

## PATHOGENIC FUNGI INTERCEPTED IN INTRODUCED OIL-SEEDS DURING 1976-98, THEIR SIGNIFICANCE AND CONTROL

USHA DEV, P.C. AGARWAL, INDRA RANI, BALESHWAR SINGH AND DINESH CHAND, National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi 110 012

Introduced germplasm of oil-seed crops is one of the key factors in doubling the oil-seed production from 10.83 million tones in 1985-86 to 22.50 million tones in 1994-95. During last twenty- three years about 39,000 oil seed germplasm accessions comprising of *Brassica* spp., safflower, soybean, sunflower, castor etc. were introduced from various countries for crop improvement programmes. A large number of pathogenic fungi of quarantine significance were intercepted. The methods employed for salvaging and the significance of these interception is discussed.

**Key words :** Oilseeds, pathogenic fungi, quarantine

India shares about 20 per cent of the global area of oilseed production while in terms of production it contributes less than 10 per cent. To achieve self-reliance in oil production, New Policy on Seed Development (NPSD) came into force on October 1st, 1988 and a free access to worldwide germplasm of oilseeds was one of the key factors in doubling the oilseed production from 10.83 million tones in 1985-86 to 22.50 million tones in 1994-95. However, such liberalization in the exchange of Plant Genetic Resources (PGR) always carried the risk of introducing exotic pests and pathogens or their more virulent strains in the country.

During the last 23 years (1976-1998) National Bureau of Plant Genetic Resources has introduced 39,169 seed-samples of different soil-seeds viz. *Brassica* spp. (6541); *Glycine* spp. (15,542); *Helianthus* spp. (6490); *Carthamus* spp. (6240); *Linum* spp. (505); *Sesamum* spp. (3,021), *Ricinus* spp. (526); *Arachis hypogaea* (235); *Cocos nucifera* (1), *Elaeis guineensis* (66) and Olive spp. (2) from different countries.

Quarantine processing of introduced germplasm begins with the examination of seeds under a stereo-binocular microscope for the presence of mal formed, deformed and discoloured seeds, fungal mycelium, fructification, rust spores and oospore crust of downy mildew etc. Seed samples of sunflower and soybeans were subjected to washing test to detect rust and downy mildew, respectively. Unhealthy looking seeds were subjected to Blotter test and incubated at 20 ( $\pm 1$ )°C for seven days. The fungal growth and colonies developed were examined on the eighth day under the stereo-binocular microscope. Infected samples were salvaged by subjecting to appropriate treatments e.g. hot water treatment, ethyl alcohol wash or pesticide dressing.

Seed samples when examined under the stereobinocular microscope showed certain characteristic symptoms indicating the presence of pathogenic fungi. Purple stain on the surface of soybean seed indicated the possible presence of *Cercospora kikuchii* (Mats. & Tomoy) M.W. Gardner. The dull white crust on the surface of

soybean seed under stereoscopic examination and washing test revealed masses of hyaline to light brown, thick and smooth walled oospores of *Peronospora manshurica* (Naum.) Syd., Out of the total 15,542 samples of soybean, 1,716 samples (11%) showed the presence of *P. manshurica* the downy mildew fungus (Mukewar *et al.*, 1980; Agarwal and Singh, 1998). The interception of *P. manshurica* is of high quarantine significance as the pathogen is yet not reported from India. Presence of 33 physiologic races in USA (Li *et al.* 1992), 21 in Poland (Marcinkowska, 1987) and long periods of viability of oospores in the seed (pathak *et al.*, 1978) further add to the problem. Its importance further amplifies with the interception of this pathogen from Malaysia (Agarwal and Khetarpal, 1985) and Indonesia (Anitha *et al.*, 1993) from where this was hitherto unreported.

Safflower seeds showed the presence of small brown coloured teleutospores of *Puccinia carthami* Corda on the seed surface (Agarwal *et al.*, 1982). Of the total safflower samples analysed, 83 per cent (5168) showed the presence of rust. *P. carthami* is reported to have caused serious epiphytotics in USA in 1949-50 (Thomas, 1952). Thus presence of *P. carthami* in safflower samples of such diverse germplasm (collected by USDA from a number of countries) is of quarantine importance as the pathogen is known to have a number of races (Thomas, 1955). Likewise, washing test revealed the presence of the uredo- and teleutospores of *Puccinia helianthi* in sunflower seeds. Out of 6490 seed samples examined, rust spores were detected in 309 samples.

Examination of plates after incubation showed a large number of fungi on various crops (Ram Nath *et al.*, 1981 and 1985; Majumdar *et al.*, 1991). The details of the infected samples and the fungi intercepted during examination is given in Tables 1-5. Some of the major fungi detected in the blotter test are *Cercospora kikuchii* (purple

Table 1. Fungi intercepted on Introduced Brassica spp.

Country	No. of samples received (found infected)	Alternaria brassicae	Alternaria brassicicola	Fusarium solani	Phoma lingam
Belgium	25 (6)	-	6	-	-
Canada	439 (128)	47	57	1	37
China	44 (6)	-	2	4	-
Denmark	46 (12)	-	11	1	-
Ethiopia	36 (2)	2	2	-	-
Finland	9 (6)	-	-	6	-
France	92 (11)	6	11	9	-
Germany	279 (2)	-	2	-	-
Holland	76 (20)	-	19	1	-
Hungary	18 (2)	-	2	-	-
Italy	187 (27)	9	-	-	18
Japan	46 (10)	-	10	-	-
Korea	37 (8)	-	8	-	-
Sweden	234 (113)	69	103	6	1
Taiwan	775 (149)	14	138	12	1
U.K.	1269 (125)	13	111	5	6
USA	2287 (601)	3	589	5	4
USSR	151 (2)	-	1	-	1
Others	491 (0)	-	-	-	-
Total	6541 (1230)	163	1072	44	67

Apart from the fungi listed, *Alternaria raphani* (7) and *Alternaria solani* (1) from Canada and *Botrytis cinerea* (1) from Sweden were also detected.

blotch), *Colletotrichum* spp. (anthracnose), *Phomopsis sojae* Lehman (pod & stem blight), *Ascochyta sojicola* Abram. (leaf spot) on *Glycine* spp; *Phoma lingam* (Tde ex Fr.) Desm. (black leg), *Alternaria brassicae* (Berk.) Sacc. and *Alternaria brassicicola* (Schw.) Wilts (leaf spot) on *Brassica* spp., *Corynespora cassicola* (Berk. & Curt.) Wei the blight fungus on safflower, *Fusarium solani* (Mart.) Sacc. the wilt fungi was detected on safflower, *Brassica* spp, soybean, sunflower,

Table 2. Pathogenic fungi intercepted in introduced seeds of *Carthamus* spp.

Country	No. of samples received (found infected)	Puccinia carthami	Alternaria carthami
Australia	94(47)	37	10
Canada	19(3)	3	-
China	50(50)	50	-
Ethiopia	14(3)	3	-
Germany	53(4)	-	-
Greece	11(5)	5	-
Italy	59(17)	16	-
Singapore	1586(1586)	1586	-
Turkey	22(9)	6	-
USA	4205(3464)	3462	-
Others	127(0)	-	-
Total	6240(5188)	5168	10

Apart from these fungi, *Fusarium solani* (3), *Pestalotia* sp. (1) from Germany; *Alternaria zinniae* (1) and *A. solani* (1) from Turkey; *Botrytis cinerea* (2) from USA and *Drechslera sorokiniana* (1) from Italy were also detected.

sesamum and oil-palm seeds. The grey mould pathogen, *Botrytis cinerea* Pers. ex. Pers., a common saprophyte in the European countries, which caused heavy losses to chickpea crops in Bihar, U.P., Haryana and Punjab (Laha and Grewal, 1983) was detected on seeds of sunflower, safflower, soybean and *Brassica*. *Drechslera sorokiniana* (Sacc.) Sub & Jain the causal organism of seedling blight in many crops was detected in soybean, a non-host crop. Most of the fungi intercepted on various crops are reported to be seed borne and seed transmitted (Richardson, 1990).

Several species of *Alternaria* were intercepted on seeds of various crops. *A. brassicae* detected on seeds of *Brassica* spp. has become a major problem due to its spread from rape-seed at harvest to seeds of other *Brassica* spp.. Chahal and Kang (1979) reported internal seed borne

Table 3. Pathogenic Fungi intercepted in introduced seeds of *Glycine* spp.

Country	No. of samples received (found infected)	Pero-nopora manshurica	Cercospora kikuchii	Colleto-trichum dematium	Colleto-trichum glycine	Phomopsis sojae
Australia	296(1)	1	-	-	-	-
Belgium	3(1)	1	-	-	-	-
Brazil	79(11)	11	-	-	-	-
Indonesia	43(1)	1	-	-	-	-
Israel	9(2)	2	-	-	-	-
Italy	21(1)	1	-	-	-	-
Japan	24(2)	2	-	-	-	-
Korea	61(18)	18	-	-	-	-
Malaysia	6(6)	-	-	-	-	-
Nigeria	860(1)	-	-	1	-	-
Poland	12(2)	2	-	-	-	-
Russia	35(5)	5	-	-	-	-
Taiwan	2330(216)	167	5	16	2	17
Thailand	20(3)	1	-	2	-	-
USA	8478(1762)	1492	116	47	9	64
USSR	90(3)	3	-	-	-	-
Zimbabwe	206(3)	3	-	-	-	-
Others	2969(0)	-	-	-	-	-
Total	15,542 (2038)	1716	121	66	11	81

In addition to the fungi listed in the table, following fungi were also intercepted. *Aschochyta sojicola* (10); *Botrytis cinerea* (2); *Colleto-trichum gloeosporioides* (2); *Fusarium solani* (9); *Pestalotia* sp. (1); *Sclerotinia sclerotiorum* (1) and *Verticillium albo-atrum* (9) from USA. *Drechslera sorokiniana* (1) from Taiwan; *Macrophomina phaseolina* from Taiwan (6) and USA (1); *Rhizoctonia solani* from Taiwan (2) and USA (3); *Fusarium poae* from Poland (1) and USA (10).

infection of *A. brassicae* at levels of 59 per cent in India. *Alternaria carthami* Chowdhury, *Alternaria solani* Sorauer and *Alternaria zinniae* M.B. Ellis the leaf spot pathogens were detected on safflower seeds.

The Purple blotch pathogen, *Cercospora kikuchii* had devastated the soybean cultivation in

Table 4. Pathogenic fungi intercepted in introduced seeds of *Helianthus* spp.

Country	No. of samples received (found infected)	<i>Puccinia helianthi</i>	<i>Botrytis cinerea</i>	<i>Verticillium albo-atrum</i>	<i>Fusarium avenaceum</i>	<i>Fusarium solani</i>
Australia	310(34)	33	-	-	-	-
Belgium	3(1)	1	-	-	1	-
Bulgaria	81(7)	6	-	1	-	-
Canda	262(13)	13	-	-	-	-
Egypt	118(8)	-	-	-	-	-
France	230(23)	21	1	1	-	1
Germany	19(1)	-	-	-	-	1
Hungary	238(15)	15	-	-	-	-
Italy	1595 (35)	7	24	2	-	1
Romania	30(1)	-	-	1	-	-
Spain	44(4)	1	-	-	-	1
USA	3295(211)	200	8	-	3	1
USSR	129(10)	10	-	-	-	-
Yugoslavia	30(2)	2	-	-	-	-
Others	106(0)	-	-	-	-	-
Total	6490(365)	309	33	5	4	6

Apart from these fungi, *Phaetrichoconis* sp. from USA (1); *Pestalotia* sp. from Australia (1); *Alternaria zinniae* from Italy (1); and *Diplodia* sp. From egypt (8) were also detected.

USA during 1978 by reducing yield to the extent of 36 million metric tonnes. Out of 8478 samples received from USA, 116 were found to be infected with this pathogen.

Apart from the interceptions listed in the tables, 16 samples of *Linum* spp. were found infected with *Colletotrichum dematium* (Pers. ex Fr) Tul. from Hungary (8 samples) and Poland (1); *Colletotrichum gloeosporioides* (Penz.) Sacc. from USA (4) : *Rhizoctonia solani* Kuhn from Poland (3). Sixty-six samples of *Ricinus* spp. were infected with *Alternaria ricini* (Yoshii) Hansford from France (1), Italy (1), USA (55) and USSR (8), and *Phomopsis* spp. from U.K. (1).

Table 5. Pathogenic fungi intercepted on introduced seeds of *Sesamum* spp.

Country	No. of samples Received (Found infected)	<i>Alternaria sesami</i>	<i>Cercospora sesamicola</i>	<i>Macrophomina phaseolina</i>	<i>Fusarium solani</i>
China	30(3)	3	-	-	-
Israel	477 (19)	11	-	4	4
Korea	14 (9)	6	-	-	-
Philippines	30 (2)	2	-	-	-
Singapore	1728 (10)	10	-	-	-
South Africa	17 (2)	-	1	1	-
Thailand	30 (10)	1	-	8	1
USA	301 (69)	62	5	-	-
Others	394 (0)	-	-	-	-
Total	3021(124)	99	6	13	5

In addition to these, other fungi intercepted were *Phoma lingam* (1) and *Rhizoctonia bataticola* (1) from USA; *Corynespora cassicola* (9) from Korea

Thus, in the quarantine processing of 39,169 samples of different oil-seeds, 9027 seed samples (23 per cent) were found infected. All the seeds showing symptoms of discolouration, deformation and presence of fungal mycelium and fungal fructifications under stereobinocular microscope were removed to minimize the inoculum. Seed samples found infected with *Alternaria brassicae*, *A. brassicicola*, *A. raphani*, *A. solani*, *A. zinniae*, *A. carthami*, *A. sesami*, *A. ricini*, *Phoma lingam* and *Fusarium solani* were salvaged using hot water treatment at 50 C for 20 minutes. Ethyl alcohol was (Agarwal *et al.* 1990) was given to seeds of safflower and sunflower infected with rusts i.e. *Puccinia carthami* and *P. helianthi*, respectively. Chemical treatment with thiram and Bavistin (1:1) mixture was given for rest of the infected samples.

Almost 23 per cent of the introduced oil-seed germplasm were found infected with various seed-borne fungi. About 81 per cent of the

infected germplasm were salvaged. Soybean seed samples infected with *P. manshurica* amounting to 4% of the total introduction were rejected and burnt. Interception of pathogenic fungi, some of which are of high quarantine significance, emphasizes the role of quarantine in introduction of germplasm.

## REFERENCES

- Agarwal, P.C., A.K. Lambat, A. Majumdar and P. Kaur. 1982. Detection of seed-borne rust spores on safflower seeds by dry examination. *Seed Research* 10: 180-182.
- Agarwal, P.C. and R.K. Khetarpal. 1985. Interception of *Peronospora manshurica* in imported soybean seeds. *F.A.O. Plant Protection Bull.* 33: 39.
- Agarwal, P.C., Ram Nath, U. Dev and A. Majumdar. 1990. Quarantine salvaging of safflower seed infected with safflower rust (*Puccinia carthami*). *FAO Pl. Protection Bull.* 38: 141-143.
- Agarwal, P.C. and Baleshwar Singh. 1998. Interception of *Peronospora manshurica* (Naum.) Syd. in soybean germplasm introduced from 1978-97. *Indian J. Pl. Genet. Resources* 11: 31-34.
- Anitha, K., P.C. Agarwal and Ram Nath. 1993. Interception of *Peronospora manshurica* (Naum) Syd. in soybean seeds imported from Indonesia. *FAO Quarterly Newsl. for Asia and Pacific Plant Prot. Commission* 36: 1.
- Chahal, A.S. and M.S. Kang. 1979. Some aspects of seed-borne infection of *Alternaria brassicae* in rape and mustard cultures in Punjab. *Indian J. Mycol. Plant Pathol.* 9: 51-55.
- Laha, S.K. and J.S. Grewal. 1983. A new technique to induce abundant sporulation of *Botrytis cinerea*. *Indian Phytopathology* 36: 409-410.
- Li, M.X. Zha, J.Z. Liu and C. Yang. 1992. A preliminary study on the physiological races of soybean downy mildew, (Naum.) Syd. *Acta Phytopathol. Sinica* 22: 71-75.
- Marcinkowska, J. 1987. Incidence and variability of *Peronospora manshurica* (Naum) Syd. on soybean and susceptibility of soybean to fungus. *Wddawnictwo ssa WAR* 67 p.
- Majumdar, A., Usha Dec, P.C. Agarwal, Ram Nath, A.K. Lambat and K.D. Sharma. 1991. Pathogenic fungi intercepted in introduced germplasm. *Indian J. Pl. Genet. Resources* 4: 34-40.
- Mukewar, P.M., Ram Nath, A.K. Lambat, Usha Kapoor, R.K. Khetarpal and Indra Rani. 1980. Interception of *Peronospora manshurica* in imported seed of soybean. *Seed Research* 8: 170-173.
- Pathak, V.K., S.B. Matur and P. Neergaard. 1978. Detection of *Peronospora manshurica* (Naum.) Syd. In seeds of soybean, *Glycine max*. European and Mediterranean plant Protection Organization Bulletin 8: 21-28.
- Ram Nath, A.K. Lambat, P.M. Mukewar and Indra Rani. 1981. Interceptions of pathogenic fungi on imported seed and planting material. *Indian Phytopath.* 34: 282-286.
- Ram Nath, P.C. Indra Rani, Agarwal, R.K. Khetarpal, P.M. Mukewar, J.L. Varshney, U. Dev, P. Kaur, A. Majumdar and A.K. Lambat. 1985. Pathogenic fungi intercepted in imported seed and planting material during 1976-81. *Indian J. Agri. Sci.* 56: 115-120.
- Richardson, M.J. 1990. An Annotated List of Seed-borne Diseases. 4<sup>th</sup> Ed. International Seed Testing Association, Zurich, Switzerland. p. 387.
- Thomas, C.A. 1952. Transmission of safflower rust on treated seeds. *Phytopathol.* 42: 108-109.
- Thomas, C.A. 1955. A new race safflower rust. *Plant Dis. Reporter* 39: 652-653.