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## Estimation of enzymatic and non-enzymatic activity of rapeseed-mustard against *Alternaria* blight

**Bhawana Mathur, NA Khan, DK Dwivedi, Adesh Kumar, Ashish Kumar and Rahul Maurya**

### Abstract

An experiment was conducted with 20 rapeseed mustard varieties Rohini, Maya, Giriraj, NDRE-7, NDRS-9-1NDYR-8, Vardan, Vaibhav, Pusa M-27, NDRE-1-11-1, Pusa Gold-45, NRCHB-101, RH-479 Pusa M-24, Kranti, NDR-8501, RH 406, NRCDR-2, RGN-48 and Basanti at Student's instructional farm of Acharya Narendra Deva University of Agriculture and Technology Kumarganj Ayodhya (U. P.) This study evaluated the enzymatic antioxidant and non-enzymatic antioxidant activity in healthy leaves and leaves exposed to *Alternaria* blight (Infected), which differed in phenol content, catalase activity and peroxidase activity. The content and activity of phenol, catalase, peroxidase were used parameter to identify the resistant genotype for breeding programmes. The finding revealed that maximum phenol content was recorded in Kranti, whereas maximum catalase activity and peroxidase activity was observed in varieties NDRS-9-1 and Kranti while minimum phenol content, catalase activity and peroxidase activity was observed Rohini in both infected and non-infected leaves of rapeseed mustard varieties.

**Keywords:** Rapeseed-mustard, *alternaria* blight, phenol, catalase and peroxidase

### Introduction

Rapeseed-mustard crops in India comprises of traditionally grown indigenous species, namely toria (*Brassica rapa* L. var. toria), brown sarson (*Brassica rapa* L. var. brown sarson), yellow sarson (*Brassica rapa* L. var. yellow sarson), Indian mustard (*Brassica juncea*), and black mustard (*Brassica nigra*), which have been grown since about 3,500 BC along with non-traditional species like gobhi sarson (*Brassica carinata*) and Ethiopian mustard or Karan rai (*Brassica carinata*).

The country witnessed yellow revolution through a phenomenal increase in production and productivity from 2.68 MT and 650 kg/ha in 1985-86 to 6.96 MT and 1022 kg/ha in 1996-1997, respectively. Despite these achievements, there is still a gap between production potential and actual realization (Shekhawat *et al.*, 2012) [1].

India grows annual oilseeds on an area of over 25.74 million hectares, producing 30.55 million tonnes, with a productivity of around 1188 kg per hectare for the quinquennium ending (QE) 2019-20.

Among these, *Alternaria* blight of rapeseed mustard caused by *Alternaria brassicae* (Berk.) Sacc. is a serious problem with huge economic consequences. This is one of the most serious and devastating disease of mustard under normal conditions.

*Alternaria* is a completely detrimental pathogen causing a widespread destruction in vegetables and other economically important crops. The best and inexpensive way to control *Alternaria* blight disease of rapeseed-mustard is to use resistant varieties. *Alternaria brassicae* species have capability to survive in seeds for numerous months at different temperatures and relative humidity.

The plants peroxidases have been involved in many defense-related processes, including the allergic responses, lignification, synthesis of phenolics, glycoprotein, suberization and phytoalexin production. Cell often use catalase to quickly breakdown hydrogen peroxide into less reactive gaseous oxygen and water molecules so as not affect the cell (Bolwell and Wojtaszek, 1997) [2].

### Material and Methods

The study was carried out with 20 rapeseed mustard varieties, namely Rohini, Maya, Giriraj, NDRE-7, NDRS-9-1NDYR-8, Vardan, Vaibhav, Pusa M-27, NDRE-1-11-1, Pusa Gold-45,

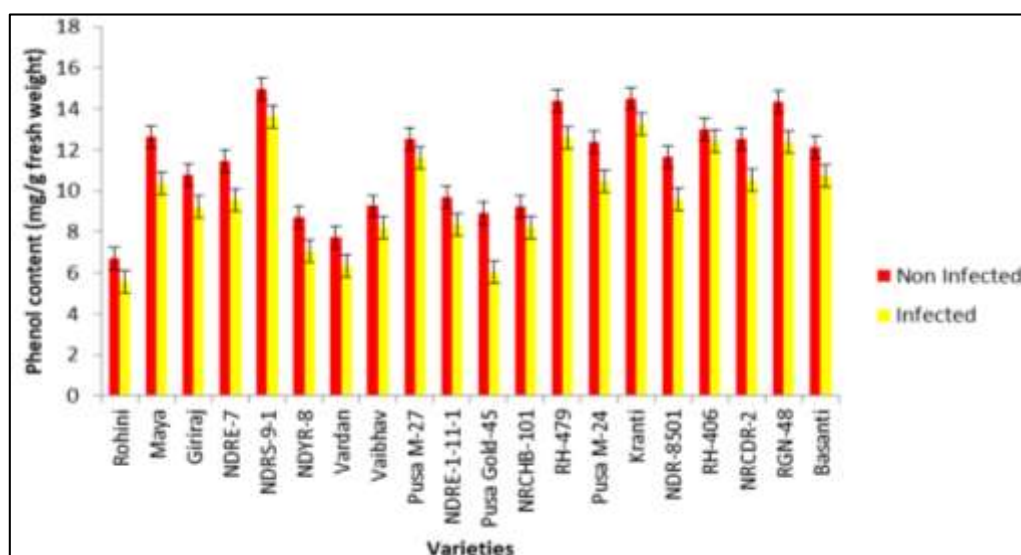
NRCHB-101, RH-479 Pusa M-24, Kranti, NDR-8501, RH 406, NRCDR-2, RGN-48 and Basanti at the student's instructional farm and a set of soil potted in greenhouse department of Plant Molecular Biology and Genetic Engineering, Acharya Narendra Deva University of Agriculture and Technology, Kumarganj Ayodhya (U.P.) in rabi season 2021-22 and 2022-2023. Geographical location of Ayodhya district lies between latitude 26° 47" north and longitude 82° 12" east, at 113 meters above sea level mean in ground nodal alluvial eastern Uttar Pradesh. This study uses 20 rapeseed mustard varieties arranged in a randomized block design (RBD) with 3 replicates. All parameters in the laboratory, such as phenol were measured by the method described (Bray *et al.*, 1954) [3], catalase activity was estimated by the method described by (Sinha *et al.*, 1972) [9] and peroxidase activity were determined using the method described in (Curne and Galston, 1959) [10].

### 1. Phenol content

In diseased free leaves of rapeseed mustard maximum phenol content was observed in NDRS-9-1 *i.e.* 14.97 mg/g fresh weight followed by Kranti *i.e.* 14.52 mg/g fresh weight were significantly increased over rest of the genotypes and lowest phenol content was recorded in Rohini *i.e.* 6.68 mg/g fresh weight followed by Pusa Gold-45 *i.e.* 8.90 mg/g fresh weight when compared to the diseases free leaves of rapeseed mustard varieties all the twenty varieties revealed differing phenol content in diseased leaves, maximum phenol content was observed in NDRS-9-1 *i.e.* 13.60 mg/g fresh weight followed by Kranti *i.e.* 13.28 mg/g fresh weight and minimum phenol content was observed in Rohini *i.e.* 5.57 mg/g fresh weight followed by Pusa Gold-45 *i.e.* 6.03 mg/g fresh weight were attained remarkable variation over reset of the genotypes. The whole data suggest that the phenol content increased with the infection of disease. Similar result was found by Neeraj *et al.*, (2010) [6].

**Table 1:** Estimation of total phenol content in diseased free leaves and diseased leaves of rapeseed mustard exposed to *Alternaria* blight.

S. No.	Varieties	(mg/g fresh weight) Disease free leaves	(mg/g fresh weight) Diseased leaves
1.	Rohini	6.68	5.57
2.	Maya	12.66	10.36
3.	Giriraj	10.80	9.24
4.	NDRE-7	11.43	9.56
5.	NDRS-9-1	14.97	13.60
6.	NDYR-8	8.71	7.05
7.	Vardan	7.74	6.32
8.	Vaibhav	9.26	8.18
9.	Pusa M-27	12.52	11.61
10.	NDRE-1-11-1	9.68	8.34
11.	Pusa Gold-45	8.90	6.03
12.	NRCHB-101	9.21	8.18
13.	RH-479	14.41	12.58
14.	Pusa M-24	12.40	10.45
15.	Kranti	14.52	13.28
16.	NDR-8501	11.66	9.59
17.	RH 406	13.01	12.42
18.	NRCDR-2	12.54	10.53
19.	RGN-48	14.35	12.39
20.	Basanti	12.11	10.72
	SEM+	0.16	0.14
	CD	0.47	0.39
	CV%	2.9	2.1



**Fig 1:** Estimation of total phenol content in diseased leaves and diseased free leaves of rapeseed mustard varieties

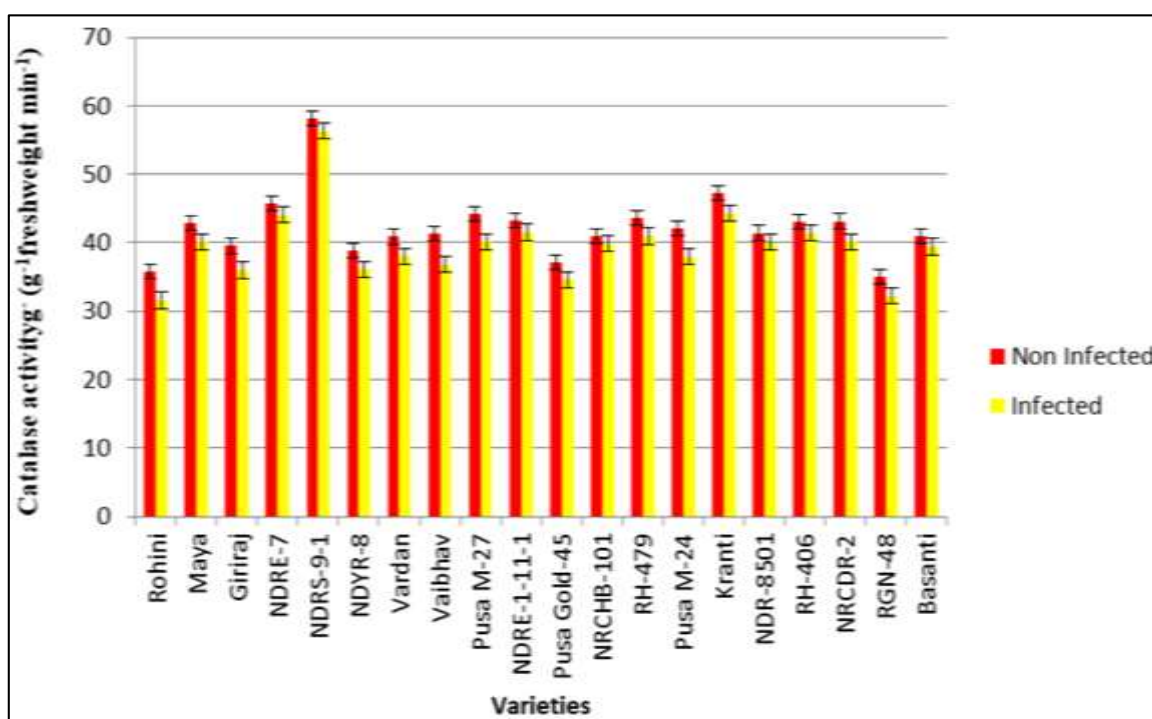
## 2. Catalase activity

Catalase enzyme activity in leaves of rapeseed mustard is depicted in table 2. The observation suggest that there was a slow decrease in catalase activity with the increase in disease. The catalase activity of disease free leaves, range between 35.08 to 58.10 in  $\text{g}^{-1}\text{freshweight min}^{-1}$ , In disease free leaves leaves of rapeseed mustard highest catalase activity was observed in NDRS-9-1 *i.e.* 58.10  $\text{g}^{-1}\text{freshweight min}^{-1}$  followed by Kranti *i.e.* 47.20  $\text{g}^{-1}\text{freshweight min}^{-1}$  whereas minimum catalase activity was noticed in Rohini *i.e.* 35.80  $\text{g}^{-1}\text{freshweight min}^{-1}$  followed by Pusa Gold-45 *i.e.* 37.06  $\text{g}^{-1}\text{freshweight min}^{-1}$  furthermore in diseased leaves of rapeseed

mustard highest catalase activity in NDRS-9-1 *i.e.* 56.33  $\text{g}^{-1}\text{freshweight min}^{-1}$  followed by Kranti *i.e.* 44.26  $\text{g}^{-1}\text{freshweight min}^{-1}$  while minimum catalase activity was noticed in Rohini *i.e.* 31.56  $\text{g}^{-1}\text{freshweight min}^{-1}$  and Pusa Gold-45 *i.e.* 34.56  $\text{g}^{-1}\text{freshweight min}^{-1}$  similar result was reported by Gupta *et al.*, (1990) [5] catalase activity was appreciably higher at initial stage, at later stages it dropped markedly. In response to infection, its activity decreased. The role of phenolics and oxidative enzymes in determining resistance in mustard against Alternaria blight disease has been highlighted catalase activity was decreased, with the progress of disease.

**Table 2:** Catalase activity in diseased free leaves and diseased leaves of Rapeseed mustard exposed to Alternaria blight.

S. No.	Varieties	( $\text{g}^{-1}\text{freshweight min}^{-1}$ ) Disease free leaves	( $\text{g}^{-1}\text{freshweight min}^{-1}$ ) Diseased leaves
1.	Rohini	35.80	31.56
2.	Maya	42.80	40.13
3.	Giriraj	39.53	35.96
4.	NDRE-7	45.76	44.06
5.	NDRS-9-1	58.10	56.33
6.	NDYR-8	38.80	36.10
7.	Vardan	40.86	38.06
8.	Vaibhav	41.36	36.83
9.	Pusa M-27	44.16	40.16
10.	NDRE-1-11-1	43.23	41.53
11.	Pusa Gold-45	37.06	34.56
12.	NRCHB-101	40.96	39.83
13.	RH-479	43.56	40.96
14.	Pusa M-24	42.10	37.96
15.	Kranti	47.20	44.26
16.	NDR-8501	41.40	40.03
17.	RH 406	43.06	41.40
18.	NRCDR-2	43.13	40.16
19.	RGN-48	35.00	32.20
20.	Basanti	40.90	39.40
	SEM+ <sub>1</sub>	0.35	0.28
	CD	0.99	0.81
	CV%	1.4	1.2



**Fig 2:** Estimation of catalase activity in diseased free leaves and diseased leaves of rapeseed mustard varieties

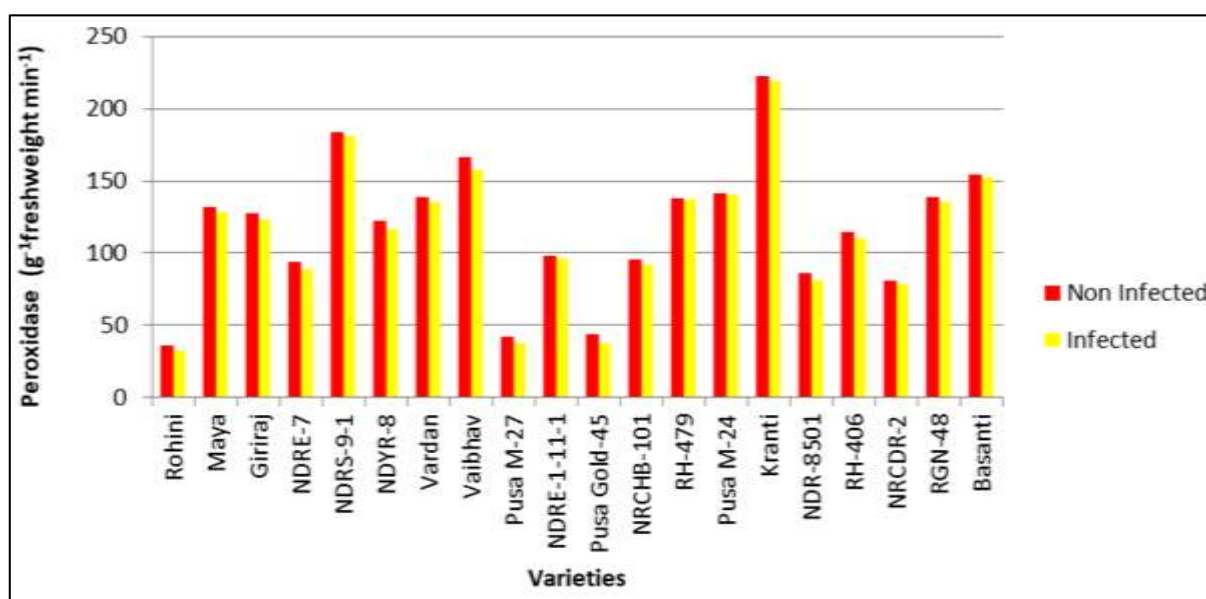
### 3. Peroxidase activity

Peroxidase enzyme activity in diseased free leaves and diseased leaves of rapeseed mustard was examined. Among all the genotypes in diseased free leaves of rapeseed mustard highest peroxidase activity was observed in Kranti *i.e.* 222.13  $\text{g}^{-1}$  fresh weight  $\text{min}^{-1}$  along with NDRS-9-1 *i.e.* 183.63  $\text{g}^{-1}$  fresh weight  $\text{min}^{-1}$  and Vaibhav *i.e.* 166.26  $\text{g}^{-1}$  fresh weight  $\text{min}^{-1}$  while minimum peroxidase activity was observed in Rohini *i.e.* 36.12  $\text{g}^{-1}$  fresh weight  $\text{min}^{-1}$  along with Pusa Gold-45 *i.e.* 44.06  $\text{g}^{-1}$  fresh weight  $\text{min}^{-1}$  coequally in diseased leaves of rapeseed mustard highest peroxidase activity was noticed in Kranti *i.e.* 218.90  $\text{g}^{-1}$  fresh weight  $\text{min}^{-1}$  along with NDRS-9-1

*i.e.* 180.66  $\text{g}^{-1}$  fresh weight  $\text{min}^{-1}$  and Vaibhav *i.e.* 158.03  $\text{g}^{-1}$  fresh weight  $\text{min}^{-1}$  while minimum peroxidase activity was recorded in Rohini *i.e.* 32.80  $\text{g}^{-1}$  fresh weight  $\text{min}^{-1}$  and Pusa Gold-45 *i.e.* 38.20  $\text{g}^{-1}$  fresh weight  $\text{min}^{-1}$ . Equivalently Gupta *et al.*, (1990) [5] reported that specific activities of polyphenol oxidase remained higher while peroxidase activities were lower in tolerant cultivars comparison to susceptible ones. In response to infection, the activity of both the enzymes increased sharply in all the cultivars but, this increase is significantly higher in susceptible cultivars than in tolerant cultivars.

**Table 3:** Peroxidase activity in diseased free leaves and diseased leaves of rapeseed mustard exposed to Alternaria blight.

S. No.	Varieties	( $\text{g}^{-1}$ freshweight $\text{min}^{-1}$ ) Non infected	( $\text{g}^{-1}$ freshweight $\text{min}^{-1}$ ) Infected
1.	Rohini	36.12	32.80
2.	Maya	131.66	128.36
3.	Giriraj	127.3	123.53
4.	NDRE-7	93.96	88.66
5.	NDRS-9-1	183.63	180.66
6.	NDYR-8	122.13	116.40
7.	Vardan	138.43	135.60
8.	Vaibhav	166.26	158.03
9.	Pusa M-27	42.06	38.13
10.	NDRE-1-11-1	98.56	96.63
11.	Pusa Gold-45	44.06	38.20
12.	NRCHB-101	95.36	91.93
13.	RH-479	137.73	136.76
14.	Pusa M-24	141.2	140.93
15.	Kranti	222.13	218.90
16.	NDR-8501	85.96	81.40
17.	RH 406	114.40	110.50
18.	NRCDR-2	80.66	78.16
19.	RGN-48	138.43	135.46
20.	Basanti	154.36	152.63
	SEM+ <sub>1</sub>	0.42	0.33
	CD	1.20	0.95
	CV%	0.6	0.5



**Fig 3:** Estimation of peroxidase activity in diseased free leaves and diseased leaves of rapeseed mustard varieties

### Conclusion

The results showed significant genotypic differences among all the different rapeseed cultivars. It has been determined that Rohini cultivar is more resistant to Alternaria brassica disease

than other cultivars. Studies have shown that phenol, catalase and peroxidase may play an important role in protecting rapeseed varieties against the pathogenesis of Alternaria blight.

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