Performance of selected phytochemicals and
 botanical nutrient on physiological features of
 okra varieties against Yellow vein clearing
 mosaic virus

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# 10 Abstract

A two factorial field experiment on okra was conducted at the Horticulture Farm of Sher-e-Bangla 11 12 Agricultural University, Dhaka during April to August, 2017. The aim of the study was to find out the 13 physiological responses of okra varieties against Yellow vein clearing mosaic virus (YVCMV), under a 14 management progam to control the disease. Four okra varieties viz. BARI dherosh-1, Green finger, 15 Orca onamika and Nuffield were selected as first factor and two phytochemicals (Imidacloprid and Sobicron) and one botanical nutrient namely Peak performance nutrients (PPN) were used as second 16 17 factor. The plants were grown and natural inoculum was relied upon the infection of YVCMV. Physiological features of okra plants were significantly affected by Yellow vein clearing mosaic virus 18 19 (YVCMV). So that normal physiological activities of okra varieties were hampered due the attack of 20 the virus. The highest net chlorophyll content, net assimilation rate, intercellular carbon-di-oxide 21 concentration, stomatal conductivity and respiration rate were recorded in Green finger and the lowest 22 in Orca onamika. Sobicron with PPN also gave the highest net chlorophyll content, net assimilation 23 rate, intercellular carbon-di-oxide concentration, stomatal conductivity and respiration rate and the 24 lowest were recorded when no phytochemicals and PPN combination was used. Green finger with the 25 application of Sobicron with PPN showed the best performance of these physiological responses 26 compared to other okra varieties and phytochemicals combinations.

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Keywords: Okra, Yellow vein clearing mosaic virus, phytochemicals, Peak performance nutrients,
 physiological features.

# 30 **1. Introduction**

31 Okra (Abelmoschus esculentus L.) is an important vegetable crop belonging to the Malvaceae 32 family. It is used for food as a vegetable and industrially as a fibre. It is also the good sources of gum, 33 starch, spice and medicinal (eg. diabetese and cancer) products. The green fruits of okra are rich sources of carbohydrate, proteins, vitamins, calcium, potassium and other minerals. The edible 34 35 portion of pod (100 gm) has moderate levels of vitamin A (0.01 mg) and vitamin C (18 gm), calcium (90mg), phosphorus and potassium. The contents of thiamine (0.07 mg), riboflavin (0.08 mg) and 36 37 niacin (0.08 mg) per 100 gm edible portion of pod are higher than many others vegetables. Okra is said to be very useful against genito-urinary disorders, spermatorrhoea and chronic dysentery. Its 38 39 medicinal value has also been reported in curing ulcers and relief from hemorrhoids,

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Yellow vein clearing mosaic virus (YVCMV) is a member of Geminivirus group which is semipersistently transmitted by whitefly (*Bemesia tabaci*) [1,3]. The virus is also transmitted through grafting, but not mechanically or through seeds [2,17]. Okra YVCMV has been considered as the most important factor of yield reduction in Bangladesh and some other okra growing regions of the sub-continent [4,5]. It is one of the most destructive diseases of okra plants. In the current study, 46 effect of okra YVCMV was assessed on plant growth and yield in naturally infected crop [7,8,9]. The 47 virus showed the significant reduction in plant height, number of leaves, flowers, fruits and over all 48 pickings and yield [19,20]. The significant reduction in plant height, flowers formation per plant, fruit 49 weight in infected plants were recorded as compared to healthy plants [6]. Interestingly, on overall 50 basis there were more numbers of leaves in infected plants as compared to healthy one [11,12]. The growth and development of okra plants depend on its normal physiological process. The pathogen 51 52 may change the normal physiological processes of the infected plants [10,13]. There are some 53 reports on biochemical changes and metabolic activities of okra plants are significantly affected due to 54 the virus infection [14,18]. Information regarding the virus infection causing tremendous yield losses of 55 okra are available [15,16]. But the study on physiological changes due to the virus in infected okra is 56 scanty.

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58 The prime objectives of the experiment were to determine the changes on physiological features and 59 cellular components due to infection of *YVCMV* in okra and to evaluate the performance of 60 phytochemicals and PPN on physiological features and cellular components of okra.

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62 2. Materials and methods

# 63 2.1 Materials used in the experiment

Four okra varieties namely BARI dherosh-1, Green finger, Nuffield and Orca onamika were used in the experiment. BARI dherosh-1 was collected from Bangladesh Agricultural Research Institute (BARI), Joydevpur, Gazipur and other three varieties were collected from the local market. The selected phytochemicals namely Imidacloprid and Sobicron were collected from the local market and PPN was collected from China through representative country dealers.

# 69 **2.2 Design and layout of the experiment**

The experiment was laid out in a randomized complete block design (RCBD) with three replications where blocks were representing the replication. Each block comprised 18 unit plot and a total number of plots were 72 (18 X 4=72). Size of each unit plot was 5 m<sup>2</sup>. The distance between plot to plot was 0.70 m and block to block was 1 m.

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# 75 **2.3 Intercultural operations and fertilizer application**

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77 Preparation of land was done as per treatment. After the establishment of seedlings, various 78 intercultural operations were accomplished for better growth and development of the okra. Proper 79 intercultural operation facilities e.g. thinning and gap filling, weeding and mulching, irrigation and 80 drainage were provided at the right time. Cow dung, TSP, MP, and Urea were applied at the rate of 81 14 ton/ha, 150 kg/ha, 150 kg/ha and 150 kg/ha respectively. The entire amount of cow dung, TSP, 82 and MP @ 100 kg/ha were applied at the time of final land preparation. The remaining TSP and MP 83 were applied after 30 days of sowing seed. Urea was applied in three equal installments at 30, 45 and 84 60 days after sowing (DAS). 85

# 86 2.4 Obsevation

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88 The virus produces typical vein yellowing and thickening of leaves forming a network of veins and 89 veinlets in the infected leaves. Initially, the leaves exhibit only vellow colored veins but under the 90 severe infection, the leaves become completely chlorotic and turn yellow. There is reduction of leaf 91 chlorophyll and the infected plants give a stunted look and produce small sized pale yellow fruits. If 92 plants are infected within 20 days after germination, their growth is retarded; few leaves and fruits are 93 formed and loss may be about 90% . The extent of damage declines with delay in infection of the 94 plants. The average chlorophyll content in the leaves of the selected plants was recorded with the 95 help of "S-pad" meter, which is an advanced technology to direct measure of the chlorophyll content 96 in plant leaf at 40, 60 and 80 days after sowing (DAS). In each reading of single leaf was recorded by 97 the machine for three times at three location of the same leaf then the machine automatically gave the 98 average data and value. But the average net assimilation rate, intercellular carbon-di-oxide 99 concentration, respiration rate and stomatal conductivity per plant were recorded from the selected 100 plants by using "LC-Pro+" machine at 40, 60 and 80 days after sowing (DAS).

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### 102 **2.5 Data collection and Statistical analysis**

A number of plants and number of infected plants from each plot at 40, 60 and 80 days after sowing (DAS) were recorded. The data were analyzed statistically by using the analysis of variance (ANOVA) and MSTAT-C software for proper interpretation. The mean value was compared according to Duncan's Multiple Range Test (DMRT) at 1% level of significance. Correlation and regression study was also done to check the relationship among the varieties, insecticides, physiological features and yield. Tables and bar graph were used to interpret the data when required.

109 110 **3. Results** 

# 3.1 Effect of varieties on Net chlorophyll content ( $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>), Net assimilation rate (g m<sup>-2</sup> d<sup>-1</sup>) and Intercellular carbon-di-oxide concentration (ppm)

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The highest observation of net chlorophyll content, net assimilation rate and also intercellular carbondi-oxide were observed for per plant in Green finger (50.39 a, 1.650 a and 5.833 a) which were just more than BARI dherosh-1 (48.24 b, 1.463 b and 4.833 b). The lowest observations of these physiological features were recorded in Orca onamika (39.11 d, 1.140 d and 4.000 c) which were just less than Nuffield (46.15 c, 1.240 c and 4.500 bc). These results are presented in Table 1.

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### Table 1: Effect of varieties on Net chlorophyll content (μ mol m-2 s-1), Net assimilation rate (g m-2 d-1) and Intercellular Carbon-di-oxide concentration (ppm)

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Variety	Net chlorophyll content (μ mol m <sup>-2</sup> s <sup>-1</sup> )	Net assimilation rate (g m <sup>-2</sup> d <sup>-1</sup> )	Intercellular Carbon- di-oxide concentration (ppm)
BARI Dherosh-1	48.24 b	1.463 b	4.833 b
Green finger	50.39 a	1.650 a	5.833 a
Orca onamika	39.11 d	1.140 d	4.000 c
Nuffield	46.15 c	1.240 c	4.500 bc
LSD 0.01	0.6521	0.02832	0.7689
CV%	1.58	1.78	17.92

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# 124 **3.2** Effect of varieties on Stomatal conductivity (mol m<sup>-2</sup> s<sup>-1</sup>) and Respiration rate (ppt/s)

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The highest stomatal conductivity and respiration rate were found in Green finger (0.2333 a and 40.82 a) which were more than BARI dherosh-1 (0.2206 a and 36.57 b). The lowest stomatal conductivity and respiration rate were found in Orca onamika (0.1867 b and 33.98 d) which were less than Nuffield (0.2083 ab and 35.77 c). The results are presented in Table 2.

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# 132 Table 2:Effect of varieties on Stomatal conductivity (mol m-2 s-1) and Respiration rate (ppt/s)

Variety	Stomatal conductivity (mol m <sup>-2</sup> s <sup>-1</sup> )	Respiration rate (ppt/s)
BARI Dherosh-1	0.2206 a	36.57 b
Green finger	0.2333 a	40.82 a
Orca onamika	0.1867 b	33.98 d
Nuffield	0.2083 ab	35.77 с

LSD 0.01	0.02832	0.2157
CV%	11.29	0.66

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### 3.3 Effect of phytochemicals and PPN on Net chlorophyll content ( $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>), Net assimilation rate (q m<sup>-2</sup> d<sup>-1</sup>) and Intercellular Carbon-di-oxide concentration (ppm) 136

137 Application of Sobicron with PPN (48.56 a, 1.405 a and 7.250 a) provided the highest net chlorophyll 138 content, net assimilation rate and also intercellular carbon-di-oxide respectively. Moderate 139 performance was observed when any one of Imidacloprid with PPN (48.06 a, 1.385 ab and 5.500 b), Imidacloprid (45.70 bc, 1.378 ab and 4.000 cd), Sobicron (46.44 b, 1.375 ab and 4.000 cd) and PPN 140 141 (45.06 c, 1.360 bc and 4.750 bc) were applied. When application with no insecticides and PPN (42.03 d, 1.337 c and 3.250 d) were applied gave the lowest performance of these physiological responses. 142 143 These results are presented in Table 3.

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#### 145 Table 3: Effect of phytochemicals and PPN on Net chlorophyll content (µ mol m-2 s-1), Net 146 assimilation rate (g m-2 d-1) and Intercellular Carbon-di-oxide concentration (ppm)

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Phytochemicals and PPN	Net chlorophyll content (μ mol m <sup>-2</sup> s <sup>-1</sup> )	Net assimilation rate (g m <sup>-2</sup> d <sup>-1</sup> )	Intercellular Carbon-di- oxide concentration (ppm)
Control	42.03 d	1.337 c	3.250 d
PPN	45.06 c	1.360 bc	4.750 bc
Imidacloprid + PPN	48.06 a	1.385 ab	5.500 b
Sobicron + PPN	48.56 a	1.405 a	7.250 a
Imidacloprid	45.70 bc	1.378 ab	4.000 cd
Sobicron	46.44 b	1.375 ab	4.000 cd
LSD 0.01	0.7986	0.03469	0.9417
CV%	1.58	1.78	17.92

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#### 3.4 Effect of phytochemicals and PPN on Stomatal conductivity (mol m<sup>-2</sup> s<sup>-1</sup>) and Respiration 149 150 rate (ppt/s)

151 The highest observations of stomatal conductivity and respiration rate were found when Sobicron 152 with PPN (0.2458 a and 39.78 a) was applied. Moderate performance were found when any one of 153 Imidacloprid with PPN (0.2183 bc and 37.28 b), Imidacloprid (0.2142 abc and 36.97 c), Sobicron 154 (0.2125 abc and 36.35 d) and PPN (0.2050 ab and 37.28 b) were applied. The lowest stomatal 155 conductivity and respiration rate were found with the application of no insecticides and PPN (0.1775 c 156 and 33.05 e). The results are presented in Table 4.

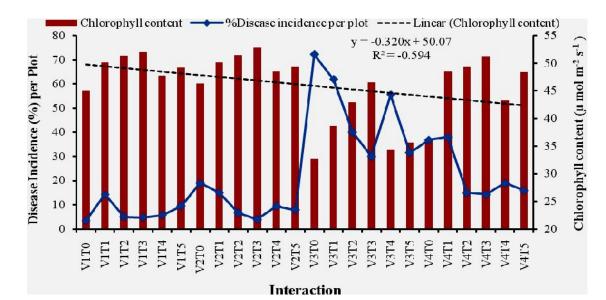
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#### Table 4: Effect of phytochemicals and PPN on Stomatal conductivity (mol m<sup>-2</sup> s<sup>-1</sup>) and 158 159 Respiration rate (ppt/s)

Phytochemicals and PPN	Stomatal conductivity (mol m-2 s-1)	Respiration rate (ppt/s)
Control	0.1775 c	33.05 e
PPN	0.2050 ab	37.28 b
Imidacloprid + PPN	0.2183 bc	37.28 b
Sobicron + PPN	0.2458 a	39.78 a
Imidacloprid	0.2142 abc	36.97 c
Sobicron	0.2125 abc	36.35 d
LSD 0.01	0.03469	0.2642
CV%	11.29	0.66

# 160 3.5 Correlation coefficient between chlorophyll content (μ mol m<sup>-2</sup> s<sup>-1</sup>) and Disease Incidence 161 (%) per Plot

162 Correlation coefficient study was done to establish the relationship between the chlorophyll content ( $\mu$ 163 mol m<sup>-2</sup> s<sup>-1</sup>) and disease incidence (%) per plot. From the study it was revealed that significant 164 correlation was observed between the parameters (Figure 1). It was evident from the Figure 1, the 165 equation y = -0.320x + 50.07 gave a good fit to the data, and the co-efficient of determination (R<sup>2</sup> = -166 0.594) showed that, fitted regression line had a significant regression co-efficient. From these 167 relations it can be concluded that the disease incidence (%) per plot was negatively (slope= -0.183) 168 correlated with the chlorophyll content of okra plants.



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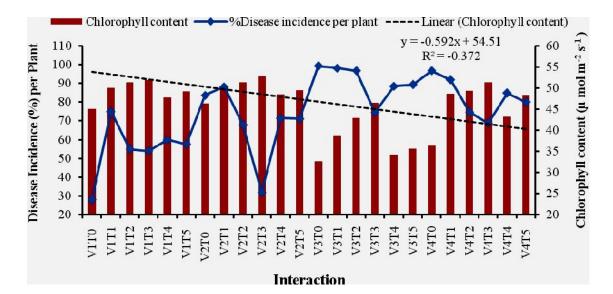
### 170 Figure 1:Relationship between Chlorophyll content and Disease incidence (%) per Plot.

### 171 Figure 1. Relationship between Chlorophyll content and Disease incidence (%) per Plot.

V1= BARI dherosh-1, V2= Green finger, V3= Orca onamika, V4= Nuffield, T0= Control (No
phytochemicals and PPN), T1= Peak Performance Nutrients (PPN), T2= Imidacloprid with PPN, T3=
Sobicron with PPN, T4= Imidacloprid, T5= Sobicron.

# 3.6 Correlation coefficient between chlorophyll content (μ mol m<sup>-2</sup> s<sup>-1</sup>) and Disease Incidence (%) per Plant

177 Correlation coefficient study was done to establish the relationship between the chlorophyll content ( $\mu$ 178 mol m<sup>-2</sup> s<sup>-1</sup>) and disease incidence (%) per plant. From the study it was revealed that significant 179 correlation was observed between the parameters (Figure 2). It was evident from the Figure 2, the 180 equation y = -0.592x + 54.51 gave a good fit to the data, and the co-efficient of determination (R<sup>2</sup> = -181 0.372) showed fitted regression line had a significant regression co-efficient. From these relations it 182 can be concluded that the disease incidence (%) per plant was negatively (slope= -0.115) correlated 183 with the chlorophyll content of okra plants.



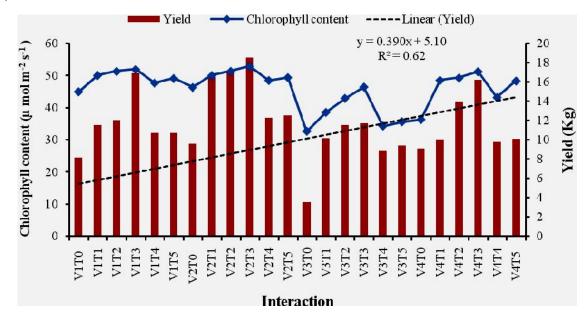
### 184 185 Figure 2. Relationship between Chlorophyll content and Disease incidence (%) per Plant.

186 V1= BARI dherosh-1, V2= Green finger, V3= Orca onamika, V4= Nuffield, T0= Control (No 187 phytochemicals and PPN), T1= Peak Performance Nutrients (PPN), T2= Imidacloprid with PPN, T3= 188 Sobicron with PPN, T4= Imidacloprid, T5= Sobicron.

# 189 3.7 Correlation coefficient between chlorophyll content (μ mol m<sup>-2</sup> s<sup>-1</sup>) and Yield (kg)

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191 Correlation coefficient study was done to establish the relationship between the chlorophyll content ( $\mu$ 192 mol m<sup>-2</sup> s<sup>-1</sup>) and Yield (kg) of okra plants. From the study it was revealed that significant correlation 193 was observed between the parameters (Figure 3). It was evident from the Figure 3, the equation y = 194 0.390x + 5.10 gave a good fit to the data, and the co-efficient of determination (R<sup>2</sup> = 0.62) showed 195 fitted regression line had a significant regression co-efficient. From these relations it can be concluded 196 that the chlorophyll content of okra was positively (slope= 0.367) correlated with the yield of okra 197 plants.



198 199 Figure 3: Relationship between Chlorophyll content and Yield

V1= BARI dherosh-1, V2= Green finger, V3= Orca onamika, V4= Nuffield, T0= Control (No
 phytochemicals and PPN), T1= Peak Performance Nutrients (PPN), T2= Imidacloprid with PPN, T3=
 Sobicron with PPN, T4= Imidacloprid, T5= Sobicron.

# 203 4. Discussion

In case of variety, all of these physiological contents mention as before, Green finger showed the best performance where Orca onamica gave very poor performance. BARI dherosh-1 and Nuffield can be considered as moderate performer response to these physiological features. Among the phytochemicals, Sobicron with PPN combinations performed as better than any other phytochemicals where minimum outcome was come from the application of no phytochemicals with PPN. Green finger with the application of Sobicron with PPN showed the best physiological performance at all growth stages as compared to other varieties and phytochemicals.

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212 Correlation coefficient and linear regression were performed to determine the relationship between 213 chlorophyll content with disease incidence per plot and plant and fruit yield. From the correlation 214 study, it was revealed that the disease incidence per plot as well as plant was negatively correlated 215 with the chlorophyll content and the chlorophyll content was positively correlated with the fruit yield. 216 Chlorophyll contents were increased with the decreases of disease incidence. Yield of okra was 217 increased with the increases of chlorophyll contents. Orca onamika application with no 218 phytochemicals and PPN showed the highest disease incidence per plot and plant with the lowest 219 chlorophyll content and lowest fruit yield.

# 220 **5. Conclusion**

221 Yield and yield contributing characters and physiological features of okra plants are changed due to 222 the infection of Yellow vein clearing mosaic virus (YVCMV), which cause serious damages of okra 223 production and reduce the market value. Development of host resistance to the virus is one of the 224 important strategy against the okra YVCMV, which is the most economical and environment friendly 225 process for reducing the yield potential of okra. But due to the sterility problem of YVCMV, it is not 226 easy to transfer the resistant gene directly among the cultivated okra varieties. Restoration of fertility 227 through colchicine treatment in the crosses between resistant wild and susceptible species could be a 228 suitable technique. Pathologists and breeders are advised to work more on evaluation of resistant 229 varieties using advanced molecular tools. The growers also should be taken preventive as well as 230 curative control measures to reduce the yield losses.

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