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# Gene Expression and Protein Profile Associated with some Common Bean Cultivars Infected with Root-Knot Nematode, *Meloidogyne incognita*

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## Abstract

The reaction of five common bean cultivars to root-knot nematode, *Meloidogyne incognita* were classified based on Damage Index (DI) were studied under greenhouse conditions. Data indicated that the cultivars were classified into three categories; one as highly susceptible (Paulista), two as susceptible (Xera, and Swiss blane), and two as moderately resistant (Nebraska and Bronco). Growth response of the tested cultivars to *Meloidogyne incognita* infection had a negative effect on plant growth, parameters, shoot fresh, and pod weights. Chlorophyll content in the leaves of Paulista cultivars in response to *Meloidogyne incognita* infection was decreased significantly during different ages of plant development compared with the control plants. Using protein electrophoresis analysis (SDS-PAGE) resulted differences in buffer soluble protein extracted from the leaves of the five tested common bean cultivars in relation to infection with *Meloidogyne incognita*. The differences in protein banding profile are expectable due to the differences in genetic composition on these cultivars. The differences in protein banding patterns among healthy and infected samples proved the difference in response of those CVs. to the infection.

**Keywords:** Common bean cultivars; *Meloidogyne incognita*; Chlorophyll content; Protein electrophoresis analysis (SDS-PAGE)

## Introduction

The common bean (*Phaseolus vulgaris* L.) is one of the most important leguminous plants worldwide and is the highly relished pulse, grain in in many developing countries, especially in Africa [1]. The common bean seeds contain an important source of dietary protein amounting to 22% of the total seed weight. In addition, N-fixation through root nodules of beans improves the soil fertility status and reduces amount of N used in intercropping systems [2]. Common bean is mostly grown in all districts of Egypt, during two seasons, spring and autumn, when the temperature is most suitable for its growth and yield. Based on it is grown on more than based on it is grown on more than 26028 ha annually in 2020 according to the official data from the Egyptian Ministry of Agriculture.

Various pathogenic nematodes have negative effects on common bean production in many parts of the world as well as in Egypt. Among them, the root-knot nematodes (*Meloidogyne* spp.) are the most important and destructive nematodes which attack common bean crops and feed on the roots causing root galls, the visible symptom of infection [3]. Root-knot nematodes are reported to cause yield losses of up to 60% [4,5]. In addition to direct pathogenic effects on plants, the nematodes act synergistically with other plant pathogens to form disease complexes that further impact negatively the crops. The nematodes also suppress nodulation and therefore affect nitrogen fixation and the photosynthetic rate and crop yield with increasing nematode inoculums level and duration of infection, in the common bean plant infected with *Meloidogyne incognita* [6,7].

This study was therefore carried out to evaluate the response of five common bean cultivars to *Meloidogyne incognita* and its effect on common bean growth under greenhouse conditions.

## Materials and Methods

Greenhouse experiments were conducted to evaluate the response of five common bean cultivars to root-knot nematode *Meloidogyne incognita* at Ismailia Agriculture Research Station of Egypt.

### Test plants

Reactions of five common bean cultivars to *Meloidogyne incognita* were evaluated under greenhouse conditions. The cultivars were Paulista, Xera, Swiss blane, Nebraska, and Bronco. These cultivars were obtained from the Vegetable Crop Research Department, Horticulture Research Institute, Agriculture Research Center and Giza, Egypt. The seeds were surface-

sterilized before sowing them into 25 cm diameter pots filled with steam-sterilized soil made up of sandy clay in the ratio of 1:4. Thinning was done one week after sowing so as one seedling per pot.

## Inoculum preparation and inoculation procedure

Two-week old seedlings were inoculated with 3,000 nematodes. The inoculum consisted of a 20 ml suspension of second-stage juvenile (J2) and eggs. The nematode was obtained from the infected common bean root system collected from infested farms in the study area. Root-knot nematode identified as *Meloidogyne incognita*, and maintained on the tomato *Solanum esculentum* cv. 888 in the greenhouse. In preparing the inoculum, the nematode was extracted from the tomato roots using the method according to Hussey and Barker [8]. The inoculum was pipetted into a depression made around the common bean roots and covered with soil. Treatments in which no nematodes were added to serve as controls (check plants). The treatments were arranged in a Randomized Complete Block Design (RCBD) and replicated 5 times. The experiment was terminated 60 days after inoculation and plant growth and disease assessment data were collected.

## Nematode assessment

The common bean plants were uprooted, and the roots were washed with a gently stream of water. Then, the having nematode galls were rated on 1-9 scale of Gall Index (GI), according to Sharma, et al. as follows: 1=no galls, 2=1 to 5 galls, 3=6 to 10 galls, 4=11 to 20 galls, 5=21 to 30 galls, 6=31 to 50 galls, 7=51 to 70 galls, 8=71 to 100 galls, and 9=>100 galls per root system. Gall Size (GS) and percent Galls Area (GA) are also, rated on a 1-9 scale. For GS: 1=no galls, 3=very small galls (about 10 % increase in root area at the galled region over non-galled normal root area), 5=small galls (about 30% increase), 7=medium galls (about 31% to 50% increase), and 9=big galls (about 51% to 100% increase). For GA: 1=no galls, 3=1% to 10% root area galled, 5=11% to 30% root area galled, 7=31% to 50% root area galled, and 9=>50% root area galled [9]. Damage Index (DI) is calculated by dividing the sum of GI, GS, and GA by 3 for each replicate. Based on DI, the host susceptibility (designation of resistance) of each plant variety is determined according to the following scheme: Plants with DI=1 is designated as highly resistant; DI=2 to 3, resistant; DI=4 to 5, moderately resistant; DI=6 to 7, susceptible; DI=8 to 9, highly susceptible. Number of egg masses/root systems as well, number of second stage juveniles in each pot were recorded [10].

## Data collection

**Plant growth assessment:** Plants were gently uprooted and the root system separated from the shoot system at the first basal node. The root systems were carefully and thoroughly washed with running water before taking their fresh weights (g), and their length (cm). Fresh and dry weights (gm plant-1), pod weights (gm plant-1), and number of nodules root-1 were obtained. The dry shoot and root weights (g) were obtained after drying them at 70°C for 72 hrs. The percentage of reduction in plant (R%) was calculated using the formula:-

$$R\% = \frac{Control plants - Infected}{Control plants} \times 100$$

**Gene expression and protein profile with root-knot nematode infection**

***Protein electrophoresis analysis (SDS-PAGE):*** The difference in buffer soluble protein extracted from the healthy and infected plants of the five tested beans cultivars (Paulista, Xera, Swiss blane, Nebraska, and Bronco) was shown by protein electrophoresis analysis (SDS-PAGE) Sodium Dodecyl Sulfate Polyacrylamide Gel Electrophoresis procedure was used according to Laemmli [11]. The products of SDS-PAGE procedure was analyzed using Protein Gel Documentation System (PGDS) program.

***Chlorophyll content:*** The effect of the infection by *Meloidogyne incognita* on the leaf chlorophyll content in the common bean cultivator Paulista was studied. Leaf chlorophyll content was estimated using SPAD-502 apparatus. Five readings were taken on the middle third of the surface lamina, the instrument itself, providing the average value given the instruments limited reading area (6 $mm^2$) in order to monitor any variations due to uneven pigment distribution.

## Statistical analysis

All experiments were performed twice in a completely randomized design with 5 replicates in each treatment. Data were subjected to Analysis of Variance (ANOVA) using MSTAT-C program version 2.10. Means were compared by Duncan's multiple range tests at $P \leq 0.05$ probabilities [12].

# Results

## Susceptibility of common bean cultivars to *Meloidogyne incognita*

The susceptibility of five common bean cultivars to root-knot nematode, *Meloidogyne incognito* was classified based on Damage Index (DI) shown in **Table 1**. Data indicated that, the cultivars were classified into 3 categories one as highly susceptible (Paulista), two as moderately resistant (Nebraska and Bronco) and two as susceptible (Xera, and Swiss blane). There were significant differences ($p \leq 0.05$) in No. of second-stage juvenile J2 in 250 g soil, No. of egg-masses/root system, and Egg mass Indices (EI) among common bean cultivars. Paulista, and Xera cultivars were the highest No. of J2/250 gm soil. While, Egg-masses Index (EI=9) of three cultivars (Paulista, Xera, and Swiss blane) while, other cultivars (Nebraska and Bronco) with EI=7 and EI=6 respectively.

**Table 1:** Relative susceptibility of five common bean cultivars infested with *Meloidogyne incognita* under greenhouse conditions.

| Cultivars | No. of root galls/root system | GI | Gall Size (GS) | Gall Area (GA) | Damage Index (DI) | Host susceptibility /Resistance | No. of J2/250 gm soil | No. of egg masses/ root system | EI |
|---|---|---|---|---|---|---|---|---|---|
| Paulista | 274$^{a}$ | 9$^{a}$ | 9.0$^{a}$ | 9.0$^{a}$ | 9$^{a}$ | HS | 360$^{a}$ | 140$^{a}$ | 9$^{a}$ |
| Xera | 260$^{b}$ | 9$^{a}$ | 7.3$^{b}$ | 7.3$^{b}$ | 7.6$^{b}$ | S | 360$^{a}$ | 121$^{b}$ | 9$^{a}$ |
| Swiss blane | 195$^{c}$ | 9$^{a}$ | 7.3$^{b}$ | 7.3$^{b}$ | 7.6$^{b}$ | S | 210$^{b}$ | 109$^{c}$ | 9$^{a}$ |
| Nebraska | 119$^{d}$ | 9$^{a}$ | 5.0$^{c}$ | 5.0$^{c}$ | 6.3$^{c}$ | MR | 180$^{c}$ | 69$^{d}$ | 7$^{b}$ |
| Bronco | 126$^{d}$ | 9$^{a}$ | 5.0$^{c}$ | 5.0$^{c}$ | 6.3$^{c}$ | MR | 120$^{d}$ | 50$^{e}$ | 6$^{b}$ |

**Note:** Data are average of 5 replicates. *Different letter (s) indicates significant differences among treatments within the same column according to Duncan's multiple range test (P ≤ 0.05). *Root Gall Index (RGI) or Egg-masses Index (EI) was determined according to Sharma, et al. [9] Gall Index (GI); and Egg-mass Indexes (EI) 1=no galls, 2=1-5 galls, 3=6-10 galls, 4=11-20 galls, 5=21-30 galls, 6=31-50 galls, 7=51-70 galls, 8=71-100 galls, and 9=>100 galls per plant. (HS) Highly Susceptible, (S) Susceptible, and (MR) Moderately Resistant.

## Plant growth

There were significant differences (p ≤ 0.05) in the plant growth parameters for infected five bean cultivars compared with the control plants **(Table 2)**.

Plant growth response as a criterion for nematode infection was recorded by calculating the fresh, dry weight of roots and shoots, and length of shoot and root of the five tested cultivars, as well as pods weight and number of nodules/plant.

**Table 2:** Plant growth response of five common bean cultivars to the infection with *Meloidogyne incognita.*

| Cultivars | Shoot | | | | | | | | | Pods weights | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Fresh weights (gm) | | | Dry weight (gm) | | | Length (cm) | | | | | |
| | C.P | I.P | R% | C.P | I.P | R% | C.P | I.P | R% | C.P | I.P | R% |
| Paulista | 6.6$^{a}$ | 4.7$^{d}$ | 29 | 4.7$^{c}$ | 2.7$^{c}$ | 42.5 | 28$^{a}$ | 22$^{b}$ | 20.8 | 5.1$^{a}$ | 3.1$^{a}$ | 39 |
| Xera | 9.0$^{a}$ | 6.9$^{b}$ | 23 | 6.2$^{b}$ | 3.3$^{a}$ | 47 | 26$^{a}$ | 20$^{c}$ | 24 | 5.4$^{a}$ | 3.3$^{a}$ | 39 |
| Swiss blane | 8.6$^{a}$ | 6.2$^{c}$ | 28 | 6.1$^{b}$ | 2.9$^{ab}$ | 52 | 24$^{a}$ | 22$^{c}$ | 9 | 5.2$^{a}$ | 4.4$^{d}$ | 15 |
| Nebraska | 8.6$^{a}$ | 6.3$^{c}$ | 26 | 6.3$^{b}$ | 3$^{b}$ | 52 | 27$^{a}$ | 26$^{a}$ | 3 | 6.2$^{a}$ | 4.46$^{b}$ | 29 |
| Bronco | 10$^{a}$ | 7.5$^{a}$ | 25 | 7.1$^{a}$ | 3.1$^{ab}$ | 56 | 29$^{a}$ | 25$^{a}$ | 11 | 7.2$^{a}$ | 5.8$^{c}$ | 19 |
| **Root** | | | | | | | | | | | | |
| | Fresh weights (gm) | | | Dry weight (gm) | | | Length(cm) | | | No. of Nodules/Plant | | |
| | C.P | I.P | R% | C.P | I.P | R% | C.P | I.P | R% | C.P | I.P | R% |
| Paulista | 3.9$^{a}$ | 3.4$^{a}$ | 12.8 | 2.7$^{a}$ | 1.7$^{a}$ | 37 | 25$^{a}$ | 21$^{b}$ | 16 | 106$^{a}$ | 15$^{a}$ | 86 |
| Xera | 2.6$^{a}$ | 2.9$^{d}$ | 9 | 1.7$^{d}$ | 0.9$^{c}$ | 47 | 26$^{a}$ | 22.7$^{a}$ | 12.8 | 49$^{a}$ | 13$^{b}$ | 74 |
| Swiss blane | 3.1$^{a}$ | 3.0$^{c}$ | 3.2 | 2$^{c}$ | 1.3$^{b}$ | 35 | 26$^{a}$ | 24$^{a}$ | 7.6 | 8$^{a}$ | 3$^{c}$ | 62.5 |
| Nebraska | 3.2$^{a}$ | 3.0$^{c}$ | 6.25 | 2$^{c}$ | 1.4$^{b}$ | 30 | 27$^{a}$ | 24$^{a}$ | 11.1 | 8$^{a}$ | 5$^{c}$ | 37 |
| Bronco | 3.6$^{a}$ | 3.4$^{b}$ | 5.5 | 2.2$^{b}$ | 1.4$^{b}$ | 36 | 23$^{a}$ | 21$^{c}$ | 10 | 9$^{a}$ | 3$^{c}$ | 66.6 |

**Note:** Data are average of 5 replicates. *Different letter (s) indicates significant differences among treatments within the same column according to Duncan's multiple range test (P ≤ 0.05). C.P=Control Plants I.P=Infected Plants. R%=Percentage of Reduction=CP-IP/ CP×100.

Generally, the data presented in **Table 2** indicated that shoot fresh weights of the all-tested cultivars were affected by the nematode infection. For instance, the percentages of reduction in shoot fresh weights ranged between 23%-29%. Paulista was the most affected cultivar, while, Xera was the least affected one. The percentage of reduction in root fresh weights ranged between 3.2%-12.8% as the highest percentage of reduction was belonging to Paulista cultivar, while the lowest was in Swiss blane. The percentages of reduction in nodules numbers ranged between 37%-86%. In **Table 2** the data showed that Paulista was the most affected cultivar, while, the lowest was for Nebraska. The percentages of reduction in pods weights ranged between 15%-39%. Paulista and Xera were the most affected cultivars, while the lowest was in Swiss blane.

## Gene expression and protein profile with root-knot nematode *Meloidogyne incognita* infection

**Protein electrophoresis analysis (SDS-PAGE):** The buffer soluble proteins extracted from the leaves of the five-tested common bean cultivars (Paulista, Xera, Swiss blane, Nebraska, and Bronco) were used as a criterion of reaction to infection with the root-knot nematode *Meloidogyne incognita*. SDS-PAGE products were illustrated in **Figure 1**, and the Protein Gel Documentation System (PGDS) program analysis was recorded in **Table 3**. Data revealed the presence of protein bands in healthy and infected plants with distinguishable differences in the number, size, molecular weights and Amt% between the five tested common bean cultivars. Data can be summarized as follows:-

- Eleven protein bands were recorded in the leaves of Paulista cultivar healthy plants (60,52,50,41,40,38,31,25,22,19 and 17 kDa) whereas there were fourteen protein bands recorded in the leaves of Paulista cultivar infected plants (63,62,57,55,53, 40,38,36,35,32,27,25,1 and 16 kDa).
- Ten protein bands were recorded in the leaves of Xera cultivar healthy plants (62,54,38,36,31,30,26,24,21 and 18 kDa), but eight protein bands were recorded in the infected plant's leaves of the Xera cultivar (62,38,36,31,29,25,22 and 20 kDa).
- Fifteen protein bands were recorded in the leaves of Swiss blane cultivar healthy plants (62,60,54,50,46,43,41,38,35, 34,31,30,26,20 and 16 kDa), but nine protein bands recorded in the infected plant's leaves of Swiss blane cultivar (63,51,36, 33,27,24,21,19 and 16 kDa).
- Twelve protein bands were recorded in the leaves of Bronco cultivar healthy plants (66,58,56,54,52,46,42,41,36,30,22 and 17 kDa) whereas nineteen protein bands were recorded in the leaves of Bronco cultivar infected plants (66,62,61,59,56,53,52, 45,42,38,36,35,33,32,31,29,24,20 and 17 kDa).
- Fourteen protein bands were recorded in the leaves of Nebraska cultivar healthy plants (66,62,54,51,42,38,36,35,32 31,29,26,22 and 19 kDa) whereas eighteen protein bands were recorded in the leaves of Nebraska cultivar infected plants (66,65,62,61,58,56,55,52,49,42,36,35,30,29,26,23,20 and 17 kDa).

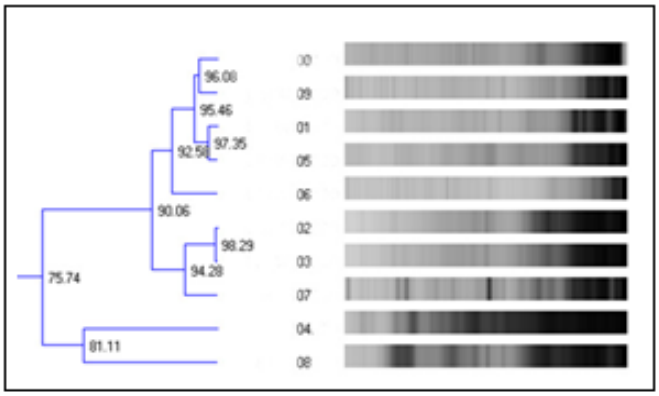


**Figure 1:** Cluster analysis showing the similarity polymorphism of protein banding patterns obtained by SDS-PAGE. **Note:** Lane 1: Leaves of healthy Paulista; Lane 2: Leaves of infected Paulista; Lane 3: Leaves of healthy Xera; Lane 4: Leaves of infected Xera; Lane 5: Leaves of healthy Swiss blane; Lane 6: Leaves of infected Swiss blane; Lane 7: Leaves of healthy Bronco; Lane 8: Leaves of infected Bronco; Lane 9: Leaves of healthy Nebraska; Lane 00: Leaves of infected Nebraska.

The data in **Figure 1** showed that the similarity between proteins banding pattern of five cultivars infected with *Meloidogyne incognita.* The similarity between Lane (3) and Lane (4) was 98.29%, Lane (2) and Lane (10) was 96.08%, Lane (2) and Lane (6) was 97.35%, Lane (5) and Lane (9) was 81.11%.

## Chlorophyll content

The effect of root-knot nematode *Meloidogyne incognita* on chlorophyll content in common bean cultivar Paulista was studied. The data in **Figure 2** showed the effects of nematode on chlorophyll content in healthy and infected plants. The measurements were taken at a different plant development period from planting (7,21,30,45,60,75 and 90 intervals days). Data revealed that there was a significant decrease in chlorophyll content in healthy and infected plants from 21 days to 90 days after planting. These results indicated there were negative effects of *Meloidogyne incognita* infection on chlorophyll content at different developmental stage of plant as compared to healthy plants.

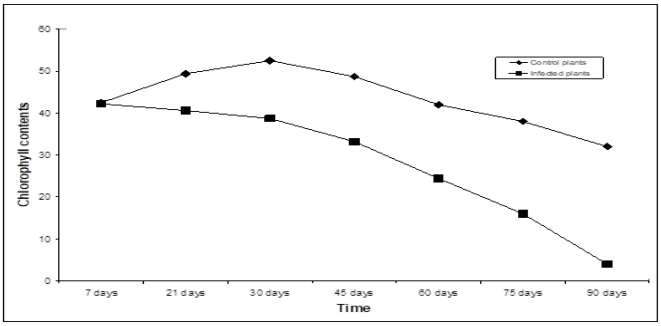


**Figure 2:** Chlorophyll content in common bean cultivar Paulista in relation to *Meloidogyne incognita* infection at different plant ages.

**Table 3:** Molecular weight (kDa) of protein bands of five common bean cultivars healthy and infected plants with *Meloidogyne incognita*.

| Pk/In# | M | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 00 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 66 | - | - | - | - | - | - | 66 | 66 | 66 | 66 |
| 2 | - | - | - | - | - | - | - | - | - | - | 65 |
| 3 | - | - | 63 | - | - | - | 63 | - | - | - | - |
| 4 | - | - | 62 | 62 | 62 | 62 | - | - | 62 | 62 | 62 |
| 5 | - | 60 | - | - | - | 60 | - | - | 61 | - | 60 |
| 6 | - | - | 57 | - | - | - | - | 58 | 59 | - | - |
| 7 | - | - | 55 | - | - | - | - | 56 | 56 | - | 56 |
| 8 | - | - | 53 | 54 | - | 54 | - | 54 | 53 | 54 | - |
| 9 | - | 52 | - | - | - | - | 51 | 52 | 52 | 51 | - |
| 10 | - | 50 | - | - | - | 50 | - | - | - | - | 49 |
| 11 | 45 | - | - | - | - | 46 | 46 | 45 | - | - | - |
| 12 | - | - | - | - | - | 43 | - | 42 | 42 | 42 | 42 |
| 13 | - | 41 | - | - | - | 41 | - | 41 | - | - | - |
| 14 | 40 | 40 | 40 | - | - | - | - | - | - | - | - |
| 15 | - | 38 | 38 | 38 | 38 | 38 | - | - | 38 | 38 | - |
| 16 | 36 | - | 36 | 36 | 36 | - | 36 | 36 | 36 | 36 | 36 |
| 17 | - | - | 35 | - | - | 35 | - | - | 35 | 35 | 35 |
| 18 | - | - | - | - | - | 34 | 33 | - | 33 | - | - |
| 19 | - | - | 32 | - | - | - | - | - | 32 | 32 | - |
| 20 | - | 31 | - | 31 | 31 | 31 | - | 30 | 31 | 31 | 31 |
| 21 | 29 | - | - | - | - | - | - | 29 | 29 | 29 | 29 |
| 22 | - | - | 27 | 26 | - | 26 | 27 | - | - | 26 | 26 |
| 23 | 24 | 25 | 25 | 24 | 25 | - | 24 | - | 24 | - | - |
| 24 | - | 22 | - | - | 22 | - | - | 22 | - | 22 | 23 |
| 25 | - | - | 21 | 21 | 20 | 20 | - | - | 20 | - | 20 |
| 26 | - | 19 | - | 18 | - | - | 19 | - | - | 19 | - |
| 27 | 16 | 17 | 16 | - | - | 16 | - | 17 | 17 | - | 17 |

**Note:** M: protein Marker; Lane 1: Leaves of healthy Paulista; Lane 2: Leaves of infected Paulista; Lane 3: Leaves of healthy Xera; Lane 4: Leaves of infected Xera; Lane 5: Leaves of healthy Swiss blane; Lane 6: Leaves of infected Swiss blane; Lane 7: Leaves of healthy Bronco; Lane 8: Leaves of infected Bronco; Lane 9: Leaves of healthy Nebraska; Lane 00: Leaves of infected Nebraska.

## Discussion

Results of our study demonstrated that all tested common bean cultivars (Paulista, Xera, Swiss blane, Nebraska, and Bronco) were susceptible to root-knot nematode *Meloidogyne incognita* infection under greenhouse. The obtained data in this study indicated that, the cultivars were classified into 3 categories one as highly susceptible (Paulista), two as susceptible (Xera and Swiss blane), and two as moderately resistant (Nebraska and Bronco). The present results agree with those obtained Ibrahim [13] who mentioned that all common bean cultivars in Egypt were susceptible to attack by *Meloidogyne incognita*, who found that all genotypes of common bean tested to *Meloidogyne incognita* and *Meloidogyne javanica* were susceptible to both nematodes in Nebraska. Reported that common bean cultivars (Bronco, Xera and Paulista) are moderately susceptible to root-knot nematode *Meloidogyne javanica* infection [14].

The present results demonstrated that there had a negative effect on plant growth parameters, *viz*. shoot and root length, fresh and dry shoot, root weights, and the number of pods affected by nematode infection when compared with their control plants. Percentages of reduction in shoot fresh weights ranged between 23%-29%. Paulista was the most affected cultivar, while, Xera was the least affected one. As well, the percentage of reduction in root, fresh weights ranged between 3.2%-12.8% as the highest percentage of reduction was belonging to Paulista cultivar, while the lowest was in Swiss blane. The percentages of reduction in nodules numbers ranged between 37%-86%. While in pods weights ranged between 15%-39%, the most affected cultivar was Paulista. These results are in line with Montasser, et al. [14] who reported that fresh weights of shoots and roots were highly significantly affected by the nematode infection when compared with their controls [14]. The percentage of reductions in shoot fresh weights on common bean cultivars arranged to (15.48%-45.79%) while (7.54%-50%) in roots, fresh weights. There were inversely proportional to number of galls and galling index because, number of galls showed more nematode intensity because of which plants fail to normally grow and hence fewer values for plant growth parameters. The results of the above parameters revealed that *Meloidogyne* spp., suppressed common bean growth with a standard level of inoculum when applied. Those varieties having more galls on their root showed more root weight compared to those having a smaller number of galls. Plant susceptibility/ resistance could be attributed to the prevailing nematode species or strain, some physiological and chemical factors of the plant, and soil temperature [15-17]. It is worth noting that the genotypes identified as susceptible, especially Paulista cultivars in this study are commercial cultivars and already have desirable agronomic characteristics, being readily available to producers.

Chlorophyll content in leaves as response of Paulista cultivar in response to root-knot nematode *Meloidogyne incognita*, infections was decreased significantly during different ages of plant development. The first noticeable decrease was 21 days after sowing compared with the control plants. The nematode effect on chlorophyll content was noticed on all different plant ages comparing with the control plants. These results are in agreement with Melakeberhan, et al. [18-19] who mentioned that *Meloidogyne incogita*, caused a significant decrease in total chlorophyll content and photosynthetic rate on *Phaseoles vulgaries* [18-19]. These results focused on the role of root-knot nematode infection in decreasing some plant contents, generally and chlorophyll content specifically and sequential the effect of plant-parasitic nematode on plant physiological processes such as photosynthesis and related functions [20]. Results indicated differences in buffer soluble protein extracted from the leaves of five tested bean cultivars in relation to infection with the root-knot nematode *Meloidogyne incognita*, these differences in protein banding pattern due to infection by *Meloidogyne incognita* may be attributed to the response of the host plant to infection. The data indicated that there were three (62-42-29 kDa) protein in moderately resistant (Nebraska and Bronco) and absent in susceptible cultivars. This result is in agreement with those of other investigators [21]. El-Moflehi who reported the differences in buffer soluble protein extracted from leaves of peach rootstocks in relation to infection with *Meloidogyne incognita*. The dynamics in plant protein expression, as reported here following infection of the susceptible and resistant common bean lines, are consistent with the emerging view of complexity in plant/nematode interaction. Alteration in plant gene expression during the initiation of root-knot nematode feeding sites, perhaps, explains the identical changes in protein patterns seen in susceptible and resistant lines following infection. The relative abundance of the differentially expressed (62-42-29 kDa) polypeptide and its localization to the root-knot nematode-induced galls led us to target this protein additional characterization. The (62-42-29 kDa) proteins will be useful in the design of oligonucleotide probes for amplification of the complementary DNA by polymerase chain reaction. Cloning and sequencing of the cDNA corresponding to the (62-42-29 kDa) response of these common bean lines to root-knot nematode.

## Conclusion

The tested common bean cultivars differed in their susceptibility/resistance to root-knot nematode *Meloidogyne incognita* depending on their Damage Index in combination with their growth. The highly resistant or moderately resistant common bean cultivars determined in this study could be recommended for the breeding programs and could be introduced in integrated pest management for controlling root-knot nematodes.

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