Chapter - VI

SUMMARY
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Dolichos bean (*Dolichos lablab*, L.), is one of the significant vegetable yielding crop of India and other countries of the world but Pakistan, Brazil, China, Egypt, Mexico, Sudan, U.S.A. and U.S.S.R. account for nearly 65.0 of the total production and having ample potential to tide over the paucity of vegetable growing in the country due to a rich source for protein, minerals, vitamins and enzymes and enriches in the soil due to fixation of atmospheric nitrogen by root module bacteria as well as used in preventing soil erosion. Dolichos bean occupies a premier place in the national economy of our country and prosperity of millions of countries, rural and urban populations, who depends upon harvest of vegetable yielding cash crop. Inspite of the presence of improved varieties and large acreage under its cultivation the total production in India is very low in comparison of other countries of the world, mainly due to off-setting of the foliar diseases of fungal origin as well as bacterial, viral and nematodal diseases. Among these, the leaf spot of Dolichos bean caused by *Alternaria alternata* (Fries.), Keissler, gradually increasing on new evolved high yielding varieties of Dolichos bean, has been recorded to be predominated in Uttar Pradesh during recent years, about which, there is no information available, requiring the immediate attention of Plant Pathologists, if the production of this crop is to be boosted in the country. So far, with a view to combat this serious malady effectively, the present investigation was taken to find out the prevalence and severity of disease in U.P., to investigate the morphological and cultural characters of the fungus, to ascertain the role of enzymes in pathogenesis in *vivo* and *vitro*, effect of pathogen on biochemical constituents of diseased parts of the host, growth and sporulation on different carbon sources and nitrogen inculture, effect of different doses of nitrogen, phosphorus and potash on the severity of disease, susceptible growth period of host, influence of climatic conditions on the development of disease, host range relationship of the pathogen, disease perpetuation as well as source of resistance, with an aim
of its control managing the strategy of disease, and the results sought are discussed here in brief.

Leaf spot of Dolichos bean caused by *Alternaria alternata* (Fries.) Keissler, was found to be moderate to heavy in severity widely prevalent under different agroclimatic conditions of Uttar Pradesh as evident from the survey conducted during the years, 2001 and 2002 in thirty localities during Kharif season. The incidence of disease at different research stations during Kharif seasons of the year 2001 and 2002 varied from 13.48 per cent to 31.57 per cent from the germplasms/cultures viz., Culture-7301, Kamgranj Selection-2, Culture-9012, Culture-7015, Culture-9118, Culture-7708, HD-4, Arka Jai, DB-1, Culture-8403, Todi-125-136, Culture-7604, Pusa Early Prolific, Goyal, JDL-79, Rajani, Culture-6801, Culture-8101, Culture-7710, Hatikan, HD-66, Culture-7210, Culture-9109, Culture-9113, Culture-8005, TDL-85, HD-93 and Kalyanpur Type - 1 and 10.25 per cent to 34.85 per cent from the germplasms/cultures viz; Culture-7301, Culture-7210, Akra Jai, Culture-9118, Culture-6801, Hatikan, Culture-8005, Culture-7015, Todi-125-126, Goya, JDL-85, Pusa Early Prolific, Culture-9102, Kamgranj Selection - 2, HD-4, Culture-9109, Culture-8101, HD-66, DPL-1, Culture-8403, DB-1, HD-93, Rajani, JDL-79, Culture-7604, Culture-7710, Culture-9113, Culture-9104 and Kalyanpur Type - 1 respectively. The maximum disease incidence 31.57 per cent and 34.85 per cent was recorded at Vegetable Research Farm, Chandra Shekhar Azad University of Agriculture and Technology, Kanpur in both the years from the germplasm/culture, "Kalyanpur Type - 1", followed by 30.80 per cent and 29.26 per cent at Government Agriculture Centre, Jalaun and Crop Research Farm, Modipuram Area, Meerut from the germplasms/cultures HD-93 and Culture-9113 and rest of locations, while minimum 13.48 per cent and 10.25 per cent disease incidence, was recorded at Agriculture Science Centre, Ganiwa, Chitrakoot in both the years.

The average disease incidence ranged from 13.48 per cent to 31.57 per cent and 10.25 to 34.85 per cent in both the years of survey at different crop Research Stations.
Disease intensity during Kharif season varied from 22.40 per cent to 38.40 per cent from the germplasms / cultures viz; Todi-125-126, JDL-79, Culture-9113, Culture-7210, Akra Jai, Culture-6801, Hatikan, Culture-9102, Culture-7301, Culture-7708, Goya, Culture-8005, Culture-9118, HD-66, Culture-7710, Culture-8403, DB-1, HD-4, Pusa Early Prolific, Kamgranj Selection-2, HD-93, Culture-7015, Culture-8101, Rajani and Kalyanpur Type-1 and 17.90 - 40.50 per cent from the germplasms / cultures viz; Culture-7710, Culture-9118, Culture-9102, Hatikan, Culture-9104, Akra Jai, Culture-7301, Culture-7604, Culture-9109, DPL-1, Culture-8005, Kamgranj Selection-2, Goya, Culture-6801, HD-93, Pusa Early Prolific, JDL-79, Rajani, Culture-8403, Culture-7015, DB-1, HD-4, JDL-85, HD-66, Culture-9113, Todi-135-136, Culture-7210, Culture-8101 and Kalyanpur Type-1 in the years 2001 and 2002 showing wide spread prevalence in nature. Maximum 38.40 per cent and 40.50 per cent disease intensity was recorded from the germplasm / culture, "Kalyanpur Type-1" at Chandra Shekhar Azad University of Agriculture and Technology, Kanpur followed by 37.80 per cent and 39.50 per cent from Oil Seed Research Farm, Kalyanpur, Kanpur and Crop Research Farm Saraimira, Farrukhabad from the germplasms/cultures 7604 and 8101 and rest of locations in both the years, 2001 and 2002, while minimum 22.40 per cent and 17.90 per cent disease intensity was recorded from Directorate of Vegetable Research Farm, Varanasi and Government Agriculture Centre, Atarra, Banda from the germplasm / cultures Todi-125-136 and Culture-7710 in both the years respectively.

In general the maximum disease intensity was recorded from Chandra Shekhar Azad University of Agriculture and Technology Kanpur.

The disease incidence as well as disease intensity could not be recorded in the years 2001 and 2002 at Crop Research Farm, Deegh, Kanpur and Regional Agriculture Research Station Dilip Nagar, Kanpur and Regional Centre, Amroha, Jhansi respectively.

The investigations on the symptomology of the disease, revealed that
under natural conditions, the disease appeared in the month of December and January in Kharif season crop and symptoms were confined on the upper surface of leaves only. The spots observed as small; brown to black; circular to oval with paler margins and yellow halo measuring 0.15-1.0 cm. in size with characteristic concentric rings and cracked centre. The lesions at first were recorded as smaller in size, while in later stage the spots were found numerous extending over the whole leaf surface due to coalescence of adjacent spots, which later became perforated due to falling away of dead tissue.

According to Koch's postulates the pathogen, was isolated on two per cent Potato dextrose agar medium by transferring surface sterilized portions of diseased leaf and subsequently, it was purified by Single Spore Culture technique. In order to test pathogenicity reisolations of the fungus, were done and the results indicated that all the isolates of fungus proved pathogenic on Dolichos bean plants. The fungus was able to cause leaf spot disease even inoculation was done without injury, thereby indicating that it was pathogenic. The infection percentage 87.50 per cent was recorded on injured leaves in comparison to un-injured leaves, which showed 35.0 infection percentage.

Different methods of inoculation proved that the disease was always more in case of pin pick inoculation than without injury indicating the fact that injured leaves provided avenues for the pathogenic attack.

The morphological characters of the pathogen, were studied on Potato dextrose agar medium, revealed that colonies were moderately fast growing, which in the beginning dull; white; fluffy; circular and later turned into dark; greenish olive with abundant sporulation. Mycelium was found as septate; branched; hyaline; later turning into black and olive buff in colour measuring 3.20-8.60 μ in width. Conidiophores arise singly or in groups usually simple; septate; straight or bent; sometimes branched; swollen terminally; geniculate and dark olive buff measuring 24.50-69.30 × 3.20 - 6.40
μ. Conidia, were formed in chains of 3-21; muriform; ovoid to obclavate; obpyriform; catenate (3-7); dark olive buff in colour; smooth sometimes verrucose with age; with 1-6 transverse septa and 0-6 longitudinal septa measuring 16.50-40.90 × 8.20-12.50 μ. Beaks were found usually short; light olive buff in colour and conical or cylindrical measuring 4.60-6.90 × 3.40-5.80 μ in size with 0-2 transversa septa. Chlamydospores were recorded sometimes terminal and intercalary and dark olive buff in colour, measuring 12.70 - 22.80 μ in diameter. On the basis of morphological characters the fungus, under study has been identified as Aternaria alternata (Fries.), Keissler causing leaf spot of Dolichos bean.

The effect of different media viz; Non-synthetic semisynthetic and synthetic on the growth and sporulation of the fungus on, was studied and recorded the best growth on Potato Dextrose agar medium measuring 92.40 mm. followed by Richard’s agar, Czapek’s agar, Coon’s agar, Leaf decoction agar, Malt salt agar, Sabouraud’s agar, Corn meal agar and Asthana and Hawker’s agar media. The least minimum growth measuring 17.46 mm. was obtained on Brown’s agar medium.

Apart from studying the radial growth and sporulation of the pathogen various other cultural characters viz; growth; shape; zonation and colour of colony; substratum colour; pigmentation; colour of hyphae; colour of conidiophores; number and septation of conidia; variation in shape; size and colour existence and size of chlamydospores are quite different on the different types of solid media belonging to Natural (non-synthetic), Semi-synthetic and Synthetic media.

The results revealed that colony growth was found good; compact and raised; good compact with downy appearence; good sparse, thick and cottony; good sparse with entire margin; average sparse with entire margin; average sparse with suppressed hairy margin; average compact; good and semi-suppressed, compact; thin and cottony; average sparse with entire margin and poor sparse with entire margin on Potato dextrose agar, Czapek’s
agar, Richard's agar; Leaf decoction agar; Coon's agar; Corn-meal agar; Malt salt agar; Sabouraud's agar; Asthana and Hawker's agar and Brown's agar media respectively. The colony shape was recorded almost circular on all the media except Czapek's agar medium, which exhibited lobe shaped colony. The colour of colony was almost recorded as dark grey; green with greenish tinge at the marginal ends; smoky grey; olivaceous black; dark black; dark greenish with darker centre; dark greenish with whitish margin; light green and creamy white on Asthana and Hawker's agar; Brown's agar; Sabouraud's agar; Malt salt agar, Richard's agar; Potato dextrose agar; Leaf decoction agar and Corn meal agar and Czapek's agar and Coon's agar media, respectively.

Substratum colour, was also found as irony grey; olive grey; light green, blackish green grey; olivaceous black; light vinaceous cinnamon; dark quaker drab and white smoky grey on Potato dextrose agar; Brown's agar; Asthana and Hawker's agar; Corn-meal agar; Czapek's agar; Leaf decoction agar, Malt extract agar! Richard's agar; Sabouraud's agar and Coon's agar media respectively. Zonation was found as distinct on Potato dextrose agar and Richard's agar media; not clear on Asthana and Hawker's agar medium; clear from upper side on Coon's agar and Czapek's agar media; clear from underside on Corn meal, Leaf decoction and Malt salt agar media, less clear from bottom side and absent from upper side on Brown's agar medium and dark quaker drab on Sabouraud's agar medium. Pigmentation was found absent on different types of media.

Colour of hyphae, was recorded as olive buff; mid olive; olive buff to greyish; pale olive buff; pale olive grey; light olive grey and colourless to greyish on Czapek's agar, Richard's agar and Sabouraud's agar media; Brown's agar; Malt salt agar; Asthana and Hawker's agar; Coon's agar; Corn meal agar; Leaf decoction agar and Potato dextrose agar media respectively. Hyphae were found septate varying in size from 2.50-8.0 μ in width on different types of media under study.
Colour of conidiophores, was recorded as olive buff to brown; midolive to brown; olivaceous brown; dark olive buff; pale to olive brown; dark olive brown and olive grey on Brown’s agar and Richard’s agar media, Czapek’s agar and Sabouraud’s agar media; Coon’s agar and Malt salt agar media; Asthana and Hawker’s agar medium, Potato dextrose agar medium, leaf decoction agar and Corn meal agar media respectively. The conidiophores varied in size from 24.20- 68.0 × 3.10- 7.80 μ on different types of media under study.

Conidia, were also found as olivaceous to dark brown; dark olive brown; olive buff to brown; light brown; dark olive grey and deep olive brown on Czapek’s agar; Malt salt agar; Richard’s agar and Sabouraud’s agar media; Potato dextrose agar; Asthana and Hawker’s agar; Brown’s agar; Coon’s agar Corn meal agar and Leaf decoction agar media. Conidia were found borne in chains of 2-4 on Asthana and Hawker’s agar, Brown’s agar and Coon’s agar media; 2-5 on Corn meal agar, Czapek’s agar and Malt salt agar media; 3-7 on Potato dextrose agar medium and 2-7 on Richard’s agar medium. Septation in conidia was also observed variable on different types of media. Transverse septa varied from 1-5 to 2-8 viz; 1-4; 1-5; 2-7 and 2-8 on Corn Meal agar medium; Asthana and Hawker’s and Brown’s agar media; Coon’s agar; Czapek’s agar; Leaf decoction agar; Malt salt agar and Sabouraud’s agar media and Potato dextrose and Richard’s agar media respectively, while longitudinal septa varied from 0-3 to 0-6 viz.; 0-3 on Asthana and Hawker’s agar; Brown’s agar, Coon’s agar and Corn meal agar media; 0-4 on Czapek’s agar; Leaf decoction; Malt salt and Sabouraud’s agar media; 0-5 on Richard’s agar medium and 0-6 on Potato dextrose agar medium. Conidia, were also found varied in size from 7.0-35.0 × 3.0- 13.80 μ in size on different types of media.

The beaks were found septate, and varied in size from 10.0-78.40 × 1.20- 6.40 μ. The transverse septa varied from 0-1 on Asthana and Hawker’s agar, Czapek’s agar, Leaf decoction agar, Malt extract agar and Sabouraud’s
agar media and 0-2 on Brown's agar, Coon's agar, Corn meal agar, Potato dextrose agar and Richard's agar media.

The beaks were also recorded as olive green; olive buff to brown; light brown; olive grey; dark olive brown; olive to dark brown and light olive buff on Asthana and Hawker's agar medium; Brown's agar medium; Coon's agar and Richard's agar media; Corn meal agar medium; Czapek's and Sabouraud's agar media; Leaf decoction agar medium; Malt salt agar medium and Potato dextrose agar medium respectively. The beaks were reported as cylindrical on corn meal, Czapek's, Leaf decoction; Richard's and Sabouraud's agar media and conical on Asthana and Hawker's; Brown's Agar; Coon's, Malt salt agar and Potato dextrose agar media.

Chlamydospores were found terminal as well as intercalary varying in size from 4.70-22.80 μ in diameter on all the different types of media but varying in size as dark brown; dark olive green; dark olive brown; olivaceous to dark brown; olive buff to brown; olive buff; light brown and olive grey on Czapek's and Sabouraud's agar medium, Asthana and Hawker's agar, Leaf decoction agar, Malt salt agar; Brown's agar, Potato dextrose agar; Coon's agar and Richard's agar media and Corn meal agar medium respectively.

The pathogen was also grown on different non-synthethic, semi-synthetic and synthetic media to select the ideal medium form, carrying out for further physiological activities of the pathogen. Out of ten liquid media studied Potato dextrose medium was found to boost the fungal growth weighing 468.30 mg. mycelial growth as well as excellent sporulation followed by Richard's medium. Brown's medium yielded poorest growth and poor sporulation.

In solid states of the media findings showed a trend almost similar to that found with their liquid forms, of course there are some variations in the order of their superiority. In present investigation Potato dextrose agar medium took first place instead of Richard's medium in terms of mycelial growth. Almost close correlation, was observed between growth and
sporulation in dry weight and linear growth of pathogen. Potato dextrose agar medium, was selected as a basal medium for physiological and enzymatic studies due to uniform best growth and sporulation. Some variations in cultural and morphological characters, were recorded on different culture media but significant differences on morphological characters were not observed.

The test organism could grow on a wide range of temperatures of 5°C to 50°C viz; 5°C (T-1), 10°C (T-2), 15°C (T-3), 20°C (T-4), 25°C (T-5), 30°C (T-6), 35°C (T-7), 40°C (T-8), 45°C (T-9) and 50°C (T-10). The optimum range being 30°C (T-6) and 35°C (T-7). The best growth and sporulation was recorded at 30°C (T-6), followed by 35°C (T-7). Sporulation, was also recorded as excellent at 25°C (T-5), 30°C (T-6), and 35°C (T-7), good at 20°C (T-4) and 40°C (T-8), fair at 15°C (T-3) and poor at 5°C (T-1) and 45°C (T-9). The pathogen failed to sporulate at 5°C (T-1) and 50°C (T-10).

The pathogen was able to grow on pH ranged from 2.50 to 12.0 viz; P-1 (2.50), P-2 (3.0), P-3 (3.50), P-4 (4.0), P-5 (4.50), P-6 (5.0), P-7 (5.50), P-8 (6.0), P-9 (6.50), P-10 (7.0), P-11 (7.50), P-12 (8.0), P-13 (8.50), P-14 (9.0), P-15 (9.5), P-16 (10.0), P-17 (10.50), P-18 (11.0), P-19 (11.50) and P-20 (12), but sporulated between the range of pH 3.50 (P-3) to pH 10.50 (P-17).

The maximum growth weighing 495.0 mg was observed at pH 6.50 (P-9), which was referred to as optimum pH. Excellent sporulation, was observed as P-9 (pH 6.50); followed by P-10 (pH 7.0); good at P-11 (pH 7.50) and P-12 (pH 8.0); fair at P-5 (pH 4.50), P-6 (pH 5.0), P-7 (pH 5.50) P-8 (pH 6.0), P-13 (pH 8.50), P-14 (pH 9.0) and P-15 (pH 9.50) and poor at P-3 (pH 3.50), P-4 (pH 4.0), P-16 (pH 10.0) and P-17 (pH 10.50). The pathogen failed to sporulate on P-1 (pH 2.50), P-2 (pH 3.0), P-18 (pH 11.0), P-19 (pH 11.50) and P-20 (pH 12.0). The pH, altered the pH of medium towards neutrality side. It was also observed that reaction of medium tended towards alkaline in cases, where the pH was on acidic side and vice-versa in the cases, where the media, was adjusted at P-14 (pH 9.0) to P-16 (pH 10.0) initially.
The studies on production of enzymes by the fungus in vitro, revealed that it produced Cellulase (CX), Polygalacturonase (PG) and Polymethylgalacturonase (PMG) enzymes. It was found that the activity of the enzymes was comparatively more in the medium supplemented with Carboxymethylcellulose (CMC), Sodium polypectate and Citrus pectin respectively; which play an important role in pathogenesis. In vivo studies, it was found that activity of Cellulase (CX), Polygalacturonase (PG) and Polymethylgalacturonase (PMG) enzymes took place in the diseased leaves inoculated with pathogen, Alternaria alternata (Fries.), Keissler and no enzymatic activity was produced in healthy leaves.

As regards the susceptible age of the host, it was observed that the pathogen may cause the disease at any stage of plant growth but the maximum susceptibility was observed in the plants, which attained the age of 50 days, followed by 60, 40 and 70 days old plants. The minimum disease intensity, was recorded from 10 days old plants. It was also concluded that plants were susceptible to disease at the age of 40-60 and 50-70 days particularly at 50 days. The susceptibility of plants towards disease decreased with the increasing age of plants and found almost as tracés at the age of 90 days and onward. In general the susceptibility of plants to the disease gradually was found decreased below or above 50 days old plants.

The environmental factors like atmospheric temperature, relative humidity and rainfall, were proved to have profound influences on the disease incidence. The disease exhibited firstly its appearence in second week of August in both the years 2001 and 2002 and increased gradually. The maximum disease development 38.84 per cent and 35.67 per cent was recorded in the second week of August during both the years 2001 and 2002 respectively, when the average temperature was 29.05°C and 28.70°C; relative humidity 86.70 per cent and 84.25 per cent and rain fall 0.29 mm. and 11.48 mm. respectively. A trend of decline of disease severity was also recorded with lowering down the temperature and relative humidity during
the month of May and finally by the third week of October, when both the atmospheric temperature and relative humidity, were unfavourable. In general the disease intensity decreased with the increase in temperature, whereas increased with the increase in relative humidity in both the years 2001 and 2002. The effect of rain fall, however was relatively more important in epidemiology of the disease as compared to distribution of rainfall 5-6 days a week accompanied by a cloudy weather.

In order to find out the best carbon source for the growth and sporulation of the pathogen, thirteen different carbon compounds were tested. Sucrose supported the best growth of the pathogen followed by galactose, maltose, raffinose, dextrose, mannose, fructose and xylose, which supported good growth. Fair growth, was observed on sorbitol, lactose and mannitol, whereas poor on dextrin and rhamnose. All the sources of carbon were significantly superior to control, which exhibited minimum growth of pathogen. Investigation on sporulation of the pathogen on different carbon sources, revealed that sucrose, maltose and raffinose supported excellent sporulation. Good sporulation was recorded on galactose dextrose, mannose, fructose, xylose and mannitol; fair on sorbitol, lactose and dextrin, whereas poor sporulation recorded on rhamnose. No sporulation was recorded in case of control.

With a view to find out the best nitrogen source for the growth and sporulation of the pathogen, A. alternata, twelve organic and inorganic nitrogenous compounds, were tested in the present study. Of these, peptone supported the best growth of fungus, whereas good growth and obtained on ammonium chloride, ammonium nitrate, calcium nitrate, potassium nitrate and sodium nitrate; fair growth on ammonium acetate, ammonium oxalate and ammonium sulphate and poor on ammonium carbonate, thiourea and urea. Excellent sporulation was recorded on peptone and sodium nitrate; good on ammonium nitrate, calcium nitrate and potassium nitrate; fair on ammonium chloride and ammonium oxalate and poor on ammonium
carbonate, ammonium sulphate and urea, whereas no sporulation was observed in case of the urea and control.

In respect of the effect of nitrogen, phosphorus and potash on disease severity, it was recorded that 60 Kg., P₂O₅ + 40 Kg. K₂O, were more effective in reducing the disease intensity as compared to 60 Kg. N + 60 Kg. P₂O₅ + 40 Kg. K₂O per hectare, whereas when 120 Kg. N per hectare was given alone. No work appears to have been done on the host nutrition in respect of the incidence of leaf spot of Dolichos bean caused by A. alternata.

Changes in biochemical constituents viz; Wax, Chlorophyll, Polyphenol, Reducing and Non-reducing sugars, Nitrogen, Phosphorus, Potassium and Sulphur, were also studied in the healthy and diseased leaves of Dolichos bean at different stages i.e. 40 days and 70 days by inoculating with Alternaria alternata. Considerable changes were observed in comparison to healthy leaves. It was also observed that contents of wax, chlorophyll a and b, polyphenols, reducing and non-reducing sugars and nitrogen, were comparatively decreased in both the categories of inoculated leaves in descending order by utilizing them for its own nutritional requirement or by destroying them through reaction. The remarkable changes regarding reduction in amount of wax, chlorophyll a and b, polyphenols and reducing sugars in the necrotic tissues of the leaves after 70 days of inoculation were observed but no changes were recorded in contents of phosphorus, potash and sulphur.

The investigation on mode of survival of the fungus revealed that the pathogen remained viable in soil, seeds and plant debris from November to June till the next sowing season. In tested seeds soil and diseased plant debris, were observed as virulent to serve as a source of primary inoculum for the pathogen. Secondary spread of disease was observed to be caused by conidia produced on the diseased spots of infected leaves and transmitted through air.

For screening the source of resistance against leaf spot of Dolichos bean
93 germplasms / cultures, were examined under natural and artificial conditions in order to examine their reactions to pathogen. In natural conditions, six germplasms / cultures viz; Arka Vijaya, Cultures 7703, 7008-B and 9101, JDL-37 and Todi-125-136 were found Tolerant being Disease Free (F); Five germplasms/cultures viz; Alatapati, Culture-7022, JDL-17, Pusa Early Prolific and Rajani were recorded as Resistant (R) having the disease intensity varied from 1.45 per cent to 2.75 per cent; twelve germplasms/cultures viz; Arka Jai, Cultures-6802, 7001, 7103, 7301, 8001, 8002, 8403, HA-3, HD-4, HD-81 and JDL-85, were recorded as Moderately Resistant (MR) having the disease intensity varied from 6.25 per cent to 9.13 per cent; thirty germplasms/cultures viz; Culture-5508, 6001, 6201, 6317, 6804, 7008-A, 7010, 7024, 7027, 7210, 7601, 7701, 7705, 7707, 8003, 8004, 8005, 8405, 9102, 9108, 9109, 9110, 9113, 9117, 9118, DPL-1, Goya, HD-10, HD-66, HD-104 and Todi-21, were found Moderately Susceptible (MS) having the disease intensity varied from 11.72 per cent to 18.29 per cent; twenty six germplasms/ cultures viz; Cultures-6022, 6023, 6701, 6801, 7006, 7007, 7012, 7015, 7016, 7019, 7020, 7101, 7205, 7206, 7603, 7702, 7710, 7711, 8101, 8401, 9104, 9105, 9114, 9116, DB-1 and Hatikan, were found Susceptible (S), having the disease intensity varied from 20.54 per cent to 29.27 per cent and thirteen germplasms/cultures viz; Cultures-6009, 6014, 6019, 7005, 7015-B, 7020-A, 7023, 7501, 7604, HD-93, JDL-79, Kalyanpur Type-1 and Kamgranj Selection-2, were found Highly Susceptible (HS), having the disease intensity varied from 51.49-62.38 per cent.

Further in artificial epiphytotic conditions of seeds and plant inoculation for their reaction to pathogen during Kharif season in the year 2001, the seeds of 23 germplasms/cultures, which were found Disease Free (F), Resistant (R) and Moderately Resistant (MR), were further examined. In seed inoculation test none of the Dolichos bean germplasms/cultures, was found to be immune and resistant. Out of 23 germplasms/cultures tested in seed inoculation test, ten germplasms/cultures viz., Arka Vijai, Cultures 6802, 7022, 7301, 8002 and 9101, HA-3, HD-81, Rajani and Todi-125-136 were
found Moderately Resistant (MR), having the disease intensity varied from 5.80 to 9.45 per cent; seven germplasms/cultures viz., Altapati, Arka Vijaya cultures 6802, 7708-B, DDL-37, HD-4 and JDL-17 were found Moderately Susceptible (MS) having the disease intensity varied from 10.36 to 18.59 per cent and remaining six germplasms/cultures (Cultures 7103, 8001, 8403, 7703, JDL-85 and DL-37 were found Susceptible (S) having the disease intensity varied from 23.82 per cent to 28.80 per cent, whereas in inoculation of potted plants, nine germplasms/cultures Arka Jai, Culture-7001, 7022, 9101, DDL-37, HD-4, JDL-85, Rajani and Todi-135-136 were found Moderately Resistant (MR), having the disease intensity varied from 6.28 per cent to 9.16 per cent; nine germplasms/cultures Altapati, Arka Vijaya, Cultures, 6802, 7301, 8001, 8002, HA-3, JDL-17 and JDL-37 were found Moderately Susceptible (MS), having the disease intensity varied from 11.92 per cent to 18.29 per cent and remaining five germplasms/cultures Cultures 7103, 7793, 7708-B, 8403 and HD-81, were found Susceptible (S) having the disease intensity varied from 21.54 per cent to 29.67 per cent.

Host range study revealed that the pathogen was able to infect the wide range of 70 host plants, both cultivated and wild belonging to 19 different families viz; Apocynaceae, Aracaceae, Chenopodiaceae, Asteraceae, Brassicaceae, Cucurbitaceae, Euphorbiaceae, Poaceae, Fabaceae, Labiatae (Lamiaceae), Liliaceae, Linaceae, Malvaceae, Myrtaceae, Papaveraceae, Pedaliaceae, Rosaceae, Solanaceae, and Apiaceae (Umbelliferae). Out of these 70 plants Abelmoschus esesulentus, Abutilon indicum, Althaea rosea, Allium cepa, Argemone mexicana, Avena savita, Arachis hypogea, Brassica campestris, B. Campestris var. dichotoma, B. juncea, B. oleracea var. botrytis, B. oleracea var. capitata, B. oleracea var. gongylodes, Carissa carandus, Carthamus tinctorius, Cajanus cajan, Capsicum annum, Chenopodium album, Chrysanthemum indicum, Colocasia antiquorum, Coriandrum sativum, Crotalaria juncea, Cucurbita maxima, Cynodon dactylon, Dahlia sp, Datura alba, Gossypium sp., Glycine max, Hibiscus rosa-sinensis, Hordeum vulgare, Linum usitatissimum, Lagenaria vulgaris, Lycopersicum esculentum,
Luffa cylindrica, Ocimum sanctum, Pisum sativum, Pennisetum typhoides, Raphanus sativus, Ricinus communis, Saccharum officinarum, Sesamum indicum, Sida acuta, Sorghum vulgare, Solanum nigrum, S. melongena, S. xanthocarpum, S. tuberosum. Tagetes erecta and Triticum aestivum, belonging to fifteen different families were found infected under artificial conditions of inoculation with spore-cum-mycelial suspension of the pathogen. It was found that the pathogen could infect the monocotyledonous and dicotyledonous plants having wide host range.

To select the suitable fungitoxicant for the control of the disease under field conditions the efficacy of twenty six fungitoxicants, Benzene (Halogenated organic compound), Inorganic Copper compound, Dithiocarbamate, Heterocyclic nitrogenous compound, Organomercury, Quinone, Systemic inorganic Sulphur compound and an Antibiotic viz; Agrosan G.N.; Bavistin, Benlate, Blitox-50, Brassicol, Calixin, Captafol. Captan, Cersan (Dry), Dichlone, Dithane M-45, Dithane Z-78, Duter, Emisan-6, Ferbam, Foltaf. 80-W, Karathane, Kavach, Hexaferb, Pancotine, Ridomil, Spergon, Suflex, Thiram, Vitavax and Ziram as well as an antibiotic (Aureofungin), were tested in the laboratory on the effect of growth of fungus, Alternaria alternata revealed that twelve of them namely Agrosan G.N.; Aureofungin, Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf 80-W, Karathane, Kavach, Pancotine, Thiram and Vitavax proved to most effective as inhibited the growth completely. The rest fungitoxicants, were found partially effective. Dithane M-45 caused the highest inhibition of fungal growth followed by Dichlone. The other fungicides in descending of efficacy, were suflex, Calixin, Blitox-50, Brassicol, Ridomil, Benlate, Ferbam, Ziram, Spergon, Bavistin, Hexaferb and Dithane Z-78. Dry weight of fungal mycelium in different concentrations of 0.05 per cent, 0.10 per cent, 0.15 per cent, 0.20 per cent, 0.40 per cent and 0.60 per cent, were significantly better in performance in comparison to control. A dosage response in different concentrations viz; 0.05 per cent, 0.10 per cent, 0.15 per cent, 0.20 per cent, 0.40 per cent and 0.60 per cent of fungitoxicants with reference to the

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pathogenic effect viz., light yellow lesion (LY), Yellowish brown lesion (YB), Light yellow brown lesion (LB), Extensive brown lesion (EB) and Phytotoxic effect (PE) on the plants were also studied. Out of the 25 fungitoxicants and an antibiotic (Aureofungins) tested Agrosan G. N., Ceresan (Dry) and Emisan-6, belonging to Organomercury, Duter, Karathane and Thiram belonging to Dithiocarbamate, Aureofungin (Antibiotic), Captan and Foltap 80-W (Captanf) belonging to Heterocyclic nitrogenous compound and Panacoline and Vitavax belonging to Systemic group were most effective as they inhibited the growth of fungus and did not produce any pathogenic effects. Other fungicides viz.; Calixin (Systemic) and Suflex (Inorganic sulphur), were also found effective in checking the growth of fungus and superior to the remaining fungicides tested. These fungitoxicants were found to be most effective as they inhibited the growth of fungus, when tested the different concentrations of 0.05 per cent, 0.10 per cent, 0.15 per cent, 0.20 per cent, 0.40 per cent, and 0.60 per cent, in respect of dry weight of mycelium. The other fungicides viz; Calixin (Systemic) and Suflex (Inorganic sulphur), were found also effective in arresting the growth of fungus upto 85.70 per cent at 0.05 per cent, 88.60 per cent at 0.10 per cent, 88.98 per cent at 0.15 per cent, 70.48 per cent at 0.20 per cent, 91.20 per cent at 0.40 per cent and 95.69 at 0.60 per cent and 88.91 per cent at 0.05 per cent, 92.44 per cent at 0.10 per cent. 93.41 per cent in 0.15 per cent. 91.33 per cent in 0.20 per cent, 94.85 per cent in 0.40 per cent and 95.79 per cent in 0.60 per cent respectively. The significant variations in inhibition in hyphal dry weight with respect to treatment of different fungitoxicants as well as different concentrations, were recorded. Apart from this these fungitoxicants were also found effective as did not produce any pathogenic effects but Brassicol (Benzene), Calixin (Systemic) and Suflex (Inorganic sulphur), were also found effective in producing Light yellow lesions (LY) only in different concentrations of fungitoxicants and established better in comparison to the remaining fungitoxicants tested except Brassicol (Benzene) in 0.30 per cent, where no pathogenic effect were shown. The number of germ tubes varied
from 5.38, 6.31 and 2.29 in 0.002 per cent, 0.0004 per cent and 0.0006 per cent concentrations of fungitoxicants, Minimum 6, 5, and 2 germ tubes were produced in treatment with Dithane M-45 (Systemic) in 0.0002, 0.0004 and 0.0006 per cent concentrations respectively, while maximum 37, 30 and 29 germs tubes in the treatment of Calixin (Systemic) and in treatment with other fungicides 10, 28, 36, 32, 11, 12, 28, 16, 30, 20, 27 and 20, 18, 23, 27, 28, 9, 11, 20, 14, 25, 18, 28 and 17, 6, 28, 27, 27, 21, 3, 16, 11, 23, 18, 28 and 14 germ tubes were found in 0.0002, 0.0004 and 0.0006 per cent concentrations. They were also effective as fungal spores failed to germinate, when fungitoxicants, were applied in 0.0002 per cent, 0.0004 per cent and 0.0006 per cent concentrations but Dithane M-45 (Dithiocarbamate) and Vitavax were less effective as produced minimum spore germination and proved better in comparison to other fungitoxicants. As far as the length of germ tube is concerned, varied from 0.005 to 0.018; 0.007 to 0.016 per cent and 0.006 to 0.014 per cent in 0.002 per cent, 0.0004 per cent and 0.0006 per cent concentrations of different fungitoxicants respectively. The minimum germ tube length measuring 0.005 mm., 0.007 mm. and 0.006 mm., were recorded in treatment with Bavistin (Systemic) in 0.0002 per cent, 0.0004 per cent and 0.0006 per cent concentrations of fungitoxicants respectively, whereas maximum number of germ tubes measuring length 0.017 mm., 0.014 mm. and 0.013 mm. with the treatment of Calixin (Systemic) in 0.0002 per cent, 0.0004 per cent and 0.0006 per cent concentrations respectively. The inhibition percentage over control varied from 24.82 per cent to 68.72 per cent, 34.74 per cent to 74.28 per cent and 45.48 per cent to 78.19 per cent in 0.0002 per cent, 0.0004 per cent and 0.0006 per cent concentrations of fungitoxicants respectively. The inhibition in all the three parameters in spore germination, germ tube length and number of germtubes, produced by spores were pronounced in higher concentrations of fungitoxicants with significant differences. Dithane M-45 (Dithiocarbamate) caused appreciable degree of inhibition in respect of germination of spore, germ tube number and germ tube length. The germ tube length was inhibited up to 78.19 per
cent at higher concentrations. At lower concentration, Blitox-50 (Inorganic Copper) and Calixin (Systemic), were not proved effective but effective at higher concentration in inhibiting spore germination.

The efficacy of selected seed dressing fungitoxicants viz., Agrosan G. N., Aureofungin, (Antibiotic), Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf-80 W (Captcha) Karathane, Kavach, Pancotine, Thiram and Vitavax as seed dressers proved effective in laboratory, were evaluated on seed treatments for studying their effects on seed germination and seed borne infection in vitro and it was found that all of them improved germination. The highest germination was recorded in seeds treated with Captan (Heterocyclic nitrogenous compound) Ceresan (Dry organomercury), Thiram (Dithiocarbamate), Agrosan G. N. (Organomercury), Aureofungin (Antibiotic), Emisan-6 (Organo-mercury), Duter (Dithiocarbamate), Pancotine (Systemic), Karathane (Dithiocarbamate), Foltaf 80-W (Heterocyclic nitrogenous compound), Kavach and Vitavax (Systemic). The seed treatment with Ceresan (Dry) resulted in elimination of the fungus but the treatment with Thiram (Dithiocarbamate), Captan (Heterocyclic nitrogenous compound) and Agroson G.N. (Organomercury), expressed their broad spectrum in nature in order mentioned and these were par in efficacy. It was also recorded that lowest infection 1.29 per cent was found in treatment with Captan. The maximum 14.55 per cent infection was recorded in treatment with Vitavax followed by Pancotine (Systemic), Thiram (Dithiocarbamate), Emisan-6 (Organomercury), Duter (Dithiocarbamate), Aureofungin (Antibiotic), Agrosan G.N. (Organomercury), Foltaf 80W (Heterocyclic nitrogenous compound), Karathane (Dithiocarbamate), Kavach and Captan (Heterocyclic Nitrogenous compound). Ceresan (Dry, organomercury), was found to be best one as it completely eliminated the infection. In general all the fungicides examined, were found significant over control in reducing seed borne infection.

The same seed dressing fungitoxicants were tested in pot experiments also ascertaining their effects on seed germination and seedling infection. The highest seed germination 95.47 per cent was recorded in treatment with Ceresan (Dry, Organomercury), followed by 92.08 per cent with Captan.
(Heterocyclic nitrogenous compound), 91.57 per cent in Thiram (Dithiocarbamate compound) and 90.45 per cent in Agrosan G. N. (Organomercury) 84.78 per cent in Emisan-6 (Organomercury), 83.75 per cent in Duter (Dithiocarbamate), 83.98 per cent in Pancotine (Systemic) 83.79 per cent in Aureofungin (Antibiotic), 81.67 per cent in Karathane (Dithiocarbamate), 79.15 per cent in Kavach and 78.60 per cent in Vitavax, while comparatively poor germination was noticed in treatment with Pancotine (Systemic), Karathane (Dithiocarbamate), Vitavax (Systemic) and Foltaf-80 W (Heterocyclic nitrogenous compound). The complete control of seedling infection in treatment with Ceresan (Dry, Organomercury), Thiram (Dithiocarbamate) and Vitavax (Systemic) had no infection. Captan (Heterocyclic nitrogenous compound) and Agrosan G. N., were found superior in elimination of seedling infection. Thus seed dressing fungitoxicants belonging to Heterocyclic nitrogenous compound (Captan), Organomercurials (Agrosan G. N. and Ceresan) compound groups were most efficacious than fungitoxicants belonging to Dithiocarbamate, Copper, Sulphur and Quinone compounds of Systemic nature controlling seeds borne infection.

In studies regarding the control of disease in the field, twelve different fungitoxicants viz., Agrosan G. N., Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf-80 W, Karathane, Kavach, Pancotine, Thiram and Vitavax as well as an antibiotic (Aureofungin), which were found completely or partially effective in bioassay test, were evaluated under pot and field in the years 2001 and 2002 in order to select out a suitable fungitoxicant reducing the incidence of disease and boosting seed yield. The best control of disease was obtained by application of Captan (Heterocyclic nitrogenous compound) followed by Thiram (Dithiocarbamate), Ceresan (Dry), Agrosan GN (organomercury), Karathane (Dithiocarbamate), Kavach, Duter (Dithiocarbamate), Aureofungin (Antibiotic), Pancotine (Systemic), Emisan-6 (Organomercury), Vitavax (Systemic), and Foltaf 80-W (Heterocyclic nitrogenous compound), differed significantly in the year 2001 and 2002 in pot experiment while in Field experiment followed by Thiram
(Dithiocarbamate), Vitavax (Systemic), Ceresan (Dry organomercury), Agrosan G.N. (Organomercury), Aureofungin (Antibiotic), Duter (Dithiocarbamate), Emisan-6 (Organomercury), Pancotine (Systemic), Foltaf 80-W (Heterocyclic nitrogenous compound), Karathane (Dithiocarbamate) and Kavach differed significantly in the year 2001 and 2002 and were significantly superior over control. Foltaf 80-W and Kavach, fungitoxicants were found poorest amongst all the fungicides controlling the disease in both the years, 2001 and 2002 in pot and field experiments respectively. Further it was also recorded the plots treated with Captan exhibited significantly better yield 31.34 and 29.08 gms. per plant than control gms. weighing 19.62 gms. and 19.82 gms. and 19.25 gms. and 17.32 gms. than control weighing 15.87 and 14.75 gms. per plate, in both the years 2001 and 2002 in pot and field experiments respectively. The next in effectiveness was Thiram followed by Agrosan G.N.; Ceresan (Dry), Aureofungin (Antibiotic), Emisan-6, Pancotine, Duter, Karathane and Kavach in Pot experiments and Thiram, Ceresan (Dry), Agrosan, G.N., Pancotine, Duter, Emisan-6, Vitavax, Karathane, Kavach, Aureofungin (Antibiotic) and Foltaf 80-W in field experiments respectively in the years 2001 and 2002. Lowest yield weighting 19.94 gms. and 20.35 and 16.08 gms. in pot experiment and 13.42 gms. per plant in field experiments in treatment with Vitavax and Foltaf 80-W in both the years. Complete control of the disease was not achieved by any of the fungitoxicans tested but significantly in the yield establishing a good correlation between Leaf infection and yield, which were lowered according to the severity.