

Revealing the effect on the nutrient content of garlic leaves after infected with *Alternaria porri*, causing purple spot disease.

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Abstract- The aim of this study was to evaluate the effect of pathogen *Alternaria porri*, who is responsible for purple blotch disease on leaves of onion in Maharashtra on nutrient contents. The experiment was conducted from year 2012 onward for successive 10 years. The result shows that the pathogen changes nutrients, the quantity of protein and amino acids elevated while remaining like Carbohydrate, reducing sugar, fibres, Vitamin C, lipid declined. The pathogenicity was confirmed when isolated pathogen was reinoculated on control and later the same symptoms are observed.

Keywords – *Alternaria*, Purple blotch, Nutrients, Disease, Allicin

1. INTRODUCTION

Garlic leaves have a long history of use in both food and traditional medicine. (Ayaz, E., and H. C. Alpsoy, 2007). Garlic was known to the ancient Egyptians, who used it in their food as well as for medicinal purposes. Garlic has been mentioned in Indian and Egyptian cultures as far back as 5,000 years, showing its ancient history of use (Petrovska, B. B., and S. Cekovska, 2010). Garlic has been used medicinally for over 7,000 years. It has been utilised for a variety of medical purposes and is renowned for its potential therapeutic properties. (L. Bayan 2014). According to historical evidence, garlic has been grown for at least 5,000 years. It has been grown all over the world and is an important part of many different cuisines. (Simon – ARS, USDA). The therapeutic benefits of garlic have been thoroughly researched in scholarly literature. Antibiotic, antiseptic, balsamic, and antihypertensive activities have been discovered in it. (Children's Health at Stanford Medicine). Cultural Significance: distinct nations have assigned distinct cultural meanings to garlic. It was placed over doorways as a kind of protection since it was thought to ward off the evil eye. Additionally, it was regarded as an aphrodisiac and utilized in magic potions. (S. Fluegel). Although garlic leaves have historical value, it's vital to remember that they are not as frequently utilized in cooking as garlic bulbs. They can still be used, though, and add a distinctive flavour to some foods.

Maharashtra is India's tenth largest producer of garlic. The state contributes for 0.48% of India's garlic production. Maharashtra produced garlic with an annual production volume of in 2019-20. Maharashtra placed 10th in garlic production in 2020-21, accounting for 0.71% of all garlic produced in India. The state planted garlic on 3,756 hectares, yielding 5,925 kg per hectare.

The content variation study was previously done on onion. (Shehu K. and Aliero A.A., 2010 and Dongre, Mayur. 2019)

2. MATERIAL AND METHOD

2.1 Samples Collection –

Maharashtra's topography is distinguished by its plateau landscape, the presence of the Sahyadri Range, and a diversified coastal area along the Arabian Sea. There is total 36 districts in Maharashtra. Geographically

Maharashtra state was divided into five zones they are Konkan, Paschim (Western) Maharashtra (Desh), North Maharashtra or Khandesh, Marathwada and Vidarbha formerly known as Central Provinces and Berar. For present study at least two samples were collected from each geographical zone (Wikipedia). The samples were collected on the spot in sterile plastic bag and kept in cool storage to check further degradation. Then the collected samples were processed at laboratory for isolation of pathogen and estimation of nutrients. (Table 1)

Table 1- Diseased plant samples collection sites Maharashtra state

Sr. no.	Place of Collection	Geographical Data
1.	Khandesh	21°04'56.8"N 74°50'33.5"E
2.	Khandesh	20°08'40.0"N 74°14'33.9"E
3.	Konkan	19°04'18.0"N 73°00'31.7"E
4.	Konkan	19°11'39.1"N 72°58'38.8"E
5.	Vidarbha	20°44'08.7"N 78°35'49.6"E
6.	Vidarbha	21°27'26.0"N 80°11'32.2"E
7.	Western Maharashtra	18°34'25.0"N 73°46'36.6"E
8.	Western Maharashtra	16°51'26.6"N 74°33'49.9"E
9.	Marathwada	19°51'46.7"N 75°20'36.7"E
10.	Marathwada	18°24'20.3"N 76°34'42.9"E

2.2 Symptoms on plant –

Purple blotch caused by *Alternaria Porri* (Ellis) was observe in the field as well as in market during early summer season. White small spots first observed on the leaves of garlic plant. As the bulb mature the severity of disease also increased. Crops of garlic from Khandesh region mostly affected by this economically important disease in month of January to March, it tries to understand that transition period of winter to summer season is more favourable for pathogen.

Leaves are susceptible to this pathogen. first symptom of this disease started as white small spots present on the upper surface of leaf, which later enlarge and converted to dark purple or brown in colour, Fig 1(A). Initially small portion covered by the disease but later spot enlarges and may occupy all leaf surface. It directly affects the photosynthetic activity. The bulb of diseased plant also affected by this disease. The size of bulb remains small or undeveloped.



Figure 1- purple blotch disease on Garlic leaf caused by *Alternaria porri*, collected from field.

2.3 Culture characters of isolated pathogen -

The pathogen isolated from diseased site were cultured on PDA later repeated subculture form a pure plate. In PDA *Alternaria porri* grow luxuriously at room temperature. Growth was started as white concentric spot. The spot acquires dark colour as the abundant conidia forms. Within 5 to 7-day full plate occupied with pathogen. From reverse side of plate dark black colour seen. Colony is dark olivaceous in colour, with cottony fluffy growth, irregular shape colony, entire margin. (Image 2 B)

2.4 Morphological characters of pathogen-

The septate hyaline hyphae on maturation form dark colour. Conidiophores are present singly or in groups, brown in colour. They are erect cylindrical and septate. Conidia ae straight, sometimes it may be curved at some point, with brown colour. Longitudinal Septae may be up to 3. Numbers of transverse Septae are 6 to 8. (Image 2 C)

Size of conidia ranges from 89 μm to 257 μm in length. The width of conidia ranges from 13 to 18 μm . Beak length of conidia was found similar to body length. But 3 or 4 times thinner in diameter. (Table-2)

Table 2- Characteristic of pathogen isolated from respective site

Sr. no.	Geographical location of sample collection site	Culture appearance on PDA plate		Conidia Length X Diameter (μm)	Beak length (μm)
		Colour of colony	Appearance		
1.	21°04'56.8"N 74°50'33.5"E	Olive green	Fluffy	198 X 16	94
2.	20°08'40.0"N 74°14'33.9"E	Dark brown	Fluffy	257 X 18	120
3.	19°04'18.0"N 73°00'31.7"E	Olivaceous brown	Fluffy	210 X 17	95
4.	19°11'39.1"N 72°58'38.8"E	Olive green	Fluffy	89 X 13	42
5.	20°44'08.7"N 78°35'49.6"E	Dark brown	Fluffy	110 X 15	54
6.	21°27'26.0"N 80°11'32.2"E	Dark brown	Fluffy	240 X 18	117
7.	18°34'25.0"N 73°46'36.6"E	Dark brown	Fluffy	230 X 16	112
8.	16°51'26.6"N 74°33'49.9"E	Dark brown	Fluffy	105 X 12	49
9.	19°51'46.7"N 75°20'36.7"E	Dark brown	Fluffy	92 X 14	44
10.	18°24'20.3"N 76°34'42.9"E	Olive green	Fluffy	195 X 17	91

2.5 Pathogenicity test of isolated pathogen on control plant leaves-

Pure culture obtained from diseased leaves of garlic were inoculated on the growing garlic plant. The plant was kept under high humidity condition. After 10 days the symptoms are seen on inoculated site. Comparison of symptoms was carried out with that of original field crop.

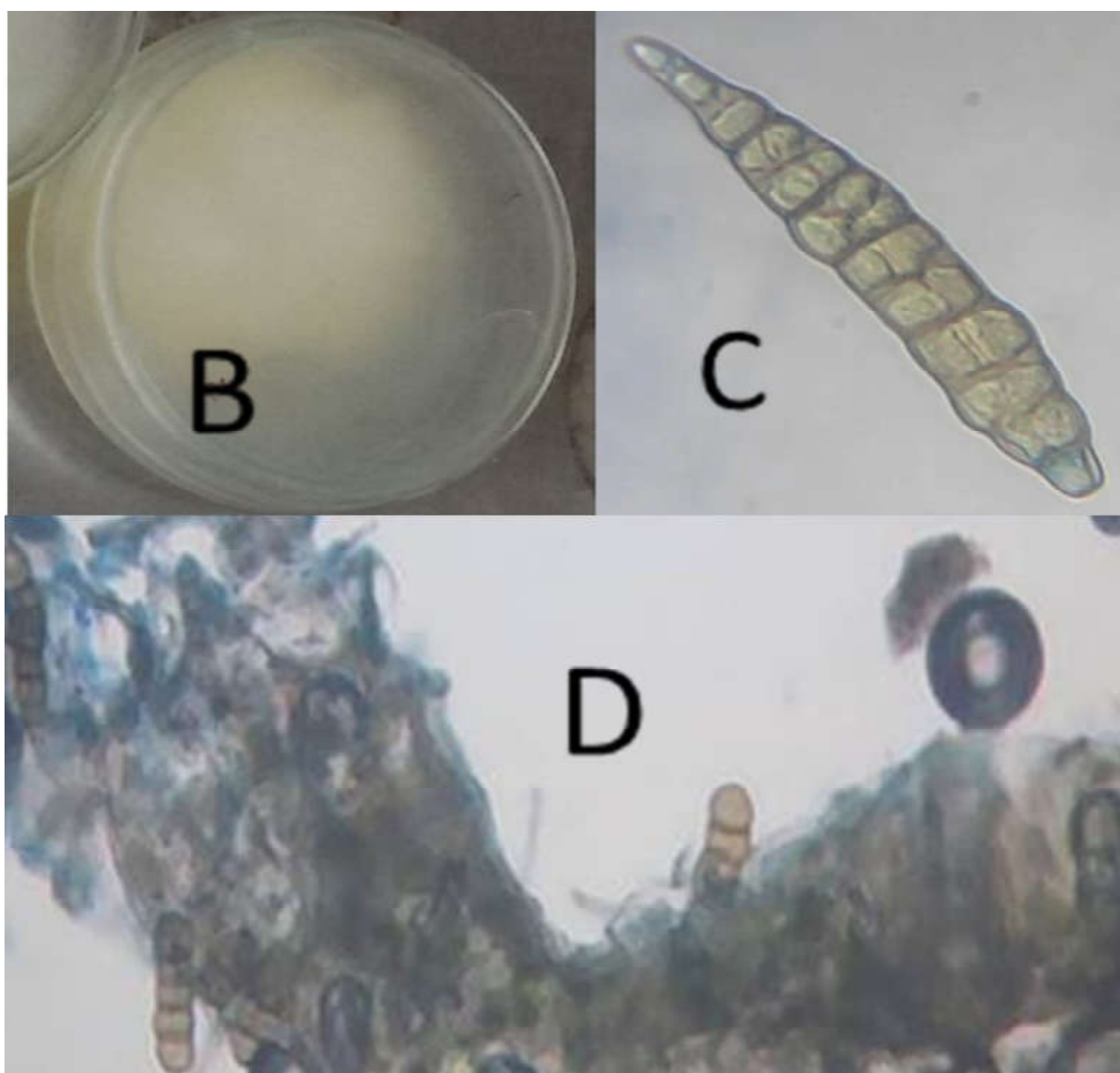


Figure 2- B-Pure culture of *Alternaria porri*, C-A conidia of *Alternaria porri*, D- Section of leaf showing conidia and hyphae.

2.6 Extraction and estimation of nutrients from control and diseased leaf-

Different nutrients are estimated utilizing methods and processes provided by various workers. Carbohydrate was calculated using the method described by Hedge, J E, and Hofreiter, B T (1962). The crude fibre content in a sample is evaluated using the acid alkali treatment method, in which the initial and final weight following ignition at 600°C give the crude fibre content. Reducing sugar was calculated using Nelson-Somogyi's arsinomolybdate reagent technique. Lowry's (1951) approach aided in protein estimation. Oil was extracted using the Soxhlet apparatus with petroleum ether as a solvent, according to the protocol described by Bligh, E.G., and Dyer, W.J. (1959). To estimate free amino acids, the Ninhydrin technique was used (Mahesha, 2012). Ruck's (1969) approach was used to determine dry matter and water content. Sadasivam and Manickam (2005) and Leslie J. Harris and Mamie Olliver, (1942) described how to quantify vitamin C levels using a 2,6- dichlorophenol-indophenol dye solution. Values were calculated using a Systronics2202 double-beam UV-visible Spectrophotometer whenever necessary.

3. Observations

The isolate pathogen is *Alternaria porri* when artificially inoculated on control leaves, they show the same symptoms, it confirms its potential to cause disease. Description provided by researchers like Ellis M.B., (1971), Bisht, I.S. and Agrawal,R.C. , (1993), Suheri H. and Price T.V (2000) and Shehu K. and Aliero A.A (2010) are helpful to confirm the

pathogen.

The diseased plant material and control plant material processed for estimation of various nutrients the changes occur in contents, these are tabulated in Table 3. The graphical representation of nutrient percentage alteration of nutrient is presented in Graph-1.

Table 3- Nutrient contents in control and diseased leaves and percentage alteration after disease

Sr. no.	Nutrients	Content in 100gram fresh material	Content in 100gram diseased vegetable	Percentage alteration due to disease
1.	Water content	69 Grams	74.724 Grams	8.295
2.	Total carbohydrate	6.8 Grams	3.0 Grams	-55.882
3.	Reducing sugar	3.8 Grams	2.6 Grams	-31.578
4.	Fibre	3.1 Grams	2.4 Grams	-22.580
5.	Protein	1.4 Grams	1.53 Grams	9.285
6.	Amino acids	2.8 Grams	3.16 Grams	12.857
7.	Lipids	0.2 Grams	00 Grams	-100
8.	Vitamin C	62 mg	6mg	-90
9.	Chlorophyll content (total)	13.4 mg	00mg	-100
10.	Dry matter	31 Grams	25.276 Grams	-18.464

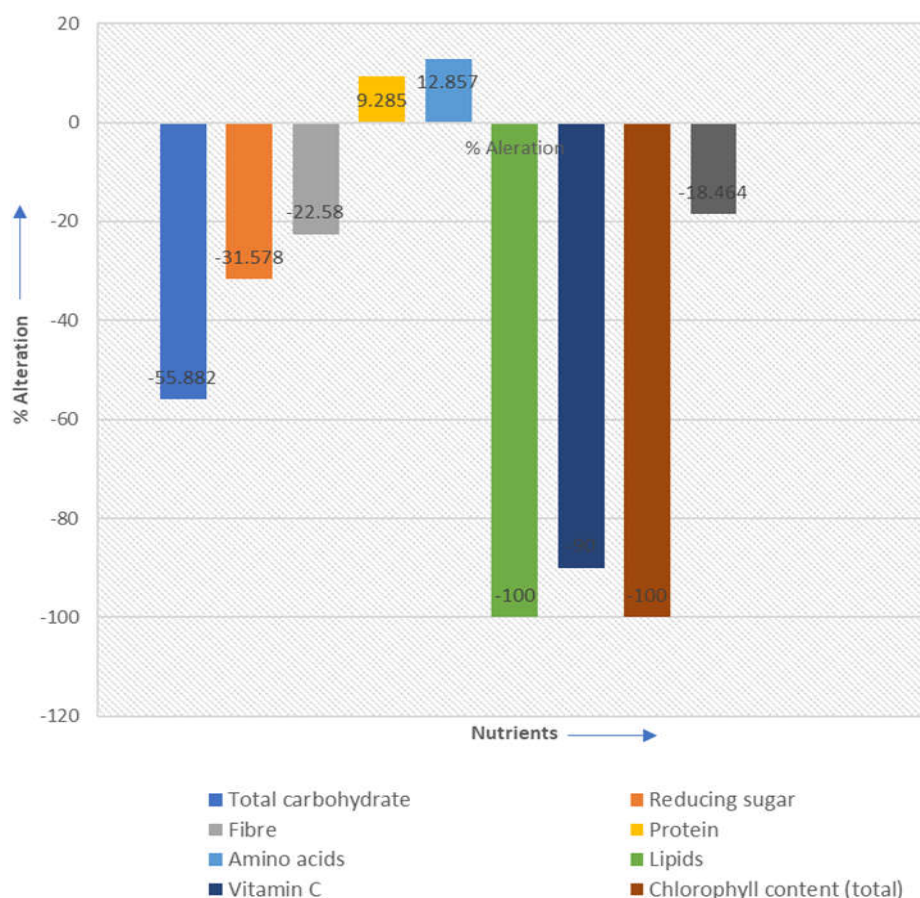
4. RESULT

From the observations, it is proven that, the amount of water content, Protein contents and free amino acids contents get increased by 8.295%, 9.285% and 12.857% respectively. While the contents like Total carbohydrate, reducing sugar, Fibres, Lipids, Vitamin C, Chlorophyll and Dry matter get reduced and their percentile loss is 55.8825%, 31.578%, 22.580%, 100%, 90%, 100% and 18.464% respectively.

5. CONCLUSION

Alternaria porri grow on plant leaves and absorb food from them, the main components which is required for growth are Carbohydrate, and water. During the growth the cell get diseased and finally dead and ultimately the Chlorophyll content started decline and at fully developed diseased state it totally vanished. The degrading activity is responsible for dietary fibre reduction. Stored vitamin C is also noticed to be fully retarded. The proteinaceous nature of fungal cell is found to be responsible for elevation of its quantity. Free amino acids are also increases and it indicate breakdown of stored protein and enzyme activity.

Graph-1- Percentile loss or gain of nutrients by *Alternaria porri* a pathogen causing purple blotch disease on Garlic leaves



Graph 1- percentile loss or gain of nutrients after infected with *A. porri* causing purple blotch disease on garlic leaves.

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