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Biological and chemical control of rice blast disease *(Pyricularia oryzae)* in Northern Greece

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Abstract. During a four-year field evaluation (1991-1994) of biological and chemical methods for the control of rice blast *(Pyricularia oryzae* Cav.*),* we studied the influence of environmental conditions on very important disease development.

Biological control of the fungus infection was attempted using Ferimzone (TF-164), which inhibited the mycelial growth by more than 90% at concentrations of 5 µg/mli and above, Flutolanil 25 with 2-2.5 µg/g in the leaf blast causing about 75% inhibition, such as the antagonists Trichoderma harzianum/CPO-80 and Chaetomium globosum/N76-1, which gave 70-88% inhibition of mycelial growth and conidial germination.

In another field experiment, the fungicides carbendazim, pyroquilon, triophanate methyl and chlobenthiazone reduced leaf but not neck blast; on the contrary, Otricyclazole was effective against neck blast.

In our area (N. Greece), the environmental conditions for blast development were favorable, particularly because the mean air temperature was between 21-28°C and wind speed 0.7-1.6 m/sec. On the contrary, relative humidity (54-84%) was adverse on the infection and disease development by the blast fungus *Pyricularia oryzae*.

I – Introduction

The discovery of several methods for the control of rice blast (*Oryza sativa* L.) caused by heterothallic Ascomycete, Magnaporthe grisea Barr (anamorph, *Pyricularia oryzae* Cav. or *Pyricularia grisea*) was the target for research in our country (Georgopoulos and Ziogas, 1992; Ntanos and Giamoustaris, 1991; Ntanos and Filippou, 1991; Thanassoulopoulos, Tzavella-Klonari and Katis, 1990).

The disease was observed mainly in our area (N. Greece) and attacks the leaves, culms, branches of the panicle and the floral structures (Georgopoulos and Ziogas, 1992; Ntanos and Giamoustaris 1991; Ntanos and Filippou, 1991; Thanassoulopoulos Tzavella-Klonari and Katis, 1990).

This study was necessary for the evaluation of biological and chemical methods for the control of this disease in relation with the influence of environmental conditions. However, the evaluation of fungal antagonists was necessary as *Trichoderma harzianum* (Ágrios 1988; Beagle-Ristaino and Papavizaw 1984; Sy, Sarr, Albertini and Moletti, 1990), *Chaetomium globosum* (Ágrios 1988; Soytong and Quimio, 1989; Sy, Sarr, Albertini and Moletti, 1990) and fungicides as Ferimzone (Okuno, Furusawa, Matsura and Shishiyama, 1989), Flutolanil (Hirooka, Miyagi, Araki and Kunoh, 1989).

Control of this disease was accomplished by using new fungicides as Fongoren (Georgopoulos and Ziogas, 1992; Moletti, Giudici, Nipoti and Villa, 1988; Naidu and Reddy, 1989), Beam (Georgopoulos and Ziogas, 1992; Mbodi, Gaye and Diaw, 1987; Moletti, Giudici, Nipoti and Villa, 1988; Okhovot, 1989) and Oryzemate (Georgopoulos and Ziogas, 1992) in relation with the influence of environmental conditions (temperature, leaf wetness, relative humidity) for blast development (Choong-Hoe, Mackenzie and Rush, 1988).

II – Materials and methods

The experiment was carried outside the farm, in Kalochori, of the Cereal Institute of Thessaloniki, during the 1991, 1992, 1993 and 1994 growing seasons. The rice variety “Rita”, partially resistant to blast, was used in the study. Seeds were planted in a soil with 18 cm row spacing at 110 kg seed/ha on 17-22 May. Fertilizers was applied at planting at a rate of 650 kg/ha of 20-10-10 (N-P2O5-K2O). The herbicide propanil was applied at the recommended rate (3.5 kg active ingredient/ha) before the permanent flood. A complete randomized block design was also used with ten treatments in four replications and the plot size was 5.3 m x 2.4 m. Five sprays were used with fungicides at 1000 litres/ha, three of which were applied up to leaf stage and the other two during panicle emergence.

In the first field experiment for biological control, the fungal antagonists, *Trichoderma harzianum* (CPO-80), *Chaetomium globosum* (N76-1), *Micromonospora* sp. (PS6-2), *Trichothecium roseum* (T372), *C. cochlioides*, *C. cuniculorum*, *Gliocladium roseum* and the fungicides Ferimzone (TF-164) and Flutolanil 25 were used.

Another experiment used the fungicides Derosal (carbendazim), Fongoren (pyroquilon), Hinosan TCP (edifenphos), Kitazin P (iprobenfos), Bla-S (blasticidin), Chlobenthiazone, Beam (tricyclazole), Orysemate (probenazole) and Neotopsin (thiophanate-methyl).

We also studied the environmental conditions (temperature, leaf wetness, relative humidity and wind speed) and their influence on disease development.

III – Results and discussion

The annual rice production and yield averages in Greece for the period 1981-1994 are shown in Table 1. The two types, Japonica and Indica, increased from 100 to 143 per 1000 ha, while the Indica type increased from 100% to 565%. Production for the two types, Japonica and Indica, increased from 100% to 200% (per 1000 tons) while yield increased from 100% to 140% (in tons/ha).

Table 1. Annual averages of the rice area, production and yield with respective indices in Greece, 1981-1994

	Area Japonica+Indica		**Area Indica**		**Production Japonica-Indica**		**Yield Japonica+Indica**	
	Ha	Index 1000	Ha	Index 1 000	Tons	Index 1 000	Tons/ ha	Index
1981	15.9	100	2.0	100	87.00	100	5.45	100
1982	15.5	98	1.0	50	83.38	96	5.37	99
1983	13.9	87	0.3	15	82.20	95	5.90	108
1984	14.0	88	0.9	45	89.84	103	6.43	118
1985	16.4	103	1.3	65	106.41	122	6.47	119
1986	17.6	111	1.6	80	119.34	137	6.79	125
1987	19.0	120	1.9	95	130.01	149	6.83	125
1988	20.6	130	4.0	200	114.10	131	5.53	102
1989	16.2	102	3.1	155	99.84	115	6.18	113
1990	16.0	101	2.3	115	95.98	110	6.00	110
1991	14.7	92	3.9	195	89.00	102	6.06	111
1992	14.5	91	4.9	245	109.51	126	7.54	138
1993	19.2	121	72.1	360	141.80	163	7.38	135
1994	22.8	143	11.3	565	174.42	200	7.64	140

For the first experiment, the biological control of rice blast by fungal antagonists and fungicides during the period 1991-94 are presented in Table 2. The mycelial growth, in the case of *Trichoderma harzianum* treatment, was 71% and conidial germination 88% (Ágrios, 1988; Beagle-Ristaino and Papavizaw, 1984; Georgopoulos and Ziogas 1992; Sy, Sarr, Albertini and Moletti, 1990). *Chaetomium globosum* was cha-

racterized by high stability profile for the control of mycelial growth (68%) and conidial germination (76%) (Ágrios, 1988; Soytong and Quimio, 1989; Sy, Sarr, Albertini and Moletti, 1990). The antagonists, *Micromonospora* sp. and *T. roseum,* did not give good results (Sy, Sarr, Albertini and Moletti, 1990) while *Chaetomium* cochlioides was a good antagonistic to the rice blast pathogen (59% and 65%). The protective capacity of the culture filtrate suggested that the fungus may produce an antagonistic substance inhibiting the development of *P. oryzae* (Soytong, 1991). On the contrary, *C. cuniculorum* had moderate results (Soytong, 1991) while the fungal antagonist *Gliocladium roseum* effectively decreased the disease development (Beagle-Ristaino and Papavizaw, 1984). Ferimzone, a new fungicide with biological properties, inhibited the mycelial growth of *P. oryzae* by 89% at 5-20 µg/ml while at 20 µg/ml it did not inhibit conidial germination, but the cytoplasm of the spores and hyphal were granulated and localized (Okuno, Furusawa, Matsura and Shishiyama, 1989). The fungicide flutolanil inhibited hyphal growth and infection-cushion formation of R. solani on rice leaf sheaths but was not effective against leaf blast *(P. oryzae)*.

Table 2. Biological control of *Pyricularia oryzae* by fungal antagonists and fungicides in Northern Greece 1991-94

Antagonists and fungicides	Rate of application	Inhibition*	
		Mycelial growth %	Conidial germination (%)
Trichoderma harzianum (GPO-80)	--	71 ab**	88 a
Chaetomium globosum (N76-1)	--	68 ab	76 ab
Micromonospora sp. (PS 6-2)	--	14 d	21 de
Trichothecium roseum (T372)	--	19 d	32 d
Chaetomium cochlioides	--	59 bc	65 b
Chaetomium cuniculorum	--	42 c	46 c
Gliocladium roseum	--	62 b	66 b
TF - 164 (ferimzone)	5-20 µg/ml	89 a	34 d
Moncut (flutalonil 25%)	2-2.5 µg/g	13 d	19 de
None control	--	5 e	7 f

* Average of four years;
** Numbers in each column followed by same letter do not differ significantly from each other at P=0.05 according to Duncan's multiple range test.

In the second experiment, the evaluation of fungicides in sprays against *P. oryzae* in the same period are shown in Tables 3 and 4. The fungicide Derosal (carbendazim) in doses of 1.5 lb/100 gallons gave satisfactory control with sprays commencing before the appearance of symptoms. The results were better against leaf blast than neck infection according to other researchers (Georgopoulos and Ziogas, 1992; Naidu and Reddy, 1989; Seneviratne de S., 1978). Fongoren (pyroquilon) in the dose of 2 kg/ha gave good results against leaf and neck blast (Georgopoulos and Ziogas, 1992; Moletti, Giudici, Nipoti and Villa 1988; Naidu and Reddy 1989) while the Beam (tricyclazole) in 0.75 kg/ha effectively decreased neck blast with one or more applications followed by pyroquilon (Georgopoulos, Ziogas 1992; Mbodi, Gaye and Diaw 1987; Moletti, Giudici, Nipoti and Villa 1988; Okhovot, 1989). The fungicides Hinosan (edifenphos) with 3.0 lb/100 gallons had moderate effectiveness (Georgopoulos and Ziogas, 1992; Mbodi, Gaye and Diaw, 1987; Naidu and Reddy, 1989; Rana and Singh, 1976; Seneviratne de S., 1978) while Kitazin (iprobenfos) (Georgopoulos and Ziogas, 1992; Naidu and Reddy, 1989; Rana and Singh, 1976), and Bla-S (blasticidin) (Georgopoulos and Ziogas, 1992) reduced leaf but not neck blast infection. Moreover, granular Chlobenthiazone in the dose of 30-40 kg/ha was better in leaf than neck infection (Naidu and Reddy, 1989) while Oryzemate (probenazole) (Georgopoulos and Ziogas, 1992) and Neotopsin (thiopharate-methyl) provide a very good control against the disease development.

Table 3. Evaluation of fungicides in sprays against rice blast in Northern Greece, 1991-94

Treatments and concentration	Rate of application	Leaves infection %* 1991	1992	1993	1994
Derosal WP (carbendazin 60%)	1.5 lb/l00gal	17.3 ab**	19.7 ab	22.1 ab	18.2 a
Fongoren WP (pyroquilon)	2 kg/ha	9.8 a	8.8 a	24.3 ab	13.4 a
Hinosan TCP 35 WP (edifenphos 0.1%)	3.0 lb/l00gal	35.5 bc	41.3 bcd	54.5 cd	42.7 bcd
Kitazin P (iprobenfos 0.1%)	750 g/ha	39.8 bcd	38.6 bc	47.8 bcd	53.9 cd
Bla-S (blasticidin)	100 µg / ml	34.1 bc	32.5 bc	35.1 abc	42.5 bcd
Chlobenthiazone	30-40 kg/ha	21.7 ab	26.1 abc	39.4 bc	20.2 ab
Beam (tricyclazole)	0.75 kg / ha	9.4 a	18.9 ab	11.2 a	8.8 a
Oryzemate (probenazole)	—	11.7 a	13.7 a	20.3 ab	12.0 a
Neotopsin WP (thiophanate-methyl 70%)	1.0 lb/l00gal	19.2 ab	34.2 bc	27.0 ab	36.3 bc
Unsprayed check	—	64.6 e	71.8 e	73.9 e	69.7 e

* Averages of four replications.
** Numbers in each column followed by same letter do not differ significantly from each other at P=0.05 according to Duncan's multiple range test.

Table 4. Evaluation of fungicides in sprays against rice blast in Northern Greece, 1991-94

Treatments and concentration	Rate of application	Leaf infection %* 1991	1992	1993	1994
Derosal WP (carbendazin 60%)	1.5 lb/l00gal	35.6 b**	23.2 b	30.6 bc	31.7 b
Fongoren WP (pyroquilon)	2 kg/ha	8.9 a	9.4 a	12.3 a	7.7 a
Hinosan TCP 35 WP (edifenphos 0.1%)	3.0 lb/l00gal	39.2 bc	37.3 cd	39.7 cd	36.1 bc
Kitazin P (iprobenfos 0.1%)	750 g/ha	49.1 d	37.5 cd	49.1 de	46.4 cde
Bla-S (blasticidin)	100 mg / ml	40.8 bc	32.4 c	38.3 cd	41.2 cd
Chlobenthiazone	30-40 kg/ha	47.5 c	33.1 c	46.5 de	37.7 c
Beam (tricyclazole)	0.75 kg / ha	8.3 a	18.7 ab	10.2 a	8.6 a
Oryzemate (probenazole)	--	12.6 a	17.5 ab	23.4 ab	10.4 a
Neotopsin WP (thiophanate-methyl 70%)	1.0 lb/l00gal	39.8 bc	36.4 cd	42.3 cde	45.2 cde
Unsprayed check	--	71.3 e	77.3 e	69.2 f	70.1 f

* Averages of four replications.
** Numbers in each column followed by same letter do not differ significantly from each other at P=0.05 according to Duncan's multiple range test.

Finally Figures 1-5 present the averages of environmental conditions in Northern Greece for the period 1991-1994.

According to Choong-Hoe Kim et al. (1988), temperatures between 19°-29°C, particularly in the range 23°-26°C and lasting more than 16 hrs at relative humidity of above 90%, are considered to be highly favorable conditions for blast development. The most favorable condition for blast development is a mean temperature between 23°-26°C, 24 hrs of leaf wetness and 24 hrs of high relative humidity (above 90%).

Especially in Northern Greece, the conditions for blast development were not favorable. The average rainfall of 0-85 mm (Figure 5) and relative humidity of 54-84% (Figure 4) were most of the time unfavorable to infection and disease development by the blast fungus *Pyricularia oryzae*.

The adverse effect of relative humidity was not influenced by the favorable temperatures in the area (average temperature: 21-28°C; min.: 12°; max.: 34°C) (Figures 1 & 2) and by the wind speed (0.5-1.7 m/sec.) (Figure 3).

Figure 1. Average temperatures of the rice area in Northern Greece (1991-1994)

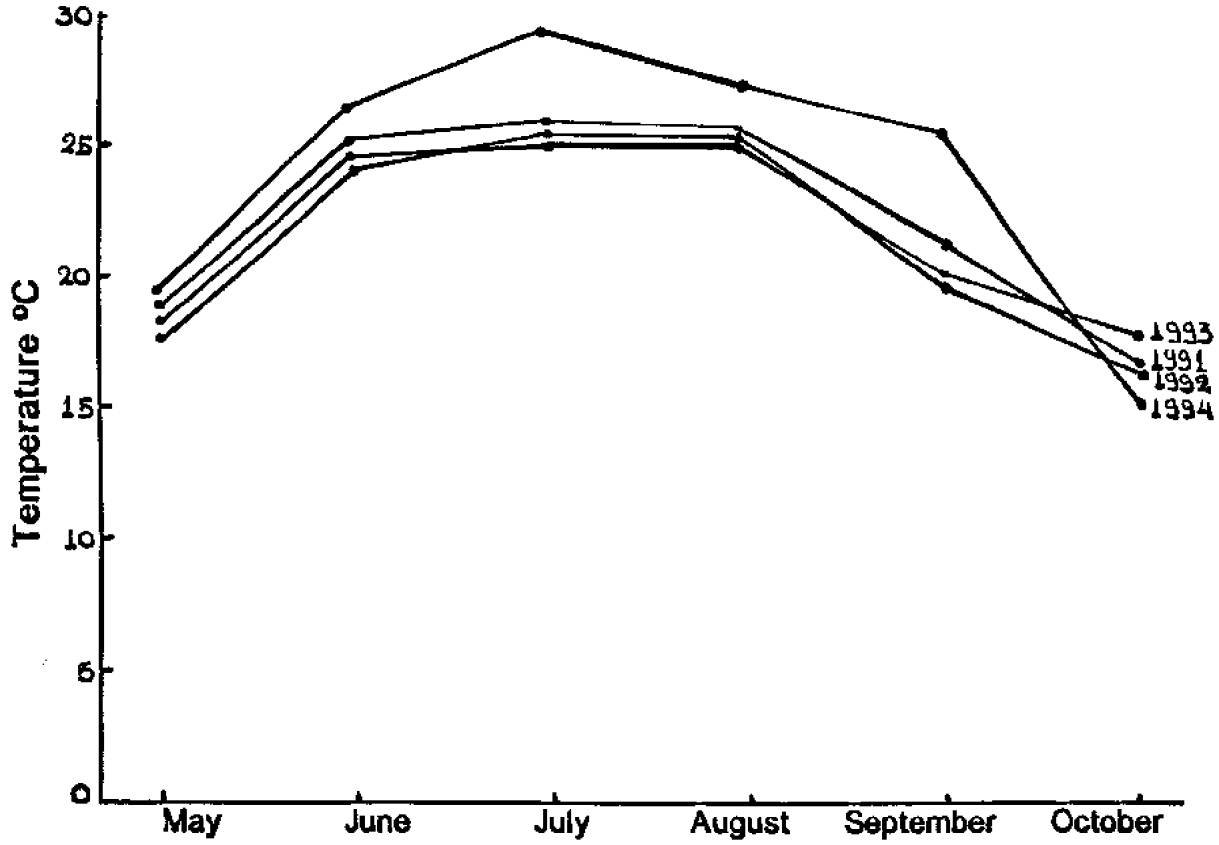


Figure 2. Average temperatures (min-max) of the rice area in Northern Greece (1991-1994)

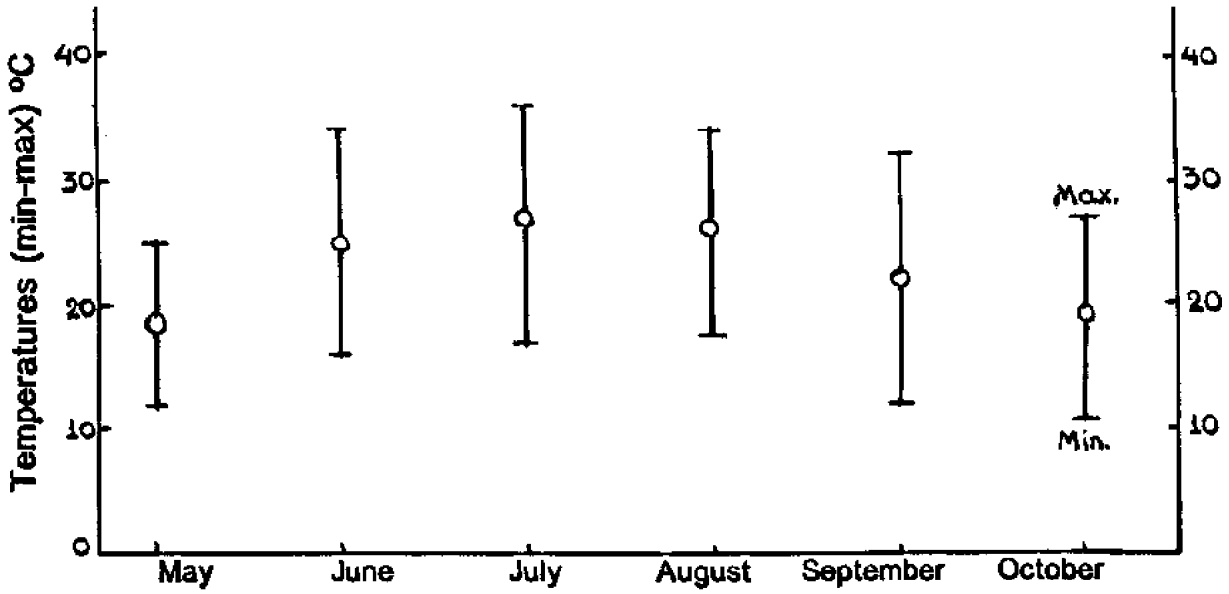


Figure 3. Average wind speed of the rice area in Northern Greece (1991-1994)

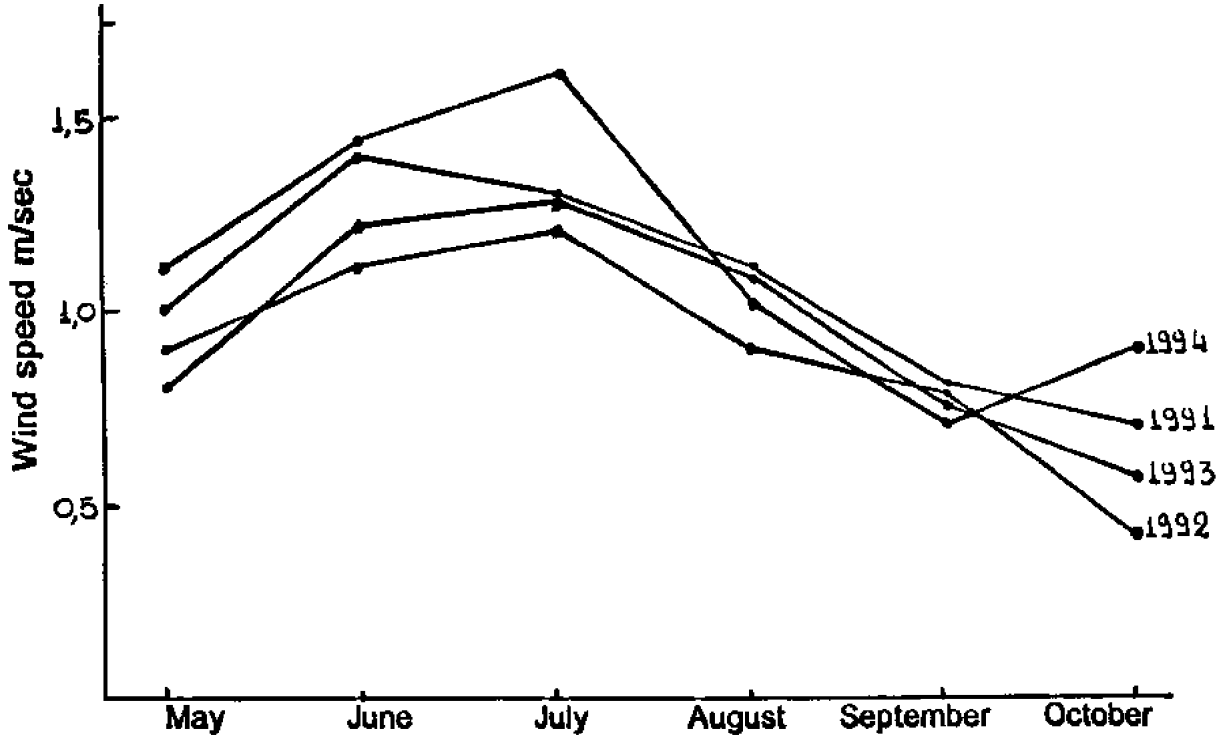


Figure 4. Average relative humidity of the rice area in Northern Greece (1991-1994)

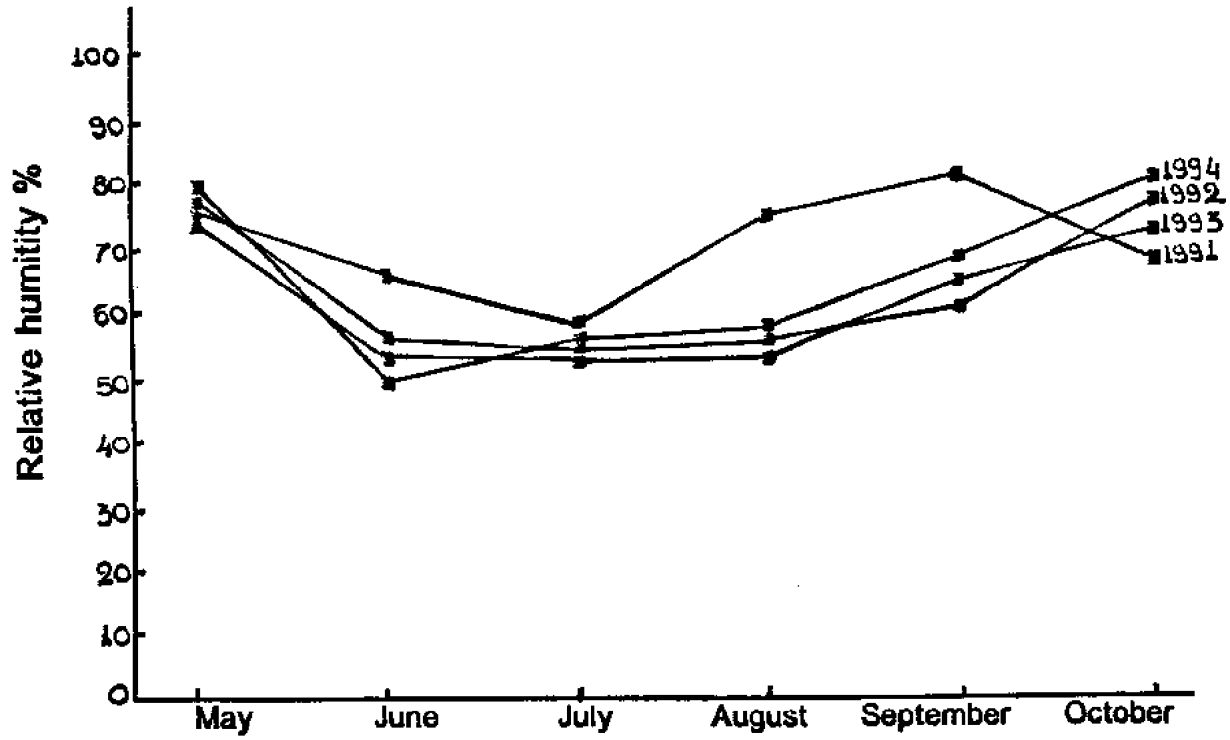
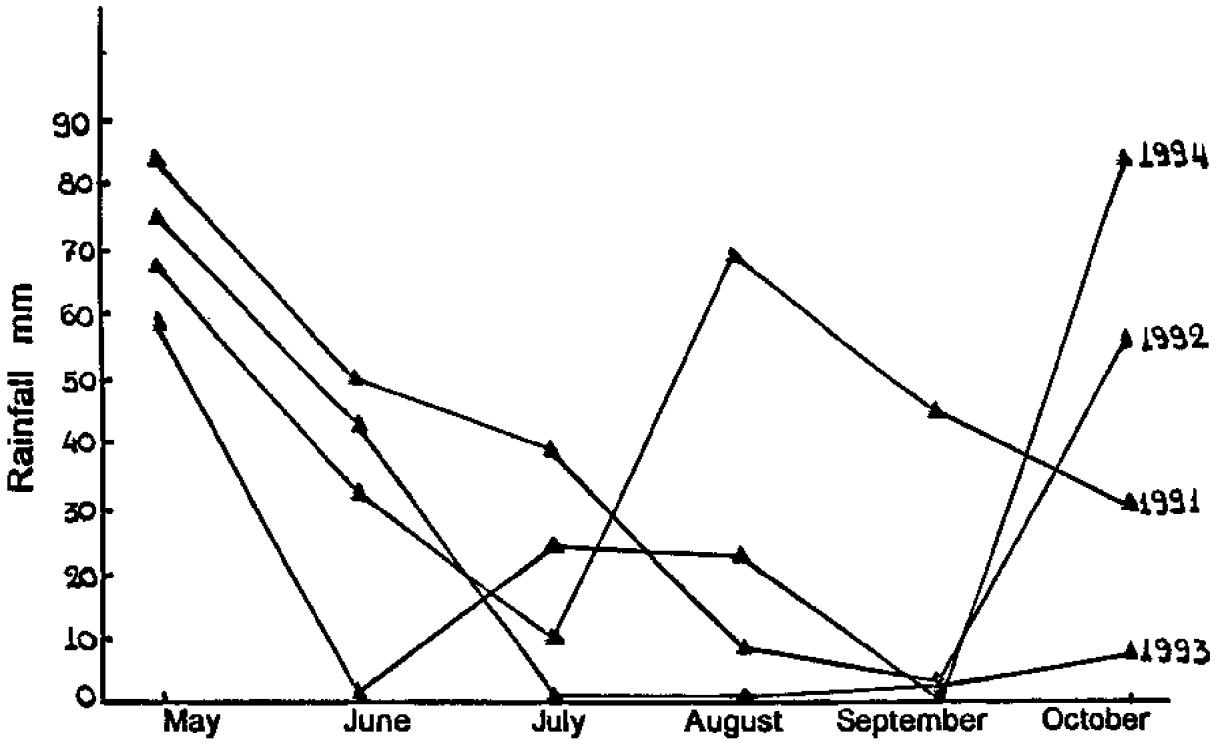


Figure 5. Average rainfall of the rice area in Northern Greece (1991-1994)



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