ١، جيزة ٥، جيزة ٦، جورجى، فيرجينيا وآر ٩٢ تحت ظروف العدوى الطبيعية بمحطة بحوث الإسماعيلية موسمى ٢٠١٦ و٢٠١٧ وكان الصنف إسماعيلية ١ أكثرها مقاومة حيث سجل أقل نسبة وشدة إصابة تلاه الصنف آر ٩٢بينما كانت الأصناف جيزة ٦ وجيزة ٥ متوسطة القابلية للإصابة، وكانت الأصناف جوروجى وفيرجينيا أكثر قابلية للإصابة حيث أعطت أعلى نسبة وشدة إصابة، أعطى الصنف آر ٩٢ أعلى محصول تلاه الصنف إسماعيلية ١ بينما أعطى الصنف جيزة ٥ أقل محصول، تم اختبار أربعة أنواع من مستحثات المقاومة الكيماوية رشاً على المجموع الخضرى وهى الأحماض العضوية، منظمات النمو، أملاح الكبريتات و أملاح السيليكات تحت ظروف العدوى الطبيعية بالحقل في مقاومة مرض تبقع الأوراق السركسبوري في الفول السوداني، أظهرت كل المستحثات المختبرة نقص معنوي في المرض مقارنة بالنباتات غير المعامله في الموسمين الزراعيين ٢٠١٦ و ٢٠١٧، حيث تم إختبار فاعلية خمسة مستحثات كيماوية وهى حمض السالسيلك، حمض النيكوتينك، حمض البيوتريك و حمض الأوكسالك بالإضافة إلى البيون باستخدام ثلاثة تركيزات ٢، ٤، ٨ مليمول لكل منهم وأوضحت النتائج أن البيون بتركيز ٨ مليمول يليه حمض السالسيلك بنفس التركيز هما الأكثر فاعلية فى هذا الصدد حيث أعطت أقل نسبة وشدة إصابة وأعلى محصول بينما أعطى حمض النيكوتينك بتركيز ٢مليمول أعلى نسبة وشدة إصابة وأقل محصول، تم اختبار أربعة منظمات نمو وهى حمض الإندول بيوتريك، حمض النافثالين، حمض الجبريلين والباكلوبترازول بثلاثة تركيزات لكل منهم (٥٠، ١٠٠ و ٢٠٠ جزء فى المليون) وكان حمض النافثالين تركيز ٢٠٠ جزء فى المليون الأكثر فاعلية حيث أعطى أقل نسبة وشدة إصابة بالمرض وأعلى محصول بينما الباكلوبترازول تركيز ٥٠ جزء فى المليون كان الأقل فاعلية حيث سجل أعلى نسبة وشدة إصابة وأقل محصول. تم إستخدام أملاح الكبريتات مثل كبريتات النحاس، كبريتات الزنك، كبريتات المغنسيوم وكبريتات المنجنيز بإستخدام ثلاثة تركيزات وهى ١، ٢ و ٤ مليمول لكل منهم وأوضحت النتائج أن كبريتات النحاس بتركيز ٤ مليمول كانت أفضل المعاملات حيث أعطت أقل مقاييس للمرض وأعلى محصول بينما كبريتات المنجنيز بتركيز ١مليمول كانت أقلهم حيث أعطت أعلى نسبة وشدة إصابة وأقل محصول، كذلك تم اختبار فاعلية أملاح السليكات مثل سليكات الألمونيوم، المغنسيوم، البوتاسيوم، الكالسيوم بالمقارنة بالمركب التجارى سيل ماتريكس بتركيزات ٢٠٠، ٤٠٠ ، ٨٠٠ و١٦٠٠ جزء فى المليون لكل منهم وكانت سليكات الكالسيوم و البوتاسيوم بتركيز ١٦٠٠ جزء فى المليون هى الأكثر فاعلية حيث أعطت أقل مقاييس للمرض وأعلى محصول بينما أعطت سليكات الألمونيوم والسيل ماتريكس أعلى نسبة وشدة إصابة بالمرض وأقل محصول، أظهرت الدراسة أن هناك علاقة بين معاملة الرش بالكيماويات المستحثة للمقاومة وحدوث تغيرات كيموحيوية في أنسجة أوراق الفول السوداني حيث زاد نشاط إنزيمات الأكسدة والإختزال (بيروكسيديز و البولي فينول أوكسيديز) وكذلك محتواها من الفينولات الكلية و الحرة والمرتبطة.

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