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Short communication

Changes in biochemical components in *Aloe vera* (L.) Burm. f. leaves infected with *Fusarium proliferatum* (Matsushima) Nirenberg

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ABSTRACT

Aloe vera (L.) Burm. f., an important medicinal plant suffers with a huge loss in its yield due to the attack of number of fungal pathogens. The aim of the present study was to analyze the biochemical changes in *A. vera* infected by *Fusarium proliferatum* (Matsushima) Nirenberg causing leaf spot disease in plant. Artificially infested leaves were examined to study the effect of fungal pathogen on chlorophyll-*a*, chlorophyll-*b*, total chlorophyll, total soluble sugar, total phenols, total anthraquinones, total flavanoids and vitamin E. Results showed a significant decrease in the contents of chlorophyll-*a*, chlorophyll-*b*, total chlorophyll and total soluble sugar in infested leaves as compared to control leaves. Whereas, total phenolics, total anthraquinones, total flavonoid and vitamin E contents were significantly increased in infested leaves as compared to control. Changes in the concentration of biochemical were directly associated with the adverse effect of pathogen during the progression of infection on *A. vera* leaves.

1. Introduction

Aloe vera (L.) Burm. f. is a succulent, evergreen, popular ornamental plant grown all around the world. It is considered as an ancient herb known from centuries for its vast role in Ayurveda, Unani, Siddha and Indian system of Medicine. Four species namely, *Aloe forbesii*, *A. inermis*, *A. ferox* and *A. barbadensis* are reported to occur in many parts of India in which, *A. barbadensis* (*A. vera*) is the most widely cultivated species grown mainly in Rajasthan, Andhra Pradesh, Gujarat and some parts of Tamil Nadu, Madhya Pradesh and Maharashtra (Maiti and Chandra, 2002). Various bioactive compounds having different pharmacological properties are found both in the gel as well as latex of the leaves. Its gel has antimicrobial properties and can be used as the treatment for cough, ulcers, muscle pain, headaches, constipation, pain relief and also curative agent for heart problems. Externally, fresh aloe gel has a very good effect in acne, pimples, eczema, burns and in the treatment of radiation dermatitis. The bitter latex has been used as laxative and to increase intestinal peristalsis (Ishii et al., 1994; Boudreau and Beland, 2006).

Being a medicinal plant, *A. vera* is prone to attack by many fungi (Harsh et al., 1990; Dubey and Pandey, 2009; Majumdar et al., 2007), bacteria (Jin et al., 1994) and nematodes (Esser et al., 1986). Amongst all, fungi are considered to be one the most destructive enemy of the plant, which diminishes the quality and quantity of gel. The phytopathogenic fungi are the chief infectious agents causing alterations in morphological characters as well as production of several secondary metabolites of the host plant.

A number of fungal pathogens are found to be associated with leaf spot disease of *A. vera*. Pritam and Kale (2007) noticed that infection of *Alternaria alternata* decreased phenols and flavonoids contents in *A. vera* gel and also diminished the antioxidant potential of the gel. *Fusarium proliferatum*, a broadly distributed fungal pathogen, can cause destructive diseases to an extremely wide range of hosts viz. *Allium sativum* (Seefelder et al., 2002), *Glycine max* (Chang et al., 2015), *Zea mays* (Peltomaa et al., 2016), *Solanum lycopersicum* (Gao et al., 2016) and *Aloe vera* (Avasthi et al., 2018). In addition, *F. proliferatum* has been reported to produce mycotoxins like fusaric acid, fumonisin, fusaproliferin and moniliformin that pose a serious threat to food safety and human health (Rheeder et al., 2002; Palmero et al., 2012; Isack et al., 2014; Gil-Serna et al., 2016). Fungal pathogens seek to manipulate the metabolism of plant for their requirement and cause an increased demand for assimilation in the plant (Swarbrick et al., 2006). Therefore, the aim of this study was to investigate the changes in various biochemical constituents of *A. vera* infected by *Fusarium proliferatum* (Matsushima) Nirenberg.

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2. Material and methods

2.1. Plant material

Healthy plants of *A. vera* were collected from the different nurseries of Gwalior city. Plant specimens were identified by Dr. Ashok Kumar Jain, Director, Center for Ethanobotany, Jiwaji University, Gwalior, Madhya Pradesh, India. The leaves of *A. vera* plants were surface sterilized with 1% sodium hypochlorite (NaOCl) solution, grown in sterilized earthen pots, filled with soil and compost (Farm Yard Manure FYM) 3:1 ratio and maintained under greenhouse conditions until they were reached at nine months old.

2.2. Fungal strain and inoculation experiment

Fusarium proliferatum was isolated from the diseased leaves of *A. vera* collected during the survey of various nurseries and botanical gardens of Gwalior city in 2013, purified by single spore technique and maintained on PDA. Identification of fungi was confirmed by Fungal Culture Collection of India (NFCCI), Pune, Maharashtra (# NFCCI Accession No. 3640). Nine plants for each treatment and control were maintained and each earthen pot contained a single *A. vera* plant. Six healthy leaves of plant were surface sterilized with 1% sodium hypochlorite (NaOCl) solution. Gentle pricks were made by sterilized needles and sprayed with conidial suspension (1×10^7 ml) of *F. proliferatum*. Leaves sprayed with sterile distilled water were taken as control. The inoculated plants along with their healthy controls were then covered with moisten polythene bags to maintain high relative humidity (RH) for 24 to 48 h. After inoculation, plants were kept in greenhouse with a minimum and maximum temperature of 22 °C and 28 °C while, relative humidity was fluctuated between 83% and 92% during experimentation. After the development of symptoms in plants, leaves were harvested on 16^{th} day and 32^{nd} day of infestation for further analysis.

2.3. Evaluation of leaf characteristics

To determine the effect of *F. proliferatum* on leaf characteristics, physical texture of leaves (color, shape, length and width) and color and consistency of gel was examined. The changes in shape and color of infested leaves were examined on visual basis while length and width by using cm scale. The outer green rind of infested leaves was peeled off, color and consistency of the gel was examined and compared with control. The infested and control leaves harvested on 16^{th} and 32^{nd} day were weighed out and compared to study the effect of fungal infestation on fresh weight.

2.4. Biochemical analysis

For biochemical analysis, *A. vera* leaves were harvested at two intervals i.e. on 16^{th} and 32^{nd} day after infestation. Three set of replicates for each treatment and control was harvested and each set having three plants. The harvested leaves were then cleaned with sterilized distilled water and blot dried in the folds of sterilized blotting paper. Leaves were cut into small pieces and dried in hot air oven at 48–50 °C. The dried leaves were grinded to fine powder and stored at room temperature in airtight containers. The powdered leaf samples were subjected to various sample preparation for biochemical estimations. All the experiments were carried out in triplicates.

2.4.1. Estimation of total soluble sugar

Total soluble sugar content was determined by phenol sulphuric acid method (Dubois et al., 1951). 100 mg oven dried powdered sample of each infested and control leaves was homogenized with 80% ethanol. Then each sample was centrifuged at 2000 rpm for 20 min, collected supernatant was evaporated to dryness and dissolved in 10 ml of distilled water. 1.0 ml aliquot from the sample was taken and 1 ml of 5% phenol reagent was added and mixed. Then 5.0 ml of 96% sulphuric acid was added rapidly. Each tube was gently shaked during the addition of acid and tubes were allowed to stand in a water bath at 26–30 °C for 20 min. Reagent blank was prepared similarly. Absorbance of the characteristic orange color was measured at 490 nm against blank using spectrophotometer (Systronics Pvt. Ltd, Model no. 639). Concentration of total soluble sugar was calculated using a standard curve of glucose and results were expressed in mg/g dry weight.

2.4.2. Estimation of total flavonoid

Total flavonoid content was determined by the aluminium chloride method (Dewanto et al., 2002). 250 mg of powdered sample was suspended in 10 ml of 80% methanol for 24 h at room temperature followed by centrifugation at 3000 rpm for 20 min. In 1.0 ml of supernatant, 0.3 ml of 5% $NaNO_2$ solution was added and left undisturbed for 3 min. After 3 min 0.3 ml of 10% $AlCl_3$ was added and was left solution undisturbed for 2 min. Then, 2 ml of 1.0 M NaOH was added and the final volume of mixture was brought to 10 ml with distilled water. Optical density of the yellowish brown color was read at 510 nm. Total flavonoid content was calculated from the calibration curve prepared from the quercetin and the results were expressed in mg/g dry weight.

2.4.3. Estimation of total phenolic content

Total phenolic content was assessed using Folin- Ciocalteau's reagent method (Mahadevan and Sridhar, 1982). 250 mg sample was suspended in 10 ml of 80% ethanol for 24 h at room temperature followed by centrifugation at 5000 rpm for 20 min. The supernatant was evaporated to dryness and residue was re-dissolved in 10 ml of distilled water. 500 µl of aliquot was taken in test tubes and 500 µl of folin-ciocalteau's reagent was added followed by the addition of 2 ml of 20% Na_2CO_3 solution. Test tubes were placed in boiling water bath for 1–2 min and ice cooled immediately. Absorbance of the characteristic blue color was measured at 650 nm against. Concentration of total phenols was expressed from a standard curve prepared from tannic acid and the results were expressed in mg/g dry weight.

2.4.4. Estimation of total anthraquinone

Total anthraquinone was evaluated by the following procedure of Sakulpanich and Gritsanapan (2008). 2.0 *g* of sample was extracted with 200 ml of 70% ethanol in a soxhlet apparatus. The pooled extract was then evaporated to dryness on water bath. 1.0 *g* dried extract was dissolved in 15 ml of distilled water and mixed. The mixture was refluxed on water bath for 15 min followed by centrifugation at 4000 rpm for 10 min. Then 10 ml of the supernatant was transferred to another flask. To this 20 ml solution of 10.5% ferric chloride hexahydrated was added and refluxed on boiling water bath for 20 min. 1.0 ml of concentrated HCl was added and the mixture was again refluxed for another 20 min, with frequent shaking. The mixture was cooled and extracted with 15 ml of diethyl ether thrice a time. The diethyl ether extract was combined and washed with 15 ml distilled water twice. 25 ml of the diethyl ether extract was then evaporated to dryness. The residue was dissolved in 10 ml of 0.5% magnesium acetate in methanol, which yielded red coloration in the solution. The absorbance of solution was read with the spectrophotometer at 515 nm. Concentration of anthraquinones was calculated using chrysophanic acid as standard and expressed in mg/g dry weight.

2.4.5. Estimation of vitamin E

Vitamin E content was determined according to method of Bieri et al. (1964). 500 mg sample was extracted with 200 ml of petroleum ether in soxhlet apparatus until it was exhausted. The extract was evaporated to dryness at 45–48 °C and dissolved in 5 ml of ethanol. 1.0 ml aliquot was taken in tubes and then added 1.0 ml of 2% alcoholic solution of 2-2′ bipyridyl. 500 µl of 1% of ethanolic ferric cholorideheхanhydrate solution was added into tubes and was mixed well.

Test tubes were immediately kept in a dark place for 15 min at room temperature. The red color developed after 15 min and absorbance of the resultant solution was measured at 520 nm. Concentration of vitamin E was calculated from standard curve prepared from α-tocopherol and the results were expressed in mg/g dry weight.

2.4.6. Chlorophyll estimation

The amount of chlorophyll was quantified by the standard method as proposed by Arnon (1949). 100 mg of freshly harvested leaves were homogenized in 80% acetone. This was followed by centrifugation at 3000 rpm for 5 min. The volume was then made up to 5 ml with 80% acetone. Thereafter, optical density of Chlorophyll-*a* and Chlorophyll-*b* were recorded at 645 and 663 nm, respectively. Chlorophyll content was calculated using formula:

$$\text{Chlorophyll 'a' (mg g}^{-1}\text{frwt)} = (0.0127 \times A_{663}) - (0.00269 \times A_{645}) \quad (1)$$

$$\text{Chlorophyll 'b' (mg g}^{-1}\text{frwt)} = (0.0229 \times A_{645}) - (0.00468 \times A_{663}) \quad (2)$$

$$\text{Total Chlorophyll (mg g}^{-1}\text{frwt)} = (0.0202 \times A_{645}) + (0.00802 \times A_{663}) \quad (3)$$

2.5. Statistical analysis

Concentration of chlorophyll was expressed in mg/g fresh weight of leaves while the other biochemical contents were expressed in mg/g on dry weight basis. Data obtained after analyses of each leaf characteristics and biochemical estimations were analyzed with the help of student *t*-test for the comparison with control.

3. Results and discussion

3.1. Disease symptomatology

In greenhouse experiment, inoculated plants showed initiation of leaf spot symptoms after fifth day of infestation. Initially symptoms were marked by small circular spots with water soaked margins. As the infection progressed spots became large, sunken, brown black in color. Later, spots were enlarged in size and aggregated to form lesions of about 1.9 × 1.6 cm in diameter. At the maturity, dark brown-black sporulation was observed on the center of the spots. Recurrence of the fungus from the infested leaves of *A. vera* was compared with the original culture of *F. proliferatum* (Fig. 1).

3.2. Changes in leaf characteristics

Results exhibited that fungal infestation also affects the quality and texture of gel as compared to control leaves. Infested leaves were light green in color, dried with twisted/distorted margins which led to decline in length and width, while, healthy leaves were dark green color, thick and fleshy. Gel of infested leaf was mushy and light creamish in color whereas, in control leaves, it was clear, viscous and transparent (Fig. 2). The leaf length in infested and control leaves was 30.20 ± 1.0 cm and 30.83 ± 0.8; 30.68 ± 0.99 and 30.83 ± 0.8 after 16 and 32 days. Similarly, the variation in width was observed as 3.19 ± 0.10 and 3.27 ± 0.10; 3.06 ± 0.06 and 3.15 ± 1.02 in control and infested leaves. The weight of infested leaves was 21.23 ± 1.18 g and 39.20 ± 1.22 g whereas, it was 26.03 ± 1.19 g and 50.21 ± 1.21 g in control leaves after 16 and 32 days of infestation (Table 1).

Fig. 1. Effect of fungal infection on growth of *Aloe vera*.

3.3. Biochemical changes

The present investigation revealed highly significant difference in the concentration of certain biochemicals in artificially infested *A. vera* leaves due to infection of *F. proliferatum* after 16 and 32 days. The significance was evaluated as marginally significant ($p \leq 0.10$), significant ($p \leq 0.05$) and highly significant ($p \leq 0.01$).

3.3.1. Total soluble sugar content

Results clearly indicated that total soluble sugar content decreased significantly in diseased *A. vera* plants as compared to control. In healthy leaves, the concentration of total soluble sugar was 38.78 ± 0.89 mg/g and 46.80 ± 0.83 mg/g while in infested leaves it reduced to 35.55 ± 0.85 mg/g and 41.51 ± 0.90 mg/g after 16 and 32days of infestation (Table 2). The reduction in sugars during disease development might be due to utilization of sugars probably for energy and synthetic reactions involved in multiplication of the pathogen (Nema, 1989). Similar findings have been reported in number of plants i.e. *Moringa* fruit infected by *Rhizopus stolonifer* (Zoadur et al., 2001), *Brassica compestris* leaves infected by *Alternaria brassicae* (Neeraj, 2010), date palm fruit infected with *Fusarium moniliforme* (Alkahtani et al., 2011), soyabean plant infected by *Phakopsora pachyrhizi* (Mengane and Kamble, 2012), palmarosa and lemongrass infected by *Puccinia nakanishikii* (Tamuli et al., 2013) and in onion bulb by *Colletotrichum gloeosporioides* (Alberto, 2014).

3.3.2. Flavonoid content

With the progression of disease a significant gradual increase in total flavonoid content was observed in infected plants as compare control. Concentration of total flavonoid in infested leaves was 8.45 ± 0.15 mg/g and 14.40 ± 0.16 mg/g, whereas, in control leaves, it was 6.60 ± 0.15 mg/g and 10.41 ± 0.15 mg/g after 16 and 32 days of infestation (Table 2). Flavonoid contents have been found increase in infected tissue, followed by stress in plants and also in response to pathogen attack (Vidyasagar and Kotresha, 2003). Similar trend of increased concentration of flavonoid has also been reported in leaves of *Terminalia arjuna*, *T. bellirica*, *T. chebula*, *T. paniculata* and *T. tomentosa* infected with *Phoma herbarum*, *Colletotrichum dematium*, *Macrophomina phaseolina* and *Pestalotiopsis versicolor* (Shivanna and Mallikarjunaswamy, 2009) and in leaves of rice infected with *Pyricularia oryzae* (Flora and Rani, 2013).

3.3.3. Total phenolic and total anthraquinone content

The total phenolic and anthraquinone content was found significantly increased in infected leaves as compared to control. Total phenol content in infested and control leaves was 10.53 ± 0.15 mg/g and 15.66 ± 0.14 mg/g; 8.83 ± 0.17 mg/g and 11.69 ± 0.16 mg/g after 16 and 32 days. Concentration of anthraquinone in infested leaves was 0.075 ± 0.01 mg/g and 0.094 ± 0.01 mg/g, however, in control it was 0.069 ± 0.01 mg/g and 0.079 ± 0.01 mg/g (Table 2). Higher accumulation of phenolic compounds in infected host tissues may be due to their release from glycoside esters by the enzymatic activity of host or pathogen (Noveroske et al., 1964), enhanced synthesis by host through the shikimic acid pathway (Neish, 1964) or due to migration of phenols from non-infected tissues (Farkas and Kiraly, 1962). Increased

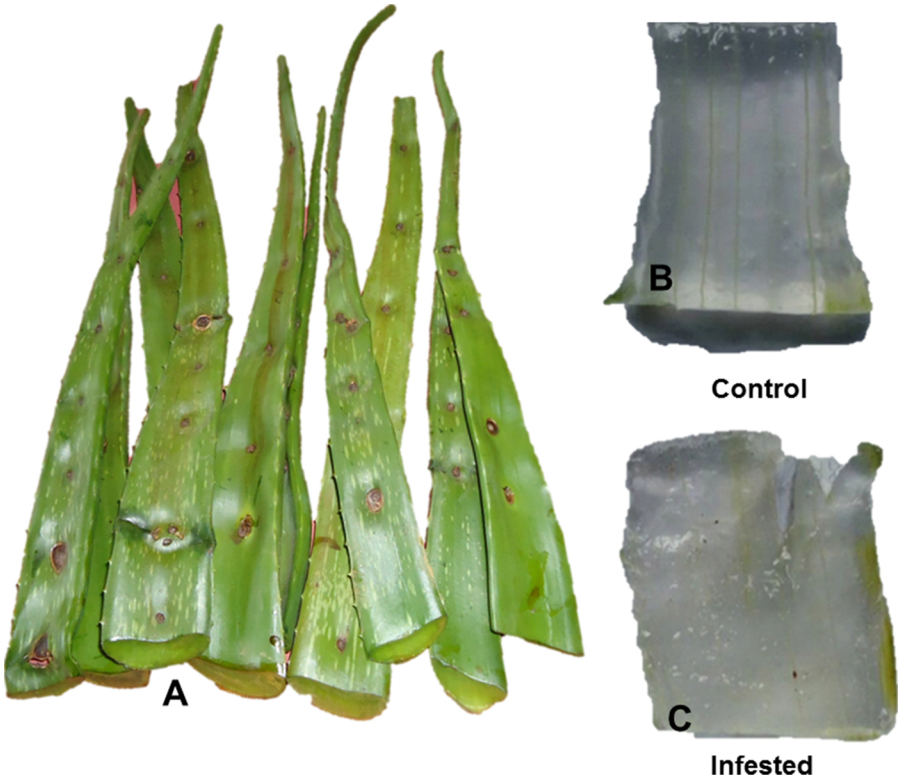

Fig. 2. Effect of fungal infestation on leaf characteristics: A) harvested leaves; B) appearance of gel (control); C) infested after 32 days of infestation.

Table 1
Effect of *Fusarium proliferatum* on the leaf characteristics of *Aloe vera*.

Leaf characteristics	After 16 Days		After 32 Days	
	Control	Infested	Control	Infested
Leaf color	light green	dark green	light green	dark green
Texture/ shape	thick and fleshy	dried with twisted margins	thick and fleshy	dried with twisted/ distorted margins
Leaf length (cm)	30.68 ± 0.99	30.20 ± 1.0*	32.74 ± 0.9	30.83 ± 0.8**
Leaf width (cm)	3.19 ± 0.10	3.06 ± 0.06*	3.27 ± 0.10	3.15 ± 1.02*
Gel color	light creamish	clear	light creamish	clear
Gel consistency	mushy	viscous and transparent	mushy	viscous and transparent
Leaf fresh weight (g)	26.03 ± 1.19	21.23 ± 1.18***	50.21 ± 1.21	39.20 ± 1.22***

Mean ± standard deviation of six replicates.
* Marginally significant ($p \leq 0.10$).
** Significant ($p \leq 0.05$).
*** Highly significant ($p \leq 0.01$).

Table 2
Effect of *Fusarium proliferatum* on the phytochemical attributes of *Aloe vera*.

Phytochemicals	After 16 Days		After 32 Days	
	Control	Infested	Control	Infested
Total soluble sugar (mg g^{-1} dr wt)	38.78 ± 0.89	35.55 ± 0.85***	46.80 ± 0.83	41.51 ± 0. 90***
Total flavonoids (mg g^{-1} dr wt)	6.60 ± 0.15	8.45 ± 0.15***	10.41 ± 0.15	14.40 ± 0.16
Total phenols (mg g^{-1} dr wt)	8.83 ± 0.17	10.53 ± 0.15***	11.69 ± 0.16	15.66 ± 0.14
Total anthraquinones (mg g^{-1} dr wt)	0.069 ± 0.01	0.075 ± 0.01	0.079 ± 0.01	0.094 ± 0.01
Vitamin E (mg g^{-1} dr wt)	50.11 ± 0.87	57.11 ± 0.87	60.0 ± 0.81	73.78 ± 0.89*
Chlorophyll-*a* (mg g^{-1} fr wt)	0.0369 ± 0.01	0.0280 ± 0.01	0.0442 ± 0.01	0.0218 ± 0.01**
Chlorophyll-*b* (mg g^{-1} fr wt)	0.0233 ± 0.01	0.0176 ± 0.01	0.0270 ± 0.01	0.0132 ± 0.01*
Total chlorophyll (mg g^{-1} fr wt)	0.0623 ± 0.01	0.0508 ± 0.01	0.0716 ± 0.01	0.0382 ± 0.01 ***

Mean ± standard deviation of three replicates.
* Marginally significant ($p \leq 0.10$).
** Significant ($p \leq 0.05$).
*** Highly significant ($p \leq 0.01$).

concentration of phenol was also found in other plant hosts like sorghum seeds infected with *Fusarium moniliforme* (Saravanan et al., 2002), taro plant infected by *Phytophthora colocasiae* (Misra et al., 2008); *Cucumi ssativus* infected by *Penicillium notatum* (Senthil et al., 2010); rapseed mustard leaves infected by *Alternaria brassicae* (Mathpal et al., 2011), in leaves of *Curcuma longa* infected by *Colletotrichum gloeosporioides* (Chawda et al., 2012) in leaves of *Centella asiatica* infected by *Cercospora centallae* (Parashurama et al., 2013) and in French green bean pod infected by *Colletotrichum lindemuthianum* (Petkovsek et al., 2014).

3.3.4. Vitamin E

Results showed significant increase in vitamin E content in infected leaves as compared to control. Concentration of vitamin E was 57.11 ± 0.87 mg/g and 73.78 ± 0.89 mg/g in infested leaves however, 50.11 ± 0.87 mg/g and 60.0 ± 0.81 mg/g was recorded in control leaves after 16 and 32 days (Table 2). Alkahtani et al. (2011) suggested that vitamin E is fat soluble, so increase in vitamin E concentration might be due to degradation of fat which, in turn, may be different lipase enzymes produced by fungi. Infection of *Fusarium moniliforme* increased the concentration of vitamin E in date palms fruits (Alkahtani et al., 2011).

3.3.5. Chlorophyll content

A gradual reduction in green pigments like chlorophyll- *a*, *b* and total chlorophyll contents was observed at different period of pathogenesis in infested leaves as compared to control. Chlorophyll-content was found to decrease in infected leaves viz. 0.0280 ± 0.01 mg/g and 0.0218 ± 0.01 mg/g, whereas, in control leaves it was 0.0369 ± 0.01 mg/g and 0.0442 ± 0.01 mg/g after 16and 32 days of infestation. Concentration of chlorophyll-*b* in infested and control leaves was 0.0176 ± 0.01 mg/g and 0.0132 ± 0.01 mg/g; 0.0233 ± 0.01 mg/g and 0.0270 ± 0.01 mg/g, respectively at both the intervals. Similarly, 0.0508 ± 0.01 mg/g and 0.0382 ± 0.01 mg/g total chlorophyll content was recorded in infested leaves however, in control leaves it was 0.0623 ± 0.01 mg/g and 0.0716 ± 0.01 mg/g (Table2). However, decrease in total chlorophyll content was more pronounced at both the intervals of infection. Decrease in photosynthesis could be attributed to the toxic effect of its metabolites on chloroplasts (Peru and Main, 1970; Chen et al., 2005), which might be due to decrease in the rate of electron transport in non-cyclic photophosphorylation (Dai et al., 2004). Similar decreasing trend was also noticed in sorghum leaves infected by *Drechslera sorghicola* (Khan et al., 2001); faba beans infected with *Fusarium moniliforme* (Elwakil et al., 2009), Mentha arvensis infected with Sclerotinia sclerotiorum (Perveen et al., 2010), *Withania somnifera* infected with *Alternaria alternata* (Pati et al., 2008; Sharma et al., 2011), and in leaves of mangrove plant (*Avicennia marina*) infected by *Alternaria alternata* (Alqarawi et al., 2013).

4. Conclusions

Aloe vera is a highly valuable medicinal plant whose pharmaceutically important constituents are declined by leaf spot diseases. It may be concluded from the present findings that infection of *Fusarium proliferatum* not only declines the quality and texture of mucilaginous gel but significantly modifies the concentration of biochemical constituents also, which results into quantitative loss of gel. Control of the fungal diseases by applying appropriate management strategies is the important thrust to protect the plant of an enormous medicinal and cosmetic value.

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References

Alberto, R.T., 2014. Pathological response and biochemical changes in *Allium cepa* L. (bulb onions) infected with anthracnose-twister disease. Plant Pathology and Quarantine 4, 23–31.
Alkahtani, M., El- Naggar, M.A., Omer, S.A., Abedl- Kareem, E.M., Ammar, M.I., 2011. Effect of toxic *Fusarium moniliforme* on some biochemical component of some date palm cultivars. American Journal of Food Technology 6, 730–741.
Alqarawi, A.A., Hashem, A., Abd-allah, E.F., 2013. Physiological alterations in *Avicennia marina* (Forski) vierh associated with leaf spot disease caused by *Alternaria alternata* (fr.) Keissler. Pakistan Journal of Botany 45, 1767–1771.
Arnon, D.I., 1949. Copper enzymes in isolated chloroplasts, polyphenol oxidase in *Beta vulgaris*. Plant Physiology 2, 1–15.
Avasthi, S., Gautam, A.K., Bhadauria, R., 2018. First report of leaf spot disease of *Aloe vera* caused by *Fusarium proliferatum* in India. Journal of Plant Protection Research 58, 109–114.
Bieri, J.G., Teets, L., Belavady, B., Andrews, E.L., 1964. Serum vitamin E levels in a normal adult population in the Washington, DC, area. Proceedings of the Society for Experimental Biology and Medicine 117, 131–133.
Boudreau, M.D., Beland, F.A., 2006. An evaluation of the biological and toxicological properties of *Aloe barbadensis* (Miller), *Aloe vera*. Journal of Environmental Science and Health 24, 103–154.
Chang, K.F., Hwang, S.F., Conner, R.L., Ahmed, H.U., Zhou, Q., Turnbull, G.D., Strelkov, S.E., Mclaren, D.L., Gossen, B.D., 2015. First report of *Fusarium proliferatum* causing root rot in soybean (*Glycinemax* L.) in Canada. Crop Protection 67, 52–58.
Chawda, S.K., Sabalpara, A.N., Pandya, J.R., 2012. Epidemiology of turmeric (*Curcuma longa* L) leaf spot caused by *Colletotrichum gloeosporioides* (Penz and Sacc). The Bioscan 7, 441–443.
Chen, S., Dai, X., Qiang, S., Tang, Y., 2005. Effect of a nonhost-selective toxin from *Alternaria alternata* on chloroplast-electron transfer activity in *Eupatorium adenophorum*. Plant Pathology 54, 671–677.
Dai, X.B., Chen, S.G., Qiang, S., An, Q.F., Zhang, R.X., 2004. Effect of toxin from *Alternaria alternata* (Fr.) Keissler on leaf photosynthesis of *Eupatorium adenophorum* Spreng. Acta Phytopathologica Sinica 34, 55–60.
Dewanto, V., Wu, X., Adom, K.K., Liu, R.H., 2002. Thermal processing enhances the nutritional value of tomatoes by increasing total antioxidant activity. Journal of Agriculture and Food Chemistry 50, 3010–3014.
Dubey, R., Pandey, A.K., 2009. A new report on collar and root rot disease of *Aloe vera* caused by *Sclerotium rolfsii*. Journal of Mycology and Plant Pathology 39, 300–301.
Dubois, M.K., Gilles, J.K., Robers, P.A., Smith, F., 1951. Colorimetric determination of sugar and related substance. Analytical Chemistry 26, 351–356.
Elwakil, M.A., El-Refai, I.M., Awadallah, O.A., El-Metwally, M.A., Mohammed, M.S., 2009. Seed-borne pathogens of faba bean in Egypt: detection and pathogencity. Plant Pathology Journal 8, 90–97.
Esser, R.P., Riherd, C.C., Harkcom, K.J., 1986. Research notes- pathogenicity of *Scutellonema brachyurum* to *Aloe vera*. Nematropica 16, 65–71.
Farkas, G.L., Kiraly, Z., 1962. Role of phenolic compounds in the physiology of plant diseases and disease resistance. Phytopathology 44, 105–150.
Flora, G., Rani, S.M.V., 2013. Effect of *Stoechospermum marginatum* (Ag). Kutz extract on controlling blast disease in rice caused by *Pyricularia oryzae* under natural field condition. African Journal of Agricultural Research 8, 3289–3293.
Gao, M.L., Luan, Y.S., Yu, H.N., Bao, Y.M., 2016. First report of tomato leaf spot caused by *Fusarium proliferatum* in China. Canadian Journal of Plant Pathology 38, 400–404.
Gil-Serna, J., Gálvez, L., París, M., Palmero, D., 2016. *Fusarium proliferatum* from rain water and rooted garlic show genetic and pathogenicity differences. European Journal of Plant Pathology 146, 199–206.
Harsh, N.S.K., Nath, V., Jamaluddin, 1990. New forest disease record for India. Journal of Tropical Forest Science 6, 175–176.
Isack, Y., Benichis, M., Gillet, D., Gamliel, A.A., 2014. Selective agar medium for isolation, enumeration and morphological identification of *Fusarium proliferatum*. Phytoparasitica 42, 541–547.
Ishii, Y., Tanizawa, H., Takino, Y., 1994. Studies of *Aloe vera*. Mechanism of cathartic effect. Biological and Pharmaceutical Bulletin 17, 651–653.
Jin, K.S., Lee, S.W., Kim, J.J., Ryu, H.Y., 1994. Identification of bacterial isolates obtained from diseased orchid and aloe plants caused by *Erwinia chrysanthemi*. RDA Journal of Agricultural Science. Crop Protection 36, 301–306.
Khan, A.J., Deadman, M.L., Srikandakumar, A., Al-Maqbali, Y.M., Rizvi, S.G., Al-Sabahi, J., 2001. Biochemical changes in sorghum leaves infected with leaf spot pathogen *Drechslera sorghicola*. Plant Pathology Journal 17, 342–346.
Mahadevan, A., Sridhar, R., 1982. Methods in Physiological Plant Pathology, second edition. Sivakami Publication, pp. 157–159.
Maiti, S., Chandra, R., 2002. Cultivation of *Aloe vera*. National Research Center for Medicinal and Aromatic Plants, Boriavi Anand, Gujrat. www.nrcmap.org.
Majumdar, V.L., Bhatnagar, K., Sharma, H., Verma, O.P., 2007. Three new diseases of *Aloe barbadensis* L. Journal of Mycology and Plant Pathology 37, 124–125.
Mathpal, P., Punetha, H., Tewari, A.K., Agrawal, S., 2011. Biochemical defense mechanism in rapeseed-mustard genotypes against *Alternaria* blight disease. Journal of Oilseed Brassica 2, 87–94.
Mengane, S.K., Kamble, S.S., 2012. Changes in biochemical composition of soybean leaves infected with resistant and sensitive isolates of *Phakopsora pachyrhizi* causing soybean rust. Deccan Current Science 7, 122–125.

Misra, R.S., Sharma, K., Mishra, A.K., Sriram, S., 2008. Biochemical alterations induced in taro in response to *Phytophthora colocasiae* infection. Advances in Natural and Applied Sciences 2 (3), 112–121.

Neeraj, Verma S., 2010. Quantitative changes in sugar and phenolic contents of Brassica leaves induced by *Alternaria brassicae* infection. International Journal of Plant Protection 3, 28–30.

Neish, A.C., 1964. Major pathways of biosynthesis of phenols. In: Harbone, J.B. (Ed.), Biochemistry of Phenolic Compounds. Academic Press, New York USA, pp. 295–359.

Nema, A.G., 1989. Sugar and phenol contents of betelvine leaves after inoculation with leaf spot bacterium. Indian Phytopathology 42 (1), 31–37.

Noveroske, R.L., Williams, E.B., Kuc, J., 1964. 7-Glucosidase and polyphenol oxidise in apple leaves and their possible relation *Venturia inaequallis*. Phytopathology 54, 98–103.

Palmero, D., De Cara, M., Nosir, W., Galvez, L., Cruz, A., Woodward, S., Gonzalez-Jaen, M.T., Tello, J.C., 2012. *Fusarium proliferatum* isolated from garlic in Spain: identification, toxigenic potential and pathogenicity on related *Allium* species. Phytopathologia Mediterranea 51, 207–218.

Parashurama, T.R., Mahishi, P., Mallikarjunaswamy, G.E., Byrappa, S.M., 2013. *Cercospora* leaf spot disease in *Centella asiatica* and its effect on pharmaceutical components. International Journal of Pharmaceutical Sciences and Research 4 (1), 64–71.

Pati, P.K., Sharma, M., Salar, R.K., Sharma, A., Gupta, A.P., Singh, B., 2008. Studies on leaf spot disease of *Withania somnifera* and its impact on secondary metabolites. Indian Journal of Microbiology 48, 432–437.

Peltomaa, R., Vaghini, S., Patiño, B., Benito-Peña, E., Moreno-Bondi, M.C., 2016. Species-specific optical genosensors for the detection of mycotoxigenic *Fusarium* fungi in food samples. Analytica Chimica Acta 935, 231–238.

Peru, R.W., Main, C.B., 1970. Chlorosis of tobacco induced by alternariol produced by *Alternaria tenuis*. Phytopathology 60, 1570–1573.

Perveen, K., Haseeb, A., Shukla, P.K., 2010. Effect of *Sclerotinia sclerotiorum* on the disease development, growth, oil yield and biochemical changes in plants of *Mentha arvensis*. Saudi Journal of Biological Sciences 17 (4), 291–294.

Petkovsek, M.M., Schmitzer, V., Jakopic, J., Cunja, V., Veberic, R., Munda, A., Stampar, F., 2014. *Colletotrichum lindemuthianum* infection causes changes in phenolic content of French green bean pods. Scientia Horticulturae 170, 211–218.

Pritam, A., Kale, P.G., 2007. Alteration in the antioxidant potential of *Aloe vera* due to fungal infection. Journal of Plant Pathology 6 (2), 169–173.

Rheeder, J.P., Marasas, W.F.O., Vismer, H.F., 2002. Production of Fumonisin analogs by *Fusarium* species. Applied and Environmental Microbiology 68, 2101–2105.

Sakulpanich, A., Gritsanapan, W., 2008. Extraction method for high content of anthraquinones from *Cassia fistula* pods. Journal of Health Research 22 (4), 167–172.

Saravanan, T., Valluvaparidasan, V., Muthusamy, M., 2002. Changes in biochemical qualities of sorghum seeds due to seed infection. Research Crops 3, 627–628.

Seefelder, W., Gossmann, M., Humpf, H.U., 2002. Analysis of fumonisin B1 in *Fusarium proliferatum* infected Asparagus Spears and Garlic Bulbs from Germany by liquid chromatography – electrospray ionization mass spectrometry. The Journal of Agricultural and Food Chemistry 50, 2778–2781.

Senthil, V., Ramasamy, P., Elaiyaraja, C., Elizabeth, R.A., 2010. Some phytochemical prosperities affected by the infection of leaf spot disease of *Cucumis sativus* (Linnaeus) caused by *Penicillium notatum*. African Journal of Basic and Applied Sciences 2, 64–70.

Sharma, A., Sharma, I., Pati, P.K., 2011. Post-infectional changes associated with the progression of leaf spot disease in *Withania somnifera*. Journal of Plant Pathology 93 (2), 397–405.

Shivanna, M.B., Mallikarjunaswamy, G.E., 2009. Fungal diseases and their effect on phytochemical constituents of medicinally important *Terminalia* species in Bhadra Wildlife Sanctuary, Karnataka, India. Indian Phytopathology 62 (1), 37–43.

Swarbrick, P.J., Schulze-Lefert, P., Scholes, J.D., 2006. Metabolic consequences of susceptibility and resistance in barley leaves challenged with powdery mildew. Plant Cell and Environment 29, 1061–1076.

Tamuli, P., Boruah, P., Samanta, R., 2013. Post-inflectional biochemical changes in *Cymbopogon martini* (Roxb.) Wats and *Cymbopogon citrates* (DC) Stapf. due to leaf rust disease. American Journal of Plant Science 4, 1666–1668.

Vidyasagar, G.M., Kotresha, D., 2003. Biochemical changes due to infection of *Phyllactinia corylea* in the leaves of certain mulberry varieties. Indian Journal of Sericulture 42, 71–74.

Zoadur, M., Saud, Z.A., Absar, N., 2001. A comparative investigation on the physiochemical properties and some selected enzymes content in the flesh of healthy and *Rhizopus stolonifer* infected *Moringa* fruit. Indian Phytopathology 54, 293–298.