ANTIFUNGAL EFFICACY OF ZANTHOXYLUM OIL AGAINST *BIPOLARIS SOROKINIANA* (SACC.) SHOEM.

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ABSTRACT

Bipolaris sorokiniana (Sacc.) Shoem. is the most economically important foliar blight pathogen of wheat in all warm zones. Essential oil of *Zanthoxylum armatum* D.C. was used to determine its efficacy against *Bipolaris sorokiniana* by calculating the percentage of mycelial growth inhibition. The oil showed 25, 37.8, 42.8, 75, 84.5 and 100 percent inhibition of mycelial growth at 5.0, 10.0, 15.0, 20.0, 30.0, and 40.0 µl ml^{-1} ppm concentrations of essential oil against the test fungus, respectively.

Keywords: Essential oil, fungitoxicity.

Green plants have been found very successful in providing less toxic, more systemic (Fawcett and Spencer 1970), easily biodegradable (Beye 1998), host metabolism stimulatory fungicide (Dixit *et al.* 1983) as well as constituting a renewable source. Certain plants contain products such as alkaloids, tannins, quinines, coumarines, phenolic compounds, in their extracts and exudates, and they are known for their antifungal activities. The presence of antifungal compounds in plant tissues has long been recognized as an important factor for disease resistance (Fawcett and Spencer 1970.)

The average production of wheat in Nepal is very low than the potential yield. This is due to some major wheat diseases. Foliar blight complex (Spot blotch) is one of them. It is commonly known as *Helminthosporium* leaf blight. It is a major disease of wheat and is caused by *H. sativum* P.R. and B (syn. *B. sorokiniana* Sacc) and *Drechslera tritici repentis* Died. The disease is extended from Terai belt to 2600 masl. The incident and severity of blight are comparatively lower in the hills than in the Terai. Considerable loss in wheat grains has been found due to this disease which ranges from 3.1 to 29.0% in a normal year in natural epidemic condition depending upon wheat varities (Mahto 1999).

The main sources of innoculum of *B. sorokiniana* are infected seeds, infected crop residues, volunteer plants, secondary host and free dormant conidia in the soil (Reis 1991). Infected seeds are one of the most important ways for the survival of this pathogen between wheat cropping season. Seed infection by *B. sorokiniana* can adversely affect germination and root system development and can kill seedlings within a few days (Mehta 1993). In Nepal the average seed infection from Terai to hill region varies from 5.0 to 89.1% irrespective of cultivars (Shrestha *et al.* 1998).Recent studies in soil health revealed the presence of *B. sorokiniana* in the soil.

Bipolaris sorokiniana: Mycelium simple, hyaline, conidiophores brown, short, unbranched, aseptate and bear 1-6 conidia. Conidia thick-

walled, ellipsoid, dark, olive brown, straight to slightly curved and having 5-9 cells and measures 6-12 µm x 12-20 µm (Zillinsky 1983). Colonies on PDA are olivaceous brown to black. In old cultures the number of conidia is so abundant that all cultures turn black and shiny.

Essential oil of *Zanthoxylum armatum* was obtained from HPPCL (Herb Production and Processing Co. Ltd.) Koteshwar, Kathmandu. *Bipolaris sorokiniana* was used as test fungus species and its culture was obtained from NARC (National Agriculture Research Council). The fungus was isolated from wheat seeds from Rampur by blotter test method. The pure culture was maintained on Potato Dextrose Agar medium. Seven days old culture of the test fungus was used to prepare inoculum discs (4 mm diameter). A single disc was aseptically placed upside down in the center of each petriplate containing essential oil loaded PDA, so as to establish a direct contact with the medium.

Acetone (80%) was used as the solvent to prepare different concentrations of essential oil (5.0, 10.0, 15.0, 20.0, 30.0 and 40.0 µl ml^{-1} ppm) for use in experiment (Rao and Srivastava 1994) to assess fungitoxicity following poison food technique (Grover and Moore 1962). A volume of 0.5 ml of each concentration of essential oil was aseptically poured into the petriplate followed by the addition of melted PDA (9.5 ml). The petriplate was kept swirling while adding PDA so as to get a thorough mixing of the contents. Essential oil was, however, replaced by an equal amount of acetone in control set. After the media solidified one inoculum disc of *B. sorokiniana* mycelium, the test fungus was aseptically placed to each petriplate and incubated at 25+2° C. The average diameter of fungal colonies was measured on 7th day after inoculation and percentage of mycelial growth inhibition was calculated (Rao and Srivastava 1994). In the experiment, three replicates of the petridishes containing PDA were used and the colony size was calculated by taking an average of the three.

$$\text{Mycelial Growth Inhibition (\%)} = \frac{gc - gt}{gc} \times 100$$

Where, g_c = Growth of mycelial colony after incubation period in control set subtracting the diameter of inoculum disc, g_t = Growth of mycelial colony after incubation period in treatment set subtracting the diameter of inoculum disc.

Table 1. Assessment of antifungal activity of essential oil of *Zanthoxylum armatum* against *Bipolaris sorokiniana*.

SN	Concentration of oil (µl ml^{-1})	Mean colony size (mm)	Inhibition of mycelial growth (%)
1	0	64.0 ± 5	-
2	5.0	49.0 ± 4	25.0
3	10.0	41.3 ± 1.53	37.8
4	15.0	38.3 ± 1.53	42.8
5	20.0	19 ± 0.58	75.0
6	30.0	13.3 ± 3.21	84.5
7	40.0	4.0	100

The results showed the mycelial growth inhibition percentage over control. The mycelial growth inhibition percentage was 25.0, 31.26, 36.16, 75.0, 84.5 and 100 percent at essential oil concentrations of 5.0, 10.0, 15.0, 20.0, 30.0 and 40.0 µl ml^{-1} ppm, respectively.

During recent years, the essential oils of a number of higher plants have been tested for their *in vitro* antifungal activities, however, the detailed fungitoxic evaluation of such oils in order to prove their usefulness as successful fungitoxicants is limited (Tripathi *et al.* 1983). The toxicity in plants is believed to be due to the presence of certain fungitoxic factors in their tissue (Fawcett and Spencer 1970). The essential oil of *Zathoxylum armatum* possesses such chemical components

which on treatment against *Bipolaris sorokiniana* exhibited absolute toxicity by complete inhibition of the growth of the test fungus.

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