

Study on Black Spot of Sunflower

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Abstract This paper deals with the black spot of sunflower cause by *Alternaria helianthi* (Hansford) Tubaki et Nishihara. The study shows its characteristics of the pathogen.

The spot of different shape, size and color ranging from light brown to black brown on leaf, stem, head and flower of sunflower.

The varieties sunflower were identified their resistance to *A. helianthi* in the greenhouse and field. As yet, the sources of immunity and resistance have not been found among the cultivated sunflower.

A. helianthi overwinters in the form of mycelia and conidia on infected harvest residues, which are the major sources of infection.

Key words: sunflower, *A. helianthi*, biology, infection sources, resistance

The black spot of sunflower is caused by *Alternaria helianthi* Tubaki et Nishihara. Since Mr Qi Piekun first reported this disease in Jilin Province in 1966, it has spread in major sunflower producing areas in China. It has become a serious epidemical disease. In general year, its pathogenicity index is about 70 and reduces sunflower yield by 20%, In epidemical year, its pathogenicity index is over 95, reduces the yield by 50%. This disease is that we are facing a serious question. So we began to study this disease from 1989 to 1992.

1. MATERIALS AND METHODS

1.1 symptoms and forms of the pathogenic fungus

The disease samples were collected from growing the fields of sunflower. The samples were isolated on PDA for obtaining *A. helianthi*. The fungus of pathogenicity was tested on the greenhouse. Through single conidium was isolated, we obtained a strain used the test.

1.2 effect of different media on fungus growth

Five different media were used in this investigation. After inoculation, the petri dishes were put in a incubator at 25°C for 15 days. Then the diame-

ters of colony were examined with 3 replicates for each treatment.

1.3 effect of temprature on fungus growth

Identical portions of the pure culture were placed in PDA. After that, the petri dishes were put in a incubator at 5, 10, 15, 20, 25, 28, 30, 32°C and 35°C respectively. The development of the fungus was determined for 5, 10 and 15 days with 3 replicates.

1.4 effect of the relative humidity on germination of the conidia

Using Cro, KNO_3 , $\text{K}_2\text{HPO}_4 \cdot 3\text{H}_2\text{O}$ seven different salts of the saturated solution controled their humidities. The water's moisture was 100%. All treatments were put at 25°C for 24 hours with microscopic examination on germination rate of conidia.

1.5 effect of different pH on germination of the conidia

Using NaOH, HCl and the acidimeter, various pH value solutions were prepared from 1 to 9. The conidia were put in each solution at 25°C for 24 hours. Germinator rates of the conidia were examined with microscope.

1.6 use of carbon and nitrogen sources

Fourteen carbon sources were used for this test, each solution contained 2% a carbon respectively. Six nitrogen sources prepared solutions that each contained 1% a nitrogen respectively. There were a small amount of the conidia in each solution, at 25°C for 24 hours, with microscopic examination on germination rate of the conidia.

1.7 primary sources of infection

After harvest in the autumn, infected the plants parts (leaves, stems, and heads) were put in surface of the ground, cold or warm house, underground 15cm and the fence respectively. We examined each treatment of the germination rates of the conidia in next June and the disease tissues were isolated on PDA. The qualities of the colony of *A. helianthi* were examined. The seedings of sunflower were inoculated, keeping their moisture with a plastic bag and after two days, transferred to the greenhouse about 25°C. After 7 d ys the infected sunflower were examined. The seeds that were collected last autumn were isolated before sowing sunflower.

1.8 Identification of the resistant varieties

500 varieties were identficated in 1989-1992. All varieties were sown

in May and each was sowed a rang. They inoculated with water suspension of the conidia on 15 - 70 July. Infected rate was investigated on 15 August and each variety examined 10 plants, then pathogenicity index was calculated.

2. RESULTS AND DISCUSSION

2.1 symptoms and forms of the pathogenic fungus

A. helianthi infected leaf, stem, stalk, head and flower and at first the leaves under plant are infected. Then the spots spread quickly in all parts of the plant. The spots differ in shape and size. Generally, the spot is round or ellipse, light brown to black brown. There is a smaller white spot in the centre of each spot. In favourable climatic conditions, the number of the spots increases significantly, many spots merge and the infected leaves wilt quickly over ten days. The spots of the stalk, stem, flower are polygonal, black brown. The spot of the stem is the biggest and reaches the length 50~60cm. The spot of the head is round and ellipse.

The color of the conidium and conidiophore is light brown to black brown. The conidiophore is single or a sheaf of 2 - 4 ones, about 40 - 110 \times 7 - 10 μ m with 0 - 4 transversal septa. The conidium is cylinder or ellipse, straight or more or less bent with 4 - 12 transversal septa with 50 - 120 \times 15 - 20 μ m Its omphalic spot is concave clearly and lie to the end of the base cell. The conidium is single or two conidia are concatenate together.

2.2 effect of different media on the fungus growth

They were evident that the test different media effected the fungus development. The weakest mycelia growth was on Sabouraud, the strongest on the PDA and V-8 medium. Table1 presents the result obtained.

Table1. Effect of different media on the fungus growth

media	diameter of the colony (mm)	produced conidia
Czopek	30.2	+
Rixhards	19.6	+
Sabouraud	13.1	+
V-8	25.4	+ + + +
PDA	24.4	+ + + +

2.3 effect of temperature on the fungus growth

The fungus can grow between 10~32°C and the most favourable temperature was at 25°C and 28°C. The fastest the colony growth was after culture 5-10 days (Table 2).

Table 2 Effect of temperature on the fungus growth

temperature (°C)	diameter of the colony (mm)		
	5 days	10 days	15 days
5	0	0	0
10	0	5.9	7.9
15	4.4	9.1	11.5
20	9.9	20.4	24.2
25	13.7	22.2	25.2
28	17.2	24.0	26.4
30	15.6	25.3	26.9
32	5.1	5.6	5.8
35	0	0	0

2.4 effect of the relative humidity on germination of the conidia

The optimal relative humidity of germination of the conidia was between 95~100%. The conidia can germinate on RH 76-100% but the most favourable moisture was in drop water (Table 3).

Table 3 Effect of moisture and pH to germination of the conidia

Salts Solution	moisture		pH	
	RH (%)	germination rate of the conidia	pH	germination rate of the conidia
Cro	35	0	4	0
KNO ₃	45	13.4	5	99.1
NaNO ₃	66	14.1	6	99.3
Na ₂ ClO ₃	75	6.9	7	99.0
(NH ₄) ₂ SO ₄	80	9.3	8	95.2
ZnSO ₄ ·7H ₂ O	90	37.2	9	93.1
K ₂ HPO ₄ ·3H ₂ O	95	78.2		
water	100	78.9		
drop water		95.8		

2.5 effect of different pH value on germination of the conidia

The result showed that the conidia can germinate between pH 5~9.

These results also showed that *A. helianthi* can be suitable a wide range to pH value (Table 3).

2.6 use of carbon and nitrogen sources

Table 4 showed that seminose and maltose were the most favourable to germination of the conidia and the worst xylose was in fourteen carbon sources. The fungus can use glutamic acid and arginine (Table 4).

Table 4 Use of carbon and nitrogen sources

carbon source				nitrogen source	
carbon source	germination rate of the conidia (%)	carbon source	germination rate of the conidia (%)	nitrogen sources	germination rate of the conidia (%)
D-seminose	98.1	arabinose	93.4	NaNO ₃	0
maltose	97.8	L-rhamnose	88.0	(NH ₄) ₂ SO ₄	0
fructose	97.1	raffinose	87.3	NH ₄ NO ₃	0
sugar	97.0	synanthrin	86.5	glutamic acid	95.1
dextran	95.9	sorbose	76.8	arginine	97.0
mannite	95.8	sorbierite	75.1	aspartic acid	0
lactose	94.0	xylose	63.0		

2.7 primary sources of infection

The mycelia and the conidia in the residues can overwinter in the warm and the cold house, on the fence, the surface of the ground and in the underground. Before the sunflowers were infected next year, their conidia and mycelia had capable of survival and pathogenicity. The experiment showed that the residues and the seeds of sunflower were the primary sources of infection of black spot of sunflower. But the residues were more important than the infected seeds. The infection rate of the testas and acicis were 0—1.2%, 0.6—1.7% respectively.

The conidia that came from overwintering residues inoculated the seeds of sunflower and the infection rates were over 90%. The underground 15cm residues had rotted and lose their survival the next May (Table 5).

Table 5 Ability of the residues survival

treatments	germination rate of the conidia (%) (in June)	solated tissue	a mount of solation	number of the colony
warm house	73.0	head	48	14
cold house	92.0	stem	92	64
fence	95.6	leaf	113	57
surface ground	89.8			
underground 15cm	rotted			

2.8 identification of the resistant varieties

These results indicated that the resistance were different between 500 varieties. There were no immunity and high resistant varieties. The middle resistance varieties were Yanan of pure seeds 9; Dehui, Early Sunflow 3, Nong an 2 fourteen varieties. The middle susceptibility was 103 varieties and the high susceptibility was 383. The oil sunflower was more susceptible than the edible one.

3. CONCLUSION

Tubaki and Nishihara (1969) Studied its systematic position and forms. Our results showed that the fungus can be suitable to temperature, pH, carbon sources in a wide rang and there were no immunity, high resistant varieties. The temperature from June to August is very suitable for *A. helianthi* of intrusion and spread in Jilin. So the key is the relative humidity and rainfall whether the disease can be epidemical. On the other hand, the residues are the major sources of infection. The rotation and fall flowing can decrease primary sources of infection and harm to sunflower next year.