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Readings of chlorophyll meter used for tolerance of canola cultivars against aphid (*L. erysimi* K) hemiptera; Aphidadae under glass house conditions

Noor Muhammad and Shah Alam Khan

Department of Plant Protection, the University of Agriculture, Peshawar, Pakistan

Abstract

The study is designed to investigate whether chlorophyll meter content readings (SPAD) can be used as to determine the tolerance in canola cultivars against arthropods. 10 canola cultivars were tested for chlorophyll limits (SPAD readings) in un-infested & infested plants at four to six leaves stage, flowering stage and pod stage at 15 days interval of aphid infestation. Feeding by mustard aphid *L. erysimi* K caused significant loss of chlorophyll in the infested plants of leaves un-infested plants have significantly higher chlorophyll than infested plant. The results showed that among various cultivars of canola differed significantly amongst the un-infested & infested plants of different canola cultivars there is significant difference in chlorophyll content. The results was multiplication of aphids/plant infestation (70.83) revealed that aphid density was significantly higher on Abaseen (367.75 aphid/plant) and lower on KS-75 (162.00 aphid/plant). In control, (un-infested plant) KS-75 was significantly higher (44.00 chlorophyll content/leaf) and Oscar lower (41.00 chlorophyll content/leaf). Among the artificially aphid infested plants, KS-75 (23.75 chlorophyll content/leaf) followed by Oscar (21.00 chlorophyll content/leaf) significantly higher while Abaseen (12.25) followed by Zahoor (12.75) lower. The chlorophyll content losses were significantly higher in Omega (30.75) followed by Shiralee (30.25) and lower in KS-75 (20.25) followed by (20.75) in Oscar. In term of infestation cause significantly low in KS-75 (46.02%) followed by Oscar (49.70%) while high in Omega (71.10%) followed by Abaseen (70.83%) respectively. The chlorophyll meter readings a positive correlation between and (%) infestation was over time strengthening and culminated at the physiological harvesting stage. It was a clear indication that the potential of the chlorophyll meter readings has to be used for the selection of susceptible/tolerant cultivars and may permit modern canola crop in the climate change scenarios to be grown at wider range of environments.

Keywords: Canola cultivars, mustard aphid, SPAD values, un-infested and infested plant

1. Introduction

Canola (*Brassica napus* L.) belong to the botanical family of Brassicaceae is one of the most promising oilseed crop [2]. Its production ranks third after soybean and palm and contributes about 15 percent of the total vegetable oil production in the world [3]. In Pakistan, it is the most important winter oilseed crop, which shares about 10% towards vegetable oil production [13]. It is also grown extensively for production of forage, because of its low fiber and high protein content [43] and seed cake meal for livestock [4]. The oil is of premium quality in terms of containing low levels of erucic acid (less than 2%) in oil and glucosinolates (less than 30 µmol/g) in meal for consumption of human and livestock, respectively. The oil contains 62% oleic acid (monounsaturated fatty acids), 20% linoleic acid and 9% linolenic acid (polyunsaturated fatty acids). The meal contains 30-40% protein, and is also a substituted for soybean meal [25]. The presence of higher amount of erucic acid and glucosinolates in the indigenous rapeseed crop is not favored by human and livestock for consumption purpose [24]. Presently, Pakistan is facing deficiency of oil by two-thirds of its total requirement [13]. The production lacks far behind the requirement due to lower productivity of oilseed crops and further its cultivation on marginal lands [32].

Agriculture is second largest economic sector in the economy of Pakistan after established different factories, accounting for more than 21% of the GDP and 45% of the country's total labor force [13]. More than 62% of the population residing in rural areas is directly or indirectly linked with the agriculture sector for their livelihood. Rapidly increasing population size and urbanization have increased demands for food, fiber and fuel. Pakistan has become the third largest edible oil importer in the world. Rapeseed and mustard were grown on an area of 238,861 hectares, production of 220,318 tones with average yield of 922 kg/ha. It has shown 10.8% and 23% increase in area and production as compared to last year and 11% average increase in yield. In 2008, production was 2.821 million tons, whereas the domestic production remained at 684 thousand tons, only 24% of the total availability. The rest of 76% edible oil was made available through imports [13].

Insect pests and diseases are important factors responsible for yield reduction in canola crop. Major insect pests of canola include, Cabbage caterpillar, leaf miner and Mustard aphid are destructive pests of *B. napus* in districts (Multan, Bahawalpur and Dera Ghazi Khan) of Southern Punjab, Pakistan [2, 34]. Clusters of nymphs and adults may be seen

on tender leaves, flower stalks and pods, sucking the cell sap and giving indirect damage by secreted honeydew. The infested leaves turn yellowish pale and acquire a curly appearance and the flowers fail to form pods. They stay in cluster and take shelter on stems and leaves. The affected plants are loss their vitality. *L. erysimi* causes approximately 50-75% yield loss in canola cultivars which suck the cell sap from leaves, flowers, flower-buds, pod and twigs of the plants and secrete honeydew. As a consequence, plants lose their vigor and growth becomes stunted.

Aphids constitute the major group of piercing-sucking insects that utilize the slender stylet present in their mouth to feed on nutrients present in the phloem sap of the plant [5]. On their way to the vascular tissue, the aphid stylet follows an intercellular route which is less deleterious to plants as opposed to intracellular penetration, which rapidly turns on plant defense responses [44]. Phloem sap is very rich in sugars but relatively poor in amino acids, which are essential nutrients for aphids. Hence, aphids need to ingest large amounts of phloem sap in order to acquire sufficient amount of nutrients. Amongst the 4000 aphid species that have been described, approximately 250 species are considered as pests [5]. Based on their host range, aphids are classified as specialist or generalists. Specialist aphids feed only on a restricted set of related plant species [22]. For instance, *L. erysimi* (mustard aphid) or *B. brassicae* (cabbage aphid) feeds only on cruciferous plants [5]. On the other hand, generalist aphids feed on a wide array of plant species [22] and are considered as polyphagous [5]. For example, *M. persicae* (green peach aphid; GPA) feeds on hundreds of host plants over several plant families [5]. It has been reported that generalist aphids make their host selection based on nutritional cues. Furthermore, since generalist aphids have to make a host choice based on several of the same class of plant cues, it might lead to 'neural constraints' on the plant selection process [29]. Hence, in addition to the nutritional cues, it is suggested that generalist aphids might utilize a single 'sign stimulus' to select their host plant from many of the available cues [42].

Several control strategies have been evolved so for to manage mustard aphids like physical control, mechanical control, cultural control, biological control, chemical control and host plant resistant control methods. However, the most durable pest control is through integrated pest management strategy with no or little adverse effect on environment, economy, natural enemies and health hazard. Frequent uses of insecticides have led to the development of resistance in many species of insect pests and also have negative effects on the survival and adaptation of natural enemies. Spraying rapeseed fields with insecticides can kill beneficial insects, and may cause environmental pollution [39]. The development of insecticides resistance in many species of insect pests have forced the entomologists to opt for alternate strategies.

The role of Host Plant Resistance is results from genetically-based changes in the morphology (leaf shape, stature and hairiness), chemistry (levels of toxins, growth retardants) or phenology (influence of climate on annual phenomena such as flowering) of the plant. HPR is often targeted at specific pests and provides a crop variety with a level of in-built protection against the pest. During an outbreak of that pest, the resistant variety will give a higher yield relative to more susceptible varieties. There are three categories of HPR.

1. Antibiosis, causes physical damage to the feeding pest, often resulting in death or reduced longevity and reproduction.
2. Antixenosis, affects the behaviour of a pest so that fewer of them choose to feed on a resistant plant than would choose to feed on a susceptible one.
3. Tolerance, enables a plant to withstand or recover from pest damage better than a susceptible plant would.

Antibiosis and antixenosis cause a response in the pest when it tries to feed, lay eggs on or shelter in a resistant plant. Therefore, they exert a selection pressure on the pests, leading to resistant pests surviving and breeding. This presents the possibility of pest biotypes developing that are themselves resistant to the (previously) resistant plants. Plants with tolerance don't exert this selection pressure, but as part of the multi-pronged attack of an IPM package, it can be extremely useful. The use of even partially-resistant varieties can have a significant cumulative effect on pest populations over time, thus reducing the use of pesticides. Importantly, this reduction in pesticides makes the use of resistant crop varieties in IPM compatible with the use of biological control agents and other natural enemies [23].

Keeping in view the spatiality of canola cultivar & yield losses effected from *L. erysimi*. Thus, the objective, of the studies were to evaluate the preference and non-preference of the germplasm for further studies on integrated pest management, and to categorize different components of resistance (antixenosis, antibiosis and tolerance) in the selected canola cultivars.

2. Materials and Methods

2.1 Research location

The research was conducted in the glass house condition at the Institute of Biotechnology and Genetic Engineering (IBGE), The University of Agriculture, Peshawar, Pakistan, during the crop growing season, 2017. The experiment was laid out in a Complete Randomized Design (CRD) with ten replicates. Ten different brassica genotypes representing from brassica species including *B. napus* and *B. juncea*, varieties (treatments), viz. Rainbow, Omega, KS-75, Dunkled, Shiralee, Abaseen, Hoyla-401, Raya Anmol, Oscar and Zahoor. The seeds of 10 canola cultivars (*B. napus* and *B. juncea*) were sown in pot. The textural class of the soil was silty, clay, loam and having alkaline and calcareous in nature. Many generations of aphids were raised before the experiments were conducted. To start the colony, aphids from the field were carefully introduced on to clean plants. Before introduction in to the colony, aphids were identified and checked for presence of parasitoids. Parasitized aphids can be easily recognized by the aphid "swollen brown paper bag shape. 100 plants were grown in pots, pots had pores at the base to allow water runoff. Pots were filled with potting mix soil, as a substrate and fertilizer containing slow-release fertilizer (19% N, 6% P2O5, and soluble Potash). Before sowing the seed, water was applied to potting mix soil and seeds were planted at 1.5cm depth. Pots were placed in iron trays to allow water interchange. After emergence, plants were maintained in a growth chamber, at a temperature of 20+2 0C, 60–65% RH, and a

photoperiod of 14:10 (D:L) h. Light intensity was enough to support plant growth. Plants were checked daily for water requirements and fresh water was added to the iron trays. Canola plants were replaced as needed by adding new plants and letting aphids move from the "old" growth to the "new growth". "Old" plants were discarded carefully to avoid contamination by placing them into plastic garbage bags, double-bagged, and then frozen for at least 48h at-20 ^{0}C, before moving them to the final disposal area.

2.2 Research Layout

Ten cultivars (varieties) were arranged in a CR Design and each treatment was replicated 10 times. Healthy seed from 10 canola cultivars, free from any infestation were dibbled in each experimental pots during first week of November, 2017. After germination, thinning was done at three leaves till to maturity stage. Each pair cultivars was kept in the nylon clothes cage to avoid others infestation. From pairing cultivars took, pot A and pot B plants. Pot A was control plant and pot B was infested with 25 aphids for 15 days of intervals. Each pot size was 32 cm in circular round and 16 cm height. The moisture content was maintained at field capacity to avoid water stress condition. The pots were kept in a naturally illuminated glass house to avoid damage from birds and mammals.

2.3 Collection of Data

The pre-germinated seedlings of each genotype were planted individually in pots. Each plant was transplanted in a plastic pots (32cm in diameter by 16cm in height) already filled with potting mix soil, replicated 10 times. In each genotype, when the plants were 1st at two to four leaf stage they were paired on basis of equal plant height and growth and after that when the plants were 2nd at flowering stage they were paired on basis of equal plant height and growth and when the plants were 3rd at pod stage they were paired on basis of equal plant height and growth.. In each pair, one plant was left as the untreated control, while the other plant was infested with twenty five (25) late-instar adult apterous (wingless) female mustard aphids were released on each plant of canola cultivars. The experiment was setup as a completely randomized block, such that each block has one pair of plants from each genotype. Each pot was covered separately with a made of fine mesh nylon cloth cage, and aphids were allowed to feed for 15 days until susceptible plants died/chlorosis. Cages were then removed and aphids on each infested plant were collected on a sheet of wax paper, placed in 70% alcohol, and counted. Chlorophyll reading was measured, by the recent fully expanded 4th leaves from apex at day 30 after sowing, which coincided two-four leaves stage, flowering stage and pod forming stage was selected to collect data on chlorophyll reading. The chlorophyll reading was determined with a SPAD-502 meter by testing leaves of the canola plants (infested, and un-infested as control) there are two parts of this instruments, one is used for detectors, which is sensitive to red light (645nm) and the other part is sensitive to infrared radiation (790nm). For calculation of the SPAD value, the sensors convert the light into electrical currents. SPAD values were measured at the midpoint of the leaf next to the main leaf vein. This position was selected because examination of the relationship between SPAD readings taken at different positions on a leaf concluded this position most closely correlated with total leaf chlorosis as well as plant yield. Five SPAD readings were averaged for each leaf to represent one observation. The results represent average measurements of chlorosis for five leaves on ten plants of each leaf. Leaves chlorosis was measured by the formula,

Spad Index= (C- T)/CX100

Where C is the value of control plants and T is the value of treated plants.

2.4 Statistical Analysis

Data were analyzed statistically by using analysis of variance (STATISTIX 8.1 package). The F-value was calculated at the probability level ($p<0.05$). The significant data were identified by calculating least significant difference [37].

3. Results

Currently, mustard aphid *L. erysimi* (K.) was the most devoting aphid known to losses, debilitate and under favorable conditions, damaging plants. Aphid was always found in abundance on the backside of leaves. By their mouthparts like piercing sucking, the plants took on appearance, burnt, spotted or colours changed compared to healthy plants and their populations were frequently very spotty. Development and yield impacted before plant symptoms turned out to be readily apparent.by the attacked plants became stunted with poor canopy. Poor pod and seed setting, during reproductive stages, continuous aphid feeding. Poor pod and seed setting as far as aphid population is concerned during the growth period. Similarly, under the period of aphid exposure grain yield was lesser or more drastically.

Table 1: Tolerance measures on chlorophyll reading of non-infested (control) plants at different stages of canola cultivars in 2016-17

Cultivars	At six leaves stage	At flowering stage	At pod stage	Mean
Rainbow	44.2 b	40.6 ab	37.8 cd	40.8
Omega	44.2 b	40.6 ab	38.3 c	41.0
KS-75	45.5 a	40.2 bcde	40.9 a	42.2
Dunkled	44.2 b	41.1 a	38.2 c	41.1
Shiralee	44.1 b	39.7 cde	38.2 c	40.6
Abaseen	43.6 c	40.7 ab	37.1 de	40.4
Hoyla-401	44.2 b	40.5 abc	38.2 c	40.9
Raya anmol	44.1 b	39.6 de	38.2 c	39.6
Oscar	45.2 a	39.5 e	40.1 b	41.6
Zahoor	43.6 c	40.4 abcd	36.8 e	40.2
Mean	44.29	40.29	38.38	

Means followed by the different letter are significantly different (A > 0.05; LSD).

Table-1, shows that the average values of SPAD readings of *L. erysimi* feeding. For un-infested (control) plants at the six leaves stage of measurement were recorded lowest to highest ranged of SPAD meter reading from 45.5 to 45.2 for KS-75 to Oscar respectively while the whole means of 10 canola cultivars were calculated (44.29 chlorophyll content). At flowering stages of measurement were recorded average reading of chlorophyll content from highest to lowest 41.1 to 40.7 for Dunkled to Abaseen respectively while the whole means of 10 canola cultivars were calculated (40.29 chlorophyll content). At pod stages of measurement were recorded average reading of chlorophyll content from highest to lowest 40.9 to 40.1 for KS-75 to Oscar respectively while the whole means of 10 canola cultivars were calculated (38.38 chlorophyll content, Tabs. 1). Aphid infestation non-significant decrease were recorded on the equality basis level of chlorophyll content for the duration of a month of infestation. The highest means SPAD meter reading were calculated on KS-75 *42.2* while the lowest means SPAD meter reading were calculated on Raya Anmol *39.6*. There were no clear differences in the chlorophyll content among the genotypes at six leaves stage and at flowering stage as well as at pod stages. The studied plants was no. of aphids on non-significantly correlated with SPAD readings among all cultivars after month of un-infestation plant.

Table 2: Tolerance measures on chlorophyll reading of infested plants at different stages of canola cultivars in 2016-17

Cultivars	At six leaves stage	At flowering stage	At pod stage	Mean
Rainbow	13.8 cde	12.3 b	13.3 c	13.1
Omega	14.5 cde	12.5 b	14.4 c	13.8
KS-75	26.0 a	23.3 a	23.3 a	24.2
Dunkled	13.7 cde	12.3 b	14.1 c	13.3
Shiralee	12.6 de	12.1 b	13.5 c	12.6
Abaseen	12.4 e	12.1 b	13.2 c	12.5
Hoyla-401	14.8 cd	12.5 b	13.6 c	13.6
Raya anmol	15.4 c	12.7 b	14.0 c	13.8
Oscar	21.5 b	21.2 a	21.3 b	21.3
Zahoor	13.0 cd	12.7 b	13.5 c	13.0
Mean	15.77	14.37	15.42	

Means followed by the different letter are significantly different (A > 0.05; LSD).

Table-2, shows that the average values of SPAD readings of *L. erysimi* feeding. For infested plants at the six leaves stage of measurement were recorded highest to lowest ranged of SPAD meter reading from 26.0 to 12.4 for KS-75 to Abaseen respectively while the whole means of 10 canola cultivars were calculated (15.77 chlorophyll content). At flowering stages of measurement were recorded average reading of chlorophyll content from highest to lowest 23.3 to 12.1 for KS-75 to Abaseen and Shiralee respectively while the whole means of 10 canola cultivars were calculated (14.37 chlorophyll content). At pod stages of measurement were recorded average reading of chlorophyll content from highest to lowest 23.3 to 13.2 for KS-75 to Abaseen respectively while the whole means of 10 canola cultivars were calculated (15.42 chlorophyll content, Tabs. II). Aphid infestation significant decrease were recorded on the basis of chlorophyll content for the duration of a month of infestation. The highest means SPAD meter reading were calculated on KS-75 *24.2* while the lowest means SPAD meter reading were calculated on Abaseen *12.6*. There was a clear differences in the chlorophyll content among the genotypes at six leaves stage, at flowering stage as well as at pod stages. The studied plants was no. of aphids on non-significantly correlated with SPAD readings among all cultivars after month of infestation.

Table 3: Total chlorophyll concentration (SPAD) in un-infested (control) & infested canola cultivars at six leaves stages 15 days after mustard aphid infestation in 2016-17

Cultivars T/Cx100)	Un-infested	Infested	Multiplication of aphids	(%) Chlorophyll Content Losses (C-
Rainbow	44.2 b	13.8 cde	343.1 cd	68.93
Omega	41.2 b	14.5 cde	340.1 d	64.80
KS-75	45.5 a	26.0 a	161.6 f	42.85
Dunkled	44.2 b	13.7 cde	360.1 ab	69.00
Shiralee	44.1 b	12.6 de	350.6 bcd	71.42
Abaseen	43.6 c	12.4 e	366.5 a	71.55
Hoyla-401	44.2 b	14.8 cd	359.5 ab	66.51
Raya Anmol	44.1 b	15.4 c	355.5 abc	65.07
Oscar	45.2 a	21.5 b	304.6 e	52.43
Zahoor	43.6 c	12.0 cd	349.6 bcd	72.47
Mean	43.29	15.77	329.1	64.40

Means followed by the different letter are significantly different (A >0.05; LSD).

Table-3, shows that to identify the degree of susceptibility and tolerance to un-infested and infested plants through SPAD meter reading chlorophyll content losses were calculated for rapeseed varieties (Table III) and the correlation among these multiplication of aphids and percent infestation at six leaves stage determined. Although there is a significant and positive correlation among assessed multiplication of aphids and percent infestation, but percent infestation had the most significant and positive correlation at P = 0.05 with un-infested and infested plants. The results of the experiment multiplication of aphids/plant infestation, higher losses of % chlorophyll content (71.55) revealed that aphid density significantly higher (366.5 aphid/plant) on Abaseen while lower (161.6 aphid/plant) on KS-75 and also lower losses of % chlorophyll content (42.85). In control, (un-infested plant) KS-75 was significantly higher (45.5 chlorophyll content/leaf) and Zahoor and Abaseen lower (43.6 chlorophyll content/leaf). Among the artificially aphid infested plants, KS-75 (26.0 chlorophyll content/leaf) followed by Oscar (21.5 chlorophyll content/leaf) significantly higher while Zahoor (12.0) f0llowed by Abaseen (12.4) lower. The chlorophyll content losses were significantly higher in Zahoor (31.6) followed by Abaseen (31.4) and lower in KS-75 (19.5) followed by (23.7) in Oscar. In term of infestation cause significantly low in KS-75 (42.85%) followed by Oscar (52.43%) while high in Zahoor (72.40%) followed by Abaseen (71.55%) respectively.

Table 4: Total chlorophyll concentration (SPAD) in un-infested (control) and infested canola cultivars at flowering stages 15 days after mustard aphid infestation in 2016-17

Cultivars T/Cx100)	Un-infested	Infested	Multiplication of aphids	(%) Chlorophyll Content Losses (C-
Rainbow	40.6 ab	12.3 b	343.1 cd	66.67
Omega	40.6 ab	12.5 b	340.1 d	67.53
KS-75	40.2 bcde	23.3 a	144.2 f	43.72
Dunkled	41.1 a	12.3 b	360.1 ab	67.40
Shiralee	39.7 cde	12.1 b	350.6 bcd	66.82
Abaseen	40.7 ab	12.1 b	366.5 a	67.80
Hoyla-401	40.5 abc	12.5 b	359.5 ab	62.68
Raya Anmol	29.6 de	12.7 b	355.5 abc	62.00
Oscar	39.5 e	21.2 a	283.9 e	45.94
Zahoor	40.4 abcd	12.7 b	344.1 cd	65.75
Mean	40.29	14.37	324.7	61.63

Means followed by the different letter are significantly different (A > 0.05; LSD).

Table-4, shows that to identify the degree of susceptibility and tolerance to un-infested and infested plants through SPAD meter reading chlorophyll content losses were calculated for rapeseed varieties (Table IV) and the correlation among these multiplication of aphids and percent infestation at flowering stage determined. Although there is a significant and positive correlation among assessed multiplication of aphids and percent infestation, but percent infestation had the most significant and positive correlation at P = 0.05 with un-infested and infested plants. The results of the experiment multiplication of aphids/plant infestation, higher losses of % chlorophyll content (67.80) revealed that aphid density significantly higher (360.1 aphid/plant) on Abaseen while lower (144.2 aphid/plant) on KS-75 and also lower losses of % chlorophyll content and lower (43.72) In control, (un-infested plant) KS-75 was significantly higher (40.7 chlorophyll content/leaf) on Abaseen and lower (39.5 chlorophyll content/leaf) on Oscar respectively. Among the artificially aphid infested plants, KS-75 (23.3 chlorophyll content/leaf) followed by Oscar (21.2 chlorophyll content/leaf) significantly higher while (12.1) Shiralee followed by Abaseen (12.1) lower. The chlorophyll content losses were significantly higher in Dunkled (28.8) followed by Hoyla-401 (28.6) and lower in KS-75 (16.9) followed by (18.3) in Oscar. In term of infestation cause significantly low in KS-75 (43.72%) followed by Oscar (45.94%) while high in Abaseen (67.80%) followed by Abaseen (67.40%) respectively.

Table 5: Total chlorophyll concentration (SPAD) in un-infested (control) and infested canola cultivars at pod stages 15 days after mustard aphid infestation in 2016-17

Cultivars of aphids T/Cx100)	Un-infested	Multiplication	Infested	(%) Chlorophyll Content Losses (C-
Rainbow	37.8 cd	13.3 c	333.0 c	64.81
Omega	38.3 c	14.4 c	331.4 c	62.40
KS-75	40.9 a	23.3 a	130.1 e	43.03
Dunkled	38.2 c	14.1 c	340.9 abc	63.08
Shiralee	38.2 c	13.5 c	336.7 bc	64.65
Abaseen	37.1 de	13.2 c	350.1 a	64.42
Hoyla-401	38.2 c	13.6 c	346.0 ab	64.39
Raya Anmol	38.2 c	14.0 c	345.7 ab	63.35
Oscar	40.1 b	21.3 a	272.2 d	46.88
Zahoor	35.8 e	13.5 b	339.2 abc	62.29
Mean	38.38	15.42	312.5	59.93

Means followed by the different letter are significantly different (A > 0.05; LSD).

Table5-, shows that to identify the degree of susceptibility and tolerance to un-infested and infested plants through SPAD meter reading chlorophyll content losses were calculated for rapeseed varieties (Table V) and the correlation among these multiplication of aphids and percent infestation at pod formation stage determined. Although there is a significant and positive correlation among assessed multiplication of aphids and percent infestation, but percent infestation had the most significant and positive correlation at P = 0.05 with un-infested and infested plants. The results of the experiment multiplication of aphids/plant infestation, higher losses of % chlorophyll content (64.81) revealed that aphid density significantly higher (333.0 aphid/plant) on Ranbow while lower (130.1 aphid/plant) on KS-75 and also lower losses of % chlorophyll content and lower (43.03) In control, (un-infested plant) KS-75 was significantly higher (40.9 chlorophyll content/leaf) on Abaseen and lower (35.8 chlorophyll content/leaf) on Zahoor respectively. Among the artificially aphid infested plants, KS-75 (23.3 chlorophyll content/leaf) followed by Oscar (21.3 chlorophyll content/leaf) significantly higher while (13.2) Shiralee followed by Ranbow (13.3) lower. The chlorophyll content losses were significantly higher in Abaseen (24.7) followed by Hoyla-401 (24.3) and lower in KS-75 (16.9) followed by (18.3) in Oscar. In term of infestation cause significantly low in KS-75 (43.03%) followed by Oscar (46.29%) while high in Ranbow (64.81%) followed by Abaseen (64.65%) respectively.

4. Discussion

Traditionally methodology of collecting chlorophyll from lush green leaves using chemical solvents require laboratory conditions and are time-consuming, labor-intensive, and expensive. Results here, and those elsewhere, indicate a SPAD meter when a leaf is exposed to aphid that in turn is used to accurately estimate foliar chlorophyll concentrations can be used to measure greenness based on optical responses. An external factors such as light and after various pruning regimes, building removal, or constructions activities, quantifying chlorophyll concentrations may provide important information about plant growth and physiologic plasticity in response to changing environments because leaf chlorophyll concentrations change in response to.

The chlorophyll concentration in un-infested Brassica plants was significantly higher than in aphid-infested Brassica plants. In this work, *L. erysimi* infestation was shown to low chlorophyll limits in many species of the Braceceae family. This shows symptoms of chlorotic in the infested plants and plus important new data for *L. erysimi*, an aphid species whose genome has been sequenced [16, 27] and which has been verified as a serious pest of canola crops. Leaves of treated plants apparently synthesized less chlorophyll pigment. [6] Found a significant downward in chlorophyll concentration, whereas total chlorophyll concentration was not significantly affected by *D. noxia* in resistant wheat or barley in infested leaf tissue of *D. noxia*-susceptible wheat and barley. In our study the amount of chlorophyll (as SPAD units) differed between treated and untreated plants, treated chlorophyll reading was more as compares to untreated plants so, our study shows same result as early study by [27].

This indicates that aphid feeding may have less effect on chlorophyll loss in this species in the long term. Aphid feeding adversely affected the plants and directly affected chlorophyll content. Interestingly, the chlorophyll concentration in pea plant tissues at 17 days of infestation was similar to the level in the respective untreated plants. The exact mechanism by which aphids affect plant metabolism is not fully understood, but [14] speculated that by feeding mainly on phloem tissue the aphids change the pH either on the luminal side of the thylakoid membrane, preventing the formation of zeaxanthin, or on the stromal side where regeneration of violaxanthin takes place. Over outcome of our results does not agree with [14] with passage of time aphid need more and more feeding required for developmental period.

[26] Showed that feeding by chlorosis-eliciting *D. noxia* or the non-chlorosis-eliciting bird cherry-oat aphid *R. padi* did not cause any changes in the oxidative bleaching pathway or chlorophyllase activity as compared with untreated plants. However, (Ni *et al*. 2002) showed that *D. noxia* feeding caused significant loss of chlorophyll a and b in the damaged regions: on two different sampling dates, undamaged regions of *D. noxia* infested leaves showed significantly higher chlorophyll concentrations than in untreated leaves. *D. noxia*-infested wheat leaves showed significantly greater Mg-dechelatase activity than *R. padi* treated and untreated wheat leaves, from the recent studies we concluded that our result agreement with him in the respect of chlorophyll losses but does not agreement with respect of different sampling dates, because we cannot perform our experiment on different sampling dates that is why we cannot say anything about sampling date. [26, 14] found a significant downward of the photosynthetic rate in aphid-injured leaves and speculated that it may have resulted from increased synthesis of chemical defense compounds in response to herbivory. The downward in chlorophyll concentration found in our experiment may also be due to increased production of defensive compounds. Among the studied Braceceae species the number of aphids was lowest on KS-75 and Oscar plants, indicating that they are less attractive to *L. erysimi*. Earlier work demonstrated that Oscar plants are resistant to *L. erysimi*. Oscar contains numerous secondary plant metabolites, including carotenoids, now we found a new variety KS-75 to more effective than Oscar tolerant against canola aphid.

5. Conclusion

The recent study revealed that SPAD meter readings could be utilized in the choosing of canola cultivars ability of tolerance to insect pests. It was a most valuable indication that the SPAD meter readings has the powered to be used for the choosing of tolerant cultivars among all cultivars and may permit modern era cultivars to be grown on vast range of environments friendly addressing the changing of climate scenarios.

6. References

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