

## Evaluation of the Effect of Morphological Traits on Blister Blight Resistance in Tea Plant (*Camellia sinensis* L.)

Thisssa Karunaratna<sup>1</sup>, Mewan Kooragoda<sup>2</sup>, Lahiru Udayanga<sup>2</sup>, Jagathpriya Weerasena<sup>3</sup>, Chandrika N. Perera<sup>4</sup> and Nishantha Edirisinghe<sup>5</sup>

### Abstract

**Background:** Blister Blight (BB) is a serious leaf disease caused by the fungus *Exobasidium vexans* Masse, damaging Sri Lankan tea plantations.

**Methods:** A morphological trait-based analysis was performed based on 14 descriptors for *Camellia sinensis* to differentiate BB resistant and BB susceptible individuals in an F1 population generated by a cross between BB resistant and BB susceptible cultivars (TRI 2043×TRI 2023). The Spearman's correlation analysis, regression modelling, Receiver Operating Characteristic (ROC) and t-test were applied in the analysis of morphological characteristics of the F1 plants.

**Results:** Leaf pubescence (SCC= - 0.530), upper leaf surface (SCC= 0.473) and length of mature leaf petiole denoted significant associations with BB disease index (P<0.05). Threshold values of the developed model to screen vulnerability of tea plant to blister blight were 1.5 for both pubescence of tea leaves and upper leaf surface.

**Conclusions:** Proposed leaf morphology-based thresholds can be successfully applied for preliminary screening of BB susceptibility, prior to further confirmation with more advanced identification techniques.

**Keywords:** Disease Control, Fungal Infection, Marker Assisted Tea Breeding, Non-Alcoholic Beverages, Plant Inherent Resistance

<sup>1</sup>Department of Bio-systems Technology, Faculty of Technology, University of Ruhuna, Kamburupitiya, Sri Lanka.


<sup>2</sup>Faculty of Agriculture and Plantation Management, Wayamba University of Sri Lanka.

<sup>3</sup> Institute of Biochemistry, Molecular Biology and Biotechnology, University of Colombo, Colombo, Sri Lanka.

<sup>4</sup>Department of Agricultural Biology, Faculty of Agriculture, University of Peradeniya, Peradeniya, 20400, Sri Lanka.

<sup>5</sup>Biochemistry Division, Tea Research Institute of Sri Lanka, Talawakelle, Sri Lanka.

\* Correspondence: [thissa@btec.ruh.ac.lk](mailto:thissa@btec.ruh.ac.lk)

 <https://orcid.org/0000-0003-4504-6451>



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## INTRODUCTION

Tea is one of the most popular non-alcoholic caffeine containing beverages in the world with high amounts of flavonoids and related bioactive compounds [1-2]. Though it remains unclear, it is considered that the tea plant (*Camellia sinensis* L.) was originated from China and spread through the other South East Asian countries as a commercial cultivation [3]. Succulent plant leaves of tea are processed to make three most popular types of tea, green (unfermented), black (fully fermented) and oolong (semi-fermented) [4].

In tea plants, a considerable crop loss results due to nutrient deficiencies, stresses from climatic variations, pests and pathogen attacks. Among diseases, Blister Blight (BB) leaf disease, is caused by the obligatory biotrophic pathogen, *Exobasidium vexans* Masee, which infects only young harvestable succulent leaves, stems and the pericarp of fruits at young stage [5]. The pathogen is spread by windborne basidiospores and infection mostly proceeds through stomata [6]. Mycelium grows intercellularly prior to the formation of basidia fruiting bodies on epidermis. When they grow further, initially small pinhole size spots become visible on young leaves. As the leaves develop the spots become transparent, larger and light brown in colour. They force up and rupture the lower epidermis to form blisters with dark green and water-soaked zones surrounding the blisters [7]. After releasing the fungal spores, the blisters become velvet and white and which subsequently turns into brown in colour [8].

The BB disease causes approximately 25% - 30% crop loss per annum in Sri Lanka. Infected harvestable leaves directly reduce crop yield, not only quantitatively but qualitatively as well, due to the changes of the composition of leaf biochemical compounds such as polyphenols, catechins, enzymes etc. [9]. As the environmental conditions such as high humidity and limited sunshine directly facilitate the infection and development of BB [10] in the field, it is important to control

these microclimatic factors, in which BB may develop.

Morphological characteristics of the infected plants can be used to develop a clear relationship between the morphological characteristics of plants and blister blight resistance using an accurate scoring system for BB susceptibility of plants. Subsequently Quantitative Trait Locus (QTL) mapping can be done for proper identification of the infected plants. Evaluating the agricultural significance of fungal leaf diseases and developing tools that enable rapid recognition of diseases are very important to eliminate these pathogens [11-12].

Various methods are used to identify different pathogens, which cause diseases in crops. Morphological and genetic analysis of infected plants are two main approaches to study plant pathogen interactions and disease development. Ponmurugan and Baby [13] conducted a study on the morphological, physiological, and biochemical changes in tea plants due to *Phomopsis* infection. Physiological parameters; photosynthetic and transpiration rates, stomatal conductance, efficiency of water usage and total chlorophyll content were scored both in susceptible TRI-2024 and tolerant TRI-2025 tea cultivars. Plant height, dry weight and plant strength were recorded as morphological characteristics, while total sugar, nitrogen, amino acids, protein, polyphenols and catechins of infected and healthy plants were studied as biochemical parameters. Results revealed all the morphological, physiological and biochemical characteristics tend remain significantly low in infected plants, compared to healthy plants [13].

Growth, photosynthetic and biochemical responses of tea cultivars to BB infection has been studied by Premkumar *et al.* [14], where susceptibility to BB infection has denoted significant strong associations with physical barriers, physiological and biochemical parameters (leaf area, shoot

length and moisture contents etc). Not only the characteristics of the plants, but also the variations in infection causing pathogens (*Exobasidium vexans* Masee) have been studied by Abeyasinghe *et al.* [15] using infected tea leaf samples. As the infection has a short (11-28 days), but multiple disease cycles with several generations within a single crop season, it requires repeated applications of fungicides to control the disease [6]. However, continuous application of fungicides can contribute to the development of new strains of *Exobasidium vexans* Masee. Morphological parameters of the pathogen such as colour, length and width of spores and DNA finger printing analysis using RAPD have revealed a high degree of genetic diversity among the samples of the *E. vexans* as an adaptation due to various conditions [16].

Plants utilize structural and chemical characteristics to prevent or reduce the spread of pathogens, which act as their first line of defence against pathogens [17]. Development of plant inherent resistance of tea cultivars against BB is the most suitable solution to control the disease, instead of applying highly toxic fungicides. In this process, marker (morphological, biochemical and molecular) assisted tea breeding is playing a key role. Among those, biochemical markers and molecular markers are more accurate and precise techniques [18-19]. However, the cost and technical requirements of molecular markers and biochemical markers are very high and the process is time consuming. Therefore, developing a simple and rapid assessment approach for early detection of BB resistance is immensely important. Hence, this study aimed to develop an inexpensive, user friendly, accurate and reliable morphological marker for preliminary screening of BB resistance traits in tea cultivars.

## METHODOLOGY

### Materials

In the present study, 300 individuals of an F<sub>1</sub> segregating population derived from a cross

between TRI 2043 (a tea cultivar resistant to BB disease) and TRI 2023 (a cultivar susceptible to BB) were used in each replicate. Three replicates were grown following Randomized Complete Block Design (RCBD) together with their parents at St. Coombs Estate (Up country wet zone of Sri Lanka), Tea Research Institute, Talawakelle, Sri Lanka.

### Assessment of Blister Blight Disease Severity of the F<sub>1</sub> Individuals

Blister Blight Disease Index (BBDI) of each F<sub>1</sub> individual was calculated using the data collected from the field assessments starting from year 2007 to 2010 at one-week intervals based on guidelines given in the BB severity assessment key [19].

### Morphological Characterization of F<sub>1</sub> Individuals

Morphological assessment of F<sub>1</sub> individuals was carried out using the descriptors for *Camellia sinensis* L. described by the International Plant Genetic Resources Institute (IPGRI) [20]. Shape of the 5<sup>th</sup> leaf, size of the 5<sup>th</sup> leaf, leaf color, apex shape of the 5<sup>th</sup> leaf, habit of the 5<sup>th</sup> leaf apex, shape of the 5<sup>th</sup> leaf base, pubescence of the 1<sup>st</sup> leaf, leaf venation, leaf vestiture, upper leaf surface, length of the 5<sup>th</sup> leaf, width of the 5<sup>th</sup> leaf, length of mature leaf petiole, leaf length to width ratio were the fourteen morphological characteristics scored in this study. Non parametric data were converted into numerical values on the scale mentioned by the IPGRI [20]. The assessment was repeated for each individual established in three different locations and the average of each parameter was used for the statistical analysis.

### Statistical Analysis

Offspring of the studied population were separated into two sets of samples based on the seed bearer (mother). The offspring produced from the seeds of TRI 2043 was considered as group 1, while group 2 consisted of the offerings from the seeds of TRI 2023. Spearman's correlation analysis

was used to assess the association between different morphological characteristics of the plant and the BBDI. Further, data obtained from morphological characteristics were subjected to regression modelling, after square root transformation in order to develop a model on susceptibility to BB. In addition, Receiver Operating Characteristic (ROC) analysis was used to define the risk thresholds for susceptibility to BB based on the significantly associated morphological characteristics of the F<sub>1</sub> plants.

## RESULTS AND DISCUSSION

### Blister Blight Disease Severity of F<sub>1</sub> Individuals

Individuals with high BB resistivity were grouped on the left-hand side of Figure 1, while high BB susceptible individuals were grouped at the right-hand side. According to the BB severity assessment key, individuals with less than 0.1 BBDI were considered as high BB resistant and individuals with a BBDI higher than 0.5 were considered as highly BB susceptible individuals. According to the analysed data, P219, P58 and P1040 were extremely resistant F<sub>1</sub> individuals. Meanwhile, P219, P1016 and P1018 were extremely susceptible F<sub>1</sub> individuals.

### Morphological Characterization of F<sub>1</sub> Individuals

The morphological characteristics of all 300 F<sub>1</sub> individuals were assessed and the results of six F<sub>1</sub> individuals from two extremes with parents are given in Table 1.

### Impact of Mother Plant on the Morphological Characteristics and Incidence of Blister Blight of the Offspring

Among the notable variations in the morphological characteristics of the two offspring groups, only eight morphological characteristics; leaf colour, leaf apex shape, leaf apex habit, leaf vestiture, upper leaf surface, length of mature leaf, width of mature leaf and length of mature leaf petiole, advocated significant differences at 95% level of significance, in accordance with the statistics of the t-test (Table 2). However, the BBDI values of the two test groups did not

show any significant variations ( $P < 0.05$ ).

### Impact of Leaf Morphology on the Incidence of Blister Blight

Among the studied leaf morphological characteristics, leaf shape, leaf apex habit, leaf pubescence, leaf vestiture and leaf length to width ratio denoted negative relationships with the BBDI, while rest of the characteristics indicated positive associations. However, only the associations of leaf pubescence, upper leaf surface and length of mature leaf petiole were significant at 95% level of confidence ( $P < 0.05$ ), in accordance with the Spearman's correlation analysis (Table 3).

Leaf pubescence advocated a significant negative moderate relationship with the BBDI (Spearman's Correlation Coefficient [SCC] = -0.530), while on the other hand, a significant positive moderate association (SCC = 0.473) was indicated by the upper leaf surface. Regardless of the significance in correlation, the impact of leaf length to width ratio on BBDI remained to be poor (SCC < 0.1).

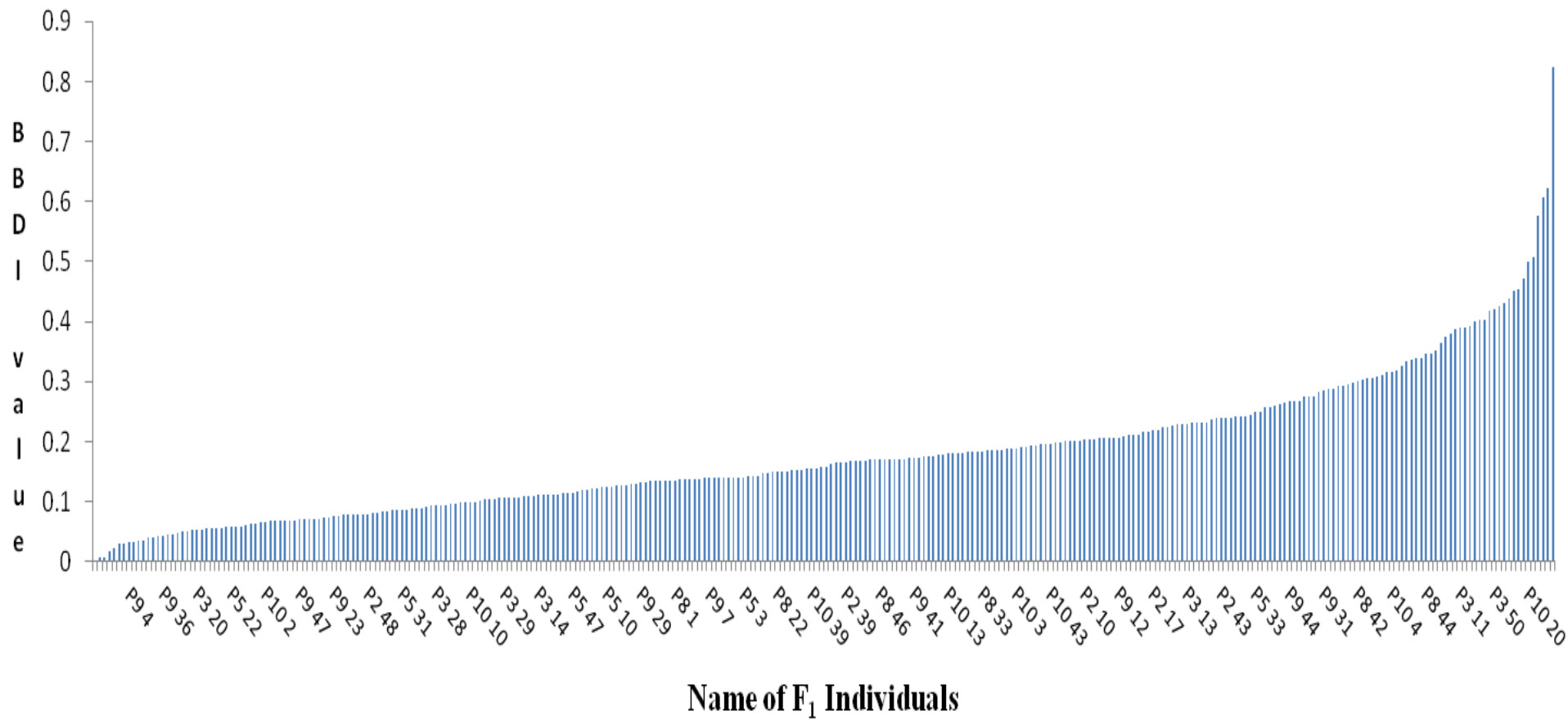
### Morphological Characteristics which Affect the Susceptibility to Blister Blight

The regression analysis based on backward elimination, yielded a simple model for the identification of susceptibility of a tea plant based on the morphological characteristics. As indicated by the model, the susceptibility of the plant remains as a function of Upper Leaf Surface (ULS) and Leaf Pubescence (LP), as indicated by the Equation 1. The model was characterized by a R<sup>2</sup> value of 0.57, followed by an adjusted R<sup>2</sup> value of 0.51.

$$\text{Susceptibility to Blister Blight Disease (1)} \\ = 0.78 + (0.60 \times \text{ULS}) - (0.25 \times \text{LP})$$

### Definition of Risk Thresholds for Blister Blight based on Leaf Morphology

The Receiver Operating Characteristic (ROC) curve analysis yielded an area coverage of 0.534 and 0.479, for upper leaf surface and leaf pubescence, respectively, while the incidence of BB was defined as BBDI > 0.1 (Figure 2).



**Figure 1:** The Bar Chart of 300 F1 Individuals against Blister Blight Disease Index

**Table 1:** Morphological Characteristics of Selected Six F<sub>1</sub> Individuals from the Two Extremes (Highly Resistant and Highly Susceptible) of the BBDI, along with Their Parents

Sam.	Leaf shape	Leaf Size	Leaf Colour	Leaf Apex Shape	Leaf Apex Habit	Leaf Base Shape	Leaf Pubescence	Leaf venation	Leaf Vestiture	Leaf Upper Surface	Leaf Length (cm)	Leaf Width (cm)	Length of Leaf Petiole (cm)	Leaf Length Width Ratio
P <sub>58</sub>	Lanceolate (4)	Oblong (2)	Greyed yellow (4)	Acute (1)	Down turned (1)	Rounded (2)	Intermediate (5)	Distinct with bullations (2)	Pubescent (3)	Rugose (2)	8.00	3.70	0.36	2.15
P <sub>920</sub>	Lanceolate (4)	Oblong (2)	Greyed green (3)	Acute (1)	Down turned (1)	Rounded (2)	Intermediate (5)	Distinct with bullations (2)	Pubescent (3)	Rugose (2)	8.30	3.10	0.33	2.70
P <sub>1040</sub>	Lanceolate (4)	Oblong (2)	Greyed yellow (4)	Acute (1)	Down turned (1)	Attenuate (1)	Sparse (3)	Distinct with bullations (2)	Pubescent (3)	Smooth (1)	8.80	3.60	0.40	2.44
P <sub>219</sub>	Lanceolate (4)	Oblong (2)	Greyed yellow (4)	Acute (1)	Down turned (1)	Attenuate (1)	Intermediate (5)	Distinct with bullations (2)	Pubescent (3)	Smooth (1)	8.30	3.60	0.38	2.31
P <sub>1016</sub>	Lanceolate (4)	Oblong (2)	Green (2)	Acute (1)	Down turned (1)	Attenuate (1)	Sparse (3)	Distinct with bullations (2)	Pubescent (3)	Rugose (2)	9.90	4.50	0.66	2.20
P <sub>1018</sub>	Lanceolate (4)	Oblong (2)	Green (2)	Acute (1)	Down turned (1)	Attenuate (1)	Sparse (3)	Distinct with bullations (2)	Pubescent (3)	Smooth (1)	6.50	3.10	0.35	2.06
TRI 2023	Lanceolate (4)	Oblong (2)	Yellow green (5)	Acute (1)	Down turned (1)	Attenuate (1)	Sparse (3)	Distinct with bullations (2)	Pubescent (3)	Smooth (1)	11.80	4.70	0.44	2.52
TRI 2043	Lanceolate (4)	Oblong (2)	Greyed yellow (4)	Acute (1)	Down turned (1)	Attenuate (1)	Dense (7)	Distinct with bullations (2)	Pubescent (3)	Rugose (2)	10.20	4.30	0.45	2.38

Note: Sam.: Sample

**Table 2:** Results of the t-Test for Significant Differences among Leaf Morphological Characteristics between the Two Groups (Highly Resistant and Highly Susceptible) of Off springs

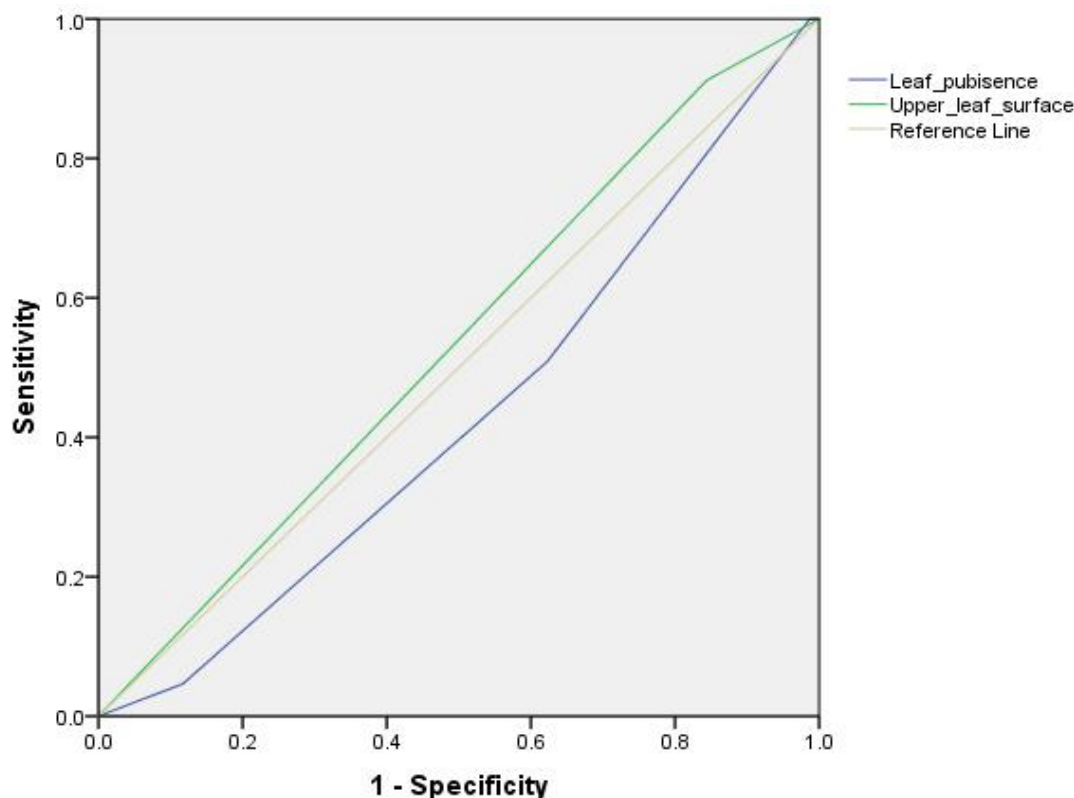
Morphological Parameters	Mean Value		F Value	t Value	df	p Value
	Group 1	Group 2				
Leaf shape	3.6	3.8	12.756	1.749	298	0.081
Leaf colour	1.15	1.30	18.983	-2.124	298	0.034*
Leaf apex shape	2.30	2.81	146.777	-5.390	298	0.001*
Leaf apex habit	1.24	1.14	21.210	2.247	298	0.025*
Leaf base habit	1.18	1.13	5.097	1.053	298	0.293
Leaf pubescence	2.54	2.66	0.561	-1.621	298	0.106
Leaf venation	2.00	2.00	0.426	-0.773	298	0.440
Leaf vestiture	4.43	4.57	68.506	3.787	298	0.001*
Upper leaf surface	1.96	1.83	0.484	-4.986	298	0.001*
Length of mature leaf	10.48	11.69	0.669	-3.705	298	0.001*
Width of mature leaf	4.38	4.71	2.663	2.210	298	0.028*
Length of mature leaf petiole	0.71	0.68	0.117	-3.131	298	0.002*
Blister Blight Severity Index	0.2	0.19	0.629	1.071	298	0.285

*Note: “\*” in the column indicates significant difference (P<0.05) among the two groups in accordance with the t-test*

**Table 3:** Results of the Correlation Analysis between Different Leaf Morphological Characteristics and Blister Blight Disease Index

Morphological Parameter	Spearman Correlation Coefficient (SCC)
Leaf shape	-0.004
Leaf colour	0.026
Leaf apex shape	0.083
Leaf apex habit	-0.053
Leaf base habit	0.047
Leaf pubescence	-0.530*
Leaf venation	0.026
Leaf vestiture	-0.022
Upper leaf surface	0.473#
Length of mature leaf	0.026
width of mature leaf	0.066
Length of mature leaf petiole	0.131*
Leaf length to width ratio	-0.036

*Note: “\*” denotes parameters that indicated a significant correlation with BBDI (P<0.05) at 5% level of significance, while “#” denotes parameters significant at 1% level of significance (P<0.01).*



**Figure 2:** ROC Curve for the Leaf Pubescence and Leaf Vestiture Associated with BBDI

Therefore, it was reassured that both of these leaf characteristics are significantly associated with the incidence of BB. Based on the distribution of the curve and the coordinates of the curve (Figure 2) (sensitivity and 1- specificity), leaf pubescence > 1.5 and upper leaf surface > 1.5, categories could be considered as risk thresholds, which can symbolize the susceptibility of tea plants to BB incidence. In the analysis of the BB severity of both F<sub>1</sub> individuals derived from TRI 2043 and TRI 2023, resistance and susceptibility characteristics have not shown any maternal segregation as denoted by the statistics of the t test.

As suggested by the overall results of Spearman's correlation analysis, morphological characteristics such as leaf shape, leaf apex habit, leaf pubescence, leaf vestiture and leaf length to width ratio have denoted negative relationships with BBDI. Narrow leaf with notable length has decreased the susceptibility to the BB. The ratio between leaf length and leaf width also have shown negative correlations with BBDI.

Therefore, lanceolate shaped leaf with higher value of ratio between leaf lengths to width can be considered as more resistant to blister blight disease, than ovate, oblong and elliptic leaf shapes. Lanceolate shaped leaves are more resistant to BB, due to the availability of a narrow space for accumulation of BB spores.

In the current study, leaf pubescence has also indicated a significant negative correlation with the BBDI. The leaf pubescence was observed under microscope and categorized as; sparse, intermediate or dense. When pubescence density increases, it can act as a physical barrier for infection [21] limiting the susceptibility to BB. In a study conducted for *Uromyces*, the presence of dense leaf pubescence has been documented to retard the germination of spores on the surface of bean leaves by trapping the spores [22], thereby reducing the probability of germ tubes reaching the penetration site [24]. A high density of trichomes can also prevent mycelial penetration and infection of other biotrophic fungi [24]. It is reported that an increased number of hydrophobic



pubescence may repel water from the leaf surfaces, thus preventing successful penetration of fungal germ tubes [24]. Alternatively, a high trichome number may simply reduce the frequency of germ tube contact points that can lead to penetration [25]. The straight leaf apex habit was more vulnerable to disease infection rather than down turned leaf, as suggested by the negative correlation between leaf apex habit and BBDI.

On the other hand, several factors such as leaf colour, leaf apex shape, leaf base habit, leaf venation, upper leaf surface, length of mature leaf, length of mature leaf petiole have shown positive correlations with BBDI. All the positively correlated characteristics increase the probability of being infected by pathogen spores, through facilitating the trapping of spores and providing more surface area to interact with the plant leaf. Increase of the leaf length and leaf width may increase the surface area of the leaf and allow the spores of pathogens to increase the chance of contamination [12].

Distinct mid rib and lateral leaf venation system with bullate has also made the leaf more vulnerable to disease infection, than indistinct sunken leaf venation in lamina. It may facilitate the trapping of spores in the wind. When considering the impact of leaf base area, the susceptibility to BB tend to increase from attenuate to blunt shapes, when the leaf base surface area increases. Leaf apex shape denoted a positive correlation with BBDI and therefore the BB severity tend to vary as acute < obtuse < attenuates in shapes, respectively.

Most outstanding leaf morphological characteristics such as upper leaf surface and length of mature leaf petiole were denoting significant positive correlations with BBDI ( $P < 0.05$  at 95% level of confidence). A rough upper leaf surface generally leads to high retention of fungal spores, while increasing the length of leaf petiole, may favour the exposure to spores of pathogen [26].

After considering the correlation of all the studied leaf morphological parameters with susceptibility to BB, a simple model to predict the vulnerability of a tea plant to BB (based on morphological features) was developed through step-wise regression analysis. However, the current model only considers the leaf morphological factors with less attention on other external environmental factors such as soil nutrients, light etc. Further, regardless of the combined effect of upper leaf surface and leaf pubescence in terms of BB susceptibility, individual thresholds for each parameter were also developed through a ROC analysis. However, it should be noted that both, upper leaf surface and leaf pubescence are non-parametric morphological parameters and the model was derived with a limited number of samples. Therefore, the current thresholds and leaf morphology-based model is recommended for preliminary screening of BB susceptibility, due to its rapid and limited resource consumptive (labour and cost) nature, prior to further confirmation with more precise molecular markers.

## CONCLUSIONS

Constitutive barriers limited or completely inhibited the penetration of tea tissues by pathogenic fungi. The resistant individuals of the analyzed  $F_1$  segregation population were characterized by a significantly higher pubescence density, than susceptible forms. In resistant individuals, upper leaf surface was smooth, which minimise the accumulation of pathogen spores.

Based on the findings, upper leaf surface and leaf pubescence can be used to evaluate the susceptibility to BB incidence in tea plants. The proposed model can be used for preliminary evaluation of BB resistant or BB susceptible traits and it should be validated with more tea cultivars in different ecological regions to enhance the reliability and accuracy.

## CONFLICTS OF INTEREST

The authors declare that there are no conflicts

of interest.

#### AUTHORS' CONTRIBUTIONS

TK: Carried out the investigations, data collection, supported the statistical analysis, and wrote the manuscript; MK and JW: Supervised the study; LU: Analysed data and wrote the manuscript; CP: supervised the study and revised the manuscript; NE: Supported data collection process. All authors read and approved the manuscript.

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#### REFERENCES

- 1 Soni RP, Katoch M, Kumar A, Ladohiya R, Verma P. Tea: production, composition, consumption and its potential as an antioxidant and antimicrobial agent. *International Journal of Food and Fermentation Technology*. 2015; 5(2):95-106.
- 2 Sharma VK, Bhattacharya A, Kumar A, Sharma HK. Health benefits of tea consumption. *Tropical Journal of Pharmaceutical Research*. 2007; 6(3):785-792.
- 3 Park DJ, Imm JY, Ku KH. Improved dispersibility of green tea powder by microparticulation and formulation. *Journal of Food Science*. 2001 Aug; 66(6):793-798.
- 4 Hayat K, Iqbal H, Malik U, Bilal U, Mushtaq S. Tea and its consumption: Benefits and risks. *Critical Reviews in Food Science and Nutrition*. 2015 Jun 7; 55(7):939-54.
- 5 Sinniah GD, Kumara KW, Karunajeewa DG, Ranatunga MA. Development of an assessment key and techniques for field screening of tea (*Camellia sinensis* L.) cultivars for resistance to blister blight. *Crop Protection*. 2016 Jan 1; 79:143-149.
- 6 Punyasiri PA, Abeysinghe IS, Kumar V. Preformed and induced chemical resistance of tea leaf against *Exobasidium vexans* infection. *Journal of Chemical Ecology*. 2005 Jul; 31(6):1315-1324.
- 7 Boekhout T. A revision of ballistoconidia-forming yeasts and fungi. *Studies in Mycology*. 1991; 33:1-94.
- 8 Keith L, Ko WH, Sato DM. Identification guide for diseases of tea (*Camellia sinensis*). *Plant Disease*. 2006; 33: 1-4
- 9 Baby UI, Balasubramanian S, Ajay D, Premkumar R. Effect of ergosterol biosynthesis inhibitors on blister blight disease, the tea plant and quality of made tea. *Crop Protection*. 2004 Sep 1; 23(9):795-800.
- 10 Agrios GN. Control of Plant Diseases. In: *Plant Pathology*, 4<sup>th</sup> Edition, Academic Press, San Diego. 1997: 200-216
- 11 Iqbal Z, Khan MA, Sharif M, Shah JH, ur Rehman MH, Javed K. An automated detection and classification of citrus plant diseases using image processing techniques: A review. *Computers and Electronics in Agriculture*. 2018 Oct 1; 153:12-32.
- 12 Jain A, Sarsaiya S, Wu Q, Lu Y, Shi J. A review of plant leaf fungal diseases and its environment speciation. *Bioengineered*. 2019 Jan 1; 10(1):409-424.
- 13 Ponmurugan P, Baby UI. Evaluation of fungicides and biocontrol agents against Phomopsis canker of tea under field conditions. *Australasian Plant Pathology*. 2007 Jan;36(1):68-72.
- 14 Premkumar R, Ponmurugan P, Manian S. Growth and photosynthetic and biochemical responses of tea cultivars to blister blight infection. *Photosynthetica*. 2008 Mar; 46(1):135-8.
- 15 Abeysinghe DC, Mewan KM, Kumari WM, Kumara KL. Morphological and molecular differences of *Exobasidium vexans* massee causing blister blight disease of tea. *Journal of the Korean Tea Society*. 2015 special issue; 21:72-76.
- 16 Baby UI, Kumar RR, Mandal AK, Balamurgan A, Muraleedharan N, Joshi SD, Premkumar R, Rahul PR. Genetic and morphological variation of tea (*Camellia*

- sinensis*) blister blight pathogen (*Exobasidium vexans*) in Southern India revealed by RAPD markers and spore morphology. *Sri Lanka Journal of Tea Science*. 2009; 74(2): 52-61.
- 17 Menezes H, Jared C. Immunity in plants and animals: Common ends through different means using similar tools. *Comp Biochem Physiol C Toxicol Pharmacol*. 2002 May; 132(1):1-7.
- 18 Bhau BS, Sharma DK, Bora M, Gosh S, Puri S, Borah B, Kumar DG, Wann SB. Molecular markers and crop improvement. In *Abiotic Stress Response in Plants*. Weinheim: Wiley-VCH Verlag GmbH and Co. KGaA.
- 19 Sinniah GD, Alagiyawadu U, Wasantha KL. An assessment key for tea blister blight: Development and validation. In: *Proceedings of 4<sup>th</sup> Symposium on Plantation Crop Research*. Colombo, Sri Lanka; 2012. p. 135-44.
- 20 International Plant Genetic Resources Institute (IPGRI). Descriptors for Tea (*Camellia sinensis*). International Plant Genetic Resources Institute, Rome, Italy. 1997.
- 21 Martin C, Glover BJ. Functional aspects of cell patterning in aerial epidermis. *Current Opinion in Plant Biology*. 2007 Feb 1; 10(1):70-82.
- 22 Mmbaga MT, Steadman JR, Roberts JJ. Interaction of bean leaf pubescence with rust urediniospore deposition and subsequent infection density. *Annals of Applied Biology*. 1994 Oct; 125(2):243-254.
- 23 Shaik M. Race non-specific resistance in bean cultivars to races of *Uromyces appendiculatus* var. and its correlation with leaf epidermal characters. *Phytopathology*. 1985; 75: 478-481
- 24 Kortekamp A, Zyprian E. Leaf hairs as a basic protective barrier against downy mildew of grape. *Journal of Phytopathology*. 1999 Jul; 147(7-8):453-459.
- 25 Niks RE, Rubiales D. Potentially durable resistance mechanisms in plants to specialised fungal pathogens. *Euphytica*. 2002 Mar; 124(2):201-216.
- 26 Jenks MA, Ashworth EN. Plant epicuticular waxes: Function, production, and genetics. *Horticultural Reviews*. 2009; 23: 1-68.