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## EFFECT OF POWDERY MILDEW INFECTION ON DPPH RADICAL SCAVENGING ACTIVITY AND FERRIC-REDUCING ANTIOXIDANT POWER OF PLANTS

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### ABSTRACT

Powdery mildews are the most harmful and destructive fungal disease to angiosperms. Pathogenicity of this disease alters metabolism of host plants. Here an attempt has been made to study changes in the Antioxidant ability of host plants during infection. Two assay techniques viz. DPPH and FRAP (methods of quantitative measurement of antioxidant activity) are used to study effect of fungal pathogenicity on *Xanthium strumarium* L., (Asteraceae) and *Dalbergia sissoo*. Roxb ex DC. (Fabaceae

**Keywords:** Powdery Mildew Fungi, DPPH, FRAP, *Xanthium strumarium* and *Dalbergia sissoo*.

### INTRODUCTION

Fungi are highly diverse and versatile organisms adapted to all types of environments. One of the interesting groups of fungi is powdery mildew fungi belongs to Family- Erysiphaceae, Order- Eryisiphales, Class- Pyrenomycetes, and Division- Ascomycotina. They reproduce asexually by enormous production of conidia giving appearance like powdery growth and hence, the name 'powdery mildew'. They are characterized by spots or patches of white to grayish, talcum-powder-like growth on plant parts.

The powdery mildews cause serious damage to angiosperms of various categories. The typical symptom of this disease is most commonly observed on upper as well as lower surface of leaves. It also affects the young stems, buds, flowers, and young fruits. Infected leaves may become distorted and fall prematurely. The infected buds may fail to open. The

pathogen of powdery mildew grows well in environment with high humidity and cool temperatures<sup>1</sup>. The powdery mildew of *X. strumarium* is caused by *Oidium xanthami* and that of the *sisso* is by *Ovulariopsis sisso* sp. nov (anamorph stage) and *Phyllactinia corylea* (telomorph stage).

With the help of Different assays, antioxidant capacity of biological samples can be measured. The concept of antioxidant capacity first originated from chemistry and then adapted to fields of biology<sup>2</sup>. There are many metabolic processes continuously going on in any living organism. As a result of normal biochemical reactions in the body, there is formation of Free radicals, which may be responsible for development of various disorders like aging, hair loss diabetes, heart disease, neurodegenerative disorders and cancer<sup>3-4</sup>. All living organisms possess natural defense mechanisms to reduce the activity of free radicals in the form of enzymes, vitamins and many others as supplementary antioxidants. Particularly in plants, bioactive compounds such as flavonoids and terpenoids play important role in the defense against free radicals<sup>5-6</sup>.

In the present study, we have evaluated effect of powdery mildew infection on the total antioxidant capacities of plants viz *X. strumarium* and *D. sissoo* from Satara, (M.S) by using a modified DPPH assay<sup>7</sup> and an improved FRAP assay<sup>8</sup>. These data may help in understanding change in antioxidant activities and severity of disease for plants.

## MATERIAL AND METHODS

For current investigation *healthy and infected leaves with powdery mildew fungi of X. strumarium* and *D. sissoo* were collected and subjected to aqueous extraction on sonicator essential for studying antioxidant capacity.

### a) DPPH free radical scavenging activity

The antioxidant activities of plant extracts and the standard were assessed on the basis of the free radical scavenging effect of the stable 1, 1-diphenyl-2-picrylhydrazyl (DPPH)-free radical activity by modified method<sup>7</sup>. The diluted working solutions of the test extracts were prepared in distilled water. DPPH (0.002%) was prepared in methanol and 3 ml of this solution was mixed with 1 ml of sample solution and standard solution separately. These mixtures were kept in dark for 30 min and optical density was measured at 517 nm using UV-VIS Spectrophotometer 119.

**b) Ferric-reducing antioxidant power (FRAP)**

The ferric reducing antioxidant power (FRAP) assay was used to measure the total antioxidant power of leaf extracts under experimentation. In the FRAP assay, reductants (antioxidants) in the sample reduce Fe<sup>3+</sup>/tripyridyltriazine complex, present in stoichiometric excess, to the blue colored ferrous form, with an increase in absorbance at 595 nm. Antioxidant activity assays were performed as per the method described by Benzie and Strain. The results are expressed as ascorbic acid equivalent antioxidant capacity.

**RESULT AND DISCUSSION**

An antioxidant can be defined as “any substance that, when present at low concentrations compared with those of an oxidizable substrate, significantly delays or prevents oxidation of that substrate”<sup>9</sup>. When we use synthetic antioxidants, they accumulate in the body which can result in liver damage and carcinogenesis. Such problems are not seen by using natural antioxidants extracted from herbs and spices, therefore, such natural antioxidants are used in food applications. These extracts are safe, potentially nutritional and have therapeutic effects. Plant material such as vegetables fruits, seeds, woods, barks, roots, and leaves have been examined as potential source of antioxidants<sup>10-14</sup>. A variety of assays have been developed to measure the concentration of specific antioxidant as well as that of all antioxidants present within the cells of an organism. The purpose of current study is to evaluate antioxidant capacity in infected and healthy leaves by DPPH assay and FRAP assay during powdery mildew infection.

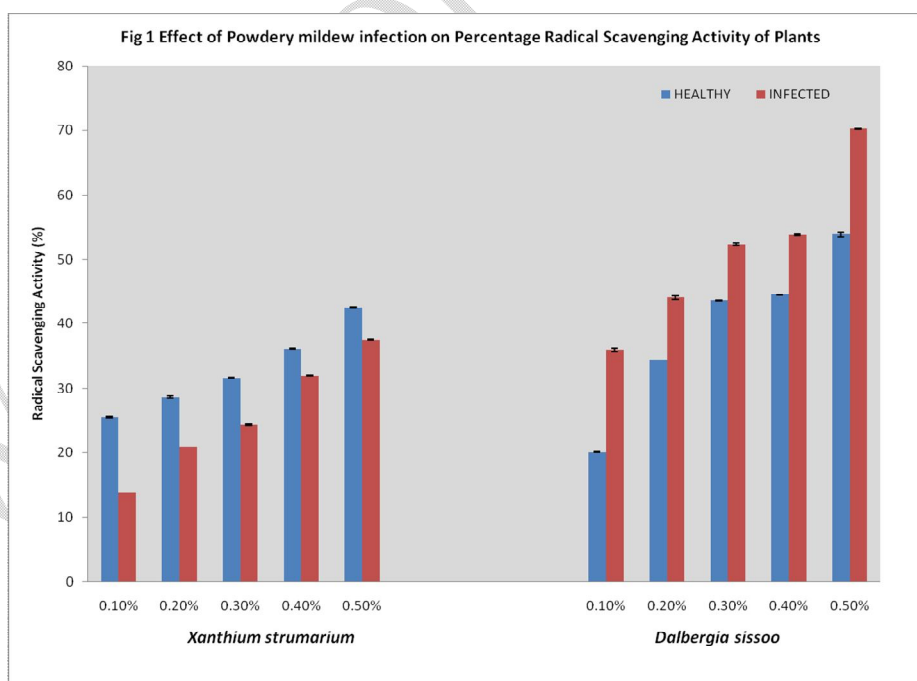
**a. DPPH radical scavenging activity**

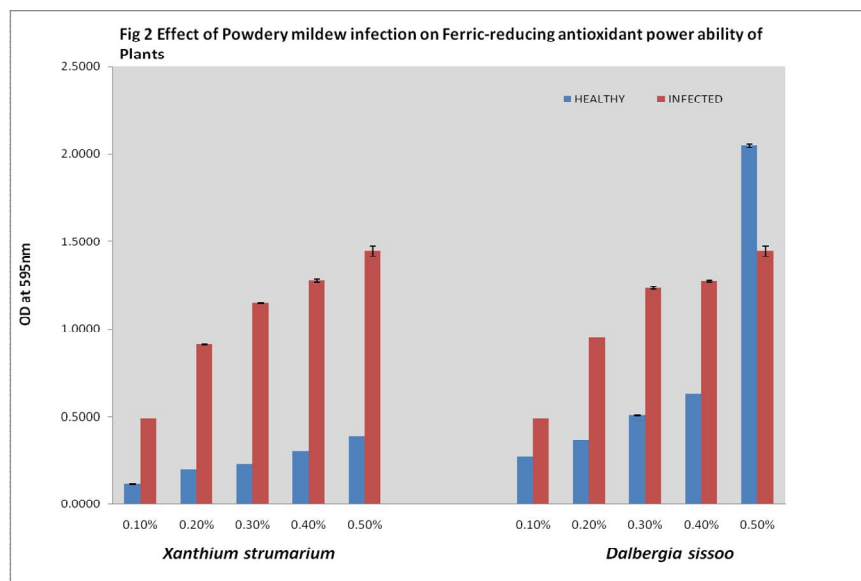
1,1-Diphenyl-2-picrylhydrazyl (DPPH), is a kind of stable organic radical. The capacity of biological reagents to scavenge DPPH radical can be expressed as its magnitude of antioxidation ability. The DPPH oxidative assay<sup>15</sup> is used worldwide in the quantification of radical-scavenging capacity. The DPPH assay has many advantages over other methods, such as good stability, credible sensitivity, simplicity and feasibility<sup>16-17</sup>. The changes in the free radical scavenging ability of aqueous extracts of healthy and powdery mildew infected leaves of *X. strumarium* and *D.sissoo* on the basis of percent inhibition is presented in Fig 1. It is evident from the figure that the healthy material of *X. strumarium* shows great ability of free radical scavenges. Due to powdery mildew infection in *X. strumarium* this ability goes down. While in case of *D. sissoo* exact reverse results are observed, that is during infection the

radical scavenging ability increases. It can be concluded that fungal pathogen alters, DPPH radical scavenging activity during powdery mildew infection. Effect of these fungi on radical scavenging activity is different for each plant species.

### b. Ferric-reducing antioxidant power (FRAP)

FRAP has been used to analyze antioxidant status in humans after hyperbaric oxygen therapy<sup>18</sup>. It has also been used to compare antioxidant activity in plants and mammals<sup>19</sup> and plant extracts<sup>20</sup>. FRAP measures the ability of the extract to donate electron to Fe(III). The higher the FRAP value, the greater is the antioxidant activity. Change in ferric reducing antioxidant power of *X. strumarium* and *D. sissoo* during stage of powdery mildew infection is presented in Fig 2. It is clear from the results that the FRAP activity of leaves of both studied species goes increasing during infection except in *D. sissoo* at higher concentration of healthy plant material (0.5%). The gradual increase in absorbance of FRAP was observed in studied species along with increasing concentration. From all these observations it can be concluded that although the plants suffers from severe infection, it does not loose it's FRAP antioxidant capacity. It is evident from results that during powdery mildew infection Ferric-reducing antioxidant power goes on increasing.





## CONCLUSION

It can be concluded from results that powdery mildew infection alters the antioxidant power of plant. The effect may be positive or negative in case of radical scavenging activity. Change in radical scavenging activity is different for every plant species. Each plant species has its own specific response for powdery mildew infection. But for change in Ferric-reducing antioxidant power, infection always shows positive response.

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