GERMPLASM PROGRAM LEGUMES

Annual Report for [illegible]

GERMPLASM PROGRAM-LEGUMES

1995 ANNUAL REPORT

1. INTRODUCTION

The cereals and legumes improvement research at the International Center for Agricultural Research in the Dry Areas (ICARDA) is done by the Germplasm Improvement Program. Among the cereals, it covers barley, durum wheat and bread wheat, while amongst the legumes it covers lentil, chickpea, forage legumes and pea. ICARDA has a global mandate for the improvement of barley, lentil and faba bean, and a regional mandate for the improvement of durum wheat, bread wheat, chickpea, pea and forage legumes. The improvement of durum and bread wheat is done jointly with the International Maize and Wheat Improvement Center (CIMMYT), Mexico, which has a global mandate for wheat improvement. Similarly, chickpea improvement is done jointly with the International Crop Research Institute for the Semi-Arid Tropics (ICRISAT), India, which has a global mandate for this crop.

To fulfill the global mandate for the improvement of barley, ICARDA has posted a barley breeder in CIMMYT-Mexico to address the needs of barley improvement for Latin America. CIMMYT has placed a durum breeder and a spring bread wheat breeder at ICARDA with a regional responsibility for West Asia and North Africa (WANA). In the case of chickpea, ICRISAT has posted a chickpea breeder at ICARDA to address the needs of the crop in WANA.

The overall objective of the Germplasm Improvement Program is to increase the productivity and sustainability of the farming systems which include barley, lentil, faba bean, durum wheat, bread wheat, chickpea, grasspea, pea and forage legumes in partnership with NARS, NGO and farmers.

This objective is being pursued through methodologies emphasizing specific adaptation through decentralized breeding, use of biotechnology, use of inputs compatible with the preservation and improvement of the resource base, and maintenance and enhancement of agricultural biodiversity.

The base for most of our research work is at the Tel Hadya, where ICARDA's headquarters are located and where additional environments are created by different planting dates. However, an increasing amount of work is done in other sites in Syria (Breda, Bouider and Jindiress) and Lebanon (Terbol and Kfardan). All these sites are directly managed by ICARDA. High elevation sites of the national programs of Syria, Turkey, Russia, Iran and Maghreb countries are used, in a collaborative mode, for developing improved winter and facultative barley, bread wheat, durum, lentil, chickpea and forage legumes adapted to cold environments. The research sites and facilities of other national programs are used jointly for developing breeding material with specific resistance to some key biotic and abiotic stress factors because of the presence of ideal screening conditions and/or expertise there. The process of decentralization of breeding work is being continued and extended with the help of national programs.

The weather conditions during the season are shown in Figures 1.1 for two typically dry sites (Bouider and Breda) and in Figure 1.2 for two wet sites (Tel Hadya and Terbol). The long-term average rainfall at these sites is 236, 267, 323, 548 mm, respectively. In all the four locations the total seasonal precipitation in the 1994-95 cropping season was less than the long term average (203, 244, 313 and 531 mm, respectively). In both Bouider and Breda there was a wet and relatively warm beginning of the winter: the winter continued warm and dry in Bouider and cold and dry in Breda. Late rains accompanied by colder than normal Spring temperatures caused a slightly longer growing season. In Tel Hadya and Terbol there was a very wet start of the season, followed by drier than usual winter and spring. The late rains in Tel Hadya partly alleviated the effects of the drought, while the cropping season ended in Terbol with an increasing moisture deficit.

There were a large number of interesting developments in all the projects and it is not possible to even summarize them in this introduction. There was a critical examination of decentralization with the conclusion that the time has come to include the farmers in a participatory fashion during the development of the major technological output of the

Program, i.e., new varieties. During 1995 this approach has been implemented successfully in our collaborative barley project in Ethiopia. There was also, as a general trend, a continuation of the efforts towards more and more precise targeting of the germplasm, including new efforts towards quality aspects in relation to human consumption. The widespread use of landraces and wild relatives continued to be one of the major characteristics of the breeding programs. A large amount of resources continued to be invested in the identification and use of sources of resistance to biotic stresses, as part of the overall commitment of the Program to sustainable development. Eventually, and this is perhaps the area which saw the largest expansion in 1995, molecular techniques started to be applied in virtually every crop the Program is dealing with, with the exception of the forage legumes.

More than 50 scientists from 25 different countries spent between few days and few months in the Germplasm Improvement Program. Their activities varied from discussions with staff members to research projects in collaboration with specific scientists. Their contributions to the achievements of the Program are reported in details in the specific sections.

This report is published in two sections, one with the results of cereal crops improvement work and one with results of the legume crops improvement work.

Most of the results reported in the two sections were obtained during the 1994-95 season, although work done in earlier years is also reported when considered important.

The training and network activities and the publications of the Program are also listed.

As mentioned earlier, much of the work reported here has been done in collaboration with our colleagues in the national programs in WANA and other developing countries and in some institutions in the industrialized countries. Space limitations prevent to mention all our collaborators individually, but to all of them goes our sincere appreciation.

S. Ceccarelli

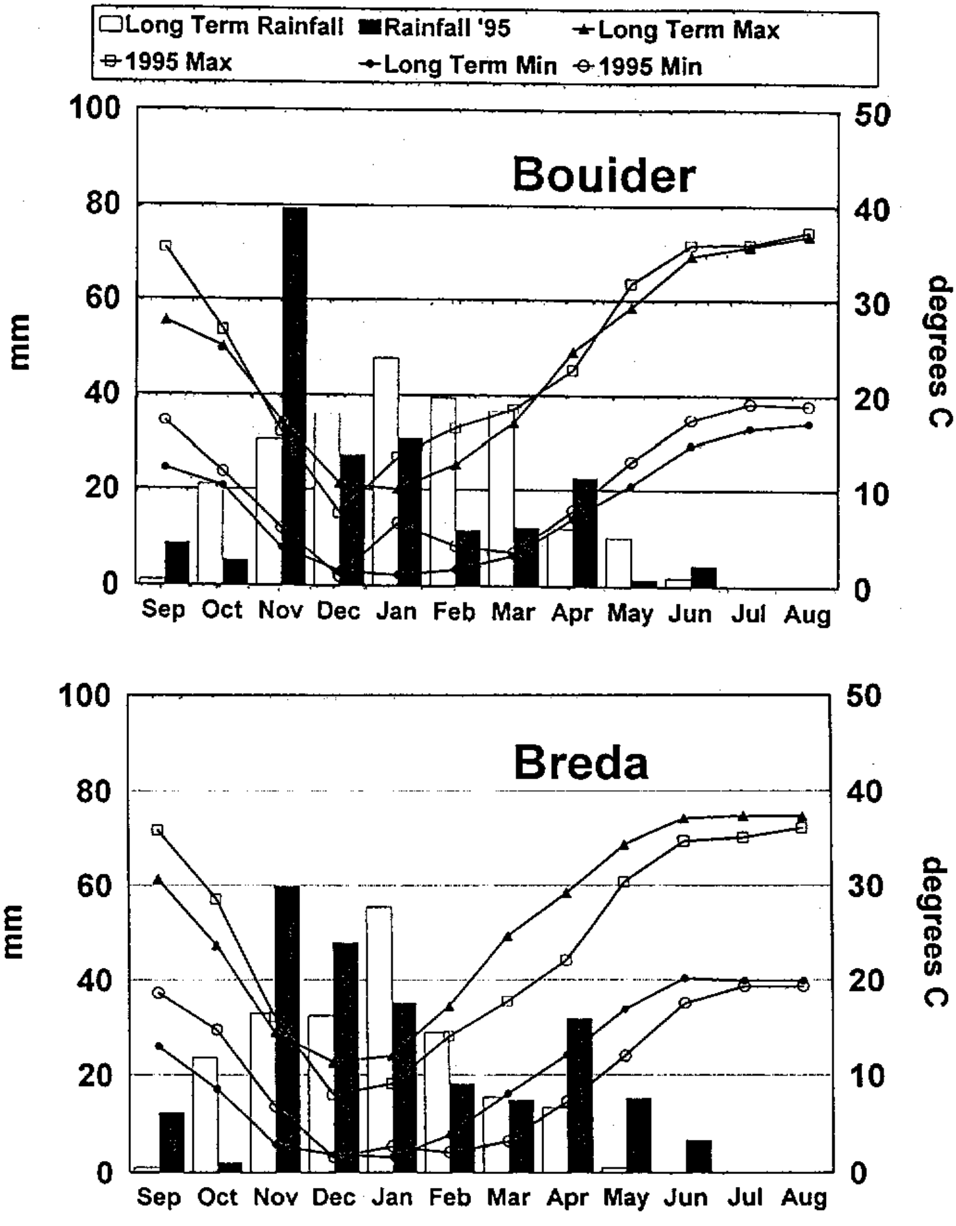

Fig. 1.1. Weather conditions at Bouider and Breda during 1994-95.

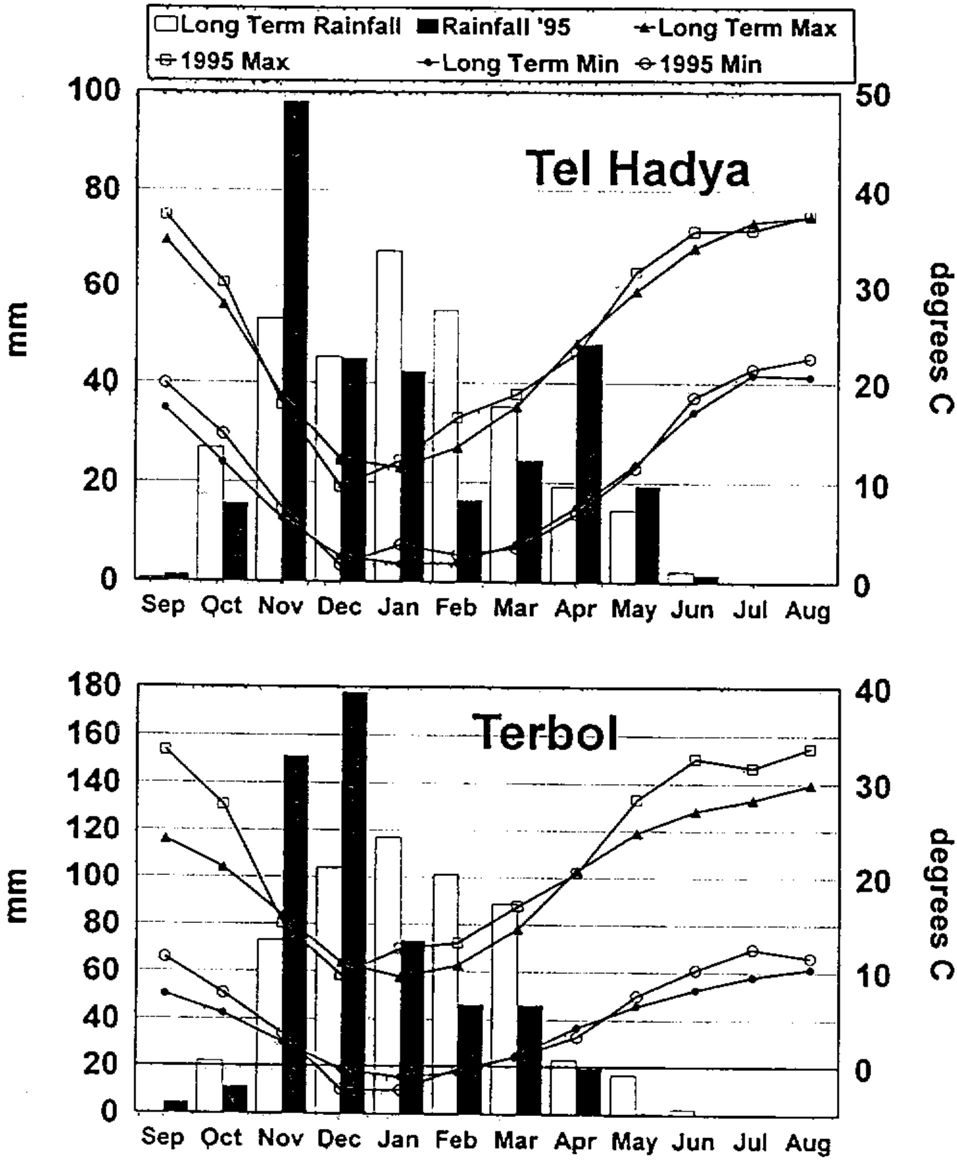

Fig. 1.2. Weather conditions at Tel Hadya and Terbol during 1994-95.

2. LENTIL IMPROVEMENT

Introduction

Average lentil yields are low because of poor crop management and the low yield potential of landraces. In South Asia and East Africa diseases are also a major constraint to production. Accordingly an integrated approach to lentil improvement is being pursued at ICARDA covering the development of both improved production technology and genetic stocks. A high priority has been placed on transferring to national programs the results of research on lentil harvest mechanization systems to reduce the high cost of harvesting by hand in the West Asia and North Africa region.

2.1. Lentil Breeding

2.1.1. Base Program

2.1.1.1. Lentil Adaptation and Breeding Scheme

The lentil is an under-exploited and under-researched annual legume. From the onset at ICARDA, we studied the variation in the world germplasm collection to understand factors affecting lentil adaptation to direct the breeding program. Such diverse factors as winter-hardiness, efficiency of iron uptake, phenology as related to the length of the growing season, the sensitivity of flowering to temperature and photoperiod, response to irrigation and disease resistance have all contributed to the pattern of variation found in the world germplasm collection.

Additional information on the specificity of adaptation within the crop has come from collaborative yield trials of common entries selected in different locations. For example, the North African Regional yield trial on lentil was established in 1990 to comprise the best lines selected in Algeria, Libya, Morocco and Tunisia. The results of this regional yield trial showed that lentils selected in the various countries of N. Africa differ substantially in phenology, indicative of the need for specific adaptation to the range of environments in the region. The requirements of Libya, Tunisia, and lowland Algeria are met by lines emanating from the ICARDA West Asian breeding program. However, late-maturing material is required for high altitude areas of Algeria and early-maturing lines are required in Morocco with resistance to rust. Armed with this understanding of the specific adaptation of the lentil crop and the various consumer/end-use quality requirements of different geographic areas, we have designed the base breeding program as a series of separate, but finely targeted, streams linked closely to national breeding programs.

Table 2.1.1. Target agro-ecological regions of production of lentil and key breeding aims.

Region	Key traits for recombination
Mediterranean low to medium elevation	
1. 300-400 mm ann.rainfall	Biomass (seed + straw), attributes for mechanical harvest & wilt resistance
2. < 300 mm ann. rainfall	Biomass, drought escape through' earliness
3.Morocco	Biomass, attributes for mechanical harvest & rust resistance
4. Egypt	Seed yield, response to irrigation, earliness & wilt resistance
High elevation	
1. Anatolian highlands	Biomass & winter hardiness
2. N. African highlands	Seed yield & low level of winter hardiness
South Asia and E. Africa	
1.India,Pakistan, Nepal& Ethiopia, Bangladesh	Seed yield, early maturity, resistance to rust, Ascochyta and wilt
	2. Seed yield, extra earliness & rust resistance

The three major target agro-ecological regions of production of lentil are 1. S. Asia and E. Africa 2. Mediterranean low to medium elevation and 3. High elevation area of West Asia and North Africa. These correspond to the maturity groups of early, medium and late maturity. Within each of these major regions there are specific target areas. Thus, for example, within the Mediterranean low to medium elevation region, specific target areas are 1. the major production area of 300-400 mm annual rainfall, 2. arid areas with < 300 mm annual rainfall, 3. Morocco, where there is the additional problem of rust and 4. Egypt, where lentil is irrigated. Each of these target areas has slightly different blends of key traits for recombination. The target areas/regions and key traits for selection/recombination are tabulated in Table 2.1.1.

Based on the premise that local selection for adaptation to a specific target area is the most efficient selection method, selection at ICARDA in West Asia is limited to adaptation to the home region - Mediterranean low to medium elevation - and for traits where we have a comparative advantage such as vascular wilt resistance. As a result, the breeding program has decentralized to work closely with national programs.

For the home region the breeding program uses a bulk-pedigree system with off-season generation advancement (at Terbol, Lebanon 950 m elevation), single plant selection in the F_4 generation and selection of progeny rows for vascular wilt resistance in the F_5 generation. For the other regions, crosses are agreed with cooperators and made at Tel Hadya; the generations advanced in the off-season and the segregating populations shipped to national cooperators for local selection. We started making specific crosses in 1985 and since then have made specific crosses for Algeria, Bangladesh, India, Jordan, Morocco, Nepal, Syria and Turkey. A total of approximately 200 crosses are made annually. One avenue for the distribution of segregating material is through these country-specific crosses. The international trial network provides another system whereby these crosses can be tested sub-regionally (see section 2.1.1.3). Selections made by NARS are fed back into the international trial system for further distribution. The results from this decentralized system are described in section 2.1.2.

2.1.1.2. Yield Trials

Selections from the breeding program for the Mediterranean low to medium elevation region are tested at three locations varying in their annual average rainfall, namely Breda (long-term average annual rainfall total 267 mm) and Tel Hadya (323 mm) in Syria and Terbol (548 mm) in Lebanon in preliminary yield trials in the F_7 generation and in advanced yield trials the following generation and season. The lines are also re-tested synchronously for vascular wilt resistance in the wilt-sick plot at Tel Hadya (see Section 2.1.1.4) to ensure that only high-yielding, wilt resistant lines are advanced in the breeding program.

Table 2.1.2. Results of the lentil yield trials (preliminary and advanced) for seed (S) and biomass (B) yields (kg/ha) at three contrasting rainfed locations, Terbol (Lebanon), Tel Hadya and Breda (Syria) during the 1994/95 season.

Location	Terbol		Tel Hadya		Breda	
	S	B	S	B	S	B
Number of trials	6	6	9	9	8	8
Number of test entries*	138	138	195	195	172	172
% of entries sig. (P<0.05) exceeding best check**	0.7	22.4	37.4	37.4	13.9	13.9
% of entries ranking above best check (excluding above)	13.0	23.8	21.0	32.8	9.9	23.8
Yield of top entry (kg/ha)	3377	8875	2142	6436	635	2349
Best check yield (kg/ha)	3008	7451	1504	3824	334	1759
Location mean (kg/ha)	2645	7379	1522	4168	248	1680
Mean C.V. (%) over trials	5.7	5.4	19.5	17.0	30.9	15.8
Mean % advantage of lattice over RCB analysis across trials	42.3	38.2	10.0	11.6	13.1	21.4
Mean % advantage of NNA*** over RCB analysis across trials	91.7	101.3	16.3	18.5	37.0	78.6

* Entries common over locations.
** Large-seeded checks: ILL 4400 long-term, ILL 5582 improved;
small-seeded checks: ILL 4401 long-term, ILL 5883 improved.
*** NNA = Nearest neighbor analysis

The 1994/95 season was drier than average with rainfall totals of 244 mm received in the growing season at Breda, 313 mm at Tel Hadya and 531 mm at Terbol. The winter was less cold than normal in Syria with 32 frost days at Breda and 23 d at Tel Hadya in the 1994/95 season compared to the long-term averages of 43 d at Breda and 37 d at Tel Hadya. By contrast the winter was cooler and longer at Terbol in Lebanon, where there were 98 frost events in the 1994/95 season.

The average seed yield varied from 2645 kg/ha at Terbol, through 1521 kg/ha at Tel Hadya to only 248 kg/ha at Breda (Table 2.1.2). The corresponding biomass yields were 7.4 t/ha in Terbol, 4.2 t/ha in Tel Hadya and 1.7 t/ha in Breda. The harvest index (HI) was strikingly lower in Breda, at HI = 0.15, compared to HI = 0.36 at the other trial sites. The crop in Breda was so severely drought-stressed prior to the arrival of rain during pod-filling that it was unable to benefit from the late rain. In contrast, the crop was able to uptake some of these late rains for seed-fill at the other two sites. The percentage of lines yielding significantly more seed than the best check was greatest in Tel Hadya and lowest in Terbol. The mean coefficient of variation over trials was the inverse of seed yield with low variability in Terbol and high variability at the dry Breda site.

All trials (with one exception) were arranged in a 5x5 lattice design. A comparison was made between the efficiency of analysis as a lattice design and as a randomized complete blocks design. Additionally, a comparison was made with the nearest neighbor algorithm in the software package AGROBASE/4. This nearest-neighbor analysis (NNA) takes the difference between the yield of a plot and the average of its two adjacent plots. For border plots, the two plots on the one side are taken as adjacent plots. Such information from the 'moving blocks' of three plots is combined for each entry across the whole trial to estimate a mean neighbors difference, then repeated till convergence. The average advantage of lattice analysis of seed yield over that of randomized complete blocks was 20 % over 21 trials (Table 2.1.2). The equivalent advantage of NNA over randomized blocks was 44 % in the same trials. Clearly, NNA was superior to lattice analysis, which was, in turn, superior to analysis as randomized complete blocks. NNA should be assessed further as it is a method of adding value to existing trial data with little extra cost.

W.Erskine

2.1.1.3. International Nurseries

The lentil international breeding nurseries have evolved in response to the needs of NARSs from the provision of un-targeted yield trials to a diversified array of crossing blocks/resistant sources, segregating populations and yield trials for each of the three major target agro-ecological regions of production (Table 2.1.3). The overall time trend of diversification and specific targeting of genetic material is illustrated in Figure 2.1.1. Since 1987, for example, we have diversified and targeted the supply of segregating material from two into four different nurseries - Cold Tolerant, Large-seeded, Small-seeded and Early. In the same period, new nurseries of stress resistant material have been launched against rust, Ascochyta blight, Fusarium wilt, drought and cold.

Table 2.1.3. Lentil international breeding nurseries showing target regions and type of nursery in the 1995 season.

Type of nursery	Region		
	Mediterranea	Lower latitudes	High elevation
Res. source/ crossing block	Large-seeded Small-seeded Fusarium wilt* Drought escape	Early* Rust* Ascochyta blight*	Cold tolerant*
Segregating population	Large-seeded* Small-seeded*	Early	Cold tolerant*
Yield trial	Large-seeded Small-seeded	Early	

* Launched since 1987.

There is a slow increase in the number of entries in international trials provided by national programs. It is our aim to increase the input of national programs into the international testing program. The line-up of the 1995 international trials included three lines from Pakistan, two from Turkey and one each from Bulgaria, Slovakian Republic, Russia and USA . Other entries have been supplied by NARSs and are in multiplication for inclusion in next season's trials.

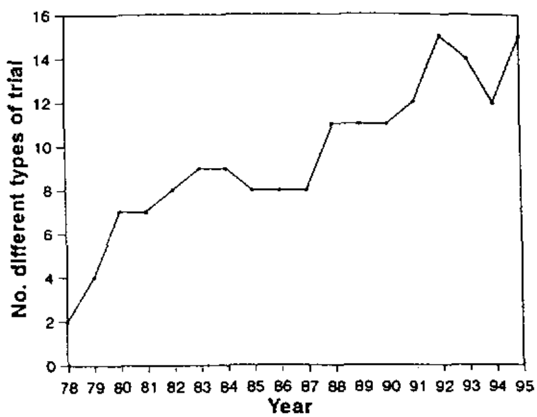

Figure 2.1.2. Time trend of the number of different types of lentil international trial/nursery despatched.

Table 2.1.4. Destination and number of seed samples of lentil breeding lines and segregating populations despatched on the request of NARSs over six years. This is additional to the material distributed through the International Legume Testing Network.

Country	1990	1991	1992	1993	1994	1995	Total	%of total
Afghanistan		10	1				11	0.28
Algeria	2					47	49	1.28
Argentina		2					2	0.05
Australia		13			173	141	327	8.56
Bangladesh			12	4		107	123	3.2
Bhutan			62				62	1.62
Canada	11		1	55	1		68	1.78
China	108						108	2.82
Colombia	13		19				32	0.83
Egypt				46		10	56	1.46
Ethiopia						66	66	1.72
Germany	2	1		11	3	1	18	0.47
India	3	47	55				105	2.74
Iran			2			29	31	0.81
Iraq			1	24	2	2	29	0.75
Italy		9	30		3	16	58	1.51
Japan		8					8	0.20
Jordan			2		4	2	8	0.20
Kenya		5					5	0.13
Kuwait					5		5	0.13
Lebanon			2				2	0.05
Libya			1	2			3	0.07
Morocco	597	42	404	4			1047	27.40
Nepal						4	4	0.10
Netherlands		10					10	0.26
New Zealand					23		23	0.60
Nicaragua					4		4	0.10
Pakistan	17	100	62		6	91	276	7.22
Palestine					4		4	0.10
Poland					13		13	0.34
Russia	22				15		37	0.96
Saudi				4			4	0.10
Slovakia		5		1			6	0.15
South					4		4	0.10
Sudan	16			6			22	0.57
Syria			55	1	1	129	186	4.86
Turkey		369	283	3		108	763	19.97
UK	28				53		81	2.12
USA		4	116	39			159	4.16
Yemen			1				1	0.02
Total	819	625	1109	200	314	753	3820	

In addition to the International Legume Testing Network, seed requests from NARSs for specific breeding lines and segregating populations have resulted in the despatch of 3820 seed samples to 40 countries in the last six years (Table 2.1.4.).

W. Erskine and R.S. Malhotra.

2.1.1.4. Screening for Vascular Wilt Resistance

Vascular wilt caused by *Fusarium oxysporum* f. sp. *lentils* is the major fungal disease of lentil in the Mediterranean region.

2.1.1.4.1. Screening of breeding material for wilt resistance

The screening of breeding material in the wilt sick plot at Tel Hadya continued in the 1994/95 season with a total of 1061 lines tested. This follows the screening of 1229 lines in 1993/94 and 897 lines in 1992/93. The tested lines may be divided into two categories: Cycle I - new untested lines and Cycle II - lines tested previously (primarily those which showed a resistant or highly resistant reaction - see below). On the basis of mean over replicates, the percentage of entries with a highly resistant (0-5% wilted plants, mean over replicates) or resistant (>5-20% wilted plants, mean over replicates) response to lentil vascular wilt were 55.7 % for Cycle I and 92.5 % for Cycle II, indicating the reliability of the screening method (Figure 2.1.2). However for breeding purposes, we have selected for advancement a specific subset of the resistant lines - those which have a maximum plot score of < 20 % wilted plants in the last score. This screening is a key and integral part of the breeding program; this is elucidated in section 2.1.1.1.

The frequency of lines resistant to wilt was generally higher among small-seeded than large-seeded germplasm in previous screening. This season showed no exception as the percentage of resistant lines in Cycle I was 50.4 % among large-seeded material and 58.1% among the small-seeded. Next season we will continue the screening of breeding material for resistance and examine the large-seeded accessions in the germplasm collection.

2.1.1.4.2. Morphological variability among Syrian isolates of *Fusarium oxysporum* f. sp. *lentils*

A collection of 32 isolates was made from all the major lentil growing areas of Syria. We examined the variation among these isolates in aggressiveness on a susceptible cultivar and in the following morphological characters: colony growth rate, colony color, presence/absence of macro- and micro-conidia and chlamydospores, dimensions of macro- and micro-conidia, the nature of hyphal growth (aerial/pionnotal). In addition to overall variability, we searched for regional differences in morphology according to the collection site and associations between morphological and pathogenic traits using one-way analyses of variance. Furthermore the association of these characters with isozymes was investigated (see following Section).

There was significant variation among isolates in aggressiveness and in all the measured morphological characters. After assigning collections sites to the three major productions areas, regional differences were not found for any of the characters. The only significant associations between pathogenicity and morphological characters were that small micro-conidial width ($\leq 3\ \mu$) was related to aggressiveness and that a bluish-white to light pink color on the upper colony surface was associated with the most aggressive isolates.

At present screening is being conducted at ICARDA Tel Hadya against only the local isolate of *Fusarium oxysporum f. sp. lentils* (isolate 31) (see above). This survey revealed considerable variation among isolates in morphological characters and isozymes (see below). Given the variability in virulence observed in chickpea and pea[1], it would be pro-active to challenge a range of lines

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Kraft, J.M., Haware, M.P., Jimenez-Diaz, R.M., Bayaa, B. and Harrabi. M. 1994. Screening techniques and sources of resistance to root rots and wilts in cool-season food legumes. Pages 268-289 in Expanding the production and use of cool season food legumes. (eds F.J. Muehlbauer and W.J. Kaiser), Kluwer resistant to the Tel Hadya isolate with the other isolates.

2.1.1.4.3. Variability in isozymes among Syrian isolates of *Fusarium oxysporum* f. sp. *lentils*

A survey was made of isozyme polymorphism of the above 32 isolates. The isolates were polymorphic for a total of seven isozyme loci, namely:

Table 2.1.5. The average and standard error (SE) for each isozyme allele of the number of days to plant death of isolates as measured in tubes in semi-solid medium of Hoaglands in the laboratory and in plastic cones in the plastic house. Means followed by the same letter are not significantly different based on Duncan's multiple range test from one-way analyses of variance by isozyme allele.

		No. days to plant death			
		Tubes in		Plastic cones	
Isozyme	Band	Mean	SE	Mean	SE
	b	32.7b	1.8	48.5b	1.6
PGM-2	a	33.1a	1.7	49.3a	1.7
	b	41.9a	2.4	57.3a	2.9
	c	20.0b	0.0	40.8b	0.0
6PGD-2	a	37.4a	4.4	55.3a	4.0
	b	34.1a	1.7	50.0a	1.7
G6PD-1	a	36.5a	5.1	53.3a	5.0
	b	34.3a	1.7	50.3a	1.6
MDH-2	a	41.0a	0.0	62.0a	0.0
	b	35.4a	6.4	51.1a	5.8
	c	34.3a	1.7	50.3a	1.6
IDH-2	a	32.9a	1.7	48.9a	1.6
	b	40.6b	3.6	57.6b	3.0
PGI-1	a	40.3a	2.5	57.5a	2.4
	b	30.4b	1.9	46.2b	1.7

* AAT-3 = Aspartate aminotransferase-3; PGM-2 = Phosphoglucomutase-2; 6PGD-2 = 6-Phosphogluconate dehydrogenase-2; G6PD-1 = Glucose 6-phosphate dehydrogenase-1; MDH-2 = Malate dehydrogenase-2; IDH-2 = Isocitrate dehydrogenase-2 and PGI-1 = Phosphate glucose isomerase-1.

Academic Publishers, Dordrecht, the Netherlands.

Aspartate aminotransferase-3 (AAT-3), Phosphoglucomutase-2 (PGM-2), 6-Phosphogluconate dehydrogenase-2 (6PGD-2), Glucose 6-phosphate dehydrogenase-1 (G6PD-1), Malate dehydrogenase-2 (MDH-2) and Isocitrate dehydrogenase-2 (IDH-2) and Phosphate glucose isomerase-1 (PGI-1). These isozymes resolved the 32 isolates into 11 different electrophoretic patterns.

The association between the isozyme status and aggressiveness of isolates was examined using one-way analysis of variance. Among the seven loci examined, there were significant associations between aggressiveness and the allelic status of the loci AA-3, PGM-2, IDH-2 and PGI-1 (Table 2.1.5). Aggressiveness was associated with the fast band *a* in IDH-2, and with the slowest band in PGM-2 (band *c*) and PGI-1 (band *b*).

2.1.1.4.4. Association of isozyme pattern with wilt reaction in lentil

A diverse range of 63 lentil lines, previously assessed for their disease reaction to lentil vascular wilt in the laboratory, plastic house and field (ICARDA, 1994), were surveyed for their isozyme patterns.

The aim was to relate isozyme pattern to the level of resistance so that the isozymes might provide a marker in selection for disease resistance. Polymorphism was found among the lentil lines for seven isozyme loci. Using these loci, the 63 lines were classified into 52 different electrophoretic patterns. A one-way analysis of variance was used to study the association of the allelic state at the isozyme loci with host plant resistance (Table 2.1.6). Among the loci studied, there were highly significant associations for Phosphoglucomutase-2 (PGM-2) and 6-Phosphogluconate dehydrogenase-2 (6PGD-2) with wilt resistance (< 20 % wilted plants) related to the fast allele (band a) at both loci. It now remains to test this association in segregating populations to determine if it is caused by a linkage that may be exploited in marker-assisted selection.

Table 2.1.6. The means and standard errors (SE) for each isozyme allele of 63 lines of lentil for disease incidence percentage measured in the field in 1994 and 1995 and disease severity from 1-9 in Hoaglands semi-solid medium in tubes in the laboratory. Means followed by the same letter are not significantly different based on Duncan's multiple range test from one-way analyses of variance by isozyme allele.

		Field 1993		Field 1994		Lab. (Hoaglands)	
Isozyme	Band	Mean	SE	Mean	SE	Mean	SE
	b	37.0b	4.6	37.5b	4.5	5.6b	0.4
	c	76.1c	3.7	77.5c	3.7	7.4c	0.2
AAT-3	a	30.2a	6.6	30.7a	7.0	4.9a	0.5
	b	40.7a	7.7	40.4a	7.8	5.4a	0.6
	c	37.8a	6.8	38.6a	6.7	5.3a	0.5
ACO-2	a	29.8a	4.0	30.0a	4.0	4.8a	0.3
	b	61.3a	9.1	61.1a	9.1	7.1b	0.4
	c	39.0b	16.2	40.9b	16.8	4.7b	1.2
ACP-1	a	23.2a	4.6	24.3a	5.0	4.6a	0.5
	b	35.0b	5.7	34.9b	5.7	5.0b	0.4
	c	62.5b	8.2	63.0b	8.2	6.9b	0.5
LAP-2	a	31.7a	5.0	31.8a	4.9	4.9a	0.4
	b	53.8b	7.7	54.1b	8.0	6.5b	0.1
	c	30.8b	8.9	32.0b	9.6	4.9b	0.8
6PGD-2	a	19.4a	3.6	18.9a	3.6	4.2a	0.4
	b	57.6b	5.3	58.9b	5.2	6.5b	0.3
ME-1	a	30.9a	5.7	31.3a	5.8	4.8a	0.4
	b	44.1a	5.2	44.6a	5.2	5.8a	0.4

* PGM-2 = Phosphoglucomutase-2; AAT-3 = Aspartate aminotransferase-3; ACO-2 = Aconitase-2; ACP-1 = Acid phosphatase-1; LAP-2 = Leucine aminopeptidase-2; 6PGD-2 = 6-Phosphogluconate dehydrogenase-2; ME-1 = Malic enzyme-1.

2.1.1.4.5. Inheritance of resistance to lentil vascular wilt

In the only reference to the genetics of resistance to lentil vascular wilt[2], it is reported that, in India, the inheritance was controlled by five independently segregating genes, based on the reaction of individual plants. We investigated the inheritance of resistance to wilt using four parents in five cross combinations. Parental disease ratings are shown in Table 2.1.7, highlighting three different types of disease reaction: ILL 6409 highly resistant, ILL 6991 and ILL 6976 moderately resistant and ILL 4605 highly susceptible.

Plants, individually randomized, of the F_1 and F_2 generations of the crosses were examined in the plastic house in trays under artificial inoculation (Experiment 1). In Experiment 2, F_2-derived F_3 progeny rows were grown under similar conditions in a replicated trial and data were collected on individual plants and averaged over the row. An examination of the parental disease scores (Table 2.1.7) indicates that individual plants of the highly susceptible line ILL 4605 were rated 7 or 9 and plants of the moderately resistant parents were rated 3 and 5. So we classified plants or rows with a score of ≤ 5 as resistant and plants or rows with a score of > 5 as susceptible.

The results of the F_2-derived F_3 generation progeny test showed a segregation pattern among progenies of 3 resistant to 1 susceptible for the three crosses involving the susceptible parent (Table 2.1.8). This is indicative of a single recessive gene for susceptibility. The results of the F_2-generation contrasted with those of the F_3 progenies, because the F_2 data of the same crosses did not fit the 3:1 ratio well (data not shown) as there was an excess of resistant over susceptible plants. The distinction between the moderately resistant parents and the susceptible parent was small on an individual plant basis, as was the case with the individual F_2 plants in Experiment 1. This opens the possibility of some individual plants being mis-classified for disease reaction. In Experiment 2, although data was collected on all plants, the experimental unit was a row with its score calculated as the mean, leading to a much lower possibility of mis-classification of disease reaction.

Kamboj, R.K., Pandey, R.K. and Chaube, H.S. (1990) Inheritance of resistance to fusarium wilt in Indian lentil germplasm (*Lens culinaris* Medik). *Euphytica* 50(2): 113-117.

Consequently row data, such as that of the F_2-derived F_3 generation progenies, are considered as reliable, whereas individual plant data is considered problematic.

Two of the five crosses were between the highly resistant parent and a moderately resistant line. Such differences in the level of resistance of the parents were significant at $P = 0.05$. The F_2-derived F_3 generation progenies of these crosses were all rated < 5 and consequently as resistant. The broad-sense heritability of the resistance was 88.7 % in Cross 4 and 84.1 % in Cross 5. This shows that although susceptibility is controlled by a single recessive major gene, there are also significant differences in the level of resistance and such differences are controlled by many genes of small effect. The relatively high heritability values suggest that some response to selection among the resistant types for a high level of resistance may be expected.

F_2-derived F_4 progeny rows of two other crosses (Cross 1: L692-15-8 (F) x L92-17-16 and Cross 2: L92-17-2 x L692-16-1(S)) of lentil between wilt resistant and susceptible parents were examined in ICARDA's wilt-sick plot in three replicates.

In the field, wilt incidence was assessed as the percentage of wilted/dead plants in a plot (row of 40 plants). The resistant parents were both rated as 3.3 % wilted plants with a standard error of ± 12.0 % in Cross 1 and a standard error of 16.0 % in Cross 2. The susceptible parents were both rated 58.3 % wilted plants with the above standard errors. There was a discontinuous distribution of the F_2-derived F_4 progenies with no progenies rated in Cross 1 between 0 and 40 % and in Cross 2 between 33 and 40 %. In Cross 1 there were 32 progenies rated < 30 % wilted plants (resistant) and 10 progenies rated > 40 % wilted (susceptible) and in Cross 2 there were 65 progenies rated resistant and 21 progenies rated susceptible. The segregation patterns of both crosses fitted a 3:1 ratio of resistance to susceptibility (Cross 1 & 2: $\chi^2 = 0$), confirmation of a single recessive gene for susceptibility.

B. Bayaa, A. Abbas (Aleppo University), I. Eujayl and W. Erskine.

Table 2.1.7. Frequency of rating scores of the parents on a single plant basis in Experiment 1 as grown with the F_1 & F_2 generations and on a row basis in Experiment 2 as grown with the F_3 generation.

Parent	Experiment 1 with F_1 & F_2						Experiment 2 with F_3				
	1*	3	5	7	9	Mean	1-3	>3-5	>5-7	>7-9	Mean
ILL 6409	36	14	-	-	-	1.5	30	-	-	-	1.2
ILL 6976	-	5	31	-	-	4.7	-	18	2	-	4.2
ILL 6991	-	3	36	-	-	4.8	-	20	-	-	4.0
ILL 4605	-	-	-	5	54	8.8	-	-	-	30	8.1

* Rating scale: 1 = No symptom/healthy to 9 = Dead/wilted plant.

Table 2.1.8. Number of F_2 progenies in five crosses from a replicated trial classified according to their disease reaction. Rating scale: 1 = No symptom/healthy to 9 = Dead/wilted plant with resistance rated 1-5 and susceptibility > 5.

Cross	Parentage	F_2 progeny rating		Expected ratio	$\chi 2$	P
		Res.	Susc.			
1	ILL 6409 x ILL 4605	32	9	3:1	0.20	0.500-0.750
2	ILL 4605 x ILL 6976	30	19	3:1	4.94	0.025-0.05
3	ILL 4605 x ILL 6991	71	21	3:1	0.22	0.500-0.750
	Total (Crosses 1-3)	133	49	3:1	0.35	0.500-0.750
4	ILL 6409 x ILL 6976	50	0	-	-	
5	ILL 6409 x ILL 6991	57	0	-	-	

* Rating scale: 1 = No symptom/healthy to 9 = Dead/wilted plant.

2.1.1.5 Effects of fungicide on lentil wet root rot

Wet root rot caused by *Rhizoctonia solani* Kuhn is a soil-borne fungus that affects lentil in most lentil-growing regions of the world. It is of economic importance in parts of India, Ethiopia, and both Columbia and Ecuador, where it caused a dramatic reduction of the lentil-growing area[3]. It also caused a failure of the ICARDA lentil summer nursery in Lebanon.

Resistance to *R. solani* in lentil germplasm was researched without success[4]. This study was conducted to determine both the minimum lethal dose of fungicide Tolcophos methyl ('Rizolex 50 WP') to control *in vitro* growth of *R. solani* and the effect of seed treatment on pre- and post-emergence damping-off and yield. Seed treatment may form one component of integrated management of the disease.

To study the effect of Tolcophos methyl on *in vitro* growth of *R. solani* two experiments were conducted. The first employed the method of poisoned media using six rates of the fungicide Tolcophos methyl from 0 - 1.0 g ai product/l and Benomyl at 1.0 g ai product/l in three replicates. The second examined the effect of dressing with Tolcophos methyl at four rates (0.0, 0.25, 0.5, 1.0 g ai/kg seeds) and with Benomyl at 1.0 g ai product/l the seeds of three cultivars (ILL 2126, ILL 5582 & ILL 5883) on *in vitro* fungal growth in three replicates. Inoculum was placed at the centre of each petri dish and the seeds around the periphery.

In the poisoned media experiment, fungal growth covered the surface (diameter: 90 mm) of the unpoisoned, control petri dishes within four days; by which time with 0.125 g ai Tolcophos methyl/l, the lowest rate examined, the diameter of the fungal colony was only 6 mm. The treatment with Benomyl was similar to the untreated

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Abawi, G. (1987) Pudriciones radiculares en leguminosas de grano. In: *Informe de consultoria, IICA-BID-PROCIANDINO*, Quito, Ecuador, pp. 8-11.

Durate, R. (1989) Avaliacao de germoplasma de lentilha para resistancia a *Rhizoctonia solani*. *Relatorio de Atividades PIEP, CNPH, EMBRAPA*, Brasiliana, Brasil 30 pp.

control and it did not affect fungal growth.

Table 2.1.9. *The effect of concentration of Tolcophos* methyl seed dressing on seed germination (%) and above-ground biological yield (g/pot) on three cultivars of lentil sown in pots artificially inoculated with *Rhizoctonia solani* in the plastic house.

	Germination %			
	Conc. fungicide dressing (g ai product/kg seed)			
Cultivar (ILL)	0	0.25	0.5	1.0
2126	15.5	100	100	93.3
5582	51.1	97.8	100	97.8
5883	31.1	100	97.8	97.8
$SE_{Cult.\ x\ Conc.}$	4.8			
	Above-ground biomass (g/pot)			
Average over cultivars	3.1	13.6	13.7	12.4
$SE_{conc.}$	1.09			

Seed dressing reduced *in vitro* fungal growth deforming it into a star shape, indicating that Tolcophos methyl has an inhibitory effect beyond the rhizoplane. The extent of inhibition varied inversely with the concentration of fungicide dressing and among the cultivars. There was no obvious phytotoxicity on seed germination. Additionally, an examination was made of the effect of Tolcophos methyl on seed-borne, saprophytic organisms. The following genera were unaffected by the fungicide: *Alternaria* spp., *Aspergillus* spp., *Penicillium* spp., *Fusarium* spp., *Cladosporium* spp., *Rhizopus* spp. and a number of bacteria.

A pot experiment was conducted in the plastic house to study the effect of the above four concentrations of Tolcophos methyl seed dressing using the same three cultivars on pre- and post-emergence damping-off and yield in three replicates with 15 seeds/pot.

Both germination % and biological yield were significantly improved by seed dressing with Tolcophos methyl with no differences observed between the fungicidal concentrations (Table 2.1.9). In the absence of seed dressing, wet root rot reduced germination % more in the local cultivar ILL 2126 than in the other cultivars.

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2.1.1.6. Breeding for Adaptation to Dry Mediterranean Environments through Drought Avoidance

In the Mediterranean region lentil is usually sown in the zone with between 300 and 400 mm annual rainfall. For adaptation to even drier areas, drought escape through early flowering and maturity has been clearly identified as the key mechanism[5]. Early material is already being produced in the Southern latitudes stream of the breeding program and this same material is now being exploited in another direction i.e. for arid Mediterranean environments. In the 1993/94 season we launched a new international screening nursery targeted toward arid Mediterranean conditions of the best early flowering/drought-avoiding lines already in the breeding program (see section 2.1.1.3).

The next logical step was to stratify the breeding program for West Asia according to flowering/maturity. Before undertaking this process, an examination of the relationship of flowering/maturity with seed yield was warranted across a wide range of environments. Each year we test in yield trials many lines across a rainfall gradient from ca 260 to 550 mm (Section 2.1.1.2). There is, thus, data available of the relationship between seed yield, on the one hand, and the time to flowering and maturity, on the other hand. Data were examined from yield trials grown at Breda, Tel Hadya and Terbol in the last three seasons (1992/93, 1993/94, 1994/95) and the means of the relevant phenotypic correlations of at least five experiments from each trial/environment were calculated. These are presented in Figure 2.1.3. together with

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Silim, S.N., Saxena, M.C. and Erskine, W. 1993. Adaptation of lentil to the Mediterranean environment. I. Factors affecting yield under drought conditions. *Experimental Agriculture* 29:9-19.

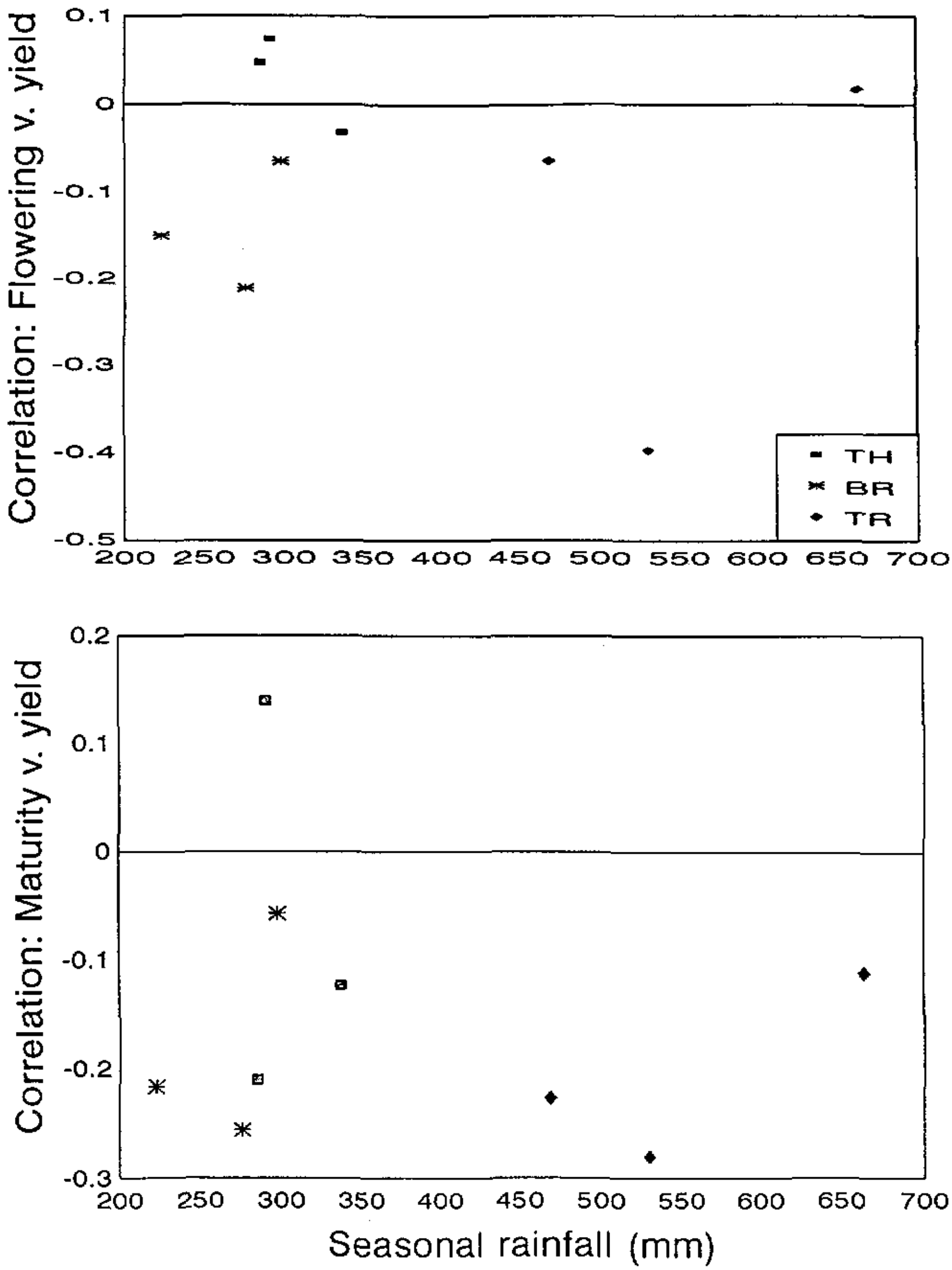

Figure 2.1.3. Graph of seasonal rainfall totals (mm) at the trial sites Breda (BR), Tel Hadya (TH) and Terbol (TR) in the 1992/93, 1993/94 and 1994/95 seasons against the average of the phenotypic correlation between seed yield (kg/ha) and time to flowering (upper graph) and maturity (lower graph) over at least five trials at each trial-season.

associated seasonal rainfall totals.It is clear that there is generally a weak negative correlation between seed yield (kg/ha) and time to both flowering and maturity, and high yields derive from early lines. However, it is equally clear that this relationship is independent of the seasonal rainfall and is not specific to the dry areas. Plainly, yield-testing for West Asia in a wide spectrum of rainfall regimes is warranted, but it is not appropriate to stratify the breeding program into a specific early-flowering stream for the dry areas.

A new project entitled 'Improvement of drought and disease resistance in lentils from the Indian sub-continent' has now been approved for funding by the Australian Centre for International Agricultural Research (ACIAR). Our partners in the project are the national programs of Nepal and Pakistan, the Victorian Institute of Dryland Agriculture and the Cooperative Research Centre for Legumes in Mediterranean Agriculture. The project will address aspects of drought in lentil.

W. Erskine

2.1.1.7. Screening for Winter Hardiness

Lentil is currently sown in spring in Turkey at elevations above approximately 850 m elevation on ca 250,000 ha. Research in Turkey has indicated that yields may be increased by up to 50 % by early sowing in late autumn with winter hardy cultivars[6]. However, the use of such cultivars is not yet widespread in Turkey, because at high elevation the level of winter hardiness is inadequate in cold winters and winter hardiness has not yet been transferred from germplasm sources into acceptable cultivars.

A major program to recombine yield with the necessary winter hardiness is underway at The Central Research Institute for Field Crops in Turkey through field

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Sakar, D., Durutan, N. and Meyveci, K. 1988. Factors which limit the productivity of cool season food legumes in Turkey. In: World Crops: Cool Season Food Legumes, (R.J. Summerfield, ed.), Kluwer, the Netherlands, pp. 137-146.

screening at Hymana. Crosses and early generation material are being produced by the ICARDA base program in Aleppo, Syria and segregating populations are sent to Hymana for selection under severe winter conditions (see section 2.1.1.1).

In the 1994/95 winter season at Hymana, Turkey the absolute minimum temperature was -16 ^{0}C. This provided good conditions to screen for winter-hardiness and cold-susceptible checks were killed in all the winter trials. There was considerable useful variability apparent in winter yield trials of 325 landrace selections from S.E. Anatolia. Separate yield trials of both red and yellow cotyledon selections from ICARDA-supplied segregating populations also yielded well under the cold winter conditions. The spring crop of 1995 yielded very poorly (<0.5 t/ha) at Hymana. Sowing was late because of a wet April, but May was very dry.

Next season on-farm evaluation of the best winter lines will be initiated in the Anatolian plateau.

As part of the thrust on winter highland lentils, a workshop was held on 12-13 December 1994 at Antalya, Turkey on 'Towards Improved Winter-sown Lentil Production for the West Asian and North African Highlands'. The workshop was designed to disseminate the findings of a two-year collaborative research programme implemented by the Central Research Institute for Field Crops, Ankara, ICARDA and The University of Reading, which investigated progress in agronomic and breeding issues of winter-hardy lentils and to postulate the potential value of autumn-sowing winter-hardy lines elsewhere in the highlands of West Asia and North Africa. Speakers came from Algeria, Iran, Morocco, Tunisia, Turkey, UK and USA, in addition to ICARDA. The proceedings have been published. The research programme was partly supported by a grant from the Overseas Development Administration, U.K.

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2.1.1.8. Exploitation of Wild Lentils for Yield Improvement

Soon after the foundation of ICARDA a few exploratory crosses were made between the cultivated lentil and its putative wild progenitor *L. culinaris* ssp. *orientalis*. In 1984 we selected 10 lines for distribution worldwide in the Lentil International Nursery Program from bulk segregating populations of these wild x cultivated crosses. Among these selections containing genes introgressed from the wild, the small-seeded ILL 5700 ranked 3rd, 1st, 7th and 2nd for average yield among 24 entries of the Lentil International Yield Trial tested in 13-15 countries from 1985 to 1988. This selection with wild parentage has since been widely used in crossing to introgress wild genes into the cultivated plant. Recently, further crosses of wild x cultivated lentil have been made to study more systematically the agronomic potential of this wide hybridization.

A line x tester mating scheme was used with three diverse cultivated lines (ILL 2501 - ex India; ILL 5582 & ILL 5674 - ICARDA) and seven male parents of *L. culinaris* ssp. *orientalis*. To ensure diversity in the wild parental population, we selected from the most diverse origins possible within the distribution of the wild material vis. Cyprus, Jordan, Syria, Turkey & former USSR. In the 1992/93 season, the parents and crosses in the F_2 generation were grown at Tel Hadya, Syria and, in the following season and generation, they were studied at the University of Amman, Jordan (in a MSc thesis).

The experiments of parents and crosses in the F_2 design with three replications. Plots were separated by a uniform border row and nylon mesh was placed on the soil to catch dehisced seed.

Biomass yields of the cultivated parents were similar over the seasons (Table 2.1.10). But the wild parents varied in yield over the seasons from 4 to 98 g m^{-2} in 1993 and from 106 to 168 g m^{-2} in 1994. The first season showed a wide gulf between the yields of wild and cultivated parents, whereas in the second season the cultivated parent from India was at par for biological yield with some of the wild parents.

Table 2.2.10. Biomass means (gm^{-2}) of parents, with their general combining abilities (GCA), and crosses of a line x tester mating system of three cultivated lentil lines and seven *L. culinaris* ssp. *orientalis* testers grown in the F_2 generation at ICARDA Tel Hadya in the 1992/93 season and in the F_3 generation at the University of Jordan in 1993/94. Accession numbers of cultivated (ILL) and wild (ILWL) accessions are given.

Testers (ILWL)	Lines						Parent mean		GCA	
	ILL 2501		ILL 5582		ILL 5674					
	1993	1994	1993	1994	1993	1994	1993	1994	1993	1994
1	165	163		71	179	118	280	13	120	-3614
11	141	191		268	200	242	224	26	106	6312
80	217	163		58	184	149	111	11	140	-14-16
89	212	174		145	225	82	184	70	168	-88
120	202	170		153	182	168	232	98	129	212
177	181	144		65	243	144	204	8	159	-244
325	204	154		77	154	173	202	4	150	-3-23
Parent mean	151	162		343	350	293	276			
GCA	35	-28		-35	2	0	25			

SED treatment means 47.8 g m^{-2} in F_2, 72.0 g m^{-2} in F_3
SE line GCA values 13.2 g m^{-2} in F_2, 8.3 gm $^{-2}$ in F_3
SE tester GCA values 28.6 g m^{-2} in F_2, 12.7 g m^{-2} in F_3

There was significant heterosis above the mid-parent in both the F_2 and F_3 generations for biomass. With substantial heterosis (deviations from the model of the mid-parent mean) for biomass, specific combining ability (SCA) was of greater importance than general combining ability, indicating the importance of non-additive gene effects.

These segregating populations are being advanced by the bulk method and single plant selections will be made within them at the F_4 generation. Meanwhile a comprehensive report on the performance of the line x tester cross is in preparation.

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2.1.1.9. Relationship of pod numbers per peduncle with grain yield

The number of flowers per inflorescence varies considerably in lentil from one flower per peduncle to four flowers per peduncle, with up to seven recorded from the glass-house. This results in different numbers of pods per peduncle at maturity. There is variation within an individual plant in flower number per inflorescence, between plants within a genotype, between genotypes and there is also a strong environmental effect on this trait. This MSc thesis at the American University of Beirut aimed to quantify the variation due to these different factors and to understand the significance of this variability to final yield.

Given the many sources of variation in flower number per inflorescence, the emphasis in the first season of experimentation was to understand the floral 'population' biology of lentil. The nodal position, date of opening and subsequent fate (flower or pod abortion) of every flower was studied in the plastic house and the field on two genotypes and sowing dates to provide an overall picture of lentil floral biology (Legume Program Annual Report 1993).

It was found impractical to track the fate of every flower on many genotypes. An assessment was made of several sampling strategies to estimate the average number of pods per peduncle per plot on different genotypes sown in different environments, using the data from the first season. Such strategies included the use of the main stem, the basal primary branch, the second primary branch and the two basal primaries together. The best sampling method utilized the two basal primary branches of five plants per plot.

In the following season 81 genotypes, representing a wide range of diversity in the lentil, were sown in a yield trial at the American University Farm, Beqaa, Lebanon and at Tel Hadya, Syria in a 9x9 lattice design with four replications.

In Lebanon the overall seed yield was low at 867 kg/ha, primarily because of a period of hot weather in mid-April, coincident with the early podding growth stage. Empty pods contributed 53 % of the overall number of pods and the average number of seeds per pods was also low at 0.57. Both these traits reflect a high level of seed abortion and a reduction in yield potential resulting from the stress conditions in the early podding growth stage.

By contrast, the average seed yield was 2076 kg/ha in Syria. Highlighting the genetic variation in the trial, time to maturity varied from 117 to 137 days and 100 seed weight varied from 1.74 to 6.83 g/100 seeds at Tel Hadya.

For the number of pods per peduncle, there were major differences between genotypes, a large location effect and a significant genotype x location interaction. The average number of pods per peduncle was 2.07 in Lebanon, whereas it was 2.4 in Syria. The genotypic range for the trait was 1.9 to 2.9 pods per peduncle in Syria and 1.5 to 2.5 pods per peduncle in Lebanon.

The broad-sense heritability (h^2) of pod number per peduncle was 0.68, lower than that for average seed weight but similar in magnitude to that for the other yield components - seed numbers per pod and pod numbers per plant (Table 2.1.11). Phenotypic correlations were calculated among yield and its components (Table 2.1.11). Grain yield (kg/ha) was positively correlated with pod number per peduncle at $P < 0.05$ in both environments.

Table 2.1.11. Phenotypic correlation coefficients between pod number per peduncle, yield components and seed yield, together with their broad-sense heritabilities (h^2), of 81 lentil lines at American University of Beirut Farm, Lebanon (L) and Tel Hadya, Syria (S) in the 1993/94 season.

Character	Country	1	2	3	4	5	h^2
Pods/peduncle	L	-0.52***	0.31**	0.44***	0.32**	0.25*	0.68
	S	-0.58***	0.45***	0.57***	0.25*	0.29**	
Seeds/pod (1)	L		-0.51***	-0.76***	-0.38***	-0.10	0.66
	S		0.35**	-0.70***	-0.64***	-0.13	
Pods/plant (2)	L			0.89***	-0.14	0.00	0.71
	S			0.86***	-0.08	0.03	
Empty pods/plant (3)	L				0.06	-0.01	0.80
	S				0.29**	0.00	
100 seed weight (4)	L					0.52***	0.87
	S					0.07	
Seed yield (5)						1.00	0.28

* < 0.05 P > 0.01, ** < 0.01 P > 0.001, *** P < 0.001

Additionally, pod number per peduncle was positively correlated with the yield components - 100 seed weight and both the total number of pods and the number of empty pods. However, it was negatively correlated to the number of seeds per pod, indicating yield component compensation. The potential for yield is laid down sequentially first through the number of potential inflorescences, then through the number of flowers per inflorescence, then through the number of seeds per pod and finally through average seed size. The timing of a stress determines which of these processes is affected and compensation between the yield components may occur at any stage. With a heritability for the number of pods per peduncle of $h^2 = 0.68$, a response to selection among genotypes for this trait may be confidently expected. However, it will probably not result in a major, correlated gain in grain yield (kg/ha), because of yield component compensation. Among yield components, the only important trait to a practical breeder is average seed weight, because of its importance in end-use quality.

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2.1.2. Use of lentil germplasm by NARSs

2.1.2.1. Advances for the Mediterranean region

The ICARDA base program provides segregating populations and breeding lines to national programs in North Africa and West Asia for elevations below c. 850 m around the Mediterranean Sea. To date, more use has been made by NARSs of lines than segregating populations and few lentil crosses are made outside ICARDA in North Africa and West Asia. Table 2.1.12. lists lentil lines released as cultivars and Table 2.1.13. gives those lines selected for pre-release multiplication and/or on-farm trials by NARSs.

In Syria the red-cotyledon line ILL 5883 will soon be submitted to the variety release committee following its testing in on-farm trials over the last five years, where it yielded significantly more grain than the local check in all geographic regions and rainfall zones.

Additionally, it has improved standing ability for harvest mechanization over the local check and resistance to vascular wilt disease, the most important disease of lentil in Syria. The spread in Syria of the earlier registered line Idlib 1, which has good standing ability and yield, is currently being monitored through an impact study on lentil harvest mechanization being conducted jointly with the Syrian General Organization of Agricultural Mechanization and the American University of Beirut. In Lebanon preliminary results from an adoption study indicate that Talya 2 is starting to spread in the Beqaa valley and that yellow cotyledon is the preferred seed type in southern Lebanon. Accordingly, FLIP 86-2L (ILL 5988), a yellow cotyledon line out-yielding Talya 2 in on-farm trials, was sown in a demonstration in South Lebanon in the 1994/95 season. Seed yields of 2000 kg/ha were realized. Farmers were impressed by the performance of FLIP 86-2L in terms of yield, seed size and colour and lodging - better than the local. A study of farmers' perspectives on the new variety concluded that there is a great potential for acceptability and adoption in the South and other lentil producing areas where large/white seed is preferred. In South-East Turkey, where winter red lentil is widely grown, ILL 1939 is in regional registration trials. Unfortunately it was not possible to conduct the necessary trials on farmers' fields last season in S.E. Anatolia. In Iraq the large-seeded, yellow cotyledon line 78S26002 was registered in 1992 as Baraka. The red cotyledon lines ILL 5883 and FLIP87-56L (ILL 6246) will be tested in on-farm trials in Iraq in the 1995/96 season as they performed well in the last two seasons and there is a demand from Iraqi consumers for both red and yellow cotyledon lentil. The food legume area in Iraq is increasing rapidly as a result of the increased food demand of the current situation. A lentil adoption study will be mounted in Iraq in the forthcoming season. In North Africa, there was a drought in Tunis in the 1994/95 season and despite the good yield of lentil in the previous droughty season lentil yields were low. In Libya the line El Safsaf 3 (78S26002), released in 1993 for cultivation for the East of the country, continues to perform well in the East but also has given high yields under central-pivot, irrigated conditions in Central Libya at Meknosa. Lentil production and area continue to decline in Algeria but the lines ILL 468, ILL 4400, LB Redjas, Setif 618 and Balkan 755 are in seed production for future use by farmers. In Morocco there was a drought in the 1994/95 season. However, there are several lentil lines in cataloque trials FLIP86-15L (ILL 6001), FLIP86-16L (ILL 6002), FLIP86-19L (ILL 6005), FLIP86-21L (ILL 6007), FLIP87-19L (ILL 6209) and FLIP87-22L (ILL 6212), all with resistance to rust.

The North African Regional yield trial on lentil was established in 1990 to comprise the best lines selected in Algeria, Libya, Morocco and Tunisia. This regional yield trial has revealed that lentils selected in the various countries of N. Africa differ substantially in phenology, indicating the need for specific adaptation to a range of environments in the region. The requirements of Libya, Tunisia, and lowland Algeria are being met by the ICARDA West Asian breeding program. However, late maturing material is required for high altitude areas of Algeria and specific crosses are being made at ICARDA for this area. In Morocco early-maturing lines are required with resistance to rust. A specific joint program of crossing is being undertaken to target this environment. In Tunisia late sowing with early maturing lines needs further testing.

In Egypt the early lines FLIP84-51L (ILL 5722) with small seeds and FLIP84-112L (ILL 5782) with large seeds are both in pre-release multiplication. The line Precoz (ILL 4605) is becoming popular in the north Sinai region, where it is known commonly as 'Shami' (from Damascus), because its early maturity avoids drought stress under the low prevailing rainfall conditions. These three early lines all have potential for inclusion in the cotton rotation in the Nile Delta region, in contrast to later maturing landraces.

National Agricultural Research Systems.

Table 2.1.12. Lentil cultivars released by national programs

Country	Cultivar name	Year of release	Specific features
Algeria	Syrie 229	1987	Yield, seed quality
	Balkan 755	1988	Yield, seed quality
	ILL 4400	1988	Yield, seed quality
Argentina	Arbolito (ILL 4605x-4349)	1991	Yield, tall & early
Australia	Aldinga (FLIP84-80L)	1989	Yield
	Digger (FLIP84-51L)	1993	Yield, red cotyledon
	Cobber (FLIP84-58L)	1993	Yield, red cotyledon
	Matilda (FLIP84-154L)	1993	Yield, yellow cotyledon
	Northfield (78S 26013)	1995	High yield, Ascochyta blight res.
Bangladesh	Falguni - BARI Masur 2 (Sel.ILL4353xILL353)	1993	Rust res. & yield
	BARI Masur 3 (Sel. L5 x FLIP84-112L)	1995	Rust and Stemphylium resistance
Canada	Indianhead (ILL 481)	1989	Green manure
Chile	Centinela (74TA470)	1989	Rust res.& yield
China	FLIP87-53L	1988	Yield in Qinghai Province
Ecuador	INIAP-406 (FLIP 84-94L)	1987	Rust res. & yield
Egypt	Precoz (ILL 4605)	1990	Intercropping in sugarcane
Ethiopia	R 186	1980	Yield
	Calew (ILL 358)	1984	Rust res. & yield
	Chikol (NEL 2704)	1994	Rust res. & yield
	FLIP84-7L	1994	Rust resistant, high yield
	Gudo (FLIP84-78L)	1995	Rust resistant, high yield
	Ada'a (FLIP86-41L)	1995	Rust resistant, high yield
Iraq	Baraka (78S26002)	1994	Yield, standing ability
Jordan	Jordan 3 (78S 26002)	1990	Yield, standing ability
Lebanon	Talya 2 (78S 26013)	1988	Yield, standing ability
	Toula (FLIP86-2L)	1995	High yield, large seeds
Libya	El Safsaf 3 (78S26002)	1993	Yield, st. ability in E. Libya
Morocco	Precoz (ILL 4605)	1990	Rust res. & yield
Nepal	Sikhar (ILL 4402)	1989	Yield
N Zealand	Rajah (FLIP87-53L)	1992	Yield, red cotyledon
Pakistan	Manserha 89 (ILL 4605)	1990	Ascochyta & rust res.
Sudan	Aribo 1 (ILL 818)	1993	Yield in Jebel Mara
Syria	Idleb 1 (78S 26002)	1987	Yield, reduced lodging
Tunisia	Nsir (ILL 4400)	1986	Large seeds & yield
	Nefza (ILL 4606)	1986	Large seeds & yield
Turkey	Firat 87 (75Kf 36062)	1987	Small seeds & yield
	Erzurum 89 (ILL 942)	1990	Spring sowing & yield
	Malazgirt89 (ILL 1384)	1990	Spring sowing & yield
	Sazak 91 (NEL 854)	1991	Winter sowing, red cotyledon
U.S.A.	Crimson (ILL 784)	1991	Yield in dry areas

Table 2.1.13. Lentil lines in pre-release multiplication or on-farm testing by NARSs.

Mediterranean region	
Algeria	ILL 468
Egypt	FLIP84-51L, FLIP84-112L
Iraq	ILL 5883, FLIP87-56L
Lebanon	ILL 2126, FLIP84-59L, FLIP87-56L
Morocco	FLIP86-15L, FLIP86-16L, FLIP87-19L,FLIP87-22L
Syria	ILL 5883, ILL 7012
Tunisia	78S26002, FLIP 84-58L,
Turkey	ILL 1939
High elevation	
Iran	ILL 842, ILL 949
Pakistan	FLIP84-4L, FLIP85-7L
S. Latitudes	
Ethiopia	FLIP87-74L
Nepal	ILL 2580
Sudan	ILL 813, FLIP88-43L
Yemen	ILL 4605, FLIP84-14L

2.1.2.2. Advances for southern latitude region This region

comprises the sub-continent of India and Ethiopia where an early flowering habit is required together with resistance to rust, ascochyta blight and wilt. The importance of foliar pathogens contrasts with other major areas of lentil production.

There are three strong lentil breeding programs in Pakistan with two in Faisalabad and the remaining program in Islamabad. Over the last five years ICARDA has worked closely with these programs in joint selection as the focus of a thrust to broaden the genetic base of lentils in South Asia. In Faisalabad a line 88503 has been released as Masur 95 with ICARDA parentage by Punjab authorities and the mutation breeding program at NIAB is based on ICARDA supplied material (see section 2.1.1.11). Three relatively salt tolerant lines - ILL 6451, ILL 6788 and ILL 6793 were identified at Bahauddin Zakariya University, Multan.

The major production problems in Bangladesh addressable through breeding are rust and Stemphylium blight. We have been making targeted crosses for Bangladesh of rust resistance sources with the local susceptible cultivar 'L5' in the base program at Tel Hadya. Selections have now been made in Bangladesh of adapted rust resistant plants from segregating populations. As a result, Falguni (BARI Masur 2) was released in 1993 as the first rust resistant lentil cultivar in Bangladesh. Another rust-resistant line (ILX 87247), locally selected from the cross of L5 x FLIP84-112L (ILL 5782), was recently released as BARI Masur 3. It also has resistance to stemphylium blight and an erect plant stature suitable for inter-cropping in sugarcane.

India has a strong lentil breeding program coordinated under the All India Coordinated Pulse Improvement Project of the Indian Council of Agricultural Research (ICAR). In India, the genetic base of the crop has been widened by the use of Precoz in crossing. Now 15% of entries in All-India Coordinated Lentil trials have Precoz as a parent. Rust resistance, selected in Morocco, was holding in Kanpur. We have established cooperation with Pantnagar Agricultural University on screening for rust resistance in breeding lines, the wild germplasm and the possibility of collaboration in the search for markers for rust resistance. Our vascular wilt resistance lines are being widely used as source parents within India.

Nepal grew around 170,000 ha of lentil spread from the Terai area adjacent to India to the lower Mid-Hills last season. ICARDA has been requested for specific targeted crosses by Nepali program. ILL 2580 is among entries being considered for release.

Bilateral interaction - ICARDA directly with the NARSs of S. Asia - has been strong in the fields of the exchange of germplasm and in the development of tailored breeding material. The value to NARSs of such bilateral interaction has fuelled the felt need for more support to regional activities on lentil improvement. At an ICARDA/ICAR sponsored seminar on 'Lentil in S. Asia' held in Delhi in 1991, participants from S. Asia were enthusiastic about the need and value of a regional network on lentil improvement and its potential for the development of the crop in their individual countries.

ICARDA has been trying to secure funding for such a regional initiative on lentil. Project submissions were made to the Asian Development Bank (ADB), Philippines and to Der Bundesminister für Wirtschaftliche Zusammenarbeit (BMZ), Germany without success. Recently, we were catalytic in securing funding for a project entitled 'Improvement of drought and disease resistance in lentils from the Indian Sub-continent' from the Australian Centre for International Agricultural Research (ACIAR). However, despite its name, the project only covers the countries of Nepal and Pakistan and is limited in scope.

It is in the context of the development of a network on lentil that the value of cooperation with ICRISAT, as a partner becomes apparent. The Cereals and Legumes Asia Network (CLAN) of ICRISAT is addressing other grain legumes such as chickpea, pigeon pea and groundnut. National grain legume teams are engaged in the improvement of the 'ICRISAT' mandate legumes as well as lentil, with staff commonly addressing all or many of the species simultaneously. It is technically and administratively efficient for both NARS and IARCs that a sub-network on lentil is formed in CLAN of ICRISAT. Additionally, donors are now pressing for increased cooperation among IARCs. At present we are submitting a revised regional project (ICARDA+ICRISAT) on lentil to ADB.

In Ethiopia Gudo = FLIP84-78L (ILL 5748) and Ada'a = FLIP86-41L (ILL 6027) were registered in 1995. Ada and Akaki are the areas where the released line NEL 358 is becoming very popular and a study on its impact is underway.

In Sudan Rubatab 1 (ILL 818) was released for cultivation in the Northern Province and Jebel Mara Region in the 1993/94 season on the basis of yields from 11-34 % more than the local check and good seed quality for splitting and dehulling. ILL 813 is in pre-release seed multiplication and the program identified FLIP88-43L (ILL 6467) as promising in the 1993/94 season in the Northern Province.

National Agricultural Research Systems.

2.1.2.3. Advances for high altitude region

The high altitude region primarily consists of those regions of Afghanistan, Iran, Pakistan and Turkey where lentil is normally grown as a spring crop because of the severe winter cold. This season at Ankara the national program of Turkey has again demonstrated that winter-sown lentil has a higher yield potential than the spring-sown crop. (see section 2.1.1.8). The spring cultivar Erzurum '89 was tested in farmer managed trials/demonstrations in the Sivas area on the eastern margin of Anatolian plateau. The mean yield of the local was 478 kg/ha compared to Erzurum '89, which yielded 738 kg/ha in the 1994 spring season. In Iran the lines ILL 842 and ILL 949 are promising and due for testing on farmers' fields.

In Balochistan (Pakistan) the Provincial Technical sub-committee gave its approval for the release of two lines Masoor-931 (FLIP84-4L) and Masoor-932 (FLIP85-7L) for highland Balochistan, subject to the approval of the Provincial Seed Council. The lines were selected at the Arid Zone Research Institute, Quetta, on the basis of their cold tolerance and a larger seed size than the local cultivar. The final release is pending.

National Agricultural Research Systems.

2.1.2.4. Advances in other areas

The New Zealand Institute for Crop and Food Research registered lentil FLIP87-53L (ILL 6243) as Rajah during 1993. It is a red cotyledon line which has out-performed the commercial standard by 15% and is well received by the lentil trade for use either whole or split. It can be harvested 2-3 weeks before the commercial standard and is more tolerant to Ascochyta blight.

In Australia there is now considerable interest in lentil. Prior to the testing of germplasm from ICARDA, lentil assessment in Australia was limited to a few lines representing phenological extremes - extra early and extra late flowering and maturity. ICARDA Mediterranean-adapted material has fitted in well into the vacuum. In Victoria the red-seeded cultivars Digger = FLIP84-51L (ILL 5722) and Cobber = FLIP84-58L (ILL 5728) and the green cultivar Matilda = FLIP84-154L (ILL 5823) are forming the basis of a viable lentil industry. Last season estimates were of a lentil crop of 8-10,000 tonnes a pool return of A$400. A four million dollar industry based on ICARDA germplasm. In New South Wales the lines FLIP84-51L (ILL 5722) and FLIP86-16L (ILL 6002) are in multi-location testing following their selection at Tamworth over several seasons.Northfield (78S 26013) as recently released in S. Australia. In Western Australia during 1994, the first commercial crops of lentil were grown. Twelve farmers grew a total of around 200 ha with average yields of 0.8 t/ha and a top yield of 1.8 t/ha from ICARDA derived lines.

In response to epidemics of lentil Ascochyta blight in the Canadian Province of Saskatoon, the Crop Development Centre, University of Saskatoon has released the first two resistant Canadian lentil cultivars; one of which, 'CDC Redwing', has resistance from an ICARDA parent (ILL 5588). Further East in Manitoba (Canada), the lentil crop suffers from Anthracnose blight. The only known resistant source to the disease is ICARDA germplasm ILL 481 = 'Indianhead'. This line is uses as a resistant source in lentil in Manitoba.

In the USA the variety Crimson (ILL 784) is now being grown on about 1000 ha in the Palouse area of Washington State. Markets have been slow to develop. Apparently the buyers want plumper seeds ('Turkish seed-type').

National Agricultural Research Systems.

2.2. Application of Molecular Techniques

2.2.1 Lentil Genetic Mapping

Breeding has received less attention in lentil than other major legumes and also the saturation of the linkage map in lentil is relatively low. Reviewing the mapping efforts in lentil a compiled linkage map has been produced based on combined linkage data and regions of homology shared with pea (*Pisum sativum* L.) including 7 morphological, 25 isozymes, 38 RFLP and 6 other loci[7]. Map construction using

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Tahir, M., C.J. Simon and F.J. Muehlbauer, 1993: Gene map of lentil: a review. LENS Newsletter 20:3-9.

RFLPs is time consuming and expensive. A promising alternative to the use of RFLP is through DNA amplification by polymerase chain reaction (PCR) using arbitrary primers. The establishment of PCR-technology is less costly than RFLP and it is more easily automated. Random Amplified Polymorphic DNA (RAPD) markers have already been used to tag disease resistances in many agricultural crops and together with RFLPs to construct genetic linkage maps. In the previous year we reported the use of RAPDs for genetic mapping and for testing of segregation distortion in F2 generation of potential mapping populations in later generations[89] (see Fig. 2.2.1). The same population has been used to develop recombinant inbred lines. The objective of this year's study was to assess the usefullness of RAPDs in building a comprehensive lentil genetic map in association with other types of markers.

Plant Material

An F2 population of the cross L92-013 (L962-15-8(F) x ILL5588) provided by Dr. F.J. Muehlbauer (WSU) was used to develop recominant inbred lines by advancing F_2 individual plants to F6 using Single Seed Descent (SSD) (Table 2.2.1). F6 plants were harvested seperately and randomly 16 seeds from each family (86) were planted as F7 lines. Their seeds were harvested to grow F8 lines. DNA for the genetic mapping was extracted from 6 to 10 plants from each line.

RAPD analysis

Total genomic DNA was isolated from fresh leaves of 100 F2 individuals plants from each cross using the protocol

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Eujayl, I., M. Baum, W. Erskine, E. Pehu and F.J Muehlbauer 1996: An early- generation evaluation of crosses for future mapping populations and RAPDs for gene mapping in lentil. TAG, submitted.

Eujayl, I., M. Baum, W. Erskine, E. Pehu and F.J Muehlbauer 1995: Development of a genetic linkage map for lentil based on RAPD markers. In: Proc. 2nd Eurp.Grain Legumes Conference, Copenhagen, Denmark. p 440-441.

previously developed by Saghai-Maroof et al.[10]. The extract was treated with RNAase to degrade the RNA. DNA was quantified using a spectrophotometer (Beckman DU-61). The quality of the extracted DNA was visually checked on a 1% agarose gel. RAPD-analysis was conducted on thermocycler Perkin Elmer 9600 system. The reaction mixture (50μl) contained, 1x PCR buffer (10mM TRIS-HCl, pH 8.3, 1.5 mM $MgCl_2$, 50 mM KCl, 0.1 mg/ml gelatine, 100 μM each of dNTP, 1 unit of Taq polymerase (all products from Boehringer Mannheim), 200 μM of primer (Operon) and 50 ng of DNA.

Table 2.2.1. Workplan for lentil mapping project including the time frame and activities already being/to be excecuted

Month/year	Activities	Facilities
1992	F_1	
W.S.University.		
Jan - May 1993	F_2	Glass house
	Isozymes analysis	GRU - laboratory
	Morphological Var.	Glass house
	RAPDS analysis	Biotech laboratory
June - Sept 1993	F_3 - SSD	Growth chamber
	RAPD analysis	Biotech laboratory
Oct.1993 - Jan. 1994	F_4 - SSD	Growth chamber
Feb. - May 1994	F_5 - SSD	Growth chamber
	F_3 seed increese	Plastic house
June - Oct.1994	F_6 - SSD	Growth chamber
Jan. - May. 1995	F_7 multiplication	Growth chamber
	Diseases screening;	Sick plot and
	F_4 families	Plastic house
June - Oct. 1995	F_8 Seed increese	Tel Hadya
	Mapping using RAPD	
	on RILs	
Dec.95 - May 1996	F_8 Field testin	Tel Hadya.
	QTL and Fusarium wilt	
	RFLP -marker analysis	
June - Dec. 1996	Markers and Bulked	
	segregant analysis	

Saghai-Maroof, M. A., K. M. Soliman, R. A. Jorgensen, and R. W. Allard, 1984: Ribosomal DNA spacer-length polimorphism in barley: Mendelian inheritance, chromosomal location, and population dynamics. Proc. Natl. Acad. Sci. USA. 81, 8014-8018.

The mixtures were overlaid with mineral oil and then subjected to the following cycle conditions: one cycle of 94°C for 2 min, followed by 40 cycles of 92°C for 1 min, 36°C for 1 min and 72°C for 2 min, followed by one cycle of 72°C for 5 min. The primer kits F to X from Operon Technologies, Alameda, Calif. were used. The PCR products were electrophoretically separated in a 2% agarose gel in 1x TBE buffer and visualized by Ethidium bromide staining.

Table 2.2.2. Segregation patteren of RAPDs loci linked at 0.0 to 7.0 cM and the chi square test.

Markers	Segragation A/B	Chi-square	Probability
1.P03e	37:47	0.96	0.33
2.O11b	38:48	0.94	0.33
3.S08b	36:49	1.68	0.19
4.H19c	36:48	1.44	0.23
5.L15b	41:45	0.10	0.75
6.R04a	43:43	0.00	1.00
7.O11d	44:42	0.00	1.00
8.G06a	41:44	0.04	0.84
9.S04b	39:42	0.04	0.84
10.O08	45:40	0.18	0.67
11.K20a	48:37	1.16	0.28
12.I20b	48:37	1.16	0.28
13.O13	49:35	2.00	0.16
14.V02	42:37	0.20	0.65
15.G18c	48:34	2.06	0.15
16.G07c	49:36	1.68	0.19
17.S04C	41:40	0.00	1.00
18.Q14e	43:42	0.00	1.00
19.O06a	42:37	0.20	0.65
20.M18a	39:44	0.18	0.67
21.U01d	43:43	0.00	1.00
22.U01a	44:42	0.00	1.00
23.P03b	47:39	0.56	0.45
24.G18a	50:35	2.66	0.10
25.S01b	50:36	1.96	0.16
26.I18b	48:37	1.16	0.28
27.MA4a	45:40	0.18	0.67
28.MA4b	45:40	0.18	0.67
29.Q14c	43:42	0.00	1.00
30.G13a	45:41	0.10	0.75

43 primers were used for scoring polymorphism in the recombinant lines. Only intense and prominant bands were scored. The 43 primers yielded 111 segregating fragments in the F6-F8 derived population. An average of 2.7 bands/primer was observed which is an eighty percent increase in polymorphism compared to segregation at the F2 generation (see Fig. 2.2.1). The markers were tested for goodness of fit to 1:1 ratio by Chi square. Out of the 111 markers, 9 markers showed skewed segregation, with predominance of the alleles of the pure *culinaris* parent in eight of them.

Statistical Analysis:
111 RAPD markers and the seed color spotting (SPC) were analysed for genetic linkage using multipoint analysis of the MAPMAKER software program. A LOD score of 3.0 and a recombination fraction of 0.25 were used.

Genetic Linkage Map
The genetic linkage map (Fig.2.2.1) consists of 90 markers falling into 14 linkage groups including linkage groups with only two or three markers limked covering 818 cM of the lentil genome. Out of the 111 markers analyzed, 21 markers were not linked at the stringent threshold. Six markers would have joined the linkage groups at LOD 3.0 and theta of 0.3. The morphological locus Scp was linked to seven RAPD markers. On average, markers were spaced at an distance of 9.0 cM along the linkage groups.

2.2.2. Characterization of *Fusarium oxysporum* f.sp *lentis* using RADP markers.

Fusarium oxysporum f.sp *lentis* isolates have been characterized in recent year using morphological differences and isozymes (see Section 2.1.1.4.2 & 2.1.1.4.3). We were interested to see if RAPD markers could also be used for their differentiation.

Fungus isolates:

Thirty-two isolates collected from different geographical areas in Syria in farmers fields were used for the analysis. Each isolate was single spored and cultured on PDA media. The isolates were tested for their virulence and typical wilt symptoms. The cultures were grown at room temperature and under continuous illumination.

Two weeks old mycelia, collected from Petri dishes (peeled from the media) were incubated in 2 ml Eppendorf tubes, immediatly frozen in liquid nitrogen and subsequent freeze dried. DNA was isolated using the CTAB method. The DNA was tested for quality and quantity using a spectrophotometer and a test-gel. Eight arbitrary 10-mer primers (Operon) were tested for PCR amplification.

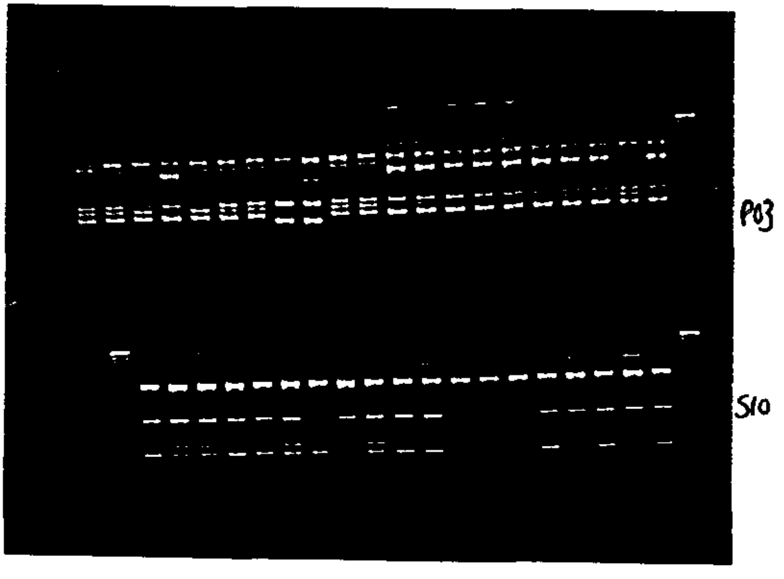

Fig.2.2.2. Screening of *Fusarium oxysporum* f.sp *lentis* using RADP markers.Top: DNA-fingerprint with primer PO3 on 21 differnt isolates, bottom Primer S10 on 19 different

Seven polymorphic primers were subsequent used to study the variation between the fungus isolates. The PCR conditions were the same as previously reported for lentil mapping. Seven primers revealed abundance of polymorphism at 51 different loci (Fig 2.2.2). According to virulance, isolates were divided into two groups i.e virulant and avirulant. From our data we are able to group the isolates into two major groups and two minor groups.

I. Eujayl, A. Abbas, B. Bayaa, M. Baum and W. Erskine

2.3. LENTIL MECHANIZATION

During the first decade of ICARDA, a major drive was made to develop economic machine harvest systems for lentil production (FLIP Annual Reports 1986-1990; Legume Program Annual Reports 1991-93). Following the introduction and use by farmers in Syria and Turkey of such systems, a moratorium was placed on further technical research at ICARDA pending the completion of an adoption survey of producers in Syria.

The survey was conducted with the Syrian General Organization of Agricultural Mechanization, the American University of Beirut and the Farm Resource Management and Germplasm Programs within ICARDA.

Seventy-seven lentil producers spread among the major lentil growing regions of North-West and North-East Syria were interviewed using a questionnaire format to elucidate the importance of lentil in the farming enterprise and issues related to harvest.

Hand harvest of lentil is still practiced by 81 % of the farmers surveyed. Nineteen percent of farmers had adopted mechanized harvest. Generalizing beyond the survey in a country where the average sown area of lentil was 92,000 ha from 1991-93 (FAO, 1994), this represents an area of adoption of harvest mechanization of approximately 17,500 ha. The adopters were all in the North-East of Syria and they had a greater area (19.2 ha) sown to lentil than the non-adopters (9.2 ha), on average. The adopters comprised those (60%) who use a self-propelled mower-swather and those (40%) who use a cereal combine harvester.

Although adoption has not moved very fast, it was concluded that the mechanical harvest of lentil is now more a problem of technology transfer than of research. To this end we will provide technical input into lentil harvest demonstrations in the forthcoming season in Syria, jointly run with the General Organization of Agricultural Mechanization and the Directorate of Extension.

The late J. Haidar (American University of Beirut), General Organization of Agricultural Mechanization (Syria), A. Salkini, A. Dakermanji & W. Erskine.

2.4 Lentil Entomology

2.4.1 Residual Effect of Carbofuran and Promet on *Sitona* and Yield

This is the third season for this expt. in which the soil was treated with Furadan 5% G at the rate of 10kgs/ha. Seeds treated with Promet at 12ml/kg and untreated check were included for comparison. Two adjacent sites were used in which lentil was alternated with cereal. The main objective was to assess the residual effect of Carbofuran which was applied in soil in season 1992/93. Nodule damage and total yields were evaluated. Results showed that Promet apparently reduced nodule damage compared with Carbofuran and the check; Carbofuran being inferior to check (Table 2.4.1). Promet and Carbofuran, however, produced almost similar yields which were significantly higher than the check. It is clear that Carbofuran has lost its insecticidal effect, but maintained its boosting effect on yield. It is, therefore, possible that Carbofuran is degraded into substances that are non-insecticidal but promote plant physiology and growth.

Table 2.4.1 Residual effect of Carbofuran and promet seed-treated on *Sitona* and total yield in lentil at Tel Hadya, (1994/95).

Treatment	%Nodule damage		Total yield kg/ha
	26.3	25.4	
Carbofuran	19.3	36.4	4528.0
Promet	13.8	36.4	4666.3
Check	15.8	32.0	3927.7
LSD at 5%	7.9	33.5	507.4
C.V.	21.5	46.7	5.1

2.5.LEGUME VIRUSES

Work on legume viruses during the growing season 1994/95 included (i) survey for legume viruses in Lebanon and Syria, and (ii) evaluation of lentil genotypes for faba bean necrotic yellows virus (FBNYV) and a lentil luteovirus resistance. Activities also included regular testing for seed-borne viruses in seeds dispatched for international nurseries and testing gene bank accessions to free them from seed-borne infections. The virology lab continued to provide ELISA kits for virus testing to a number of NARSs laboratories of WANA upon request.

2.5.1. Survey for legume viruses in Lebanon and Syria

A survey was conducted during April, 1995 to study the spectrum of viruses that are causing yellowing/stunting/necrosis in faba bean, chickpea and lentil. A total of 510 legume samples (157 faba bean, 100 chickpea, 242 lentil and 11 *Vicia ervilia*) were collected from 32 fields surveyed and covered the Bekaa valley in Lebanon from Baalbek to Zahle including the Terbol Station. The survey in Syria included the area between Damascus and Dara'a, the coastal plains and Ghab regions.

Table 2.5.1. Viruses in chickpea, faba bean, lentil and *Vicia ervilia* samples with yellowing symptoms collected from Lebanon and Syria during 20-22 April, 1995. Identification was based on serological reaction (TBIA).

COUNTRY Crop	No. of fields visited	No. of samples collected	No. of plants found positive to FBNYV	No. of plants found positive to Luteovirus[a]	No. of plants with BBMV, BYMV BBSV, PSbMV	No. of which did not react single infection	No. of plant samples with all antisera used
LEBANON							
Chickpea	3	63	1	10	0	10	52
Faba bean	1	30	11	3	0	10	18
Lentil	4	31	2	0	0	2	29
Sub-total	**8**	**124**	**14**	**13**	**0**	**22**	**99**
SYRIA							
Chickpea	5	37	0	20	0	20	17
Faba bean	11	127	56	52	0	67	39
Lentil	7	211	38	68	5	109	102
Vicia ervilia	1	11	0	11	0	11	0
Sub-total	**24**	**386**	**94**	**151**	**5**	**207**	**158**
Total	**32**	**510**	**108**	**164**	**5**	**229**	**257**

[a]A monoclonal antibody (5G4) which reacts with a large number of luteoviruses was used.

Fields observation indicated that 50% of the fields surveyed had, at the time of the survey, a virus disease incidence of 1% or less. In 34.3% of the fields, virus disease incidence was in the range 2-20%, and in 9.4% of the fields had an incidence of 21-50%. Only in 6.2% of the fields surveyed, virus disease incidence was more than 50%.

The collected samples were tested by the tissue-blot immunoassay (TBIA) and using antisera for the following viruses: monoclonal antibodies for faba bean necrotic yellows virus (FBNYV) and 5G4 which has a broad specificity for luteoviruses and polyclonal antibodies against broad bean stain (BBSV), broad bean mottle (BBMV), bean yellow mosaic (BYMV) and pea-seed borne mosaic (PSbMV) viruses. Results obtained from laboratory tests are presented in Table 2.5.1. The viruses detected were luteoviruses (32.1%), FBNYV (21.1%) and any of BBSV, BBMV, BYMV or PSbMV (0.9%).

When the 68 field isolates which reacted with the broad-spectrum luteovirus monoclonal antibody were tested by another set of four specific monoclonals by the TBIA test, five different types of reaction patterns were obtained (Table 2.5.2). Such results suggested the existence of five different luteovirus strains/viruses which need further characterization.

Table 2.5.2. Reaction patterns of selected legume virus isolates with monoclonal antibodies known for their differential reaction with luteoviruses

Number of isolates	Reaction with monoclonal antibodies[a]				
	5G4	4B10	3B11	6F9	6G4
15	+	-	-	+	+
7	+	-	-	+	-
20	+	+	+	+	+
1	+	+	+	+	-
25	+	-	-	-	-

[a] Provided by Dr. Lina Katul, BBA, Braunschweig, Germany

2.5.2. Screening for faba bean necrotic yellows virus (FBNYV) and a lentil luteovirus resistance in lentil

Fifty nine lentil genotypes were evaluated for their resistance to a local isolate of FBNYV (*SV292-88*) and a lentil luteovirus (SL 1-94) using artificial inoculation by aphids. Genotypes tested were planted in the field in two replicates, each of two one meter rows, with 25 plants per meter in a randomized complete block design for both the inoculated and non-inoculated treatments. Yield loss (%) and disease incidence (%) based on symptoms produced were determined for all the genotypes tested and are summarized in Tables 2.5.3 and 2.5.4.

Table 2.5.3. Variability in yield loss (%) and disease incidence on symptoms produced among lentil genotypes in response to infection with faba bean necrotic yellows virus (FBNYV) evaluated during the 1994/1995 growing season

Lentil genotypes	Yield loss (%)
ILL 71, 74, 75, 86, 291, 2581, 6193, 6198, 6245, 6458, 6994, 7217.	0.0
ILL 85, 203, 213, 222, 292, 323, 478, 4400, 5816, 6435, 7216.	0.1-10
ILL 204, 221, 265, 324, 344, 346, 3433, 6811, 7157, 7165, 7184, 7188, Crimson.	10.1-20
ILL 112, 212, 214, 5871, 6024, 6258, 7181.	20.1-30
ILL 105, 271, 6229, 6778, 6972, 7183, 7185, Chilean 78.	30.1-40
ILL 5750, 6789, 7193, Red Chief, Palouse.	40.1-80
ILL 6015, 7201, 7212.	80.1-100
	FBNYV incidence (%)
ILL 71, 74, 75, 86, 214, 221, 291, 2581, 5816, 6193, 6245, 6994, 7217	0.0
ILL 85, 112, 203, 204, 213, 222, 265, 323, 3433, 4400, 5871, 6024, 6198, 6435, 6458, 6778, 6789, 7184, 7185, 7188.	0.1-10
ILL 7157, 7165, 7181, 7183.	10.1-20
ILL 478, 5750, 6811, Crimson	20.1-30
ILL 271, 292, 344, 346, 6972	30.1-40
ILL 105, 212, 324, 6229, 6258, 7193, 7216, Red Chief, Chilean 78, Palouse	40.1-80
ILL 6015, 7201, 7212.	80.1-100

Table 2.5.4. Variability in yield loss (%) and disease incidence (%) based on symptoms produced among lentil genotypes in response to infection with a lentil luteovirus (not fully characterized) evaluated during the 1994/1995 growing season.

Lentil genotypes (%)	Yield loss
None	0.0
ILL 75.	0.1-10
None	10.1-20
ILL 204, 214, 3433, 6458.	20.1-30
ILL 85, 212, 213, 222, 6024, 6193.	30.1-40
ILL 71, 74, 86, 105, 112, 203, 221, 265, 271, 291, 292, 323, 344, 346, 478, 2581, 6015, 6198, 6258, 6435, 6811, 6994, 7157, 7165, 7183, 7184, 7185, 7188, 7193, 7201, 7216, 6229, 7217, Crimson, Palouse.	40.1-80
ILL 324, 4400, 5750, 5816, 5871, 6245, 6778, 6789, 6972, 7181, 7212, Red chief, Chilean 78	80.1-100

	Lentil luteovirus incidence (%)
ILL 75, 214, 3433.	0.0
ILL 74, 85, 221, 265.	0.1-10
ILL 213, 6778, 7217.	10.1-20
ILL 86, 6435.	20.1-30
ILL 6811	30.1-40
ILL 71, 105, 203, 204, 212, 222, 271, 291, 344, 346, 478, 2581, 6458, 7157, 7181, 7183, 7185, 7188, 7193, Crimson, Chilean 78.	40.1-80
ILL 112, 292, 323, 324, 4400, 5756, 5816, 5871, 6015, 6024, 6193, 6198, 6229, 6245, 6258, 6789, 6972, 6994, 7165, 7184, 7201, 7212, 7216, Red Chief, Palouse.	80.1-100

Based on these results it was possible to divide the genotypes tested into five categories: (1) Highly resistant: genotypes which did not produce disease symptoms and without loss in grain yield; (2) Resistant: genotypes where disease incidence did not exceed 20% and grain yield loss was between 0-20%; (3) Moderately resistant: genotypes with disease incidence (%) and grain yield loss around 40%; (4) Susceptible genotypes: which had a yield loss (%) and disease incidence (%) around 40.1-80%; (5) Highly susceptible: genotypes which had a yield loss (%) and disease incidence (%) around 80.1-100%. Results obtained clearly indicated the presence of useful genetic diversity in *Lens culinaris* with respect to FBNYV and a lentil luteovirus resistance.

K.M. Makkouk and S.G. Kumari

2.5.3. Testing for Seed-borne Viruses

2.5.3.1. Testing lentil international nurseries seed lots

A total of 684 accessions of lentil seed lots destined for international nurseries were tested serologically for the presence of seed-borne virus(es) infection. 139 lots were rejected on the basis of detecting a virus in at least one seed in a sample of 400 seeds per seed lot.

2.5.3.2. Cleaning germplasm in the gene bank from seed-borne infections

450 lentil accessions planted for germplasm multiplication and evaluation purposes, were purified from seed-borne infection by eliminating infected plants during the late flowering-early podding stage, and only seeds from healthy plants were harvested and deposited in the gene Bank

K.M. Makkouk and N. Attar

2.5.4. Production of ELISA Kits

During 1995, antisera to lentil luteovirus isolate (SL1-94), pea early-browning tobravirus (Algerian isolate) and a legume isolate of cucumber mosaic cucumovirus were produced. ELISA kits were prepared and made available for laboratories of the national programs in WANA. In each kit there is enough immunoglobulins and enzyme conjugate to test 2000 samples. Accordingly, ELISA kits for the following legume viruses are now available at the Virology Laboratory of the Germplasm Program.

Legume viruses

- Alfalfa mosaic alfamovirus
- Bean yellow mosaic potyvirus
- Broad bean mottle bromovirus
- Broad bean stain comovirus
- Broad ben wilt fabavirus
- Chickpea luteovirus
- Cucumber mosaic cucumovirus
- Lentil luteovirus
- Pea early-browning tobravirus
- Pea seed-borne mosaic potyvirus

K.M. Makkouk and S.G. Kumari

2.6. INTERNATIONAL TESTING PROGRAM

The international testing program on lentil is a vehicle for the dissemination of genetic materials and improved production practices, in the form of international nurseries and trials, to the national programs in and outside the WANA region. The genetic materials comprise early segregating populations in F_3 and F_4 generations, and elite lines with wide or specific adaptation, special morphological or quality traits, and resistance to common biotic and abiotic stresses. Nurseries are only sent on request and often include germplasm specifically developed for a particular region or a national program. A list of trials supplied in the 1995/96 season is given in Table 3.6.1.

The testing program helps in identification of genotypes with specific and wide adaptation. The performance data permit assessment of genotype x environment interaction and help in targeting breeding efforts for specific agro-ecological conditions.

The salient features of the 1993/94 international nursery results, received from cooperators until 31 October 1995, are presented here. The stability analyses of some of the trials were done using Eberhart and Russell (1966) model.

In lentil there are three different yield trials but for 1993/94, the seed for two trials (large seeded and small seeded) could not be supplied as most of the entries in the seed increase plots at Tel Hadya were severely damaged by Fusarium wilt. This was the first year when we had the Fusarium wilt severity in our fields at Tel Hadya, indicating that Fusarium wilt may be a major problem in the region. Thus only the trials which had the resistance to Fusarium wilt were supplied to cooperators. The results received from the cooperators for the 1993/94 are reported here.

For Lentil International Yield Trial-Early (LIYT-E) data were analyzed for seed yield for 13 locations. At Shanxi-1 and Shanxi-2 in China, and Rawdat Harma in Qatar, 5, 12 and 13 of the test entries exceeded the respective local check in seed yield by a significant margin ($P \leq 0.05$). On basis of mean across locations the five heaviest yielding lines wereFLIP 87-70L, 74TA 441 x Pant L 639, FLIP 89-53L, ILL 7162 and FLIP 89-67L. Stability analysis for seed yield revealed that only three of the 23 entries, namely FLIP 89-53L exhibited specific adaptation to the high yielding environments, and another two namely, FLIP 87-66L and FLIP 88-43L exhibited general adaptation over different environments. The remaining 20 entries were unpredictable across environments.

Table 3.6.1. Distribution of Legume International Nurseries to cooperators for the 1995/96 season.

International Trial/Nursery	No. of sets
Yield Trial, Tall (LIYT-L-96)	44
Yield Trial, Small (LIYT-S-96)	28
Yield Trial, Early (LIYT-E-96)	26
Screening Nursery, Large-Seed (LISN-L-96)	35
Screening Nursery, Small-Seed (LISN-S-96)	27
Screening Nursery, Early (LISN-E-96)	29
Screening Nursery, Drought Tolerance (LISN-DT-96)	35
F_5 Nursery, Large Seed (LIF_5N-L-96)	22
F_5 Nursery, Small Seed (LIF_5N-S-96)	15
F_5 Nursery, Early (LIF_5N-E-96)	11
F_5 Nursery, Cold Tolerance (LIF_5N-CT-96)	6
Cold Tolerance Nursery (LICTN-96)	27
Ascochyta Blight Nursery (LIABN-96)	14
Fusarium Wilt Nursery (LIFWN-96)	29
Rust Nursery (LIRN-96)	16
Total	364

For Lentil International Screening Nursery - Large (LISN-L), Small (LISN-S), and Early (LISN-E), the data for seed yield were reported from 33, 22, and 11 locations, respectively. The analyses of data revealed that at 9 locations in LISN-L (Chengdu in China; Gallina in Italy; Rabba and Mushaggar in Jordan; Izra'a, Gelline, Hama and Tel Hadya in Syria; and Erzurum in Turkey), one location in LISN-S (Aleppo in Syria), and 5 locations in LISN-E (Dingxi in China, Ghinchi in Ethiopia, Maragheh and Sanandag in Iran and Tel Hadya in Syria), some of the test entries exceeded the respective local check by a significant margin ($P \leq 0.05$). The five heaviest yielding lines across locations in these nurseries are given in Table 3.6.2.

Table 3.6.2. The five heaviest yielding lines across locations in different lentil screening nurseries, 1993/94.

Rank	LISN-L	LISN-S	LISN-E
1	FLIP 90-13L	FLIP 90-41L	FLIP 92-50L
2	78S 26002	81S15 (ILL 5883)	FLIP 92-42L
3	FLIP 88- 1L	FLIP 89-31L	FLIP 93-36L
4	FLIP 92- 8L	FLIP 93- 1L	FLIP 93-44L
5	FLIP 90- 3L	FLIP 92-34L	FLIP 93-45L

The results of Lentil International F_6-Nursery Large (LIF_6N-L), F_6-Nursery Small (LIF_6N-S), F_6-Nursery Early (LIF_6N-E), and F_6-Nursery Cold Tolerance (LIF_6N-CT) were received from 10, 6, 6 and 4 locations, respectively. At 6, 3, 2 and 1 locations the cooperators made some individual plant selections for their use.

The results of Lentil International Cold Tolerance Nursery were reported from seven locations. There was no cold at Ghinchi, and Chefe Donsa in Ethiopia, and Hymana in Turkey. None of the test entries at Almora in India, 13 test entries at Toshevo in Bulgaria, all the test entries at Tel Hadya in Syria and Erzurum in Turkey exhibited tolerance reaction (rating $\leq$4).

The results of Lentil International Ascochyta Blight Nursery were received from 5 locations. All the test entries at Toshevo in Bulgaria showed a reaction of $\leq$ 5 (on 1-9 scale, where 1=no infection and 9=killed). At Jammu in India, all the test entries with the exception of 2 showed a reaction of $\leq$ 3. At Tel Hadya in Syria, all the test entries were rated as 1 and susceptible check as 2, and at Guelma in Algeria and Jema'a Shain in Morocco there was no disease infestation.

The results of Lentil International Rust Nursery were reported from 7 locations. The test entries showed a reaction of 1-3 at Akaki in Ethiopia, 1-5 at Pant Nagar in India, 3-9 at Jammu in India, 1-7 at Kanpur in India, and 1-5 at Jema'a Shain in Morocco. The tolerant lines with highest frequency across locations included, UJL 81-129, Lenka, FLIP84-76L, ILL5883, FLIP86-16L, FLIP87-17L, FLIP87-74L, and FLIP88-32L.

The results of Lentil International Fusarium Wilt Nursery were reported from 10 locations. There was no disease infestation at Guelma in Algeria and Heimo in

Syria. At Toshevo in Bulgaria, Debre Zeit and Akaki in Ethiopia, Pant Nagar in India, Khumaltar in Nepal, Piestany in Slovakia, Izra'a and Hama in Syria, 8, 19, 26, 21, 4, 28, 25 and 29 entries, respectively, showed resistance reaction (rating $\leq$ 5). Five entries (FLIP85-33L, FLIP86-38L, FLIP89-60L, FLIP90-7L and FLIP90-22L) were resistant across 7 out of 8 locations.

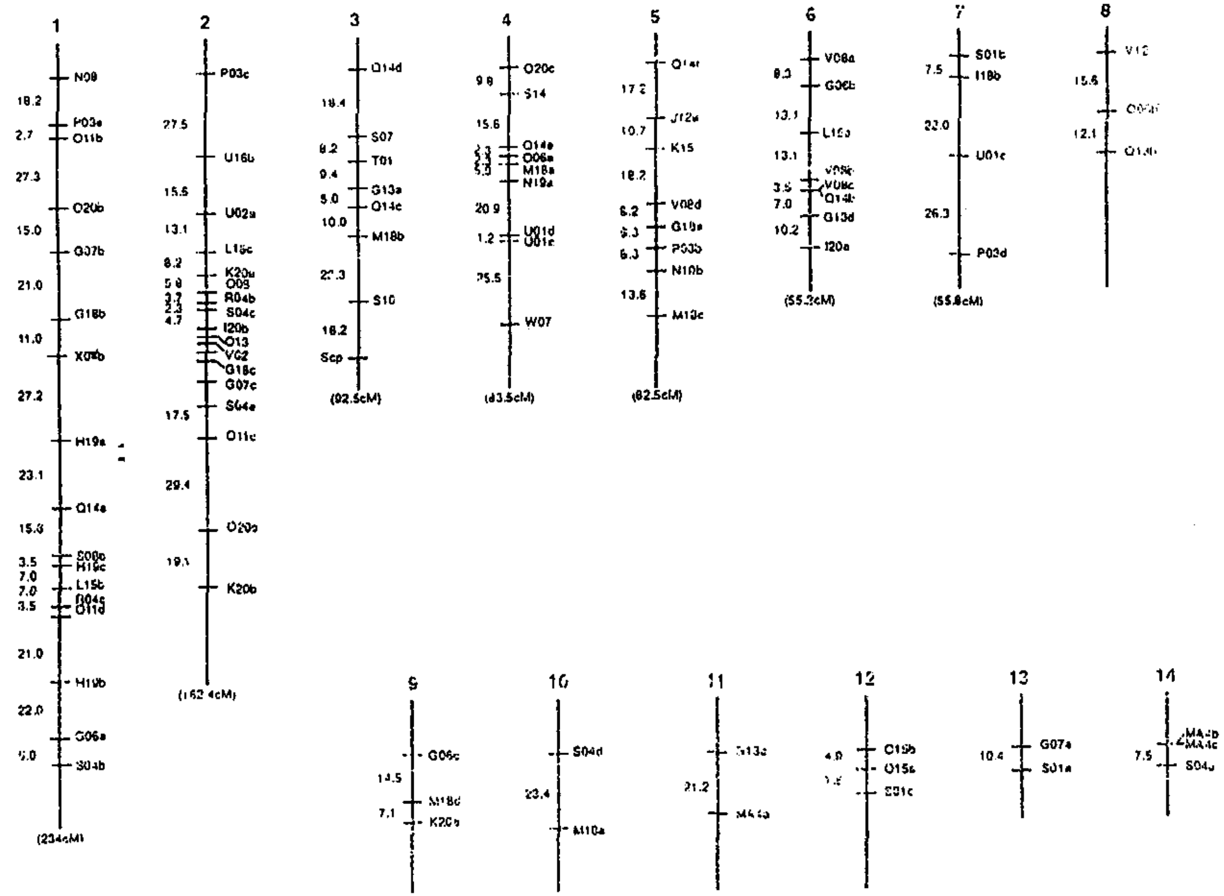

Fig. 2.2.1. Linkage map of lentil established in a recombinant inbred population using 88 RAPD and 1 morphological marker.

3. KABULI CHICKPEA IMPROVEMENT

The kabuli chickpea improvement is a joint program with ICRISAT, India. The main objective of the program is to increase and stabilize kabuli chickpea production in the developing world. Of the five main regions, where chickpea is grown, the Mediterranean region and Latin America produce mostly kabuli-type chickpea. Five to ten percent of the area in the other three main production regions (Indian subcontinent, East Africa, and Australia) is also devoted to the production of the kabuli type. The kabuli chickpea is also grown at high elevation areas (>1000 m elevation) in West Asia, especially in Afghanistan, Iran, Iraq, and Turkey; and in the Atlas mountains of North Africa.

Ascochyta blight and fusarium wilt are the two major diseases of chickpea. Leaf miner in the Mediterranean region and pod borer in other regions are major insect pests. Drought is the major abiotic stress throughout the chickpea growing areas and cold assumes importance in Mediterranean environments and the temperate region especially for winter sowing. The kabuli chickpea is mainly grown as a rainfed crop in the wheat-based farming system in areas receiving between 350 mm and 600 mm annual rainfall in the West Asia and North Africa region. In Egypt and Sudan, the crop is only grown with supplemental irrigation and in South Asia, West Asia and Central America, a small part of area is grown with supplemental irrigation.

In West Asia and North Africa, where the crop is currently spring-sown, yield can be increased substantially by advancing sowing date from spring to early winter. With the introduction of winter sowing, chickpea cultivation can be extended to lower rainfall (to 300 mm) regions. Increasing plant density and reducing row width can also increase yield significantly, especially in winter-sown crop. Winter sowing allows the crop to be harvested by machine.

Major efforts are underway to stabilize chickpea productivity by breeding cultivars resistant to various stresses, such as the diseases (ascochyta blight and fusarium wilt), insect pest (leaf miner), parasite (cyst nematode), and physical stresses (cold and drought). The exploitation of wild *Cicer* species for transfer of genes for resistance to different stresses and transfer of yield genes is another area receiving high research priority at the Center. DNA fingerprinting in *Ascochyta rabiei* is being pursued with great promise.

During 1995, several collaborative projects continued to operate. In the project "Development of chickpea germplasm with combined resistance to ascochyta blight and fusarium wilt using wild and cultivated species", four Italian institutions collaborated with ICARDA. Screening for cyst nematode was carried out in association with the Istituto di Nematologia Agraria, C.N.R., Bari, Italy. Fusarium wilt resistance screening was done in association with the Institut de la Recherche Agronomique de Tunisie (IRAT), Tunisia and the Departamento de Patologia Vegetal, Cordoba, Spain. Screening for tolerance to cold was done in cooperation with agricultural research institutes in Turkey. The University of Saskatchewan, Canada is collaborating in studies on genetic diversity of kabuli chickpea. Studies on mechanism of drought and cold tolerance and aspects of biological nitrogen fixation are being conducted in collaboration with INRA, Montpellier, France. Studies on leaf miner resistance and application of restriction fragment length polymorphism (RFLP) in characterization of chickpea genotypes and *Ascochyta rabiei* isolates are carried out in collaboration with the University of Frankfurt, Germany. Research on the development of irrigation-responsive cultivars is being conducted with the Agriculture Research Centre, Giza, Egypt. Studies on the development of drought-tolerant chickpeas are carried out in collaboration with the University of Addis Ababa, Debre Zeit, Ethiopia.

3.1. Chickpea Breeding

Major objectives of the breeding are (1) to develop cultivars and genetic stocks with high and stable yield and segregating populations to support National Agricultural Research Systems (NARSs) and (2) to conduct strategic research to complement Objective 1. Specific objectives in the development of improved germplasm for different regions are:

1. Mediterranean region: (a) winter sowing: resistance to ascochyta blight, tolerance to cold, suitability for machine harvesting, medium to large seed size; (b) spring sowing (30% of resources): cold tolerance at seedling stage, resistance to ascochyta blight and fusarium wilt, tolerance of drought, early maturity, medium to large seed size;

2. High elevation areas: spring sowing, cold tolerance at seedling stage, resistance to ascochyta blight, terminal drought tolerance, early maturity, and medium to large seed size.

3. Indian subcontinent and East Africa: resistance to ascochyta blight and/or fusarium wilt, drought tolerance, early maturity, small to medium seed size, response to supplemental irrigation;

4. Latin America: resistance to fusarium wilt, root rot and virus diseases, large seed size.

Major strategic research projects are:

1. Exploitation of wild *Cicer* species for (a) transfer of genes for resistance to cold and cyst nematode and (b) transfer of "yield genes".
2. Pyramiding of genes for resistance to ascochyta blight.
3. Identification of races of fusarium wilt in the WANA region.
4. Increasing shoot biomass yield in chickpea.

K.B. Singh

3.1.1. Use of Improved Germplasm by NARSs

3.1.1.1. International nurseries/trials and other breeding lines

During 1995, 14,275 chickpea entries were shipped to NARS through international nurseries (370 sets) and special requests to 45 countries. Eighty-five percent of international trials and nurseries were furnished to developing countries and the remaining to industrialized countries. The nurseries were in demand from six continents. Despite our encouragement to national programs to accept more segregating and crossing block materials, the demand for finished material isincreasing, suggesting that breeders have found ICARDA-ICRISAT chickpea lines useful for their direct exploitation.

K.B. Singh, R.S. Malhotra and M.C. Saxena

3.1.1.2. On-farm trials in Syria

Three chickpea lines along with an improved check Ghab 3 were selected by the Directorate of Agriculture and Scientific Research (DASR), Ministry of Agriculture and Agrarian Reforms, from the ICARDA/ICRISAT international trials for evaluation in on-farm trials in 15 locations throughout Syria in 1994/95. At Jableh, FLIP 89-29C and at Atareb FLIP 88-85C was significantly superior in seed yield compared to the improved check (Ghab 3). At most locations, FLIP 88-85C was superior to check in seed yield with an overall increase of 10.7%, but the differences were statistically non-significant. FLIP 88-85C also had 16.5% larger seed size than Ghab 3. Like Ghab 3, the new cultivar is tall and is thus suitable for combine harvest, an important attribute for the large-scale introduction of winter chickpea. This line has potential to produce over 4 t ha^{-1} seed yield under favorable conditions.

The ICARDA-ICRISAT Kabuli Chickpea Project was involved in the conduct of on-farm trials in many other countries including Algeria, Iraq, Jordan, Lebanon, Morocco, Syria, Tunisia, and Turkey. The degree of our involvement varied from complete association (e.g. Lebanon) to only providing advice (e.g. Turkey). Results have been encouraging as demonstrated by a large number of releases of cultivars and their adoption by farmers.

NARSs scientists and K.B. Singh

3.1.1.3. Pre-release multiplication of cultivars by national programs

Fifty-two lines have been chosen by 14 NARSs during 1994 from the ICARDA/ICRISAT international trials for the pre-release multiplication and on-farm testing. This is an incomplete list because we do not have full information. All new lines have resistance to ascochyta blight and tolerance of cold. They have large seed size, thus they meet consumers' demands. If grown in winter, they attain a minimum height of 40 cm and can be thus harvested by machine. Seeds of some of the promising lines are being multiplied at ICARDA to meet the potential initial demand of NARSs.

NARS Scientists, K.B. Singh, R.S. Malhotra and M.C. Saxena

3.1.1.4. Release of cultivars by NARSs

Thirteen chickpea cultivars were released by five national programs during 1995. These included ILC 482, ILC 3279, and FLIP 84-48C in Iran; Pascia (FLIP 86-5C) and Otello (ICC 6306/NEC 206) in Italy; Farihane (FLIP 84-79C), Moubarak (FLIP 84-145C), and Zahor (FLIP 84-182C) in Morocco; FLIP 87-45C and FLIP 89-130C in Oman; Athenas (ILC 72 x CA 2156), Bagda (ILC 72 x CA 2156), and Kairo (ILC 72 x CA 2156) in Spain. To date, NARSs in 21 countries have released 69 lines as cultivars from the improved germplasm furnished by ICARDA. Forty-nine of these have been released for winter sowing in the Mediterranean region, 14 for spring sowing including four in China, and six for winter sowing in more southerly latitudes. With these releases chickpea cultivars bred at the Center have been released in the four major regions of the chickpea production.

NARS Scientists, K.B. Singh, R.S. Malhotra and M.C. Saxena

3.1.2. Screening for Stress Tolerance

3.1.2.1. Cultivated species

3.1.2.1.1. Wilt resistance

Diseases are the major constraints to chickpea production. Among the 50 diseases reported, fusarium wilt induced by *Fusarium oxysporum* Schlecht. emend. Snyd. & Hans. f.sp. *ciceri* (Padwick) Snyd. & Hans. is the second most important disease worldwide. In WANA, it is prevalent in parts of North Africa and in the Nile Valley. *F. oxysporum* f. sp. *ciceri* is both soil-borne and seed transmitted. It can survive in the soil for long periods. Inoculum in infected seeds can be eradicated by seed dressing with fungicide. The most practical and economical method for the control of fusarium wilt is the use of resistant cultivars. Accordingly, resistance breeding has been one of the main objectives in chickpea improvement. In this effort, the major bottleneck has been the presence of different races of the pathogen. The occurrence of seven races of the pathogen has been reported. Therefore, more efforts are needed to increase the effectiveness and stability of disease resistance.

Screening in the field must be conducted in a wilt-sick plot. A wilt-sick plot has been developed at ICARDA. During 1994/95, 2155 breeding lines developed at ICARDA were evaluated for fusarium wilt in the wilt-sick plot at Tel Hadya. Six and 251 lines were rated 0 and 1, respectively, where 0 = no plants killed and 1 = 1-20% plants killed. These 257 lines were evaluated in pots in the green house. It was observed that 4 lines, FLIP 85-124C, FLIP 88-1C, FLIP 92-48C, and FLIP 93-211C, had a 0 rating and were free from any damage from fusarium wilt. In addition, 59 lines with 1 rating were resistant. The evaluation data of 2155 lines is summarized in Table 3.1.1.

K.B. Singh and R.S. Malhotra

Table 3.1.1. Reaction of chickpea breeding lines to fusarium wilt under field sick-plot and greenhouse conditions at Tel Hadya during 1994/95.

Accessions	Accessions with score 0-4					
	0	1	2	3	4	Total
FLIP lines	4	59	523	436	1133	2155

3.1.2.1. Sources of resistance to Wilt

The following three types of breeding material were tested in our wilt-sick plot at Tel Hadya:

1. thirty lines which were found resistant in our collaborative projects with IRAT, Tunisia and Departamento de Patologia, Universidad de Cordoba, Spain, and included in the Chickpea International Fusarium Wilt Nursery and furnished to over 15 countries.
2. 64 breeding lines.
3. One hundred seventy-one lines obtained from crosses between early maturing lines and fusarium wilt-resistant

lines.

The susceptible check sown after every two test entries was uniformly killed by the wilt. The results are summarized in Table 3.1.2. where the three types of breeding material are referred to as A, B, and C.

K.B. Singh

3.1.2.1.5. Segregating material

Three F_4 bulks and 15 F_2 bulks were grown in the wilt-sick plot. Wherever seed was available, 1000 plants were grown for each bulk. Resistant plants were selected and will be grown the following season.

K.B. Singh

Table 3.1.2. Reaction of entries in the Chickpea International Fusarium Wilt Nursery (CIFWN) in Tel Hadya, 1995.

Rating scale	% of plant killed	Type of breeding material[a]		
		A	B	C
1	0	0	0	50
2	1-5	0	8	50
3	6-10	4	6	9
4	11-20	4	8	18
5	21-40	2	1	25
6	41-60	3	11	14
7	61-80	0	3	4
8	81-99	0	2	0
9	100	0	25	1
Total		30	64	171

3.1.2.2. Ascochyta blight

Ascochyta blight caused by *Ascochyta rabiei* is the most serious foliar disease of chickpea in the West Asia and North Africa region, particularly where low temperatures (15-25^0 C) prevail during the crop season. Its occurrence is irregular and is weather dependent. However, a good season for the chickpea crop is often favorable to ascochyta blight. Winter sowing of chickpea provides an opportunity to increase chickpea yield by almost 100%; unfortunately, it also increases the risk of ascochyta blight devastation. Therefore, control of ascochyta blight is essential to increase chickpea production and yield stability. Host resistance is the most practical and economic way to manage the ascochyta blight problem, but its level has to improve.

3.1.2.2.1. Evaluation of segregating populations for resistance to a mixture of six races

The reaction of F_2 to F_5 generations to the mixture of six races of ascochyta blight in the field at Tel Hadya is given in Table 3.1.3. The disease developed in epidemic form as was evident from the death of check lines throughout the nursery. No progeny was rated 1 or 2, but 159 progenies were rated 3. Another 1534 F_5 progenies were rated 4. These results indicate that there were sufficient resistant materials from which high yielding lines could be selected.

K.B. Singh

Table 3.1.3. eaction of F_2 to F_5 generations to Ascochyta blight at Tel Hadya, 1994/95.

Generation	Reaction on 1-9 scale									
	1	2	3	4	5	6	7	8	9	Total
F_2 Bulk	0	0	2	22	33	44	13	10	0	124
F_4 Bulk-L	0	0	0	18	24	18	1	0	0	61
F_4 Bulk-T	0	0	0	9	1	0	0	0	0	10
F_4 Bulk	0	0	0	7	13	13	0	0	0	33
F_5 Progenies-T	0	0	158	1058	766	234	14	1	1	2232
F_5 Progenies-E	0	0	0	187	588	317	28	0	0	1120
F_5 Progenies	0	0	1	289	2160	1588	92	16	2	4148

Scale: 1 = free from damage; 2 = highly resistant; 3 = resistant; 4 = moderately resistant; 5 = intermediate; 6 = moderately susceptible; 7 = susceptible; 8 = highly susceptible; and 9 = all plants killed.

3.1.2.2.2. Evaluation of breeding lines for resistance to a mixture of six races

Lines included in different trials were evaluated in the field against a mixture of six races during the 1994/95 season (Table 3.1.4). No line was free, but two lines had 2 rating and 18 lines had a rating of 3. Another 224 lines had 4 rating. Likewise, more than hundred lines were tolerant with 5 rating. There were about 10% lines from 490 lines with ratings of 6-9, indicating that the majority of lines identified resistant in the past were holding their resistance.

K.B. Singh

Table 3.1.4. Reaction of breeding lines in differenttrials to six races of *A. rabiei* at Tel Hadya, 1994/95.

	Disease Reaction on 1-9 scale									
Trial Name	1	2	3	4	5	6	7	8	9	Total
CIYT	0	0	5	16	0	0	0	0	0	21
CIYT-SPR	0	0	2	11	4	3	1	0	0	21
CIYT-SL1	0	0	3	9	7	2	0	0	0	21
CISN-W	0	0	10	34	11	6	0	0	0	61
CISN-SPR	0	0	0	48	10	1	2	0	0	61
CISN-SL1	0	0	0	27	11	7	1	0	0	46
PYT	0	0	0	79	114	54	8	3	1	259
Total	0	0	20	224	157	73	12	3	1	490

3.1.2.2.3. Reaction of the entries in the International Ascochyta Blight Nursery at Tel Hadya

Fifty lines were included in this nursery and their reaction is shown in the Table 3.1.5.

Table 3.1.5. International Ascochyta blight Nursery in the field at Tel Hadya, 1994/95.

	Disease Reaction on 1-9 scale									
	1	2	3	4	5	6	7	8	9	Total
No. of accessions		0	0	13	28	9	0	0	0	050

The susceptible check sown after every two test entries was uniformly killed throughout the nursery. Thirteen, 28, and 9 lines had a rating of 3, 4, and 5, respectively. Lines with 3 rating were FLIP 84-182C, FLIP 88-83C, FLIP 91-149C, FLIP 92-179C, FLIP 92-189C, FLIP 93-160C, FLIP 93-176C, ICC 4475, ICC 12004, ICC 13269, ICC 13416 and ICC 13555. These lines would be useful in breeding programs.

Table 3.1.6. Number of progenies / plants selected for ascochyta blight resistance in the gene pyramiding project at Tel Hadya, 1994/95.

Generation	Material sown	No. of plants with 1-3 ratings selected
F_5 progenies	99	0
F_3 progenies	100	243
F_2 bulks	5	201

The principal mechanism is to cross resistance sources of diverse origin assuming that they have different genes for resistance to pyramid the resistance genes. This program was initiated in 1989/90 and materials developed by the end of the 1994/95 season are shown in Table 3.1.6. During 1994/95 season five crosses were made, and 444 plants from F_2, F_3, and F_5 generations were selected. All plants had a rating of 3 or less on 1-9 scale, where 1 = free from damage and 9 = all plants killed. Three hundred and ninety-nine lines developed from the gene pyramiding project in the previou eason were evaluated in the greenhouse against the Pathotype III (Isolate no. 13) maintained by the Biotechnology Section of ICARDA. We found 61 lines with a rating of 2-4 at the seedling stage, whereas only four lines with a rating of 4 at the podding stage. These were S 94605, S 94627, S 94655 and ICC 13729.

K.B. Singh

3.1.2.1.3. Drought tolerance

Drought causes severe yield loss in chickpea. Lack of screening technique and rating scale for drought tolerance have restricted plant breeders from making progress. Therefore, a study was initiated in 1990 to develop a screening technique and rating scale, and to evaluate germplasm for drought tolerance.

Based on a screening technique involving (1) delayed sowing by three weeks during spring at a relatively dry site, (2) preliminary evaluation of materials on 1 (=esistant) to 9 (=susceptible) scale to discard susceptible lines, and (3) final evaluation of promising lines under stress (drought) and non-stress (supplemental irrigation) conditions to select drought-tolerant lines which perform well under both conditions a total of 4165 germplasm lines were evaluated in two stages in a period of six years.

Based on these criteria, 19 lines were considered drought tolerant (Table 3.1.7). These lines are capable of producing over one tonne seed yield per hectare under drought conditions and over two tonnes yield under non-stress conditions.

Table 3.1.7. Yield performance of previously identified drought-tolerant lines under rainfed and irrigated conditions at Tel Hadya during spring 1995.

Entry name	Days to flower (rainfed)	Rainfed yield (kg/ha)	Score[a]	Irrigated yield (kg/ha)
Resistant lines				
ILC 142	48	1426	3	2212
ILC 391	56	1166	5	1988
ILC 588	50	1113	4	2176
ILC 1306	54	1352	4	1950
ILC 1799	54	1135	4	2376
ILC 2216	51	1141	6	1952
ILC 2516	51	1171	4	1858
ILC 3550	50	1135	4	2055
ILC 3764	53	1246	4	2310
ILC 3832	52	1200	4	2022
ILC 3843	49	1332	4	2532
ILC 4236	50	1064	4	2325
FLIP 87-7C	49	1100	4	2520
FLIP 87-8C	49	1016	4	2368
FLIP 87-58C	46	1085	4	2197
FLIP 87-59C	48	1191	4	2245
FLIP 87-85C	49	1028	4	2075
FLIP 88-42C	49	1392	4	1858
ICC 4958	49	1194	5	1931
Check lines				
ILC 72	68	74	9	1445
ILC 1171	65	161	8	931
Mean		961.4		1854.7
SE±		119.2		214.4
C.V. (%)		21.5		20.0
LSD at $P \leq 0.05$		330.3		594.1

[a] 1 = free from damage, 5 = intermediate, 9 = all plants dried without producing any seed.

Out of these 19 lines, one line (ILC 142) was rated 3, 14 lines rated 4, one line rated 5, and one line had a rating of 6. Our results showed that all drought-tolerant lines were early flowering, but not all early lines were drought tolerant. What factor differentiate between these two groups will be an interesting topic for the future research.

These lines have been supplied to cooperators in West Asia, North Africa, and southern Europe for evaluation and selection under their conditions through the Chickpea International Drought Tolerant Nursery. At ICARDA, we are utilizing these lines to generate material with high yield, drought tolerance and disease resistance.

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3.1.2.1.3. Combined evaluation to seven stresses

The number of lines evaluated between 1978 and 1995 for different stresses are shown in Figure 3.1.2. The results include evaluations in the 1994/95 season for wilt and drought. Resistant sources have been identified for all stresses except to seed beetle and cyst nematode. Further evaluation of lines will continue only for wilt and drought because we believe there is scope to identify improved sources of resistance to these stresses.

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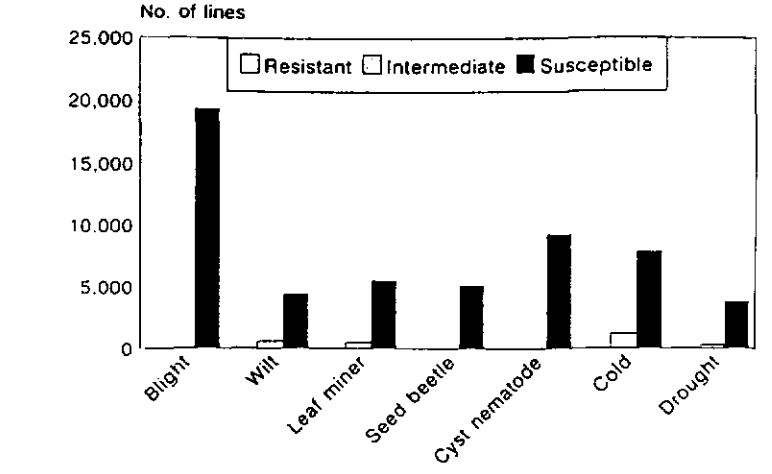

Resistant	32	110	8	0	0	13	85
Intermediate	9	591	485	0	20	1,191	265
Susceptible	19,329	4,473	5,532	5,153	9,237	7,891	3,815

Figure 3.1.2. Reaction of chickpea germplasm accessions to biotic and abiotic stresses at Tel Hadya, 1987-1995.

3.1.2.1.4. Multiple-Stress Resistant Chickpeas

Three germplasm lines, FLIP 91-178C, FLIP 93-53C and FLIP 93-98C, of kabuli chickpea with combined resistance to ascochyta blight, fusarium wilt, and cold have been bred by the ICRISAT-ICARDA Kabuli Chickpea Project in Syria. All lines have resistance to six races of *Ascochyta rabiei* pathogen from Syria, to Tel Hadya isolate of *Fusarium oxysporum* f. *ciceri* and to cold during winter sowing at low to medium altitude in Mediterranean environments.

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3.1.2.2. Wild species

3.1.2.2.1. Fusarium wilt

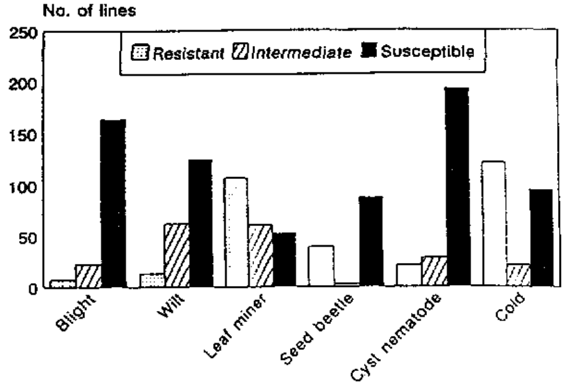
At ICARDA, 200 accessions of seven wild *Cicer* species were evaluated in the wilt-sick plot. Accessions with rating 0 and 1 (on 0 to 4 scale) were tested in pots in the green house. The results of two evaluations are given in Table 3.1.8. Only 3 accessions from *C. bijugum*, 7 from *C. judaicum*, 2 from *C. pinnatifidum* and 1 of *C. cuneatum* were resistant with rating 1 and all others rated two or more.

Table 3.1.8. Reaction of accessions of wild *Cicer* species to fusarium wilt under field sick-plot and greenhouse conditions at Tel Hadya during 1994/95.

Cicer species	Accessions with score 0-4					Total
	0	1	2	3	4	
bijugum	-	3	19	8	1	31
cuneatum	-	1	-	2	-	3
echinospermum	-	-	-	-	9	9
judaicum	-	7	30	20	4	61
pinnatifidum	-	2	11	17	13	43
reticulatum	-	-	1	2	46	49
yamashitae	-	-	1	2	1	4
Total	-	13	62	51	74	200

3.1.2.2.2. Combined evaluation for six stresses

Evaluation of eight annual wild *Cicer* species has been conducted to identify sources of resistance to multiple stresses. The results are summarized in Figure 3.1.3. Sources of resistance were found for all six stress factors. Wild species were the only source of resistance so far found to seed beetle and cyst nematode and had higher level of resistance than the cultivated species for fusarium wilt, leaf miner, and cold. The most important source for resistance to different stress factors was found in *C. bijugum*, while *C. yamashitae* was the least important.



	Blight	Wilt	Leaf miner	Seed beetle	Cyst nematode	Cold
Resistant	7	13	107	39	21	121
Intermediate	22	62	61	3	28	21
Susceptible	164	125	53	88	193	96

Figure 3.1.3. Reaction of wild Cicer accessions to seven stresses at Tel Hadya, 1987-1995.

3.1.2.2.3. Development of sources of resistance to biotic and abiotic stresses in chickpea and *Cicer* species

Known sources of resistance are shown in Table 3.1.9. Sources of resistance are available individually to all seven stresses for which research is being conducted. Sources with multiple stress-resistance have been identified in *Cicer* species. These sources have been shared with NARS through the distribution in the Legumes International Testing Program. They have been used in crossing programs at ICARDA and many national programs. High yielding lines with combined resistance to ascochyta blight and cold have been bred at ICARDA and shared with the national programs. Research at ICARDA has acted as a catalyst and many national programs are engaged in resistance breeding.

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Table 3.1.9. Resistance sources to biotic and abiotic stresses in chickpea developed at ICARDA.

Stresses	Sources of resistance
Single stresses	
Ascochyta blight	ILC (20 6482) ICC (4475,6328, 12004)FLIP(AB)(90-98C, 91-2C, 91-18C, 91-22C, 91-24C, 91-46C, 91-50C, 91-54C)
Fusarium wilt (FW)	ILC (267, 1278, 1300), FLIP (86-93C, 87-33C, 87-38C)
Leaf miner (LM)	ILC 3800, ILC 5901, ILC 7738
Seed beetle (SB)	ILWC 39, ILWC 181
Cyst nematode (CN)	ILWC 292
Cold (CO)	ILC 8262, ILC 8617
Drought (DR)	FLIP 87-59C
Multiple stresses	
AB, FW, CO	FLIP 91-178C, FLIP 93-53C, FLIP 93-98C
AB, LM, CO	ILWC 37 (*C. cuneatum*)
FW, LM, SB,CO	ILWC 39 (*C. echinospermum*)
AB, FW, SB, CN, CO	ILWC 62, ILWC 70, ILWC 73 (*C.bijugum*)
FW, LM, SB	ILWC 98, ILWC 102 (*C. judaicum*)
FW, CO	ILWC 141 (*C. reticulatum*)
FW, SB, CO	ILWC 112 (*C. reticulatum*), ILWC179, ILWC 181 (*C. echinospermum*)
AB, LM, CN	ILWC 250 (*C. pinnatifidum*)

3.1.3. Germplasm Enhancement

3.1.3.1. Improvement in shoot biomass yield

Low shoot biomass in the Mediterranean basin is an important reason among others for poor yield. Our previous results show that seed yield in chickpea is highly correlated with biomass yield (above ground plant parts). In the last decade the maximum biomass yield under Tel Hadya rainfed conditions was less than seven tonnes per hectare during winter sowing. Mostly the maximum biomass yield was around five tonnes per hectare. Another observation was that the biomass yield was usually greater in tall genotypes than in short bushy type.

Therefore, a project was initiated during 1989/90 to increase the biomass yield. The segregating material from six crosses was advanced by bulk-pedigree method and eight progenies were bulked in F_5 generation and these were tested for biomass yield and other agronomic characters during 1993/94 at Tel Hadya and 1994/95 at Tel Hadya and Terbol. The results are summarized in Table 3.1.10. Two lines, S 92249 and S 92260, produced biomass yield of 7135 and 7353 kg ha^{-1}, respectively compared to 6225 kg ha^{-1} produced by the tall check (ILC 3279). Although these two lines produced only 15 and 18% more biomass yield than the check, these yields were perhaps the largest ever produced under Tel Hadya rainfed conditions. These lines also produced about 20% higher seed yield than the check.

We continued making about six crosses anually. The F_5 progenies during 1994/95 were grown in the ascochyta blight nursery where about 200 mm water was provided by mist-irrigation during March and April to create the disease in epidemic form. Additional water not only helped disease development but also helped the resistant plants to grow taller and produce higher yield. We bulked 33 F_5 progenies and recorded a few observations.

The results of 10 selected lines are given in Table 3.1.11. Although these are the results from an unreplicated 4 m-long plots, yet they are indicative of the yield potential of this material. All lines produced more than 10 tonnes per hectare and one line produced over 13 tonnes biomass yield, a record under Tel Hadya conditions.

Table 3.1.10. The biological yield (kg ha^{-1}) and seed yield (kg ha^{-1}) of tall breeding lines at Tel Hadya and Terbol.

Entry	Tel Hadya		Terbol	Mean	% increase over check
	1993/94	1994/95	1994/95		
Biological yield					
S 92217	5428	6760	7161	6450	3.6
S 92218	6244	7224	6911	6793	9.1
S 92249	6297	6826	7351	7135	14.6
S 92260	7317	6757	7714	7353	18.1
S 92307	5889	6114	6554	6604	6.0
S 92310	5511	6852	6696	6545	5.1
S 92312	5000	6223	7042	6359	2.1
S 92440	6311	6521	7337	6796	9.2
ILC 3279 (ch.)	6328	6658	6690	6225	-
Mean	5925	6770	7050		
SE ±	312.5	370.7	180.5		
C.V. (%)	10.4	10.9	5.1		
LSD 5%	912.1	1082.2	527.0		
Seed yield					
S 92217	1951	2122	2054	2042	-2.0
S 92218	2122	2139	2071	2111	1.3
S 92249	2620	2276	2338	2487	19.4
S 92260	2848	2157	2357	2470	18.6
S 92307	2346	2434	2173	2349	12.8
S 92310	2101	2212	2030	2181	4.1
S 92312	1896	2080	2119	2125	2.0
S 92440	2081	2141	2172	2157	3.6
ILC 3279 (ch.)	1996	2238	2079	2083	-
Mean	2215	2200	2136		
SE ±	92.4	114.4	86.2		
C.V. (%)	8.3	10.4	8.1		
LSD 5%	269.7	434.0	251.6		

All these lines were nearly 100 cm tall. They also produced very high seed yield; one line produced 4.28 tonnes per hectare. These lines are being evaluated in a replicated test during 1995/96.

Table 3.1.11. Biomass yield along with other agronomic characters of 10 selected F_5 progenies at Tel Hadya, Syria, 1994/95.

Entry no. Sel. 95TH	Pedigree	PTHT[a]	PrBr	SecBr	BYLD	SYLD
3149	X92TH82/FLIP 86-10C x FLIP 89-77C	98	2.3	15.0	11111	3189
3727	X92TH182/(FLIP 88-62C x FLIP 85-15C) x S 91167	97	3.3	15.7	12000	3011
3744	X92TH182/(FLIP 88-62C x FLIP 85-15C) x S 91167	97	4.3	21.7	10667	3084
3752	X92TH182/(FLIP 88-62C x FLIP 85-15C) x S 91167	98	3.3	13.7	10222	2507
3822	X92TH183/(FLIP 88-70C x FLIP 85-15C) x S 91167	98	4.0	15.0	11556	2978
3828	X92TH183/(FLIP 88-70C x FLIP 85-15C) x S 91167	100	4.0	13.0	10501	2447
3851	X92TH183/(FLIP 88-70C x FLIP 85-15C) x S 91167	100	3.0	13.0	10370	2765
4097	X92TH188/(FLIP 89-63C x FLIP 85-45C) x FLIP 85-18C	98	2.7	15.0	13043	4280
4132	X92TH188/(FLIP 89-63C x FLIP 85-45C) x FLIP 85-18C	100	3.0	9.7	11556	2365
4187	X92TH190/(FLIP 89-67C x FLIP 83-46C) x S89TH 78998	97	3.3	15.7	12444	3689
ILC 3279 (Check)	Stepnoj-1	90	3.1	18.0	6721	1780

[a] PTHT = plant height, PrBr = primary branches/plant, SecBr = secondary branches/plant, BYLD = biological yield (kg/ha), SYLD = seed yield (kg/ha)

3.1.4. Development of Improved Germplasm

3.1.4.1. Breeding Methods:

Four breeding schemes are operating to develop improved germplasm for different agroclimatic zones. They are:

1. **The bulk-pedigree method of breeding for blight- and cold-resistance**. This method was adopted during the mid 1980s and it takes four years from crossing to bulking of new lines. It allows effective screening for cold and ascochyta blight during the main season. This type of material is suitable primarily in West Asia and North Africa for winter and spring sowing. Some of these materials are also useful in the Indian subcontinent.

2. **Two cycle selection for ascochyta blight and fusarium wilt**. After the development of wilt-sick plot at Tel Hadya, we have developed a two-cycle selection for ascochyta blight and fusarium wilt resistance. It allows selection for blight in one generation and for wilt in the next. This type of material is in demand for North Africa.

3. **Bulk-pedigree method of breeding for wilt- and drought-resistance**. This breeding scheme consists in delayed sowing in wilt-sick plot and harvesting in off-season when part of the crop has matured. Output from this project benefits East Africa.

4. **Generation of large-seeded, wilt-resistant germplasm**. We sieve to select large-seeded material while advancing method through bulking up to F_4 generation. The focus of this project is Latin America.

3.1.4.2. Performance of newly bred lines at ICARDA sites

Two hundred and forty one newly-bred lines were evaluated in four preliminary yield trials (PYTs) at two locations (Tel Hadya and Terbol) and in two seasons (winter and spring). Several lines were superior to checks in yield (Ghab 1 in winter and ILC 1929 in spring), although only a few were significantly better yielding (Table 3.1.12).

Table 3.1.12. Performance of newly developed lines during winter and spring lantings at Tel Hadya and Terbol, 1994/95.

Location and season	No. of trials	No. of entries: Tested	Exceeding check	Sig. exceeding check	Yield: Mean of location	Mean of highest yield (kg ha^{-1})	Range for: C.V. (%)	LSD ($P \leq 0.05$) (kg ha^{-1})
Tel Hadya								
-Winter	4	241	22	1	1988	2766	13-16	491-632
-Spring	4	241	5	0	545	968	15-21	307-399
Terbol								
-Winter	4	241	7	0	1889	2479	7-11	184-227
-Spring	4	241	6	0	1161	1996	14-20	318-418
Overall								
-Winter	-	-	-	-	1939	-	-	-
-Spring	-	-	-	-	853	-	-	-

The 1994/95 was a normal season, but rainfall distribution was poor. However, the yield in winter plantings was higher than previous years, and was lower in spring plantings as compared to previous years. Over the two locations, winter chickpea produced 1939 kg ha^{-1}, giving an increase of 127% over spring planting.

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3.1.5. Strategic Research

3.1.5.1. evelopment of early flowering and podding lines under low temperature

Chickpea is highly sensitive to low temperature as compared to faba bean, lentil and pea. The latter crops start flowering and podding much before chickpea when planted at the same time. This may be a distinct disadvantage to chickpea especially during dry seasons or seasons with low rainfall during the spring months. Therefore, an effort has been underway to develop early flowering lines under low temperatures at Tel Hadya since 1992/93. To achieve this goal, we made intraspecific and interspecific crosses and also made selections from early flowering germplasm. During the 1994/95 season, 118 plants selected from germplasm accessions and 125 plants selected from intraspecific hybridization were grown. Thirty plants from germplasm accessions and 69 plants from intraspecific hybridization flowered 10 days earlier than ILC 482 (early flowering check) and these were selected and harvested individually (Table 3.1.13).

Table 3.1.13. Materials sown for the development of early flowering lines under low temperature at Tel Hadya, 1994/95.

Material	Sown	Harvested
Early flowering germplasm	118 plants	30
Intraspecific hybridization	125 lines	69

3.5.1.2. Analysis of a decade of winter/spring chickpea

Chickpea is traditionally grown as a spring-sown crop in the Mediterranean region. The crop therefore suffers from heat and moisture stress as the season advances toward maturity, giving low and unstable yields. In contrast, sowing the crop in winter, with ascochyta blight-resistant and cold-tolerant cultivars, both reduces the effect of terminal heat and drought stresses, and increases and stabilizes productivity. Therefore, two studies were conducted to establish the gain in seed yield and other advantages adopting winter sowing in a Mediterranean environment.

In the first study, a comparison of spring versus winter sowing was made over 12 years (1983/84 to 1994/95) at three sites (Tel Hadya, Jindiress and Terbol), except in 1994/995 when the testing was done at Tel Hadya and Terbol using breeding lines (testing between 72 and 486 lines). The winters of 1984/85, 1988/89, 1989/90, 1991/92 were colder than normal and the springs of 1983/84, 1988/89, 1989/90, 1990/91, and 1992/93 (especially at Tel Hadya) were drier than normal.

The seed yield data in Figure 3.1.5 showed that winter-sown trials on average produced 1731 kg/ha against 1009 kg of spring-sown trials, giving 72% or 722 kg/ha more yield. The yield differences between winter and spring were larger during dry seasons than in normal or wet seasons. During an abnormally cold year (1984/85), yields of winter-sown trials were lower than spring-sown trials. This trend, however, was reversed after the very cold 1988/89 season, because of selection for cold tolerance since 1984/85. Many lines produced more than 4 t ha^{-1} seed yield during winter, especially in the favorable environment at Terbol. Clearly, there is a big advantage of winter sowing over spring.

In the second study, over 20 ascochyta blight-resistant and cold-tolerant kabuli chickpea breeding lines were compared in winter and spring sowing for 10 years (1983-1984 to 1992-1993). Trials were held at two locations in Syria (Tel Hadya and Jindiress), and one in Lebanon (Terbol). These 22 to 24 entries were sown rainfed in both winter and spring at all three locations, in randomized complete block designs with two replications. Correlation between different traits, and stepwise regression of seed yield on other traits, were determined using SAS package.

On a 10-year average, winter-sown chickpeas produced

70% (or 692 kg ha^{-1}) more seed yield than spring-sown crop. The longer growing period of winter-sown chickpea resulted in higher biomass production, which mainly contributed towards increased seed yield. The yield potential of lines sown during winter was approximately 4000 kg ha^{-1}, and yields were more stable than in the spring-sown crop. The correlation between seasonal rainfall and seed yield was positive and significant ($P \leq 0.01$) in both seasons. The values were slightly higher in spring ($r = 0.46$, $P = 0.01$) than in the winter-sown crop ($r = 0.43$, $P = 0.02$). In 1988-1989, when the Tel Hadya site experienced severe drought (seasonal precipitation 234 mm), spring-sown crop resulted in virtually no seed yield, whereas winter-sown crop produced an average yield of 542 kg ha^{-1} by escaping partially the severe drought. Winter-sown crop has higher symbiotic nitrogen fixation; and the plants are taller than in spring sowing, permitting harvesting by combines. Because of these advantages, the winter sowing of chickpea is gaining popularity in the region around the Mediterranean sea.

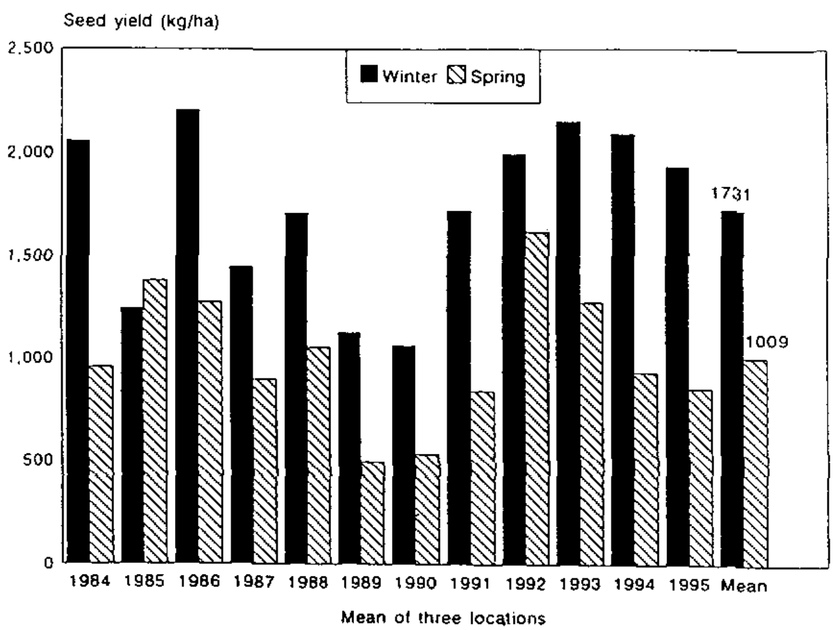

Figure 3.1.5. Mean seed yield (kg ha-1) of chickpea grown in winter and spring at three locations in eleven years and two locations in the twelfth year.

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3.1.5.3. Analysis of data on the effect of supplemental irrigation on winter-sown chickpeas

In the dry Mediterranean environments of the West Asia and North Africa region, the use of limited irrigation to supplement rainfall (supplemental irrigation) to augment productivity and yield stability of field crops is becoming increasingly popular. Chickpea, an important pulse crop of the region, often suffers from drought and can benefit from such a practice. To investigate the differences in response to supplemental irrigation, field experiments were conducted at Tel Hadya, Syria, from 1985-1986 to 1987-1988, using 24 improved chickpea genotypes sown in winter. Scheduling of irrigation was done using daily water balance computed from rainfall and pan evaporation. The seasonal rainfall was 316, 358, and 504 mm and supplemental irrigation was 130, 120, and 80 mm in 1985-1986, 1986-1987, and 1987-1988, respectively. Averaged over three seasons, supplemental irrigation increased the mean seed yield by 916 kg ha^{-1} (44.0%) over rainfed condition. Genotypes ILC 464, ILC 1272, ILC 3256, ILC 4291, and ILC 147, with respective mean seed yields of 3877, 3726, 3208, 3266, and 3246 kg ha^{-1} under supplemental irrigation had high response to the irrigation and their response to irrigation over years was predictable. Above ground biomass contributed 49% of total variation in seed yield under irrigated condition, followed by plant height (26%) and early maturity (16%). The study showed that it is possible to breed chickpea for improved response to supplemental irrigation, and this practice can help in maximizing the realization of yield potential in winter sowing of chickpea in the lowland Mediterranean drylands.

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3.5.1.4. Genetic diversity and phylogenetic relationships among the annual *Cicer* species as revealed by isozyme polymorphism

There are few estimates of genetic variability within and among populations of the nine annual *Cicer* species and for the wild species this information is based on few accessions. The present study was undertaken to examine genetic variation within and between annual *Cicer* species. One hundred and thirty-nine accessions of nine annual *Cicer* species were used for electrophoretic analysis at ICARDA. High polymorphism in all wild annual *Cicer* species was found. However, for the cultigen, among 14 loci assayed, only two were polymorphic, ADH and PGD2. This is in contrast to earlier research which had shown high polymorphism only in *C. reticulatum*. *Cicer reticulatum* had the highest proportion of polymorphic loci. The nine species formed four phylogenetic groups based on genetic distances. The first group comprised *C. arietinum*, *C. reticulatum* and *C. echinospermum*, the second *C. bijugum*, *C. judaicum* and *C. pinnatifidum*, the third *C. chorassanicum* and *C. yamashitae*; the fourth group consisted of one species, *C. cuneatum*. Genetic distance data supported a closer relationship of the cultigen to *C. reticulatum* than to *C. echinospermum*. Genetic diversity data showed that the greatest diversity was within *C. reticulatum* and the lowest with the cultigen, *C. arietinum*. With the exception of *C. reticulatum*, genetic diversity increased with genetic distance from the cultigen. Little geographic variation in genetic diversity was found.

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3.5.1.5. Race identification of *Fusarium oxysporum* f. sp. *ciceri* in chickpea in the Mediterranean region

Wilt caused by *Fusarium oxysporum* f.sp. *ciceri* is the second most important disease of chickpea in the Mediterranean region. In countries, such as Morocco, Spain and Tunisia, it can cause serious damage to the crop. ICRISAT has conducted extensive research in India on this disease. But the progress in cultivar development has been seriously hampered due to presence of different races of the pathogen at different locations. Research done at Cordoba, Spain has shown the presence of three races (race 0, 5, 6) in Spain alone. It is expected that many more exist in the region. Systematic evaluation of germplasm accessions in Tunisia and Spain has resulted in identification of a number of chickpea genotypes resistant to wilt. These resistance sources have been made available to scientists in the region and are being used in breeding programs. Breeders and pathologists may eventually develop resistant cultivars, but they may undergo the same fate as those cultivars developed at ICRISAT. Therefore, it is important that we know the distribution of the various races in the Mediterranean region. Therefore, the objective of this study is to characterize races of *Fusarium oxysporum* f. sp. *ciceri* prevailing in Algeria, Egypt, Ethiopia, Lebanon, Mexico, Morocco, Spain, Sudan, Syria and Tunisia.

Research activities in the collaborative project have dealt with the characterization of isolates of *Fusarium oxysporum* form chickpea in Algeria, Morocco and Tunisia by means of biological pathotyping and genetic fingerprinting using RAPD-PCR in Spain. Isolates were provided by collaborators in Algeria (7), Morocco (7), and Tunisia (20) either as pure cultures or as stem pieces from affected plants. Isolations were carried out from stem pieces and all cultures of *F. oxysporum* were single spored. Pathogenicity studies were carried out with 17 of the 34 isolates by means of standardized pot-culture inoculation method. Isolates pathogenic to chickpea were further characterized to pathogenic race by means of standardized inoculation of race differential lines.

All isolates from Algeria and one each from Morocco and Tunisia were nonpathogenic. Pathogenic isolates from Morocco (6) were characterized as races 1 and 6, while those of Tunisia (2) were characterized as races 0 and 1. Studies are *in progress* to further characterize isolates from Tunisia.

R. Jimenez-Diaz and K.B. Singh

3.1.6. Interspecific Hybridization

3.1.6.1. Yield Improvement

Wild species have significantly contributed to crop improvement including seed yield. However, exploitation of wild species is confined to major cereal and cash crops. In chickpea, efforts to exploit wild *Cicer* species are recent. Therefore, an attempt was made to increase seed yield in chickpea through the introgression of genes from wild relatives and results of eight years study are summarized here.

Two crosses, ILC 482 (*C. arietinum*) x ILWC 179 (*C. echinospermum*) and ILC 482 x ILWC 124 (*C. reticulatum*), between carefully chosen parents were made during 1987-88. Also, two reciprocal crosses between them were made. Pedigree method was followed to advance the segregating generations. Heterosis was visually observed in F_1 and the single plant measurement for seed yield was recorded in F_2 populations. Selections were made in subsequent generations for characters like the cultigen. Promising and uniform progenies were bulked in F_5 generations. Ninety-six F_6 lines were evaluated for seed yield and other agronomic characters in replicated test. In F_7, 22 lines were evaluated in a replicated trial for seed yield and many agronomic and morphological characters.

Abundant heterosis was present in F_1. Several F_2 plants produced two to three times more seed yield than the best plant from the cultigen. Twenty lines produced more seed yield than the cultigen parent in F_6.

Nine F_7 lines produced higher seed yield than the cultigen (Table 3.1.14). However, only one line, which produced 39% higher yield than the cultigen (ILC 482), was significantly different. Sixteen lines had higher biological yield than the cultigen. Fifteen lines were heavier in seed weight by 15 to 39 % than the cultigen. Most of the new lines were phenotypically similar to ILC 482 in maturity, plant height, harvest index, and seed protein content. The wild relatives had significantly lower seed yield and harvest index, and also they had shorter plant height and smaller seed weight than derivatives from interspecific crosses.

All derived lines were similar to the cultigen parent in growth habit (erect to semi-spreading), uniformity in maturity, and non-pod dehiscence (Table 3.1.15). Whereas, the two wild species had spreading growth habit, non-uniform maturity and dehisced pod.

Accessions of wild *Cicer* species have seed dormancy of varying length, though the wild progenitor, *C. reticulatum*, have partial seed dormancy. When the test was conducted three months after harvest, the derived lines had the same germination percentage as that of the cultigen parent. On the other hand, *C. echinospermum* parent had 0 % germination and *C. reticulatum* parent had

13 % germination (Table 3.1.16). All derived lines had seed quality like that of cultigen parent.

Table 3.1.14. Seed yield and yield related characters of 22 F_7 lines derived from interspecific crosses involving *C. arietinum* (ILC 482) and *C. reticulatum* (ILWC 124) and *C. echinospermum* (ILWC 179) ranked in order of decreasing seed yield at Tel Hadya, 1994/95.

Line	Cross	DFL[d]	DMA	HGT cm	BYD kg/ha	SYD kg/ha	HI %	SW g
55	RA[c]	107	161	43	7415	3420	46	25.2
126	AE	103	161	42	6925	3160	46	31.1
41	AE	110	163	47	6762	2850	42	35.9
63	RA	109	159	42	6258	2711	43	25.3
90	AE	106	157	40	5265	2706	51	35.0
118	AE	110	159	49	6350	2687	43	33.0
86	AE	106	158	44	5664	2585	46	27.9
49	RA	111	160	43	5755	2584	44	22.3
95	AE	108	159	40	5510	2487	45	16.2
482[a]	A	106	158	42	4966	2458	50	26.7
50	RA	109	161	42	5538	2457	44	29.7
17	AR	114	160	50	6476	2450	38	26.7
5	RA	100	163	42	4966	2416	48	26.9
79	AE	107	163	53	6408	2371	37	30.8
82	AE	114	160	38	5660	2367	41	33.3
67	RA	110	163	45	6490	2355	36	26.9
80	AE	110	162	47	6041	2343	38	26.3
81	AE	110	161	50	5633	2324	41	32.3
53	RA	106	158	43	4673	2226	46	28.7
130	AE	110	160	47	5003	2077	41	24.8
51	RA	110	163	47	4871	2034	46	37.1
96	AE	110	162	43	5286	1997	38	27.1
18	AR	109	160	39	4599	1819	40	29.5
124[b]	R	106	160	27	6289	662	14	12.3
179[b]	E	120	157	13	1644	424	30	13.3
S.E.		1.1	1.2	4.4	201.2	331.3	2.7	1.31
C.V.		1.8	1.3	17.9	25.3	23.2	11.4	8.32

[a] ILC line
[b] ILWC line
[c] A = *C. arietinum* (ILC 482), R = *C. reticulatum* (ILWC 124), E = *C. echinospermum* (ILWC 179), AE = A × E and AR = A × R.
[d] DFL = days to flower, DMA = days to maturity, HGT = plant height, BYD = biological yield' SYD = seed yield' HI = harvest index, SW = 100-seed weight.

Results of two years trials of 22 test lines and three parents for seed yield, 100-seed weight, and plant height are shown in Figure 3.1.6. The performance of lines was different in two years because of the climatic differences. However, 13 lines produced higher seed yield than ILC 482. There were several test lines which surpassed ILC 482 in 100-seed weight and plant height. Of course, the wild relatives were poor in all three attributes.

Deleterious genes introduced from *C. echinospermum* and *C. reticulatum* to the cultigen were less of a problem than expected. Therefore, backcross was not considered imperative.

Table 3.1.15. Wild species related characters of 22 F_7 lines derived from interspecific crosses involving *C. arietinum* (ILC 482) and *C. reticulatum* (ILWC 124) and *C. echinospermum* (ILWC 179) ranked in order of decreasing seed yield at Tel Hadya, 1994/95.

Line	Cross	Characters evaluated			
		UMAT[c]	GRH	PDD	GRM
55	RA	u	semi-erect	nd	97
126	AE	u	erect	nd	95
41	AE	u	semi-erect	nd	94
63	RA	u	erect	nd	97
90	AE	u	semi-erect	nd	93
118	AE	u	erect	nd	89
86	AE	u	erect	nd	99
49	RA	u	semi-spreading	nd	98
95	AE	u	erect	nd	96
482[a]	A	u	erect	nd	99
50	RA	u	erect	nd	89
17	AR	u	erect	nd	52
5	RA	u	semi-spreading	nd	81
79	AE	u	semi-erect	nd	67
82	AE	u	semi-spreading	nd	90
67	RA	u	semi-erect	nd	75
80	AE	u	erect	nd	100
81	AE	u	erect	nd	89
53	RA	u	erect	nd	99
130	AE	u	erect	nd	70
51	RA	u	erect	nd	74
96	AE	u	semi-erect	nd	65
18	AR	u	semi-spreading	nd	81
124[b]	R	nu	spreading	d	13
179[b]	E	nu	spreading	d	0
S.E.					7.5
C.V.					11.5

[a] ILC line
[b] ILWC line
[c] UMAT = uniformity in maturity, GRH = growth habit, PDD = pod dehiscence, GRM = percent of germination, u = uniform, nu = non-unifom, nd = non-dehiscent, d = dehiscent.

Table 3.1.16. Quality characters of 22 F_7 lines derived from interspecific crosses involving *C. arietinum* (ILC 482) and *C. reticulatum* (ILWC 124) and *C. echinospermum* (ILWC 179) ranked in order of decreasing seed yield at Tel Hadya, 1994/95.

Line	Cross	Characters evaluated					
		PRO[c] %	SSH	STY	SCOL	SW g	CT min.
55	RA	18.1	owl	K	beige	25.2	140
126	AE	18.6	owl	K	beige	31.1	148
41	AE	18.5	owl	K	beige	35.9	143
63	RA	18.2	owl	K	beige	25.3	130
90	AE	19.4	owl	K	beige	35.0	138
118	AE	18.6	round	I	beige	33.0	132
86	AE	18.0	owl	K	beige	27.9	135
49	RA	18.0	owl	K	beige	22.3	147
95	AE	18.7	round	I	beige	16.2	115
482[a]	A	17.9	owl	K	beige	26.7	143
50	RA	18.6	round	I	beige	29.7	138
17	AR	18.7	round	I	beige	26.7	133
5	RA	17.9	round	I	copper	26.9	120
79	AE	18.3	round	I	orange	30.8	138
82	AE	18.5	owl	K	beige	33.3	143
67	RA	19.9	round	I	copper	26.9	125
80	AE	18.3	owl	K	beige	26.3	142
81	AE	18.3	round	I	beige	32.3	138
53	RA	17.8	owl	K	beige	28.7	138
130	AE	18.5	owl	I	brown	24.8	128
51	RA	18.4	owl	K	beige	37.1	127
96	AE	18.4	owl	K	beige	27.1	132
18	AR	18.7	round	I	brown	29.5	145
124[b]	R	20.1	owl	D	brown	12.3	115
179[b]	E	18.5	owl	D	brown	13.3	138
S.E.		0.33				1.31	5.2
C.V.		2.96				8.32	6.7

[a] ILC line
[b] ILWC line
[c] PRO = protein content, SHH = seed shape, STY = seed type, SCOL = seed color, SW = 100-seed weight, CT = cooking time, K = kabuli, D = desi, I = intermediate.

Comparisons between reciprocals of crosses of cultigen with *C. echinospermum* showed that positive transgressive segregants for seed yield were recovered only from the cross using cultigen as a female parent, suggesting strong maternal effect of this species. But no such difference was noticed in crosses between cultigen and *C. reticulatum* and their reciprocal. In general, both wild species contributed equally towards the recovery of superior lines and the differences between the species were non-significant.

The presence of transgressive segregation for seed yield and other agronomic traits would suggest genetic complementarity between *C. arietinum*, *C. echinospermum* and *C. reticulatum*. This would foster hope of better segregants compared to those already obtained.

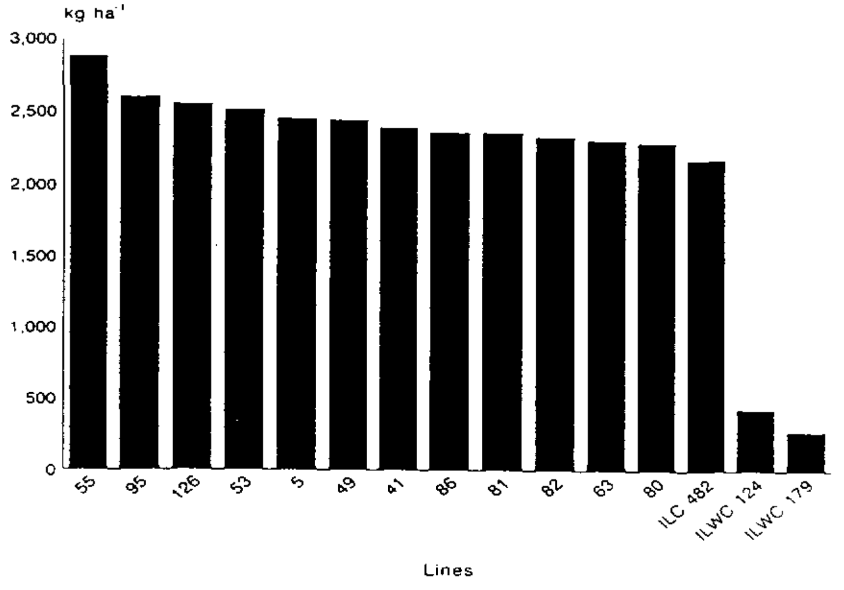

Figure 3.1.6. Averge seed yield of F_6 and F_7 lines grown during 1993/94 and 1994/95 at Tel Hadya, Syria.

3.1.6.2. Cyst nematode resistance

No sources of resistance to cyst nematode were found in 9,257 accessions of cultivated species which were evaluated at ICARDA. But when wild *Cicer* accessions were evaluated, 22 accessions of *C. bijugum*, five of *C. pinnatifidum* and one of *C. reticulatum* were found resistant to cyst nematode. Since cyst nematode is a serious problem in parts of the chickpea growing area and crosses between cultigen and resistant accession of *C.*

reticulatum were made during 1990/91 to transfer genes for cyst nematode resistance to cultigen. Materials screened during 1994/95 are shown in Table 3.1.17.

Seventeen plants were selected from F_2 backcross. One hundred sixty-six plants were selected from F_5 and 25 plants from F_7. Observations in F_7 were recorded along with checks on growth habit, days to flower, maturity, pod dehiscence, plant height, seed color, seed shape, and seed size. Most of the characters in F_7-derived lines were similar to those of the cultigen parent except seed color, shape and size, which were intermediate between desi and kabuli types.

After we identified ILWC 119 (*Cicer reticulatum*) resistant to cyst nematode, we discovered this line to be a mixture. Therefore, 220 plants were sown in the plastic house for purification and from these 27 resistant plants were selected. Resistance in these plants were confirmed and a new accession number ILWC 292 was assigned to bulk of 27 resistant plants.

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3.1.6.3. Cold tolerance

Materials sown during 1994/95 for cold tolerance study included Chickpea International Cold Tolerance Nursery, segregating populations of intraspecific crosses (50 F_7-F_{10}, 59 F_6, 124 F_5, 116 F_4 lines, and 109 F_3 bulks) and interspecific crosses (102 F_7-F_8, 82 F_6, 36 F_5, 193 F_4, 35 F_3, 135 F_6 of backcross one, and 213 F_3 bulk of backcross two). The 1994/95 season was warm, hence no selection for cold tolerance was possible. Some material from all generations were harvested for the next season.

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3.1.7. Protein quality

It is our endeavor to develop cultivars with the same or higher protein content as of the check cultivar. To meet this objective, we analyze newly developed lines for protein content.

Table 3.1.17. Reaction of plants from interspecific crosses in F_2 backcross, F_5, and F_7 generations to cyst nematode in the greenhouse at Tel Hadya, 1994/95.

Generation	Cross no.	Parents	Scale[1]						
			0	1	2	3	4	5	Total
F_2 backcross	X 94TH188	(ILC 482 x ILWC 119)x ILC 482	0	0	13	157	74	30	274
	X 94TH189	(FLIP 87-69C x ILWC 119)x FLIP 87-69C	0	0	4	34	49	25	112
	X 94TH190	(ILC 482 x ILWC 119)x ILC 482	0	0	0	3	83	118	204
	Total		0	0	17	194	206	173	590
F_5	X 90TH571	ILC 482 x ILWC 119	10	85	452	796	69	0	1412
	X 90TH572	FLIP 87-69C x ILWC 119	3	35	216	601	37	0	892
	X 93TH163	ILWC 119 x FLIP 88-85C	7	26	37	48	0	0	118
	Total		20	146	705	1445	106	0	2422
F_7	X 90TH572	FLIP 87-69C x ILWC 119	3	22	51	117	0	0	193

1/ Scale 0 = no cyst formation on roots, 5 = $\geq$ 50 cysts on roots.

Since earlier studies had indicated no significant effect of seasons on the protein content, only newly developed lines grown at Tel Hadya during winter were tested for protein content in seed. The protein contents of the 239 newly developed lines were similar to that of the check cultivar.

K.B. Singh

3.2. Chickpea Pathology

The objectives of chickpea pathology are to (1) assist chickpea breeders in the development of high yielding, disease-resistant cultivars and evaluate breeding material for resistance to the major diseases of chickpea; (2) collect information on disease epidemiology and pathogenic variability and develop disease management strategies for ascochyta blight; (3) collect information and monitor disease incidence and severity in the WANA region in collaboration with the national scientists; and (4) develop research collaboration with national programs on disease management of ascochyta blight and wilt. The screening for disease resistance has been discussed in the breeding section of the project report. The screening for Ascochyta blight resistance is discussed in the breeding section of the project report.

3.2.1. Disease management

Host-plant resistance is the cheapest and most practicalway controlling ascochyta blight. While laying major emphasis on the development of blight-resistant germplasm, efforts are under way to find other methods of disease control. Therefore, an experiment, including known methods of disease control, was conducted at Tel Hadya. The plot size was 4 m, 8 rows spaced at 0.45 m apart. Randomized complete block design was followed with three replications. Tecto was used for seed dressing and Bravo 500 for fungicide spray.

The results are summarized here in Table 3.2.1. Sowing dates did not influence the disease incidence. The cultivar Ghab 3 had a mean rating of 3.04 compared to 9 of ILC 1929, which was killed. The seed dressing with Tecto and fungicide spray at flowering and podding made no difference on disease development. The seed yield was significantly higher during the December sowing than the January sowing. On average, Ghab 3 produced 1909 kg ha^{-1} seed yield, whereas susceptible cultivar produced none. This experiment will be repeated with certain modifications, such as variation in number of fungicidal application.

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Table 3.2.1. Influence of sowing date, variety, seed dressing and fungicide spraying on disease development and seed yield at Tel Hadya, 1994/95.

Treatment		Rating 1-9 Scale	Yield (kg ha^{-1})
Sowing:	December	6.04	1035
	January	6.00	872
Variety:	Ghab 3	3.04	1909
	ILC 1929	9.00	0
Seed dressing:	Tecto	6.04	952
	Control	6.00	957
Spraying:	At flowering	6.04	972
	At podding	6.00	937
S.E.		± 0.029	± 70.9
L.S.D. at 5%		0.059	144.7

Scale: 1 = free from any damage, 9 = all plants killed.

3.2.2. Evaluation of lines developed by pyramiding of genes for resistance to ascochyta blight in the greenhouse

Three hundred and ninety-nine lines were evaluated along with a susceptible check, ILC 263, against pathotype III (Isolate no. 13) supplied by the biotechnology section. Lines developed earlier were evaluated against a mixture of six races from Syria under field conditions. The materials were evaluated at seedling and podding stages. For each stage the evaluation was done twice and the higher rating between the two was taken as the average. The experiment was conducted following the alpha design. The results are presented in Table 3.2.2. At seedling stage, 10, 23 and 28 lines were rated 2 (highly resistant), 3 (resistant) and 4 (moderately resistant), respectively. Whereas at podding stage, only four lines had moderately resistant reaction. These lines were S 94605, S 94627, S 94655 and ICC 13729. They should be used extensively in breeding germplasm for durable resistance.

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Table 3.2.2. Evaluation of lines developed by pyramiding of genes for resistance to ascochyta blight in the greenhouse, 1994/95.

Rating scale	Seedling stage	Podding stage
1	0	0
2	10	0
3	23	0
4	28	4
5	60	19
6	168	169
7	102	167
8	8	35
9	0	5
Total	399	399

3.2.3. Development of differentials to identify races of *A. rabiei*

In an attempt to develop differentials to identify ascochyta blight races, 26 genotypes with known differential reactions were chosen and grown in the greenhouse with three replications using randomized complete design. All genotypes were evaluated against six races individually and in mixture. At the end, genotypes were classified as resistant (R = $\leq$ 4), intermediate (I = >4 to < 6), and susceptible (S = $\geq$ 6). The results are shown in Table 3.2.3. The differences in reaction of genotypes are clear. Since this is the first year, it is not possible to suggest this set to be used as a standard differential set for identification of races in *Ascochyta rabiei*.

K.B. Singh

Table 3.2.3. The reaction of genotypes used as differentials for identification of Ascochyta rabiei races expressed as the average disease severity rating and the resistance group (R,I,S) in the greenhouse, 1994/95.

Entry name	Race 1	Race 2	Race 3	Race 4	Race 5	Race 6	Mixture 6 races
ILC 72	R(2.6)	R(2.0)	I(5.4)	R(2.2)	R(2.1)	R(2.8)	I(5.1)
ILC 182	R(3.7)	R(2.5)	I(5.1)	R(3.7)	R(2.5)	R(3.6)	I(4.8)
ILC 190	S(7.1)	R(2.9)	S(6.8)	S(6.5)	R(2.4)	S(6.0)	S(6.1)
ILC 194	R(3.7)	R(3.5)	S(6.0)	S(6.6)	R(3.7)	I(4.2)	S(6.9)
ILC 195	I(5.0)	R(2.0)	I(4.5)	R(2.7)	I(5.1)	R(3.6)	I(5.9)
ILC 200	R(3.6)	R(2.1)	R(3.7)	R(2.1)	R(2.6)	R(1.3)	I(4.1)
ILC 202	R(1.3)	R(2.3)	I(5.5)	R(2.2)	R(2.1)	R(2.4)	I(4.9)
ILC 215	S(6.2)	I(5.7)	S(7.3)	S(7.9)	I(4.5)	S(6.3)	S(7.8)
ILC 482	S(6.5)	R(2.2)	S(6.3)	I(5.9)	R(2.3)	I(5.5)	S(6.6)
ILC 1929	S(8.1)	S(8.6)	I(5.7)	S(8.5)	S(8.1)	S(7.3)	S(8.1)
ILC 2506	R(3.2)	R(2.8)	I(5.5)	R(1.4)	I(5.1)	R(2.9)	I(4.6)
ILC 2956	R(3.8)	R(2.2)	R(3.9)	R(1.6)	R(3.2)	R(2.7)	I(4.9)
ILC 3279	R(3.1)	R(2.4)	I(4.1)	R(2.3)	R(2.5)	R(3.6)	I(4.5)
ILC 3856	R(2.8)	I(3.2)	I(5.5)	R(2.3)	I(4.1)	R(2.7)	I(5.4)
ILC 5263	R(2.9)	R(3.7)	R(3.9)	R(2.1)	R(3.7)	R(3.4)	I(5.1)
ILC 5894	R(2.3)	R(2.6)	I(4.6)	R(2.1)	R(2.5)	I(4.2)	I(5.0)
ILC 5928	R(2.1)	R(2.6)	I(5.2)	R(2.0)	I(4.8)	R(1.9)	I(5.1)
ILC 6260	R(2.3)	-	-	-	-	-	-
ILC 6287	R(2.9)	R(1.3)	I(5.6)	R(2.8)	R(3.1)	R(2.1)	I(5.5)
ILC 7374	R(3.4)	R(2.1)	R(3.8)	R(2.1)	R(2.7)	R(3.4)	I(5.3)
ILC 7795	R(3.1)	R(2.3)	I(5.0)	R(2.1)	R(2.1)	R(2.5)	I(4.7)
ICC 1903	R(4.0)	R(2.7)	I(5.5)	I(4.5)	S(6.2)	S(6.0)	I(5.1)
ICC 3996	R(2.)1	R(2.0)	R(2.0)	R(2.1)	R(2.0)	I(4.3)	R(2.9)
ICC 4475	I(4.3)	R(2.0)	R(2.0)	R(2.6)	R(2.4)	I(5.4)	I(4.7)
ICC 12004	R(3.1)	R(2.0)	R(2.1)	I(4.6)	R(2.0)	R(3.3)	R(3.4)
F8	I(4.1)	R(3.3)	S(6.9)	I(5.8)	R(3.3)	I(5.1)	I(5.3)
PCH 15	S(6.9)	R(3.0)	S(6.1)	S(7.2)	I(5.0)	S(6.4)	S(6.3)

R = resistant ≤ 4; I = intermediate > 4 < 6; S = susceptible ≥ 6.

3.3. Application of Molecular Techniques

3.3.1. Pathogenicity Survey of Ckickpea Ascochyta Blight in Syria Using DNA Markers

Periodical pathogenicity survey is important to monitor the evolution of new pathotypes in chickpea growing regions. A pathogenicity survey helps in the identification and deployment of suitable genes of resistance and also in the formulation of suitable breeding strategies for efficient management of the disease (also see under the section 3.1.1.). Conventionally, pathogenicity of the pathogen is measured as an interaction effect on a set of differential chickpea cultivars with different levels of resistance. Based on the pathogenicity test, the pathogen is classified into pathotypes. This method has drawbacks: a) the results are very often influenced by the environment, i.e. epiphytotic condition prevailed/provided during inoculation and subsequent disease development. b) Often it is not possible to compare pathogenicity of isolates of different countries of origin simultaneously, because of quarantine restrictions. These limitations can be over come by the use of DNA markers, namely, (RFLP) or Restriction Frogment Length Polymorphisms polymerase chain reaction (PCR)-based the Random Amplified Polymerphic DNA (RAPDs). Since DNA markers detect variation directly at the DNA level, they are not influenced by the environment (test conditions). Moreover, PCR-based RAPD is fast, less cumbersome, and very little template DNA is required for the analysis; and therefore a very large number of samples can be handled in a very short period of time. These properties make DNA markers, especially RAPDs, very useful for studying pathogenic variability. Previously we have established the relationship between genotype (RAPD and RFLP pattern) and pathotype (phenotype) of the pathogen, using a set of periodically sampled isolates (see Ann. Report, LP, ICARDA, 1993 and 1994). Using this information, the pathogenicity survey was conducted 1994 and 1995, as follows:

i) Sampling sites in Syria

The stem symptoms (lesion on stem) were collected from ascochyta blight infected fields scattered all over the chickpea growing regions of Syria. The sampling sites are presented in Table 3.3.1.

ii) Genotyping of the isolates using DNA markers

Pure cultures of the fungus was isolated from individual stem lesions. The pure cultures were further single spore purified. DNA was extracted from the single spore-derived cultures. The DNA was used for RFLP analysis and for RAPD analysis using pathotype specific primers.

Table 3.3.1. Occurrence of genotype H in different chickpea growing regions of Syria.

sampling site	No.of isolates		other genotype
	analyzed	genotype H	
Year 1994			
Tel Hadya	6	5	1
Haasund/Malkieh	4	4	0
Ein Diven/Malkieh	3	3	0
Sachie/Malkieh	3	3	0
Rouge	1	1	0
Hemo/Malkieh	6	6	0
Alkamia	3	2	1
Afrin*	9	9	0
Jinderess	2	2	0
Ghab (ILC 482)	5	5	0
Ghab (ILC 3279)	7	5	2
Ghab (F 84-15C)	26	20	6
Afrin* (GHAB 3)	6	5	1
Year 1995			
Idleb	5	5	0
Gellin	6	6	0
Jable	2	2	0
Homs	3	2	1
Izraa	5	4	1
Ghab	7	2	5
Rouge	7	7	0
Hama	6	6	0
Hemo (spring local)	2	2	0
Hemo (Ghab 1)	2	1	1
Hemo (Ghab 2)	2	2	0
Tel Hadya*	3	3	0
Tel Hadya	11	11	0
Sqielbiye	6	6	0
Aljameleh	5	4	1
Jinderess	2	2	0
Alkamia	2	2	0
Alkamia	1	0	1
Azaz	5	1	4
Total	163	136	27
Percentage (%)	100	83.4	16.6

* farmer's field.

The primer OPJ-01 was used to identify the genotype H (see previous Ann. Report, LP, ICARDA, 1994) of the pathogen (Table 3.3.2; Figure 3.3.1). The salient findings are as follows:

1. The genotype H of the pathogen predominates (83.4%) in

both 1994 and 1995 in all the sampling sites, indicating clonal origin of the genotype, most possibly disseminated through infected seeds. Seed treatment can help in better management of thedisease.

2. The isolates from a single lesion have similar genotypes. This result shows that a lesion is formed by a single invading conidia.

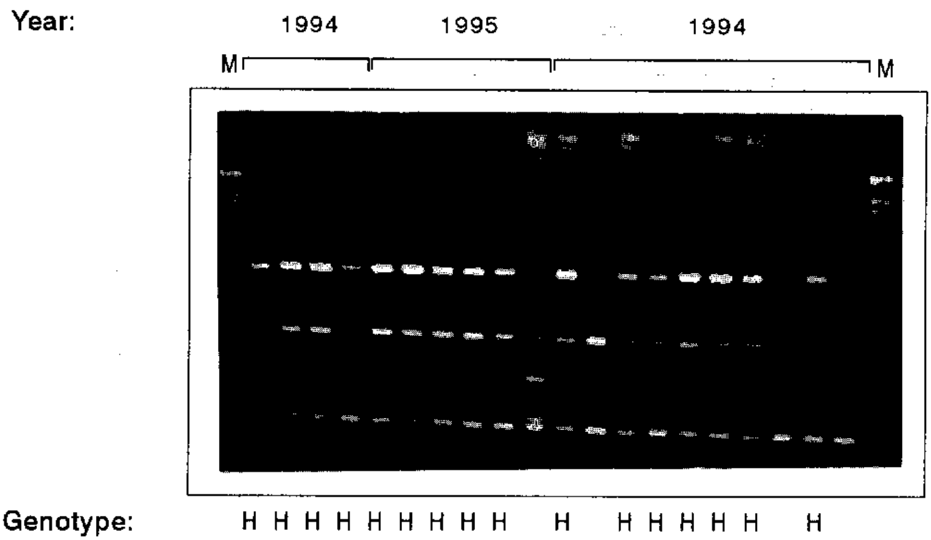

Figure 3.3.1. Genotyping of Ascochyta rabieiisolates of Syria using RAPD marker, OPJ-01. The lanes showRAPD pattern of the isolates collected during 1994 and 1995. Lane M contains lambda EcoRI-HindIII digested DNA.

3. Genotype H of the pathogen was also recovered from infected chickpea fields where varieties with different levels of resistance were grown. Therefore there is a need to further increase the level of resistance in chickpea cultivars.

4. Genotype H was observed in both spring and winter sown chickpea fields. This means that both winter and spring sown chickpeas can be infected by the pathogen.

5. Genotype H was recovered from farmers fields and also from the fields of research stations.

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3.3.2. Comparative Genetic Analysis of Indian and Syrian Isolates of *Ascochyta rabiei*

Chickpea crop in the North-western parts of India are often infected by ascochyta blight. A collaborative research project has been initiated with ICRISAT for genetic characterization of the pathogen prevailing in India. Since the isolates can not exchanged because of quarantine restrictions, DNA samples of the Indian isolates were received by ICARDA from ICRISAT for RAPD analysis. Four of the received isolates were analyzed with RAPD markers using pathotype specific primers and compared with Syrian isolates (Figure 3.3.2.). The study revealed that, the genotypes of the Indian isolates are similar to genotypes of pathotype II isolates of Syria and are not related to the predominant genotype H of Syria.

J.P. Moss (ICRISAT), S.M. Udupa, F. Weigand

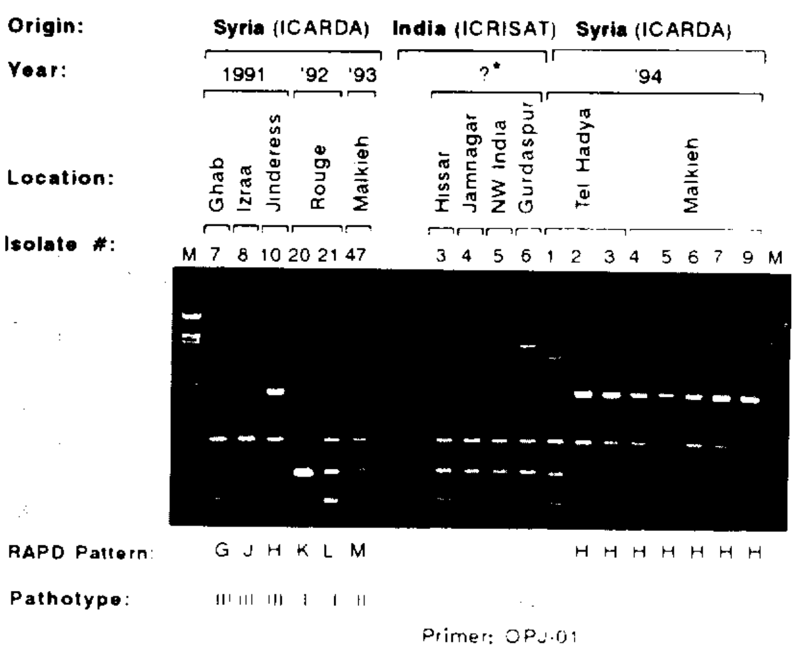

Figure 3.3.2. Comparative genetic analysis of Ascochyta rabiei isolates of Syria and India using RAPDs.

3.3.3. Sexual Mating and Segregation Analysis of Microsatellite Markers in A. rabiei

A sexual cross between the low aggressive isolate Mat USA and the aggressive isolate = 6 from Syria was performed in USA. A subset of a population derived from the cross was analyzed for segregation of microsatellite markers at the University of Frankfurt. One such marker analysis is shown in Figure 3.3.3.

A markers are segregating in Mendelian fashion. The future study aims to develop of a microsatellite based map of A. rebiei.

J. Geistlinger and G. Kahl (University ofFrankfurt), F. Weigand

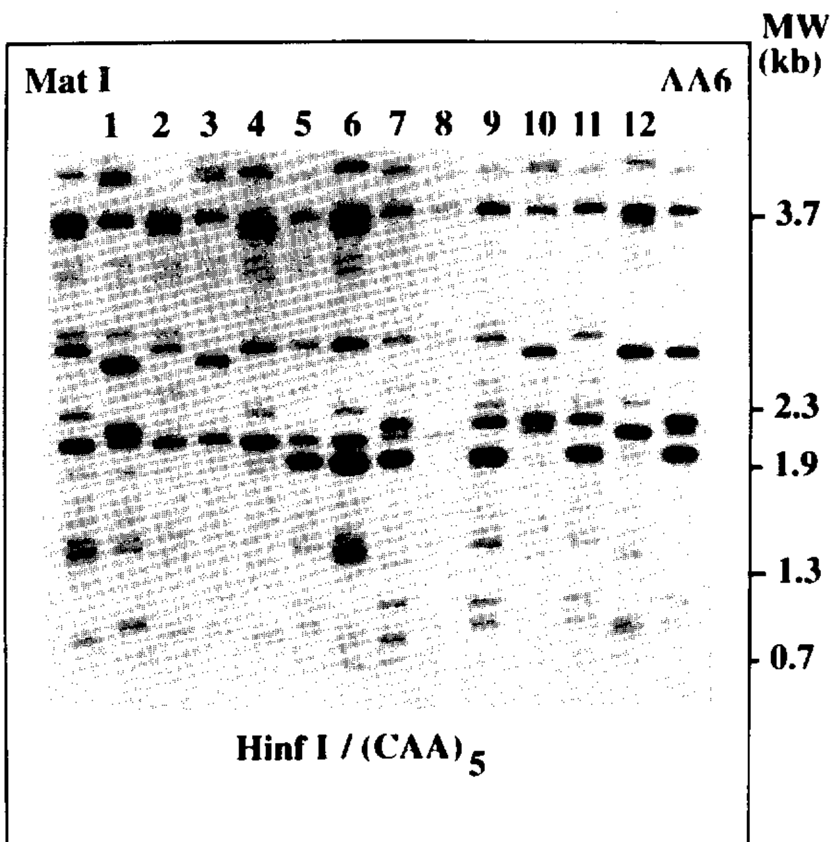

Figure 3.3.3. Segregation of microsatellite markers in a population derived from a sexual cross between Mat I (USA) and Isolate # 6 (Syria) of Ascochyta rabiei.

3.3.4. Breeding for Resistance in Chickpea to the most Predominant Genotype (Pathotype III) of *A. rabiei* of Syria.

Identification of sources of resistance in germplasm accessions and further improvement of the level of resistance through pyramiding (stacking) of resistance genes are important components of disease resistance breeding programs. Normally, large number of land races and breeding lines are screened under field conditions and resistant lines are selected. Under field conditions, it is often very difficult to provide suitable epiphytotic condition for disease development and to have control on the pathotype or genotype of the pathogen interacting with the host. Therefore, there is a need to screen breeding lines under controlled environment conditions. A set of 400 chickpea lines which showed reactions of 3-5 on a rating scale of 1-9 under field conditions were further screened for resistance to the most predominant genotype (H) of the pathogen under growth chamber conditions. The tested material included 14 land races and more than 300 breeding lines derived from crosses between resistant x resistant lines (gene pyramiding). The screening was conducted in two stages. In the first stage four plants of each line were inoculated with genotype H (pathotype III, isolate No. 13). Nineteen promising lines were identified for further screening. The 19 promising lines along with additional 15 new lines were tested in four replications and each replication consisted of four plants. ILC 3279 which is susceptible to pathotype III served as a check. The performance of the top 13 lines are presented in Table 3.3.2.

The conclusions are as follows:

1. The results of field and growth chamber screening differed from each other. Many lines which showed ratings of 3-5 in the field were highly susceptible to genotype H in the growth chamber screening. The reasons for the susceptible reactions under growth chamber condition is due to the controlled use of the aggressive genotype H (pathotype III) and also due to the proper epiphytotic condition provided for the disease development. In the growth chamber screening the susceptible check was rated 7.00 while in the field screening the same susceptible check was rated

only 4-5.

2. Sources of resistance to genotype H (pathotype III) are available in desi type chickpea (ICC 3991 and ICC 3912).
3. Several breeding lines developed through gene pyramiding showed significantly higher level of resistance than their parents and the standard check (ILC 3279). Hence the conventional approach to pyramid resistance genes is efficient in improving the level of resistance in chickpea.
4. Future work on gene pyramiding should be carried out with the most predominant genotype H (pathotype III) under controlled growth chamber conditions.

Table 3.3.2. Sources of resistance to the genotype H (pathotype III) of *Ascochyta rabiei* of Syria.

Genotype	Disease reaction (1-9 scale)		
	Growth chamber condition		**Field** condition
	Mean ± SD		Mean
S 94673	3.19±0.24	(1.78)	3.00
S 94674	3.25±1.04	(1.79)	3.00
S 94664	3.50±0.46	(1.87)	3.00
ICC 3991	3.56±0.13	(1.89)	4.00
ICC 3912	3.18±0.13	(1.95)	4.00
S 94654	3.94±1.39	(1.96)	3.00
S 94683	4.25±0.74	(2.06)	3.00
S 94685	4.15±0.31	(2.04)	3.00
ICC 3919	4.88±0.75	(2.20)	4.00
S 94667	4.25±0.35	(2.06)	3.00
S 94655	4.06±0.80	(2.00)	3.00
S 94656	3.88±0.48	(1.97)	3.00
Flip 93-179C	4.94±0.24	(2.22)	4.00
ILC3279(Check)	7.00±0.00	(2.65)	3.00
C.V. (%)	5.29	(3.09)	
S.E.		(0.06)	
LSD at 1%		(0.14)	

Notes:

a) S series breeding lines were developed through gene pyramiding. Example of a typical pedigree:
ILC 482 x (ILC 72 x ILC 215) x (ILC 72 x ILC 215)

b) ICC series are desi type chickpea landraces.

c) Numbers in parenthesis are square-root transformed values.

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3.3.5. Development of Populations for Gene Tagging, Marker-Assisted Selection and Genetic Studies

Two F_2 populations of the cross between ILC 1272 x ILC 3279 were grown under plastic house conditions. During initial growth period, comparatively lower temperature conditions (15-20°C) and short photoperiod (10 hr light) were provided to promote vegetative growth. Later on higher temperature (23-26°C) and longer photoperiod (16 hr) were provided to promote reproductive phase of the plants. The plants were continuously nourished with nutrient solution. By doing so we could harvest approximately 10 g of leaf per plant for DNA extraction without affecting further plant growth and seed yield. Number of seeds harvested per plant ranged from 11 to 172. In addition to this cross, the crosses ILC 1929 x ILC 200, ICC 12004 x ILC 1929 and ICC 13718 x ILC 1929 were carried forward from F_1 to F_2 generation.

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3.3.6. Screening of Parental Lines for Resistance to Pathotype I, II and III of A. rabiei

The parental lines used for development of gene tagging populations were screened for resistance to pathotypes prevailing in Syria. The isolate Nos. 51 (pathotype I), 47 (pathotype II) and 13 (pathotype III) were used in the study (Table 3.3.3).

Table 3.3.3. Screening of the parental lines for resistance to ascochyta blight.

Genotype	Disease reaction (1-9 scale)		
	Pathotype I	PathotypeII	Pathotype III
ILC 191	3.00±0.00	9.00±0.00	8.25±0.42
ILC 6482	3.14±0.69	9.00±0.00	8.75±0.46
ICC 12004	2.38±0.74	3.00±0.92	2.25±0.46
ICC 13718	2.13±0.35	3.00±0.76	4.13±1.36
ILC 482(check)	3.00±0.50	8.52±0.51	8.42±0.50
ILC 3279(check)	-	5.52±1.05	8.67±0.48
ILC 1929(check)	8.90±0.31	-	-

The resistance to pathotype III was found only in desi type cultivars, ICC 12004 (2.25±0.46) and ICC 13718 (4.13±1.36). The kabuli chickpea cultivars, ILC 191 and ILC 6482 showed resistance to pathotype I.

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3.3.7. Intra Accession Variability in Chickpea

Intraspecific variability existing in germplasm collections can be measured at inter and intra accession levels. Intra accession variability will provide information on the extent of genetic diversity existing within an accession and if an accession is identified as genetically very diverse (heterogeneous) then there is more scope for plant genetic improvement through pure line selection.

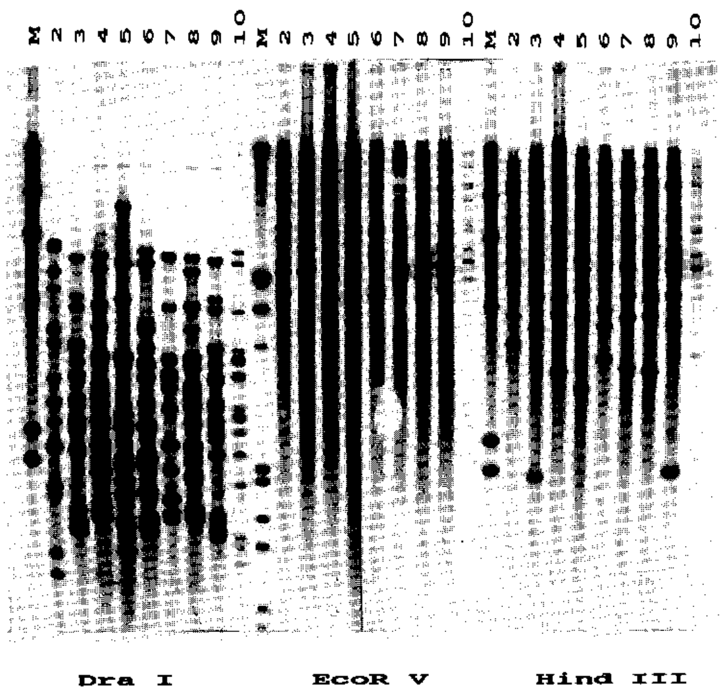

Figure 3.3.4. Intra accession variability as detected by microsatellite-based RFLP analysis. Nine individual plants from Afghanistan (ILC 1354) were analyzed with the probe (GATA)4 and the enzymes DraI, EcoRV and HindIII. The lane M contains lambda EcoRI-HindIII digest.

To identify the level of diversity in a germplasm accessions is part of germplasm conservation efforts. Periodic monitoring of intra-accession variability may help in safe-guarding the level of diversity in an accession which might change due to mechanical mixtures during handling and due to outcrossing during multiplication. Since it is often very difficult to asses intra-accession variability with morphological traits microsatellite-based DNA markers were tested to study polymorphism within an accession. Very high degree of inter-accession polymorphism could only be detected with microsatellite-based markers but not with RAPDs (see Ann. Report, LP, ICARDA, 1994). The identification of suitable probe-enzyme combinations which detect a high degree of polymorphism is a first step towards assessing the variability at DNA level. Therefore the DNA from 9 individual plants of the accession ILC 1354 (collected in Afghanistan) was digested with EcoRI, DraI, EcoRV and HindIII and probed with the microsatellite sequence (GATA)4 (Figure 3.3.4). The analysis revealed that all the three probe/enzyme combinations are equally efficient in detecting polymorphism within this accession as indicated by the same diversity index (0.94).

Since the enzyme EcoRI is cheaper than the others the combination (GATA)4/EcoRI was used for further studies (Section 3.3.8.).

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3.3.8. Estimation of Genetic Diversity in Chickpea Collected from Different Geographical Regions

A total of 30 accessions of *Cicer arietinum* L. from the germplasm collection held at ICARDA were selected for this study. These accessions originated from 11 countries representing basically to two regions: Near East and Indian Subcontinent (Table 3.3.4.). To estimate the genetic diversity and genetic distances within and between accessions, microsatellite-based RFLP analysis (DNA fingerprinting) were performed using the probe/enzyme combination, $(GATA)_4$/EcoRI. Eight individual plants from an accession were randomly selected for this purpose. The results of this analysis are presented in Table 3.3.4. In general, the microsatellite marker was able to detect high levels of genetic diversity within an accession. The highest genetic diversity value (1) was estimated in two accessions from Afghanistan, Pakistan (Fig. 3.3.5.) and one accession from Russia, Iraq and Palestine. The lowest genetic diversity (0.643) was observed in one accession of Palestine, Jordan and Syria.

Table 3.3.4. Geographic origin and diversity index of 30 cultivated chickpea accessions.

Origin	Accession number	Diversity index
Afghanistan	ILC 1354	1.00
	ILC 1596	0.86
	ILC 1669	1.00
India	ILC 1963	0.89
	ILC 2413	0.93
Pakistan	ILC 5637	0.96
	ILC 5649	1.00
	ILC 5652	1.00
Iran	ILC 286	0.93
	ILC 322	0.86
	ILC 2350	0.82
Russia	ILC 197	0.93
	ILC 200	1.00
	ILC 2506	0.93
Turkey	ILC 502	0.96
	ILC 3560	0.96
Iraq	ILC 59	1.00
	ILC 67	0.96
Syria	ILC 4540	0.64
	ILC 4569	0.96
	ILC 4649	0.93
Jordan	ILC 1789	0.93
	ILC 1793	0.64
	ILC 4346	0.71
Lebanon	ILC 573	0.93
	ILC 1782	0.96
	ILC 1783	0.93
Palestine	ILC 241	1.00
	ILC 2299	0.86
	ILC 2300	0.64

The range of similarity index was highest for the accessions from Afghanistan and lowest for the accessions from Palestine. Results show that a high degree of inter and intra accession diversity exists in the chickpea germplasm accessions. The high degree of intra accession variability observed suggests that insitu conservation of chickpea germplasm may be necessary to conserve the existing genetic diversity of chickpea. The results also indicate the existence of several centers of diversity for this crop. This information may guide future collection missions into regions where the estimated genetic diversity is not fully represented by the genetic diversity within the existing collection.

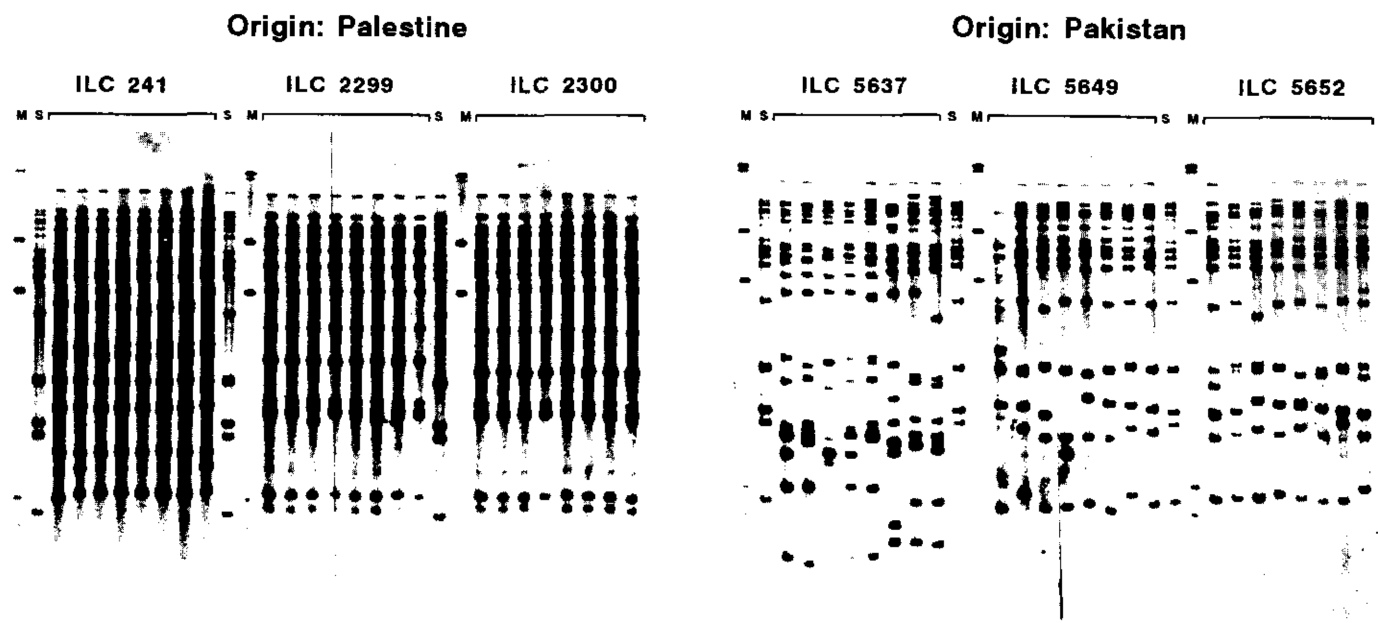

Figure 3.3.5. DNA fingerprints detected by the microsatellite probe $(GATA)_4$ in EcoRI-digested genomic DNA of chickpea accessions showing a wide range of genetic diversity for the accessions from Palestine and a very high level of genetic diversity for the accessions from Pakistan.

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The genetic diversity data DNA fingerprinting should be digitalized. This could help to conduct the diversity study on a wider basis and the information could be compared with data from other germplasm collections.

3.3.9. DNA Amplification Fingerprinting (DAF)

DAF is very similar to the RAPDs and also uses short, random oligonucleotide primers, but in comparison to RAPD it needs higher amounts of magnesium, polymerase and primer and lower amounts of DNA. Separation and detection of DAF products is done on silver stained polyacrylamide gels. Because of special DAF conditions and staining, many more products were generated and detected compared to RAPD. Also the reproducability of the reaction was much better. Intra- and interspecific polymorphism of DAF products were tested between one accession of *Cicer reticulatum* and seven of *Cicer arietinum* accessions including the desi variety WR 315. As can be seen from Table 3.3.5. the polymorphism between *C. reticulatum* and its cultivated relatives was generally three times higher than the intraspecific polymorphism within the cultivated chickpea. The desi variety did not differ from other cultivated chickpea accessions in this aspect.

Table 3.3.5. Inter- and intraspecific DAF polymorphisms per primer. The number of tested primers is given in parentheses.

	C. reticulatum	ILC 191	ILC 200	ILC 1929	ILC 1272	ILC 3279	ILC 315
ILC 191	7.3 (36)						
ILC 200	6.7 (35)	2.3 (38)					
ILC 1929	6.8 (36)	1.9 (37)	1.5 (35)				
ILC 1272	6.7 (35)	2.2 (36)	1.6 (35)	0.8 (36)			
ILC 3279	6.7 (35)	2.8 (37)	1.8 (36)	1.9 (37)	1.8 (36)		
WR 315	6.5 (35)	2.4 (37)	2.1 (36)	1.9 (37)	2.0 (36)	2.4 (37)	
C 104	6.6 (35)	2.5 (36)	1.9 (35)	1.6 (36)	1.7 (35)	2.0 (36)	1.8 (36)

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3.3.10. Multiple Arbitrary Amplicon Profiling (MAAP) of DAF Products

A set of accessions were tested with RAPD primers. To further increase the amount of polymorphism DAF products were restricted with 3 different mixtures of restriction enzymes to produce leaved amplified polymorphic sequences (CAPS) (Figure 3.3.6.). This raised the amount of polymorphism from an average of 2.2 for DAF products to 3.5 for restriction mix I. As can be seen from the Table 3.3.6. the gain in level of polymorphism was highest for mix I (AluI, RsaI and HpaII), lower for mix II (TaqI and HinfI) and lowest for mix III (SspI). Most of the (CAPS) could be easily scored and were repeatabl

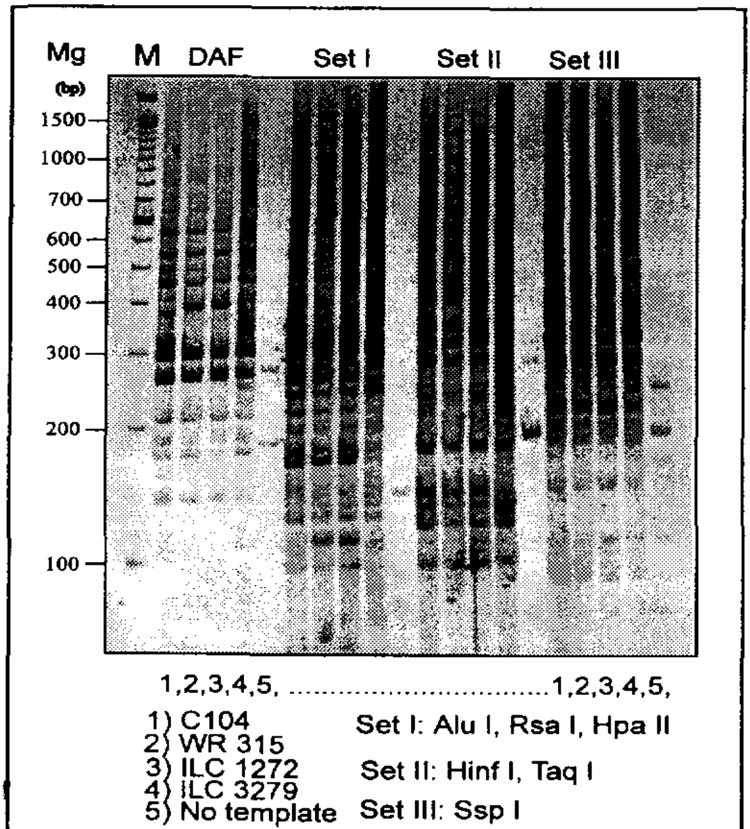

Figure 3.3.6. Separation of DAF products and CAPS from amplification of the indicated DNAs with primer 5'-AGCGTCACTC-3' on a denaturing, 4% poly-acrylamide gel. The gel was stained with silver.

Table 3.3.6. Polymorphism per primer with respect to post-digestion of DAF products with different sets of restriction enzymes.

Treatment	Acc.#	WR 315	ILC 1272	ILC 3279	Treatment mean±SD
DAF	C 104	2.2	2.1	2.1	2.2±0.3
	WR 315	-	2.6	1.7	
	ILC 1272	-	-	2.5	
Set I	C 104	4.5	2.8	3.7	3.5±0.7
	WR 315	-	3.8	2.8	
	ILC 1272	-	-	3.4	
Set II	C 104	4.0	2.2	3.7	3.3±0.7
	WR 315	-	4.0	2.9	
	ILC 1272	-	-	2.9	
Set III	C 104	3.4	1.9	2.3	2.5±0.6
	WR 315	-	2.9	1.8	
	ILC 1272	-	-	2.6	

Note:
Set I = AluI, RsaI, HpaI
Set II = TaqI, HinfI
Set III = SspI

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3.3.11. Repetitive elements in chickpea

Repetitive elements have been used for phylogenetic studies, *insitu* characterization of chromosomes and identity testing in tissue culture. A survey of repetitive elements was made in *Cicer arietinum* and the related wild annuals *C. reticulatum*, *C. echinospermum*, *C. bijugum*, *C. cuneatum*, *C. chorassanicum* *C. pinatifidam* and *C. judaicum*, of which the first two give rise to fertile offspring in interspecific crosses with *C. arietinum*. From an ordered small-insert library of 1100 EcoRI-clones in 248 colonies highly repetitive elements were identified, which could be grouped into 6 classes. Two of the classes could be identified as satellite DNA. These classes were present in all *Cicer* species in similar amounts but not in pea, faba bean or tomato. The other four classes probably consists of dispersely distributed elements. Two of these could be used to differente between three groups of *Cicer* species. The first group includes *C. arietinum* and the two crossable species, the second group contains *C. bijugum*, *C. cuneatum*, *C. judaicum* and *C. pinatifidum*. The third contains only *C. chorassanicum.* These results are in good agreement with the current opinion about relatedness between annual *Cicer* species.

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3.3.12. Generation of Sequence-Tagged Microsatellite Site (STMS) Marker

Sequence tagged microsatellite markers are PCR-based, site-specific, and highly informative molecular markers. The generation of these site-specific microsatellite markers requires cloning of target DNA, screening of libraries for microsatellite sequences, sequencing of repeat containing fragments and design of repeat-flanking primers. As a first step size-selected genomic DNA libraries (250-600 bp) were constructed consisting of approximately 30000 recombinant clones (=1.5% of the chickpea genome). These clone libraries were hybridized to a variety of radioactively labelled probes (eg. $(GA)_8$, $(GT)_8$, $(TAA)_5$). Approximately 350 microsatellite-containing clones have been identified up to October 1995. Positive bacterial colonies are currently re-screened to ensure the single origin of recombinant clones.

Some primer pairs flanking repetitive chickpea loci were evaluated for polymorphism between accessions. STMS-mediated polymorphism can only be scored if small electrophoretic mobility differences are considered and therefore a 6% polyacrylamide/7M urea separation matrix was used. The result obtained applying a primer pair (CA27AB) flanking an $(AT)_n$-repeat to PCR analysis of chickpea accessions are shown in Figure 3.3.7. Four different alleles could be detected in 10 different accessions. As expected for an inbreeding species like chickpea no allele heterozygosity was observed.

B. Hüttel, P. Winter and G. Kahl (University of Frankfurt), S.M. Udupa, F. Weigand

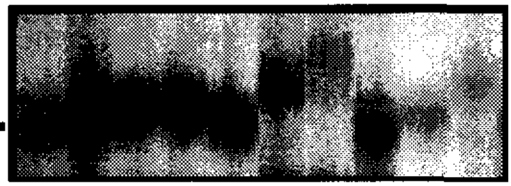

Figure 3.3.7. Evaluation of polymorphism in chickpea using STMS. Individual chickpea DNA (A: ILC 190, B: ILC 191, C: ILC 200, D: ILC 202, E: ILC 482, F: ILC 1272, G: ILC 1929, H: ILC 3279, I: ILC 4978, K: ILC 6298) was analyzed by PCR. The amplification products were separated on 6% polyacrylamide/7M urea. One primer was end-labeled with ^{32}P-gamma ATP.

3.3.13. Construction of chickpea genomic DNA library

The first step in construction of sequence tagged microsatellite site markers (STMS) is the construction of a gene library with small chickpea DNA fragments. Therefore genomic DNA of chickpea was digested with Sau3A restriction enzyme and resulting DNA fragments were fractionated by agarose gel electrophoresis. The fragments of approximately 100-300 base-pair size were eluted from the gel and ligated onto an BamH1 digested and BAP treated plasmid vector, pUC 19. The ligated plasmid was transformed into bacteria *(Escherichia coli)*. The resulting transformed bacteria was screened for recombinants by plating on LB medium containing ampicillin (antibiotic), IPTG and x-gal. The isolated recombinants (white colonies) were further confirmed by replating onto the above media. Plasmid DNA was isolated from the transgenic bacteria and digested with EcoRI and PstI restriction enzymes to know the size of chickpea DNA inserts. The constructed gene library was screened for microsatellite sequences using $(GGAT)_4$ and $(GTG)_5$. Out of 80 recombinants screened none of them contained the microsatellite sequences. Recent literature on microsatellites in plants shows that only 1% of the genomic library contains microsatellite sequences. Hence more genomic clones have to be screened to isolate microsatellite sequences. To visualize the small inserts of the cloned chickpea DNA fragments PCR amplification of the inserts were performed using universal forward and reverse primers of M13 bacteriophage. The electrophoregram of the analysis is presented in Figure 3.3.8.

Figure 3.3.8. PCR amplification of the inserts of chickpea genomic clones using M13 forward and reverse primers. Lane M contains lambda EcoRI-HindIII digest.

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3.3.14 Interspecific crosses in chickpea

The wild annual *Cicer* species have been screened for biotic and abiotic resistance genes at Tel Hadya in recent years (see Table 3.3.7). Some of these species carry resistance genes which cannot be found in the cultivated *Cicer* or multiple stress resistance or resistance of higher level compared to the cultivated species. Unfortunately, sexual hybrids have so far only been reported between *C. arietinum* and *C. recticulatum* and *C. echinospermum*.

Table 3.3.7. Evaluation for resistance and tolerance for biotic and biotic stresses, (1=resistance 9=suceptible)

ILWC	Ascoc. blight	Fusarium wilt	Leaf miner	Seed beetle	Cyst nematode	Cold
32	7	1	7	4	2	2
62	4	1	7	2	2	2
95	9	4	6	4	4	3
203	5	4	7	9	4	4
236	9	-	5	-	1	4

(D-vac sampling) towards the end of April (Table 3.4.1 and Fig. 3.4.1). Grain yield was lowest in the control and was not significantly different to the other treatments (Table 3.4.1 & Fig. 3.4.2). It was clear that Thiodan gave the best control followed by the Neem extract whereas the Neem powder was comparable to the control.

Table 3.4.1. Effect of 3 sprays of neem-seed extract and Neem powder and one spray of Thiodan on leaf miner infestation, pod borer damage and yield at Tel Hadya in winter sown chickpea (1994/95).

Treatment	VDS		%infested leaflets		% Pod damage	Seed yield (kg/ha)
Date of assay	30.4	15.5	12.4	27.4		
Neem Extract	2.7	3.4	22.8	20.8	0.3	1655
Neem Product	3.8	4.2	20.0	22.3	2.5	1667
Thiodan	2.6	2.5	21.8	26.5	0.3	1696
Water	3.6	5.3	20.1	23.8	2.5	1589
LSD 5%	0.64	1.64	5.20	2.64	1.23	204.8
C.V.	12.7	26.9	15.3	7.1	55.2	7.8

In the spring sown chickpea, the infestation was higher than in the winter and the infestation was about 100 adults, 37% infested leaflets and 5.8 on VDS. The parasitoid followed a similar trend and recorded the highest level of 65 adults (D-Vac) by mid-May (Tables 3.4.2, 3.4.3 & Figs. 3.4.1, 3.4.2). The overall average yield of grain was about 70% less than the winter production, partly because of the heavier infestation with the pest in spring and of the longer period for the development of the corp in winter (Table 3.4.2 & Fig. 3.4.2) also in the spring planting, yield was about 50% higher at Sheikh Yousif than at T. Hadya. However, the 4 attributes followed a similar trend under the different insecticidal treatments as in winter.

Of the infested leaflets kept in the lab.in petri-dishes with moistened filter paper, only few developed to pupae. However, extremely low percentage of these developed into adult miners and parasitoids; the reason for this is not clear but perhaps due to insects reared in petri-dishes with low relative humidity (Fig. 3.4.3).

Table 3.4.2. Effect of 3 sprays of neem-seed extract and Neem powder ndone spray of Thiodan on leaf miner infestation, pod borer damage and seed yield at Tel Hadya in spring sown chickpea (1994/95).

Treatment	VDS			%infested leaflets		% Pod damage	Seed yield (kg/ha)
	1.5	16.5	29.5	13.4	16.5		
Neem Extract	3.5	3.7	3.0	19.8	25.3	0.0	483
Neem Product	4.3	5.9	4.2	17.5	35.0	2.6	484
Thiodan	4.7	2.2	2.5	18.0	2.0	0.6	491
Water	4.7	5.9	4.9	16.5	37.0	1.5	425
LSD 5%	0.93	0.81	0.49	3.52	6.57	1.86	73.4
C.V.	13.7	11.5	8.5	12.3	16.6	99.2	9.8

Table 3.4.3. Effect of 3 sprays of neem-seed extract and Neem powder and one spray of Thiodan on leaf miner infestation, pod borer damage and seed yield at Shiekh Yousef in spring sown chickpea (1994/95).

Treatment	VDS				%infested leaflets		% Pod damage	Seed yield (kg/ha)
	4.5	18.5	1.6	23.4	4.5	18.5		
Neem Extract	3.1	3.3	3.5	31.3	25.3	27.5	2.0	1135
Neem Product	3.8	4.0	5.5	32.0	24.5	26.0	5.8	1052
Thiodan	3.6	2.1	4.5	30.0	29.8	8.3	3.0	1221
Water	4.3	4.2	6.5	31.3	29.3	32.8	2.8	1075
LSD 5%	1.06	0.84	2.29	2.21	11.12	5.36	2.053314	
C.V.	18.1	15.5	28.7	4.5	25.6	14.2	37.9	185

Screening New Breeding Lines

Ninety four breeding lines of chickpea were planted at Tel Hadya on 5 March to screen against the leaf miner. Damage was assessed twice on 30 April and 23 May using VDS on 1-9 scale. All entries which scored 4 and less were considered resistant and these will be further tested with new breeding lines next season. The ILC lines were as followed according to superiority.

1. 5901	16. 394	31. 2769	46. 1216
2. 3800	17. 809	32. 2839	47. 2383
3. 799	18. 822	33. 2840	48. 2755
4. 515	19. 826	34. 2959	49. 2759
5. 1784	20. 1075	35. 2961	50. 3470
6. 726	21. 1169	36. 3350	51. 3476
7. 319	22. 1431	37. 878	52. 4414
8. 1008	23. 2211	38. 930	53. 5289
9. 2435	24. 2221	39. 5285	54. 5290
10. 3805	25. 2250	40. 5297	55. 5294
11. 3828	26. 2319	41. 5600	56. 5307
12. 5283	27. 2334	42. 831	57. 5555
13. 5309	28. 2436	43. 869	58. 5594
14. 5550	29. 2618	44. 967	59. 5608
15. 316	30. 2763	45. 1000	60. 5615

Almost all the entries defined as resistant were early maturing.

3.4.2 The Podborer, *Helicoverpa armigera* Hb

3.4.2.1 Selective Chemical Control

Efficacy of the same insecticidal treatments described for the leaf miner were assessed against the pod borer based on larva collection on trays, pod damage and grain yield. Results showed that Neem extract and Thiodan significantly reduced the number of larvae collected and pod damage. Accordingly, yields were comparatively but not significantly higher (Tables 3.4.1, 3.4.2, 3.4.3 and Figs. 3.4.1, 3.4.2).

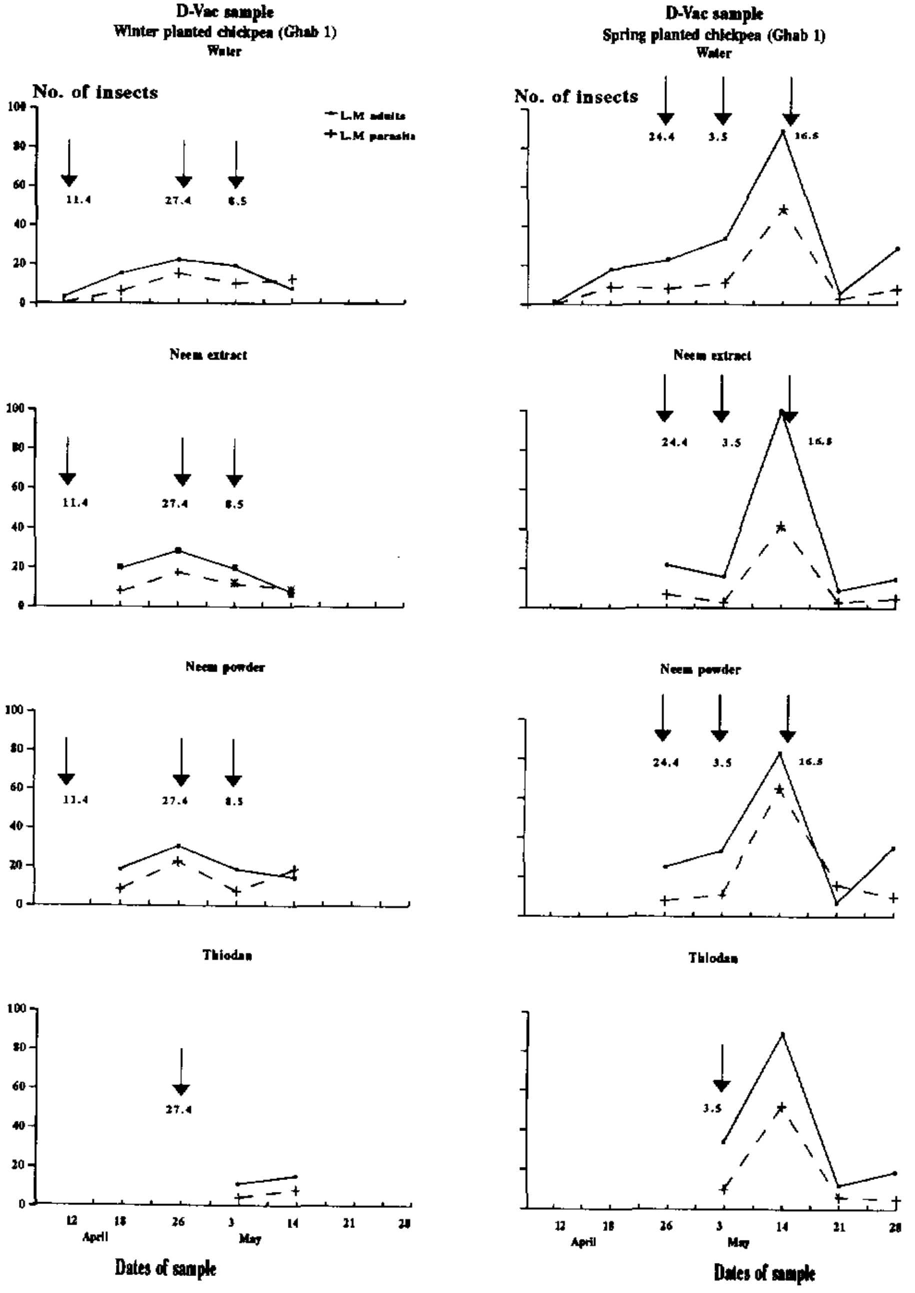

Fig. 3.4.1. Effect of one application of Thiodan and three of Neem extract and Neem powder on adult population of the leaf miner and parasitoides at Tel Hadya (1994/95)

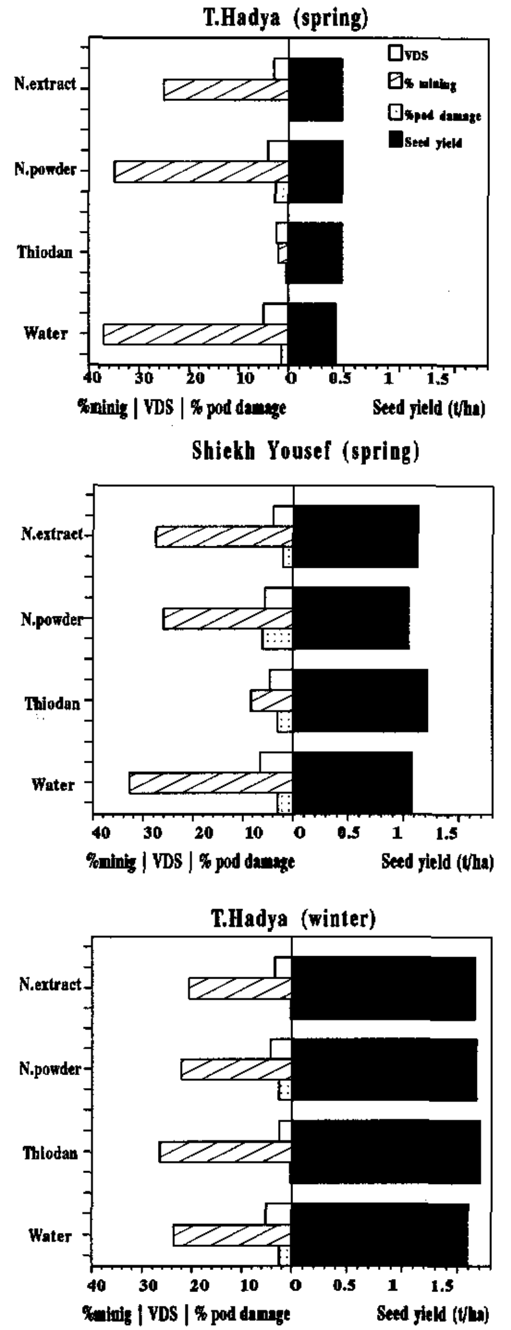

Fig. 3.4.2. Effect of 3 sprays of Neem seed extract and Neem powder and one of Thiodan on leaf miner infestation, pod borer damage and yield in chickpea (1994/95)

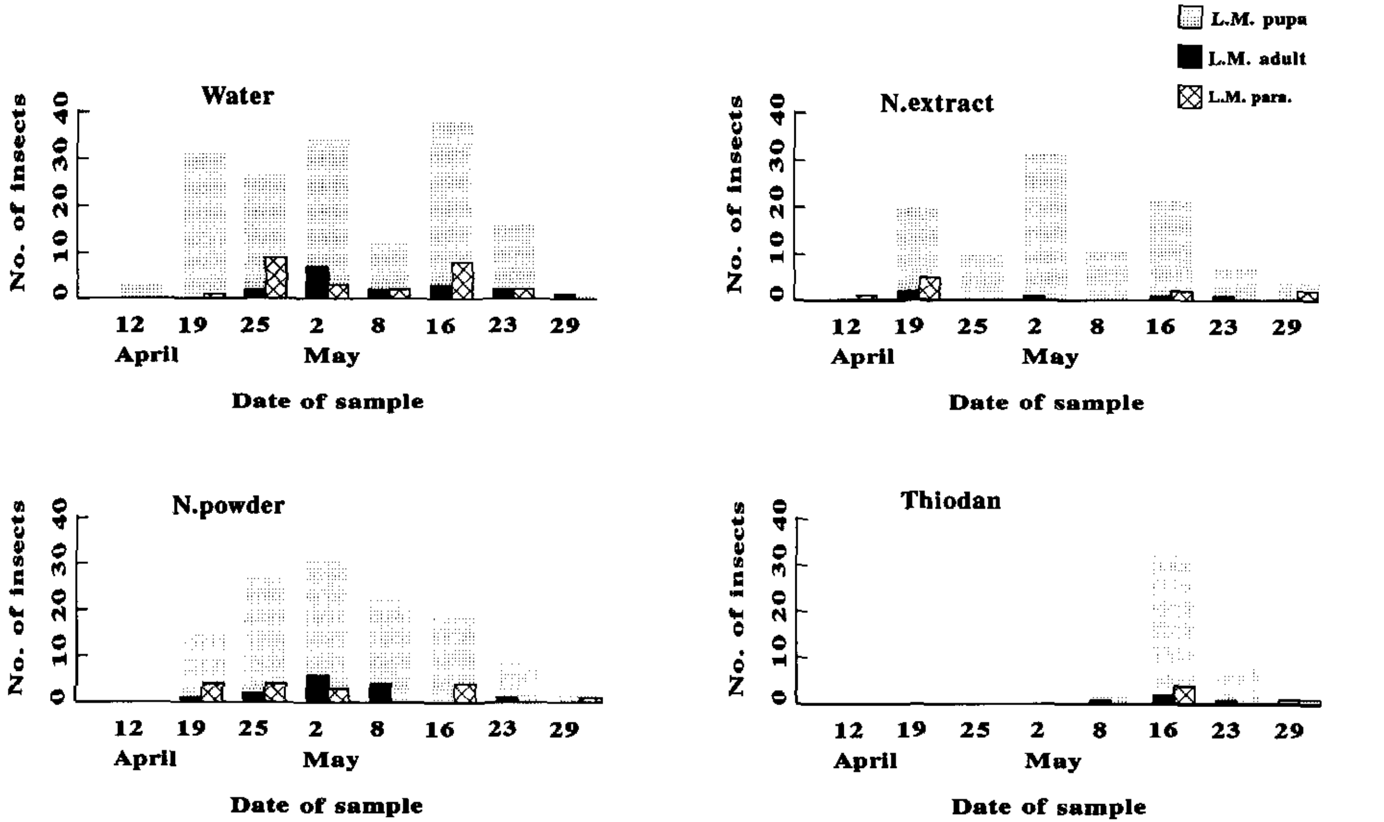

Fig. 3.4.3. Number of leaf miner and parasitoid adults emerging from 40 infested leaflet samples from spring sown chickpea receiving different insecticidal treatments (1994/95)

3.4.2.2 Host-Plant Resistance

ILC 5901 was rescreened with the susceptible check ILC 1929 for confirmation of resistance to pod borer. The larva number and pod damage assessment clearly showed that ILC 5901 was more susceptible to pod borer attack and sustained higher number of the larvae and greater pod damage than ILC 1929. This is perhaps due to the small leaflets which form an open canopy for the pest to move and find the pods, or due to longer growth period and this can be verified by more frequent damage assessment (Table 3.4.4).

3.4.2.3 Monitoring

A number of phermone traps were operated through the season in different locations in the country to monitor the distribution of the pod borer in chickpea, cotton and tomatoes. Collections were made at 15 day-intervals and the catches were sorted out and *H. armigera* numbers were recorded. Although catches were inconsistent from one year to another still some areas like Tel Hadya B fields, Sqielbiye, Sheikh Yousif, Gib Kass and T. Hadya seem to be the more affected areas (Fig. 3.4.1). The traps did not cover other parts of the country such as the South (Sham area) and the East such as Hassaka, Dier EZ-Zour (Myadeen area) and Raqqa and monitoring will be extended to some of these areas with few selected ones of the old sites. This will give better coverage of all the country and will indicate the relative importance of *Heliothis* in the different areas. The survey will also include from next year collection of larvae of pod borers from different crops in the country to identify the species of borers and parasitoids prevalent in different crops for an overall management of this pest in the agrosystem.

3.5. CHICKPEA NEMATOLOGY

During the last decade ICARDA has studied the nematodes of cool season food legumes to identify constraints for these crops in the Mediterranean basin. These studies have identified several nematodes which are very damaging to host crops. Moreover, it is now clear that control of these nematodes can be based exclusively on agronomic practices and on resistant cultivars. Unfortunately no cultivar of legumes with resistance to major nematodes occurring in the Mediterranean area is available. However, resistance to *Heterodera ciceri* was identified in the wild species *Cicer reticulatum*. This resistance is now being transferred to the cultivated chickpea through several crosses between resistant *C. reticulatum* and susceptible *C.arietinum* lines. In 1994-1995 the evaluation of the progenies of these crosses has continued. Several nematodes possess pathotypes and the available resistance may be specific to only some pathotypes of the same nematode species. Therefore, an investigation was initiated to explore the occurrence of pathotypes in populations of *H. ciceri* from Syria and Turkey.

3.5.1. Nematode survey of legumes in Jordan

Twenty three soil and root samples were collected in fields of chickpea (11) and lentil (12) in early May. At sampling chickpea was at the flowering stage while lentil was maturing. The sampled areas were between Amman, Madaba and Karak, south of Amman, and the Irbid Governorate, near the Syrian border. Nematodes were extracted from the soil combining the sieving and decanting method with the Baermann's method and from the root samples with the incubation method.

Analysis of root samples (Table 3.5.1) showed that the most common endoparasitic nematode was the root-lesion *Pratylenchus* spp., which were found in all chickpea fields and nine lentil fields. However, with the exception of a chickpea field at Marue, numbers of nematode in the roots were rather low. Another root-lesion nematode, *Pratylenchoides* sp.,was found in three fields of chickpea and two of lentil. The chickpea cyst nematode, *Heterodera ciceri*,was present in large numbers on chickpea at Husan, Irbid Governorate, confirming the importance of this nematode.

Table 3.5.1. Endoparasitic nematodes recovered from 5 g roots of chickpea and lentil collected in Jordan in 1995.

No.	Location	Crop	Nematode		
			Pratylenchus	*Pratylenchoides*	Heterodera
1	Madaba	Chickpea	44		
2	"	Lentil	1		
3	"	"			
4	Shehan	Chickpea	148		
5	"	"	152	5	
6	Al-Qasen	Lentil		20	
7	Simakyya	"	15	10	
8	"	Chickpea	599	5	
9	"	"	51		
10	Al-Rabba	Lentil	104		
11	"	"	1		
12	"	"	58		
13	"	"			
14	Huson	"	9		
15	"	"	136		
16	"	"	450		
17	"	Chickpea	5		
18	"	"	120		
19	"	"	125		64
20	"	Chickpea	167		
21	Marue	"	96	244	
22	"	"	1100		
23	"	Lentil	131		

In the soil (Table 3.5.2) the most common and abundant nematodes were *Tylenchorynchus* spp., which were present in all fields. In seven fields (one of chickpea and six of lentil) populations of the nematode were more than 1,000 specimens/500cm^3 soil and might be responsible for crop damage. *Tylenchus* spp. were also present in all soil samples but in rather small numbers. Among the other nematode groups, the most common were *Pratylenchus* sp., in 14 samples, and *Helicotylenchus* sp., 15 samples. Other nematodes, found in only a few samples, were *Pratylenchus* sp. (4 samples), *Rotylenchulus* sp. (5 samples), *Paratylenchus* sp. (2 samples), *Heterodera* sp. (4 samples), and *Criconemoides* sp. and *Ditylenchus dipsaci* in one sample each.

Table 3.5.2. Nematodes extracted from 500 Cm² soil from soil sampels collected in Jordan in 1995.

N°	Locality	Crop	Nematode									
			Pra.	Prat.	Het.	Hel.	Tyle	Tyl.	Rot.	Para	Cric.	Ddt.
1	Madaba	Chickpea	9			84	1892	150				
2	"	Lentil					519	70				
3	"	"					3060	60				
4	Shehan	Chickpea	7			7	727	382				
5	Al-Qasen	"	8				175	152				
6	Cimex	Lentil		944		16	136	320				
7	"	"				8	1075	185				
8	"	Chickpea	77			6	66	26				
9	Al-Rabba	"	41			61	143	204				
10	"	Lentil	113				214	76				
11	"	"				84	1444	312			4	
12	"	"	15				730	76				
13	"	"	9				1040	902				18
14	Huson	"				1033	2251	386				
15	"	"				36	1086	330				
16	"	"	9		35		898	361	26	9		
17	"	Chickpea				304	198	84		30		
18	"	"	8			638	106	99	532			
19	"	"	55		483	13	84	67	59			
20	"	"	405				110	101	18			
21	Marue	"	479	76	4	236	40	15				
22	"	"	18			41	110	18	212			
23	"	Lentil				5	340	64				

* Pra=*Pratylenchus*; Prat=*Pratylenchoides*; Het.=*Heterodera*; Tyle=*Tylenchorynchus*; Tyl=*Tylenchus*; Rot.*Rotylenchulus*; Para.=*Paratylenchus*; Cric.=*Criconemoides*; Ddt.=*Ditylenchus*.

3.5.2. Screening of chickpea breeding material to *Heterodera ciceri*

The breeding program to transfer the resistance to *H. ciceri* in chickpea continued last season. F_6 and F_7 material (2613 plants) were evaluated in the plastic house at Tel Hadya (Syria). Plastic pots, containing 3.5 dm^3 soil infested with eggs of the nematode/g, were arranged on benches and three 200 cm^3 pots made of organic matter were sunken into them. One seed in each of the organic pots. Fifty days after germination the plants were uprooted, gently washed free of soil and the infestation of the roots by the nematode evaluated on a 0 to 5 scale, as in previous years, according to the numbers of females and cysts on each root. Roots with 0-2 ratings were considered resistant, those with 3 moderately resistant and those with 4-5 susceptible to the nematode. Among the tested plants 23 (0.9%) were free of nematode, 168 (6.4%) were rated as 1, 751 (28.7%) as 2, 1573 (60%)as 3 and only 98 (3.7%) as 4-5. In total 36% of plants were highly resistant and 60% moderately resistant.

Backcrosses of *C. arietinum* (susceptible) x *C. reticulatum* (resistant) were obtained at Tel Hadya (Syria) in spring 1994 and seeds increased at Terbol (Lebanon) in summer the same year. The F_1 seeds were sown in soil infested with *H. ciceri* as described and the nematode evaluated. The results showed that of the 591 plants evaluated none was free of the nematode, 16(2.7%) were resistant, 190 (32.1%) moderately and 385 (65%) susceptible to the nematode.

The line ILWC 119 of *C. reticulatum*, resistant to *H. ciceri*, is a mixed population containing the mostly resistant seeds but also some which are susceptible to the nematode. To obtain pure material for breeding purpose, 220 seeds of this line were sown in infested soil, as above, and the derived plants selected for their resistance to the nematode.

Plants which were free the nematode (4) or rated 1 (32) were used to obtain seeds which were later increased at Terbol. These seeds will be used in the breeding program.

3.5.3. Identification of pathotypes of Heterodera ciceri

The investigation was conducted at ICARDA Tel Hadya , in a plastic-house maintained at 15-25^0 C. The inoculum was extracted with a large can from infested soil from four localities in Turkey and two in Syria, reported in Tables 3.5.1. and 3.5.2. The population from each locality was used to infest the soil of plastic pot containing 1000 cm^3 soil. Infestation level was 20 eggs of the nematode/g soil. Pots were planted with 22 plant species or lines (Table 3.5.3) and there were 8 pots of each entry. When most of the plant species were at flowering to early podding stage, about 50 days after germination, four plants of each entry were uprooted. The roots were then gently washed free of adhering soil, weighed and nematodes extracted by the centrifugation method and collected in about 50 ml water suspension. Nematodes were counted and separated according to life stage. The remaining four plants per species were left to grow for a further month to allow most of the nematodes to reach the cyst stage. Plants were then cut at soil level, the soil left to dry and then 200 g per plot used to extract cysts by the Fenwick can. Cysts were counted to determine their egg content and the reproduction rate of the nematode on each plant specie calculated .

All populations reproduced very well on chickpea, lentil, grass pea and only that from Tel Hadya also on annual medics and alfalfa. All populations gave some reproduction on the line ILWC 119 of *C. reticulatum*, and all except the population from Bismil (Turkey) also on ILWC 62 of *C. bijugum*. Reproduction on the line ILWC 71 of *C. bijugum* was observed only in a few pots inoculated with populations of the nematode from Idleb and Tel Hadia (Syria) and Kiziltepe and Kirbasi (Turkey). The population from Tel Hadya reproduced also in some pots planted to ILWC 213 and ILWC 252 of *C. pinnatifidum*. No reproduction occurred on all other tested plants, including those which are known to be hosts for other cyst nematode species of the *H. trifolii* group.

In summary, this experiment has confirmed that H. ciceri reproduces only on a few leguminous plant species and, moreover, that some differences may occur in the reproduction of population of the nematode of different geographic origin.

Table 3.5.3. Reaction of 22 host plants to six population of *Heterodera ciceri* in plastic-house.

Host Plant	Population					
	1	2	3	4	5	6
Chickpea	+**	+	+	+	+	+
Lentil	+	+	+	+	+	+
Grass pea	+	+	+	+	+	+
Pea	+	+	+	+	+	+
Faba bean	-	-	-	-	-	-
Bean	-	-	-	-	-	-
Cowpea	-	-	-	-	-	-
Lupin	-	-	-	-	-	-
Soybean	-	-	-	-	-	-
Vetch	-	-	-	-	-	-
Annual medics	-	-	-	-	-	+
Alfalfa	-	-	-	-	-	+
Spanish espercet	-	-	-	-	-	-
Crimson clover	-	-	-	-	-	-
Red Clover	-	-	-	-	-	-
White Clover	-	-	-	-	-	-
Cicer reticulatum	+	+	+	+	+	+
ILWC 119	+	-	+	+	+	+
Cicer bijugum ILWC	+/-	-	+/-	+	-	+/-
62	-	-	-	-	-	+/-
Cicer bijugum ILWC	-	-	-	-	-	+/-
71	-	-	-	-	-	-
Cicer pinnatifidum						
ILWC 213						
Cicer pinnatifidum						
ILWC 252						
Carnation						

* 1= Idleb (Syria); 2 = Bismil 12 (Turkey); 3 = Kiziltepe (Turkey); 4 = Kirbasi 23/91, 5 = Kadinhani (Turkey); 6 = Tel Hadya (Syria).
** + = Final/initial population (Pf/Pi)>1; - = Pf/pi< 1; +/- = reproduction observed in some populations

3.6. International Testing Program

The international testing program on Kabuli chickpea is a vehicle for the dissemination of genetic materials and improved production practices, in the form of international nurseries and trials, to the national programs in and outside the WANA region. The genetic materials comprise early segregating populations in F_3 and F_4 generations, and elite lines with wide or specific adaptation, special morphological or quality traits, and resistance to common biotic and abiotic stresses. Nurseries are only sent on request and often include germplasm specifically developed for a particular region or a national program. A list of trials supplied in the 1995/96 season is given in Table 3.6.1.

The testing program helps in identification of genotypes with specific and wide adaptation. The performance data permit assessment of genotype x environment interaction and help in targeting breeding efforts for specific agro-ecological conditions.

The salient features of the 1993/94 international nursery results, received from cooperators until 31 October 1995, are presented here. The stability analyses of some of the trials were done using Eberhart and Russell (1966) model.

The Chickpea International Yield Trial-Spring (CIYT-SP) was reported from 24 locations in 14 countries. A number of test entries exceeded the respective local check by a significant margin ($P \leq 0.05$) at Khroub in Algeria; Toshevo in Bulgaria; Saskatoon in Canada; Tolentino in Italy; Terbol in Lebanon; Hama in Syria; and Ankara, Erzurum, and Menemen in Turkey. The five heaviest yielding entries across the locations were FLIP 91-203C, FLIP 91-186C, ILC 482, FLIP 88-70C, and FLIP 90-173C, respectively, with seed yield of 1188, 1185, 1176, 1119 and 1119 kg/ha. The stability analysis revealed that the entries, FLIP91-203C, FLIP88-70C, FLIP82-150C, FLIP91-188C, FLIP89-67C and FLIP89-24C were relatively stable across environment as compared to others.

For Chickpea International Yield Trial-Winter-Mediterranean (CIYT-W-MR) data were reported from 26 locations in 11 countries. At Dahmouni and Setif in Algeria; Gorgan in Iran; Ramtha in Jordan; Jindiress, Gelline, Hama, Al-Ghab, Idleb, Heimo and Al-Jammasah in Syria; and Bornova and Menemen in Turkey, some of the test entries exceeded the respective local check by a significant margin ($P \leq 0.05$). The stability analysis for seed yield indicated that the five heaviest yielders across locations included FLIP88-85C, FLIP91-220C, FLIP89-29C, FLIP82-150C, and FLIP90-96C gave seed yield of 1924, 1891, 1890, 1881, and 1832 kg/ha, respectively. The entries FLIP91-220C, FLIP89-29C, FLIP82-150C, FLIP91-61C and FLIP90-76C with above average yield were superior to other lines in adaptation.

Table 3.6.1. Distribution of Legume International Nurseries to cooperators for the 1995/96 season.

International Trial/Nursery	No. of sets
Yield Trial Spring (CIYT-Sp-96)	44
Yield Trial Winter, Medit. Region (CIYT-W-MR-96)	50
Yield Trial Southerly Latitudes-1 (CIYT-SL1-96)	11
Yield Trial Southerly Latitudes-2 (CIYT-SL2-96)	12
Yield Trial Latin America (CIYT-LA-96)	8
Screening Nursery Winter (CISN-W-96)	35
Screening Nursery Spring (CISN-Sp-96)	30
Screening Nursery, South. Latitudes-1(CISN-SL1-96)	9
Screening Nursery, South. Latitudes-2(CISN-SL2-96)	7
Screening Nursery, Latin America (CISN-LA-96)	9
F_4 Nursery, Mediterranean Region (CIF$_4$N-MR-96)	27
F_4 Nursery, Southerly Latitudes (CI$_4$N-SL-96)	10
Ascochyta Blight Nursery: Kabuli (CIABN-A-96)	31
Ascochyta Blight Nursery: K. & D. (CIABN-B-96)	16
Fusarium Wilt Nursery (CIFWN-96)	33
Cold Tolerance Nursery (CICTN-96)	31
Drought Tolerance Nursery (CIDTN-96)	40
Total	403

The results of Chickpea International Yield Trial Southerly Latitudes-1 (CIYT-SL1) was reported from 4 locations in 3 countries. At Shanxi in China and Tel Hadya in Syria some of the test entries exceeded the local check by a significant margin. The five heaviest yielding entries across locations included, FLIP90-62C, FLIP90-14C, FLIP91-29C, FLIP90-27C and FLIP90-63C with seed yields of 2568, 2200, 2168, 2128 and 2058 kg/ha, respectively. The stability analysis for seed yield revealed that seven entries namely, FLIP90-14C, FLIP91-29C, FLIP90-27C, FLIP90-63C, FLIP91-10C, FLIP91-33C and FLIP90-85C were relatively adaptable across environments.

The results for Chickpea International Yield Trial Southerly Latitudes-2 (CIYT-SL2) were reported from 11 locations in 7 countries. None of the test entries exceeded the local check in seed yield by a significant margin at any of the southern latitude locations. The five heaviest yielding entries across locations included FLIP90-126C, FLIP88-42C, FLIP90-125C, FLIP89-82C and FLIP88-6C with seed yields of 2375, 2362, 2321, 2312, and 2265 kg/ha, respectively. The stability analysis for seed yield revealed that the entries namely, FLIP88-42C, FLIP90-125C, FLIP88-66C, and FLIP89-120C, were adaptable across environments.

The results for Chickpea International Yield Trial

Latin America (CIYT-LA) were reported from 7 locations in 6 countries. The ANOVA for seed yield revealed that at Saskatoon in Canada, Lincoln in New Zealand and Tel Hadya in Syria, 15, 9, and 10 test entries exceeded the local check in seed yield by a significant margin ($P \leq 0.05$). The five heaviest yielders across locations included FLIP91-132C, FLIP91-93C, FLIP88-6C, FLIP90-32C and FLIP91-98C with seed yields of 2901, 2586, 2584, 2578 and 2558 kg/ha, respectively. The stability analysis for seed yield revealed that a large number of lines namely, FLIP91-132C, FLIP88-6C, FLIP90-32C, ILC97, FLIP89-121C, and ILC 4184 with above average mean and non-significant deviation from regression were adaptable across environments.

The data on seed yield of Chickpea International Screening Nurseries -Winter (CISN-W), -Spring (CISN-SP), -Southerly Latitudes-1 (CISN-SL1), -Southerly Latitudes-2 (CISN-SL2) and -Latin America (CISN-LA) were reported from 26, 19, 3, 4, and 6 locations, respectively. Some of the test entries exceeded the local check by significant margins at 9, 5, 2, 2 and 4 locations, in CISN-W, CISN-SP, CISN-SL1, CISN-SL2 and CISN-LA, respectively. The five heaviest yielding entries across locations are given in Table 3.6.2.

Chickpea International F_4 Nurseries for Mediterranean (CIF_4N-MR) and for Southerly Latitudes (CIF_4N-SL) were supplied to cooperators at 25 and 12 locations, respectively, and only 7 and 3 locations reported the usefulness of these nurseries under their own environmental conditions and made individual plant selections for use in their own breeding materials.

The Chickpea International Ascochyta Blight Nursery results for kabuli type (CIABN-A) were reported from 8 locations and for desi+kabuli type (CIABN-B) from 7 locations. None of the entries (kabuli or desi) was tolerant to Ascochyta blight infestation across all locations. Considering the frequency of occurrence of an entry among the tolerant group (with rating $\leq$ 5 on 1-9 scale) in Kabuli types, entry FLIP 90-85C, showed tolerance at 12 out of 15 locations and appeared best, and was followed by FLIP88-83C, FLIP91-8C, FLIP91-14C, FLIP92-16C, FLIP92-139C and FLIP92-152C, which occurred 11 times. Similarly, among desi lines FLIP87-506C and ICC 4475, ICC 12004, ICC 13416, ICC 135008, ICC 13555, and FLIP 87-507C were tolerant at 4 out of 7 locations. These entries thus exhibited relatively broad-based resistance to Ascochyta blight as compared to others in this nursery. The differential reaction of lines at various places further revealed the presence of variability in the pathogen. This nursery has been very useful in the identification of resistant sources to ascochyta blight and several NARSs have used these resistant sources in their breeding programs.

Table 3.6.2. The five heaviest yielding lines across locations in different chickpea screening nurseries, 1993/94.

Rank	CISN-W	CISN-SP	CISN-SL1	CISN-SL2	CISN-LA
1	F[a] 92-40C	F 92-41C	F 92-77C	ILC 482	F 91-136C
2	F 92-169C	F 9-20C	F 92-176C	F 82-150C	F 91-82C
3	F 92-162C	F 92-47C	F 92-38C	F 92-17C	F 90-119C
4	F 92-120C	F 92-160C	F 91-199C	F 92-91C	F 92-184C
5	ILC 482	F 92-183C	F 92-158C	F 92-94C	F 92-106C

[a] F = FLIP

The results of Chickpea International Leaf miner Nursery (CILMN) were reported from five locations (Tarquinia in Italy, Tel Hadya in Syria, Terbol in Lebanon, and Bornova and Izmir in Turkey). The susceptible check took a score of 9 at Tarquinia in Italy, 7 at Tel Hadya in Syria, 5 at Terbol in Lebanon, and 7 to 9 at other two places (on 1-9 scale, 1=free, 9=highly susceptible). Out of 30 test entries none of the test entries was tolerant to leaf miner at Tarquinia in Italy and one entry ILC 1048 was tolerant to leaf miner at Bornova and Izmir in Turkey where the susceptible check took 9 rating. Across different environments ILC 1048 was tolerant at 4 out of 5 locations and possessed relatively broad-based tolerance to leaf miner.

The results of Chickpea International Cold Tolerance Nursery (CICTN) were reported from 2 locations (Tel Hadya in Syria and Erzurum in Turkey). At Tel Hadya the susceptible check took rating between 7 and 9 on 1-9 scale (where 1=free, 9-killed) and all the other entries were rated between 2 and 3. At Erzurum in Turkey, however, all the test entries including the susceptible check were rated between 1 and 3. This nursery has generated useful information about the level of cold tolerance required for different geographical areas.

4. FORAGE LEGUME IMPROVEMENT

The West Asia and North Africa region is experiencing increased pressure on its agricultural resource base due to rapidly growing livestock and human population. Shortage of animal feed is inflicting a heavy burden on the range lands which are deteriorating.

Severe feed and food deficit also triggered replacement of fallow-barley rotation with continuous barley in the dry-lands and increased cropping on marginal lands with consequent degradation of the soil resource base. Expansion of the cultivation of forage legumes such as vetches (*Vicia* spp.) and chicklings (*Lathyrus* spp.), which are indigenous to the Mediterranean basin, may augment feed supply when sown either to interrupt the barley monoculture or to replace fallow in the fallow-barley rotation[1].

These species are sown and harvested in a single year and can be used for grazing during winter, harvested for hay in spring either in pure stand or in mixtures with cereals (oat, barley or triticale), or for grain and straw at full maturity. They differ from food legume crops only in the end use. They are used mainly to feed livestock, whereas food legumes are for human consumption. Therefore, flexibility in forage legume crops to meet different types of utilization in different agro-ecological zones is always of great importance in producing new adapted cultivars. The introduction of *Vicia* spp. and *Lathyrus* spp. in the rotation also increases the production of feed resources and subsequently, the carrying capacity of the land in a sustainable manner. This is because of the maintenance of organic matter and nitrogen status of soil, improved soil physical conditions and better control of diseases and pests as compared to continuous culture of cereals rotations.

Abd El Moniem, A.M., P.S. Cocks and Y.Swedan, 1988: Tield stability of selected forage vetches (*Vicia* spp.) under rainfed conditions in west Asia. Journal of Agriculture Science, Cambridge. 11,295.301.

Abd El Moniem, A.M., P.S. Cocks and B. Mawlawy, 1990: Genotype - environment interactions and stability analysis for herbage and seed yields of forage peas under rainfed conditions - Plant Breeding. 104, 231-240.

4.1. Environmental Adaptation

Although there is a huge diversity of species of *Vicia* and *Lathyrus* in the Mediterranean region, only a few have been used as feed crops and these have received little attention in the past from plant breeders and agronomists.

There are at least three species of *Lathyrus* and nine species of *Vicia* of potential importantance. We focus only on those species within the two *genera* which are annual and adapted to areas where annual rainfall is between a total of 250 and 400 mm. Table 4.1.1 summarizes the use and environmental adaptation of the species of interest. In areas where rainfall is less than 300 mm *Lathyrus* spp. are common, whereas in higher rainfall areas *V.* spp. are better adapted. *V. narbonensis* is adapted to drier sites, whereas *V. sativa* and *V. ervilia* perform better with a more assured moisture. *V. ervilia, V. villosa* spp. *dasycarpa* and *V. Panonica* are better adapted to cold environments in the highlands than the other Vicia species and those of *Lathyrus*. Underground vetch (*V. sativa* spp. *amphicarpa*) and underground chickling (*Lathyrus ciliolatus*) are adapted to areas with marginal lands, hilly rocky lands and low rainfall.

4.1.1. Germplasm Enhancement

The general objective of the forage breeding program is to develop and produce improved cultivars of feed legume crops, mainly vetches (*Vicia* spp.) and chicklings (*Lathyrus* spp.), and to target these crops to feed livestock in areas with less than 400 mm, rainfall, either in arable lands or marginal non-arable lands. It is also highly desirable to have cultivars that can be recommended for different locations with similar agro-ecological conditions. While attempting to improve yield potential and adaptation, emphasis is laid on ensuring that the quality components such as palatability, nutritive value, protein content, intake of herbage, hay, grain and straw are acceptable by animals. This work is being done in close collaboration with the Pasture, Forage and Livestock Program (PFLP). To develop improved lines of *Vicia* spp. and *Lathyrus* spp. two approaches are adopted. In one, selection is done in the wild accessions to develop improved cultivated types. In the second, hybridization is done to introgress desirable traits from wild accessions. The research is carried out by a multidisciplinary team involving breeder, pathologist, physiologist, entomologist and animal nutritionist.

In 1994/95, 280 accession of *V. ervilia* and 250 accession of *V. narbonensis* were evaluated in collaboration *with the* Genetic Resources Unit (GRU). Selected genotypes of *V. sativa, V. narbonensis* and *L. sativus* were evaluated in observation rows at Tel Hadya. Promising genotypes of *V. sativa, V. panonica, L. sativus* and *L. cicera* were tested in preliminary microplot yield trials at Tel Hadya. Promising lines of *V. sativa, V. narbonensis, V. dasycarpa, V. ervilia, V. palaestina, L. sativus, L. cicera and L. ochrus* were tested in advanced yield trials at Tel Hadya, Breda, Terbol and Kfardan (seasonal rainfall totals 313, 244, 531 and 365 mm, respectively). F_4 lines of *L. sativus* crosses for low neurotoxin B-N-Oxalylamino-L-alanin (BOAA) content and other desirable agronomic traits were evaluated at Tel Hadya and Breda. The diallel crosses of *V. sativa* for nematode resistance and F_4 families of *V. sativa* spp. *sativa* and *V. sativa* spp. *amphicarpa* crosses were tested at Tel Hadya.

A study to investigate the potential of underground vetch in rotation with barley, and the hard seeded breakdown and seed bank dynamic was continued. The response of certain species of *Vicia* and *Lathyrus* to simulated grazing (clipping) and regrowth rates was studies at Tel Hadya and Kfardan. The reaction of promising lines against major foliar diseases was monitored in the disease nurseries at Tel Hadya Table 4.1.2 lists the locations and number of forage legume species/entries studied in 1994/95.

Table 4.1.1. Use and evironmental adaptation of different species of *Lathyrus* and *Vicia* in west Asia and north Africa region and priority objectives for these crops at ICARDA.

Species objectives	Use*	Adaptation	Priority research
L. sativus (common chickling or grasspea)	GZ,G,S	<300mm rain, moderate cold	Resistance to *Orobanche* and foliar diseases; High HI; low BOAA content.
L. cicera (dwarf vetch)	G,S	<300mm rain, moderate cold	Resistance to *Orobanche* and foliar diseases; High HI; low BOAA content.
L. ochrus (ochrus chickling)	G,S	<300mm rain, mild winters	Improve cold tolerance.
V. sativa ssp. *amphicarpa* (underground chickling)	GZ	250mm rain, cold, marginal	improve biomass yield, hard seededness.
L. ciliolantus (underground chickling)	GZ	" " " "	" " " "
V. sativa (common vetch)	GZ,G,S,H	>300mm rain, moderate cold	Leafiness, non shattering pods; resistance to foliar diseases and nematodes; low B-cyano-alanin (BCA)
V. narbonesis (narbon vetch)	G,S	<300mm rain,moderate cold	Earliness, improve HI, Botrytis, downy mildew resistance, low tanin content.
V. *villosa* ssp. dasycarpa (wooly-pod vetch)	GZ,H	>300mm rain, high elevation,	Earliness, increase leaf retention, cold tolerance and improve HI.
V. ervilia (bitter vetch)	G,S	>300mm rain, cold	Reduce pod shattering
V. panonica (Hungarian vetch)	GZ,G,S	>300mm rain high elevation, severe cold	Improve HI, Ascochyta blight resistance
V. hybrida (hybrid vetch)	GZ	<300mm rain high elevation, severe cold	Improve HI
V. palestina (Palaestinian vetch)	GZ,H	>300mm rainfall, mild winter	Improve pod-shattering and cold tolerance

** G = grain; GZ = grazing; H = hay; S = straw.*

All the research was done under rainfed conditions without supplementary irrigation.

As international center with major responsibility for WANA, we aim to serve the national programs through (1) assembling, classifying, evaluating, maintaining and distributing germplasm (2) developing and supplying NARS with breeding populations with adequate diversity to be used in different environments and (3) coordinate international trials to facilitate multilocation testing and identification of widely adapted genotypes.

Table 4.1.2. Trial, location and number of entries of forage legume species studied in 1994/95.

Trial/species	Location	Entries
Nurseries		
Vicia sativa	Tel Hadya	144
V. narbonensis	Tel Hadya	81
Lathyrus sativus	Tel Hadya	36
Microplot Yield Trials		
V. sativa	Tel Hadya	25
V. panonica	Tel Hadya	25
L. sativus	Tel Hadya	36
L. Cicera	Tel Hadya	25
Advanced Yield Trials		
Vicia sativa	Tel Hadya	16
V. sativa	TH-Kfardan-Terbol	25
V. narbonensis	TH-Breda-Kfardan	36
V. dasycarpa	TH-Breda-Kfardan	25
V. ervilia	TH-Kfardan-Terbol	25
V. palaestina	TH-Br-Kfr-Tr	16
L. sativus	TH-Breda-Kfr	25
L. cicera	TH-Breda-Kfr	25
L. ochrus	TH-Breda-Kfr	16
F4 families		
L. sativus crosses	Tel Hadya-Breda	110
F4 families of V. sativax V. amphicarpa		
Crosses	Tel Hadya	36
V. sativa, crosses	Tel Hadya	21
Response to cutting	Tel Hadya-Kfardan	710
Total	758	

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4.1.1.1. Germplasm Evaluation of bitter vetch and narbon vetch.

280 accessions of *Vicia ervilia* and 250 accessions of *V. narbonensis* were evaluated in nursery rows in two separate trials in collaboration with the Genetic Resources Unit (GRU). A total of 49 entries from each trial were selected on the basis of seedling vigour, winter and spring growth, cold effect, leafiness, erect growth habit, and earliness to flowering and maturity.

Special attention will be given to these genotypes in further evaluation of their herbage and seed yields and reaction against major foliar and root diseases during 1995/96. Detailed results of this part are reported in 1995 annual report of the GRU.

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4.1.1.2. Evaluation of two vetches and common chickling for adaptation.

Three experiments using cubic lattice design with three replicates (5 rows each) were conducted; 144 accessions of common vetch (*Vicia sativa*) from different origins were evaluated in the first, 81 accessions of narbon vetch (*V. narbonensis*) were evaluated in the second, and 36 accessions of common chickling (*Lathyrus sativus*) were evaluated in the third. All experiments were fertilized with 40 kg P_2O_5/ha. In these trials the accessions were visually scored at 1-9 scale (1=poor;and 9=very good) for seedling vigour, winter growth, spring growth, leafiness and leaf retention. The two middle rows were harvested at full maturity to estimate grain yield. Times to start flowering, 100% flowering and full maturity were recorded.

A broad variation was observed between the three species and within the same species (Table 4.1.3).

Table 4.1.3. Range, mean, standard error and coefficient of variation (CV%) for eight characters of 144 accessions of common vetch, 81 accessions of narbon vetch and 36 accessions of common chickling (grasspea).

Character[+]	Common vetch				Narbon vetch				Common chickling			
	Range	Mean	SEM±	CV%	Range	Mean	SEM+	CV%	Range	Mean	SEM±	CV%
Seedling vigour[+]	3.7-9.1	7.27	0.95	18.5	1.89-9.0	5.76	0.59	14.1	3.3-9.08	6.1	0.58	16.0
Winter growth[+]	6.0-8.0	6.5	1.02	22.0	2.00-9.0	5.54	0.66	17	3.3-9.0	5.6	0.54	16.4
Spring growth[+]	5.3-9.2	7.9	0.72	12.0	2.00-9.0	6.75	0.62	12.9	5.6-9.0	7.6	0.48	10.9
Leaf-retention[+]	4.3-8.7	5.2	0.92	15.0	2.00-7.0	5.40	0.50	14.9	4.3-8.0	6.1	0.60	14.0
Days to 1st flowering	79-100	90.0	1.09	1.7	79-95	86	1.6	2.7	83-97	91.0	0.98	1.9
Days 100% flowering	92-108	99.0	1.08	1.6	88-105	95	1.9	2.9	94-106	101	0.72	1.2
Days to maturity	127-143	133.0	0.89	0.9	123-138	130	1.4	1.5	128-144	136	0.74	0.8
Grain yield (kg/ha)	328-2012	1107	189	24.0	105-2484	1347	178	18	599-1400	1049	95	15.7

(+) On visual score where 1=poor; 9=very good.

The results show that there is a wide range of adaptation which has been fully documented for reference and future exploitation. Accessions showing good adaptation were identified as promising genotypes for future breeding program.

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4.1.2. Preliminary Evaluation in Microplot Yield Trials (MYT)

The availability of an appropriate genetic base is an indispensable prerequisite of breeding programs. Therefore, special attention is given to the material evaluated in observation rows for adaptation. The following agronomic characters are considered important: rapid winter growth, cold tolerance, early flowering and maturity, leafiness, erect habit, non-shattering pods, high herbage and grain yields and resistance to biotic and abiotic stresses.

Selection for desirable expression of these traits begins in microplot yield trials in the year following nursery-row evaluation and continues through advanced yield trials at Tel Hadya, Breda (Syria), Terbol and Kfardan (Lebanon) locations before regional testing of selected promising lines.

In 1994/95 microplots trials of two *Vicia* species i.e., *Vicia sativa* and *V. panonica*, and two *Lathyrus* spp.i.e., *L. sativus* and *L. cicera* were grown at Tel Hadya in 3.5m^2 plots arranged in triple lattice design. Number of entries for each trial are shown in Table 4.1.2. Seed rate was 100 kg/ha and fertilizers were applied at 40 kg P_2O_5/ha. These microplots were in two sets, one was harvested at 100% flowering to determine the herbage yield (DM) and the other was harvested at maturity to measure seed and straw yields and other agronomic traits.

4.1.2.1. Common Vetch (*Vicia sativa*)

A total of twenty-five selections were tested at Tel-Hadya, and 8 selections which combined both high seed yield and herbage(DM) production were identified for more critical evaluation in (Table 4.1.4). Herbage yield (DM) at 100% flowering varied from 2094 to 3640 kg/ha, total biological yield at maturity from 4342 6007 kg/ha, the grain yield from 841 to 2114 kg/ha and the harvest index from 17 to 38%.

The moderate and high temperatures in winter and spring accompanied with high rainfall at Tel Hadya facilitated the development of foliar diseases such as downy mildew (*Peronospora viciae*), powdery mildew (*Erisiphi pisi*), ascochyta blight (*Ascochyta* spp.) and chocolate spot/blight (*Botrytis fabae*). Natural infection on leaves led to low herbage and grain yields. Selected genotypes showed a relatively high level of natural resistance to the above mentioned diseases are shown in Table 4.1.4. These are identified for further tests under artificial infection in the disease nurseries.

Table 4.1.4. Winter growth (WG), herbage (H),biological (B) and grain (G) yields (kg/ha), harrest index (HI) and days to flower and mature of the top 8 selections of common vetch in preliminary yield trials at Tel Hadya.

		Yield (kg/ha)				Days to	
IFLVS	WG+	H	B	G	HI(%)	Flower	Mature
2608	8.7	2455	5274	1638	31	90	129
2609	7.3	3114	5605	1771	31	99	136
2611	6.0	3066	5758	1539	27	92	130
2617	7.6	2446	5655	1897	34	89	125
2621	6.7	2355	5396	1618	30	91	130
2624	8.7	2739	6007	2114	35	89	127
2626	8.3	2434	5506	1828	33	87	124
2627	9.0	2844	5809	1782	31	87	125
Mean++	7.02	2691	5140	1562	30	91	129
SEM±	0.53	252	322	98	1.7	1.08	0.74
LSD (P=0.05)	1.51	719	924	281	4.7	3.1	2.10
CV(%)	11.0	17	10.9	12	9.6	2.05	0.99

(+) On a scale 1 to 9, where 1 is slow; and 9 very rapid growth meseared in mid-February,1995.

(++) Mean for all 25 selections.

4.1.2.2. Hungarian Vetch (*V. panonica*)

Hungarian vetch is characterized as a cold tolerance species having slow winter growth, followed by rapid spring growth and a long flowering period.

Twenty-five selections were evaluated in microplot field trials at Tel Hadya. Herbage yield varied from 457 to 952 kg/ha. One of the reasons of low grain yield is the mild winter of 1994/95. It initiated flowers and pods when temperatures were relatively high, resulting in a short reproductive period and consequently low yield. Table 4.1.5 shows the performance of top 10 selections for both herbage and grain yields.

Table 4.1.5. Winter growth (WG), herbage (H), biological (B) and grain (G) yields (kg/ha), harvest index (HI) and days to flower and mature of the top 10 selections of Hungarian vetch in preliminary yield trials at Tel Hadya.

		Yield (kg/ha)				Days to	
IFLVS	WG+	H	B	G	HI(%)	Flower	Mature
2655	6.3	2207	4231	773	18	95	144
2656	5.2	2453	4121	884	21	96	141
2658	7.8	2727	3582	796	22	93	138
2660	5.4	2779	4570	952	20	96	139
2664	6.2	2230	4073	709	18	98	142
2666	7.2	2317	4033	739	18	98	141
2670	5.8	2434	4276	769	18	98	144
2673	7.6	2796	4445	853	19	97	142
2675	7.2	2573	4164	884	21	94	139
2676	5.9	2606	3021	750	24	93	139
Mean+	6.5	2453	3845	715	18.5	96	141
SEM±	0.75	165	308	82	1.07	0.99	1.41
LSD (P=0.05)	2.01	469	885	233	3.07	2.82	3.30
CV(%)	18.4	11.0	13.9	20.0	10.02	1.77	1.4

(+) Mean for all 25 selections.

4.1.2.3. Common chickling (*Lathyrus sativus*)

Thirty-six selections were tested at Tel Hadya. Results of the top 10 selections are shown in Table 4.1.6 Herbage yield varied from 1723 to 2801 kg/ha, whereas, the grain yield from

339 to 1093 kg/ha. The late rain favored attack by powdery mildew (*Erisiphe pisi*) when pods were formed.

Table 4.1.6. Winter growth (WG), herbage (H), biological (B) and grain (G) yields (kg/ha), harvest index (HI) and days to flower and mature of the top 10 selections of common chickling in preliminary yield trials at Tel Hadya.

		Yield (kg/ha)				Days to	
IFLLS	WG	H	B	G	HI(%)	Flower	Mature
594	7.9	2713	4914	747	15	94	136
596	7.3	2323	4457	659	14	95	138
598	7.6	2074	4373	1021	23	94	137
600	8.7	2478	4849	899	19	95	137
612	7.6	2430	4876	716	15	97	139
618	6.9	1967	3642	785	21	94	136
624	7.6	2556	3272	651	20	92	137
625	8.0	2523	4495	895	20	94	137
627	6.9	2415	4297	884	20	93	139
587	5.8	2361	3764	1093	29	84	132
Mean[+]	7.20	2286	3987	595	14.9	94	138
SEM±	0.66	216	443	110	1.62	1.39	1.29
LSD (P=0.05)	1.89	610	1250	312	1.58	3.39	3.65
CV(%)	16.00	16.00	19.00	32	19.14	2.55	1.61

(+) Mean for all 36 selections.

4.1.2.4. Dwarf chickling (*Lathyrus cicera*)

Twenty-five selections were tested in microplot yield trials at Tel Hadya. Herbage yield varied from 2399 to 4094 kg/ha, whereas grain yields ranged from 1189 to 2030 kg/ha, and harvest index from 26 to 36%. Table 4.1.7 shows the performance of the top 10 selections. In contrast to *L. sativus*, *L. cicera* is a cold and drought tolerant species. These results indicate the clear need to collect and evaluate native genotypes of *L. cicera* which might show desirable levels of cold and drought tolerance, early winter and spring growth as well as high yield potential.

Table 4.1.8 is a summary of the results of the microplot field trials in 1994/95. As forage legume species can be used for grazing during winter, harvested for hay in spring or harvested for grain and straw at maturity, one may see how the various species will fit into various farming systems.

Table 4.1.7. Winter growth (WG), herbage (H), biological (B) and grain (G) yields (kg/ha), harvest index (HI) and days to flower and mature of the top 10 selections of dwarf chickling in preliminary yield trials at Tel Hadya.

		Yield (kg/ha)				Days to	
IFLIC	WG	H	B	G	HI(%)	Flower	Mature
88	7.0	2873	5384	1707	32	88	128
629	8.0	3435	5040	1649	33	90	128
632	9.0	2902	4849	1657	34	88	129
634	8.0	3126	5013	1554	31	90	131
635	7.0	4094	5760	2030	35	89	126
643	8.0	3302	5005	1459	29	89	130
647	7.3	2509	4750	1691	35	91	127
649	6.0	2399	4324	1588	35	89	126
650	6.0	2777	4929	1665	33	90	128
651	7.0	2796	4785	1722	36	100	127
Mean+	7.5	2937	4905	1563	31.8	89	129
SEM±	0.52	288	479	243	2.9	0.58	0.97
LSD(P=0.05)	1.48	819	13.63	691	8.2	1.68	2.80
CV(%)	11.2	16	17.0	15.3	15.9	1.13	1.30

(+) Mean for all 25 selections.

The high harvest index, early maturity and high level of harvest index of *L. cicera* suggest that it can be used for straw and grain production, whereas, *V. sativa* would be recommended for hay, straw and grain production. *V. panonica* is more suitable for cold areas, because of its cold tolerance and possibly for requirement for vernalization. *L. sativus* can be used for grazing in late winter or for grain and straw production at full maturity.

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4.1.3. Advanced Yield Trials (AYT)

Experiments were carried out to test promising lines of Vicia spp. and Lathyrus spp. at Tel Hadya (TH), Breda (Br), Terbol (T) and Kfardan (Kfr) (Table 4.1.2). Materials used in these trials are either progenies of single plants (selections or pure lines) selected from the wild types or selected F3 and

F4 families of intra-specific crosses.

These lines are selected on the basis of their performance in microplot yield trials for two years. The trials were sown and managed as microplots but with larger plot size ($28m^2$).

4.1.3.1. Advanced yield trials of common-vetch (*V. sativa*)

Sixteen new lines were tested at Tel Hadya for the first year. There were large differences in winter growth, days to flowering and maturity, herbage yield at 100% flowering (DM), degree of leaf retention and harvest index. Herbage yield varied from 1933 to 2909 kg/ha and grain yield from 295 to 2141 kg/ha.

Seed yield was negatively correlated with time to reach maturity ($r= -0.890, P< 0.01$). The results indicate a clear need to continue the search for early maturing genotypes combining high and stable herbage yield with high seed production. Table 4.1.9 shows the major attributes of the tested lines.

Table 4.1.8. Variation in major attributes of two *Vicia* spp. and two *Lathyrus* spp. evaluated in microplot field trials at Tel Hadya.

Attributes	Species			
	V. sativa	*V.panonica*	*L.sativus*	*L.cicera*
Seedling vigour	4.9-9.0	4.9-7.3	5.6-8.6	5.2-8.7
Witer growth	3.6-8.7	5.2-7.9	5.3-9.0	5.6-9.0
Cold effect	4.0-8.0	7.0-9.5	4.2-8.0	6.2-9.5
Spring growth	6.6-9.0	6.6-8.7	5.6-8.6	6.3-8.3
Leafiness	*4.0-9.0*	*4.5-6.5*	*6.0-8.3*	*4.5-7.5*
Days to lowering	93-108	93-101	94-109	87-96
Days to maturity	122-135	137-145	132-143	126-136
Herbage yield(kg/ha)	2094-3640	1944-2796	1723-2801	2399-4094
Grain yield (kg/ha)	841-2114	457-952	339-1093	1189-2030
HI (%)	17-39	13-24	8.0-29	26-36

4.1.3.2. Advanced yield trials of common vetch (*V. sativa*)

Twenty-five promising lines were tested at Tel Hadya, Terbol and Kfardan. The three locations were chosen to sample the environmental conditions of the cereal zone in Syria and Lebanon.

There was large variation between lines within the same location, and between location for winter and spring growth, cold effect, herbage, biological and grain yields and days to flowering and maturity.

Table 4.1.9. Winter growth (WG), herbage (H), biological (B) and grain (G) yields (kg/ha), harrest index (HI) and days to flower and mature of the 16 new lines of common vetch in AYT at Tel Hadya.

		Yield (kg/ha)				Days to	
IFLVS Lines	WG+	H	B	G	HI(%)	Flower	Mature
2560	6.3	2576	4586	1602	35	85	128
2637	9.0	2549	5331	2141	40	78	123
2638	7.3	2440	2025	1655	33	84	130
2639	8.0	2104	5205	1693	32	82	133
2640	9.0	2545	5213	2013	39	78	123
2641	7.3	1990	4524	1606	35	84	128
2642	8.6	2500	4859	1501	31	85	131
2643	8.6	2521	5238	1781	34	83	131
2604	8.3	2492	5133	1558	30	83	132
1429	7.3	2162	4849	1680	35	86	136
2025	5.0	2909	2922	480	17	98	136
2023	6.6	2800	3874	724	19	91	136
2003	7.6	2120	4725	1444	31	89	132
2083	7.3	1940	3626	1057	29	88	132
2564	9.0	2245	3316	295	9	107	142
2679	8.6	1933	3727	632	17	101	143
Mean	7.7	2364	4508	1366	30	87.6	132
SEM+	0.42	185	282	87.0	1.3	0.42	1.2
LSD(P=0.05)	1.22	534	813	251	3.8	1.22	3.4
CV(%)	9.4	14.0	11.0	10.9	7.8	2.3	1.5

(+) On 1 to 9 visual scale basis, where 1 = poor and 9= excellent growth on 15 February, 1995.

The large variations in both herbage and biological yields between locations (Table 4.1.10) were mainly due to the variation among tested lines in their winter and spring growth as indicated by the highly significant correlation between winter growth and total biological yield of r = + 0.527, r = + 0.766 and r=+0.829 at Tel

Hadya, Terbol and Kfardan, respectively.

Table 4.1.10. Location means of winter growth (WG), herbage (H), biological (B) and grain (G) yield (kg/ha), harvest index (HI) and days to flower and mature for 25 lines of common vetch in AYT.

Location	WG	Yield (kg/ha) H	B	G	HI (%)	Days to Flower	Mature
Tel Hadya	7.6 (±0.42)	2174 (±131)	4654 (±228)	1780 (±116)	38.3 (±1.6)	86 (±0.79)	127 (±1.01)
Kfardan	4.5 (±0.44)	2714 (±223)	5190 (±252)	1276 (±115)	25 (±2.0)	110 (±1.6)	163 (±1.83)
Terbol	6.4 (±0.42)	6677 (±440)	1081 (±497)	3377 (±497)	32 (±1.0)	120 (±1.85)	175 (±0.50)

Table 4.1.11. shows the most promising and adapted lines at each location. IFLVS 2483 showed high adaptation and was the most promising across the three location as indicated by its high herbage, grain and biological yields. Its exploitation in future breeding program in case of high herbage and grain yields would be most desirable.

Table 4.1.11. The most promising and adapted lines of common vetch at Tel Hadya, Kfardan and Terbol.

Location	Promising Lines					
Tel Hadya	2483+,	2484,	2485,	2499,	2493,	2499,
	2500,	2503+,	2504,	2505,	2506	
Kfardan	2483,	2487,	2488,	2495,	2503,	2506
Terbol	2483,	2485,	2487,	2488,	2490,	2493,
	2494,	2499,	2503			

(+) The most adapted line across the three locations.

4.1.3.3. Advanced yield trials of narbon vetch (*Vicia narbonensis*)

Thirty-six promising lines of narbon vetch were evaluated under rainfed conditions at Tel Hadya, and Breda in Syria and at Kfardan in Lebanon to determine the relative value of these locations for development of high and stable yielding narbon vetch cultivars.

Winter growth, biological and grain yield and harvest index were measured at each location (Table 4.1.12). There were differences in the performance of the lines at the three locations for all the traits. Yields were greater at Kfardan than Tel Hadya and Breda both for biological and grain yield. The harvest index followed the same trend of the biological and grain yields. The low biological and grain yields at Breda were mainly due late planting and some herbicide damage.

The high yielding lines at each location are shown in Table 4.1.13. None of the lines showed high yield across all the three locations. Lines IFLVN 2478 and 2381 were promising at Tel Hadya and Breda, whereas, IFLVN 2561 was promising at both Tel Hadya and Kfardan.

Table 4.1.12. Location means of Winter growth (WG), biological (B) and grain (G) yields, harvest index (HI) and days to flower and mature for 36 lines of narbon vetch in AYT.

		Yield (kg/ha)			Days to	
Location	WG	B	G	HI(%)	Flower	Mature
Tel Hadya	7.4 (±0.52)	3633 (±386)	1154 (±170)	29.9 (±2.15)	81 (±1.4)	127 (±2.2)
Breda	7.6 (±0.45)	2549 (±200)	532 (±147)	20.8 (±3.36)	91 (±0.36)	131 (±0.84)
Kfardan	4.6 (±0.56)	6992 (±517)	2588 (±218)	36.9 (±1.9)	118 (±0.90)	158 (±1.09)

Table 4.1.13. The most promising and adapted lines of narbon vetch at Tel Hadya, Breda, Kfardan.

Location	Promising Lines					
Tel Hadya	2561, 2470,	2376, 2478	2377,	2381,	2389,	2598,
Breda	2377,	2378,	2381,	2386,	2463,	2470
Kfardan	2561, 2389,	2376, 2388,	2381, 2391,	2382, 2461	2384,	2385,

4.1.3.4. Advanced yield trials of wooly-pod vetch (*V. villosa* spp. *dasycarpa*)

Twenty-five lines were tested at Tel Hadya, Breda and Kfardan. There were differences between lines at the three locations and also between locations (Table 4.1.14).

Table 4.1.14. Location means of winter growth (WG), herbage (H), biological (B) and grain (G) yields, harvest index (HI) and days to flower and mature for 25 lines of Wooly-pod vetch in AYT.

		Yield (kg/ha)				Days to	
Location	WG	H	B	G	HI(%)	Flower	Mature
Tel Hadya	7.7	3244	4590	999	21.8	91	134
	(0.48)	(230)	(359)	(92)	(1.6)	(0.9)	(0.73)
Breda	8.3	2281	1701	328	19.0	88	130
	(0.45)	(207)	(229)	(49)	(2.1)	(0.7)	(0.74)
Kfardan	4.8	4257	4908	709	14.5	105	160
	(0.61)	(577)	(415)	(88)	(1.4)	(0.9)	(0.87)

The low herbage and seed yields at Breda were mainly due to the severe damage by herbicides, which resulted a great reduction in stand in the field. Seed yield was higher at Tel Hadya, whereas, the herbage yields was higher at Kfardan (Table 4.1.14). In contrast to other Vicia spp., wooly-pod vetch is characterized by a long flowering period, high herbage and straw yields and low seed yields. These characteristics make the species most suitable for grazing or hay making. Because we consider leafiness and leaf retention as important selection criteria for wooly-pod vetch, the high herbage yielding lines are characterized by high leaf retention which is a desirable character for high quality hay. Our results also confirmed that wooly-pod vetch is resistant to broomrape (*Orobanche crenata* Forsk.).

Table 4.1.15 shows the most promising lines at the three locations. IFLVD 2431 showed promise at all three locations. More emphasis has to be given to reduced young pod abortion, pod shattering, high leaf retention and earliness to flowering and maturity to improve the productivity of wooly-pod vetch.

Table 4.1.15. The most promising and adapted lines of wooly-pod vetch at Tel Hadya, Breda and Kfardan.

Location	Promising Lines					
Tel Hadya	2431+,	2435,	2441,	2442,	2456,	2565
Breda	2431,	2439,	2443,	2456,	2452	
Kfardan	2562,	2431,	2440,	2443,	2454,	2546, 2452

(+) The most promising and adapted line over the three locations.

4.1.3.5. Advanced yield trials of bitter vetch *(V. ervilia)*

Twenty-five selections were tested at Tel Hadya, Terbol and Kfardan. Herbage yield varied from 1782 to 2974 kg/ha, at Tel Hadya 4126 to 7215 kg/ha at Terbol and from 1627 to 3630 kg/ha at Kfardan. The total biological yield varied from 4086 to 5920 kg/ha at Tel Hadya, 7695 to 11733 kg/ha at Terbol and from 3833 to 5750 kg/ha at Kfardan. Herbage, biological and grain yields were higher at Terbol than Tel Hadya and Kfardan. (Table 4.1.16.)

Table 4.1.16. Location means of winter growth (WG), herbage (H), biological (B) and grain (G) yields, harvest index (HI) and days to flower and mature for 25 lines of bitter vetch in AYT.

		Yield (kg/ha)				Days to	
Location	WG	H	B	G	HI(%)	Flower	Mature
Tel Hadya	7.8 (0.35)	2440 (165)	5096 (246)	1900 (118)	37 (1.8)	81 (0.9)	127 (128)
Kfardan	4.9 (0.48)	2551 (443)	4920 (280)	902 (182)	18 (3.9)	105 (2.7)	144 (1.86)
Terbol	6.2 (0.32)	5485 (540)	9860 (708)	2713 (178)	27 (1.3)	110 (1.9)	150 (1.98)

Bitter vetch showed cold tolerance, and rapid winter and spring growth as compared to other vetches. Its yield was substantially better than other vetches. No symptoms of downy mildew, powdery mildew or ascochyta blight appeared, but the late maturing selections were affected by *Orobanche*. Table 4.1.17 shows the most promising and adapted selections at each locations. IVLVE 2515 was promising at all locations with consistent rank.

Table 4.1.17. The most promising and adapted lines of bitter vetch at Tel Hadya, Kfardan and Terbol.

Location	Promising Lines					
Tel Hadya	2508,	2509,	2510,	2514,	2515+,	2516,
	2518,	2520,	2648,	2649		
Kfardan	2563,	2515,	2522,	2644,	2646,	2651
Terbol	2512,	2515,	2521,	2644,	2648,	2650,
	2651					

(+) The most promising and adapted lines over the three locations.

1.1.3.6. Advanced yield trials of palestine vetch *(Vicia palaestina)*

Sixteen promising lines of *Vicia palaestina* were tested at Tel Hadya, Breda, Kfardan and Terbol Table 4.1.18. The herbage yield varied from 1030 to 2090 kg/ha, 1059 to 1638 kg/ha, 833 to 1489 kg/ha and from 891 to 2028 kg/ha at Tel Hadya, Breda, Kfardan and Terbol, respectively. At maturity, the grain yield ranged from 1176 to 1749 kg/ha, 343 to 859 kg/ha, 146 to 572 kg/ha and from 907 to 2924 kg/ha at Tel Hadya, Breda, Kfardan and Terbol, respectively.

Palestine vetch at Terbol and Tel Hadya was higher yielding than at Breda and Kfardan. Palestine vetch grows slowly in winter but rapidly in the Spring. The plants are vigorous, tall, erect and compete well with weeds during vegetative period. It produces high straw yields due to the tall plants and its leaf retention. The disadvantage of palaestine vetch is its high pod-shattering at maturity. So far no genotypes have been identified as non-shattering.

Table 4.1.18. Herbage, biological and grain yields (kg/ha) for the 16 lines of Palaestine vetch (*Vicia palaestina*) in AYT at Tel Hadya (TH), Breda (Br), Kfardan(Kfr) and Terbol(T).

IFLVPA	Herbage yield (kg/ha)				Biological yield (kg/ha)				Grain yield (kg/ha)			
	TH	Br.	Kfr.	T	TH	Br.	Kfr.	T	TH	Br.	Kfr.	T
2523	1030	1059	1159	1281	3364	2557	2687	6784	1380	777	415	907
2524	1682	1099	1234	1628	3572	2298	2869	7628	1384	828	377	1260
2525	1801	1638	1088	1375	4673	2487	2476	7220	1549	600	189	1007
2526	1955	1624	833	1177	3279	1941	2024	5286	1312	343	182	1928
2527	1535	1453	1126	1346	3520	2764	2147	6658	1382	615	213	1479
2528	2090	1251	1175	1491	4282	2331	2555	7307	1749	669	294	1280
2529	1185	1258	842	891	3623	2643	2369	6126	1435	700	321	1029
2530	2014	1455	1320	2028	4448	2725	2981	6650	1472	859	440	1090
2531	1757	1162	1246	1181	3831	2499	2855	7113	1495	735	450	1520
2532	1949	1624	1161	1927	4631	2893	2724	7432	1542	625	572	1195
2533	1417	1456	1012	1501	3285	2292	2271	6047	1207	580	232	1466
2534	1514	1254	1385	1482	3754	2718	2612	6776	1443	718	238	1547
2535	1416	1243	1489	1617	3527	2160	2179	6201	1333	499	177	1378
2536	1596	1386	1194	1960	3491	2395	2315	8165	1513	580	146	2924
2537	1750	1263	995	1473	3307	2487	2875	7200	1176	663	490	2249
2538	1416	1203	875	1318	3868	2361	2430	5950	1422	471	383	1398
Mean	1632	1339	1134	1480	3778	2472	2523	6784	1425	624	319	1475
SEM±	192	90	138	193	284	172	201	501	119	78	90	210
LSD (P=0.05)	554	283	400	569	835	505	590	1475	349	225	266	617
CV%	20.4	19	21	22	13	18	14	13	15	31	30	25

4.1.3.7. Advanced yield trials of three *Lathyrus* spp.

Promising lines of three *Lathyrus* spp.i.e. *Lathyrus sativus,* (common chickling or grass pea), *L. cicera* (dwarf chickling) and *L. ochrus* (Ochrus chickling) were tested at Tel Hadya, Breda and Kfardan. The numbers of lines tested at each location are shown in Table 4.1.2.

Common chickling

Table 4.1.19 shows herbage, biological and grain yields at the three locations for the top five lines. Herbage and biological yields were greater at Kfardan than Tel Hadya and Breda, whereas the grain yields was greater at Tel Hadya than Breda and Kfardan. The low grain yield at Breda was mainly due to the effect of herbicides and late planting. The top five lines were identified for more multilocation testing in the international nurseries program.

Dwarf chickling

Twenty five lines of dwarf chickling were evaluated at Tel Hadya, Breda and Kfardan. The crop at Kfardan was higher in herbage, biological and grain yields than at Tel Hadya and Breda (Table 4.1.20). Mean herbage yield varied from 1380 to 2511 kg/ha at Tel Hadya, 836 to 1287 kg/ha at Breda and from 3109 to 5243 kg/ha at Kfardan, whereas, the grain yield ranged from 1406 to 2084 kg/ha at Tel Hadya, 216 to 1092 kg/ha at Breda and from 1574 to 2145 kg/ha at Kfardan. Table 4.1.20 shows the most promising lines combining both high herbage and grain yields. Dwarf chickling showed more cold tolerance than common chickling and ochrus chickling at Kfardan.

Table 4.1.19 Herbage, biological and grain yields (kg/ha) for the top 5 lines of common chickling (*Lathirus sativus*) in advanced yield trials at Tel Hadya (TH), Breda (Br) and Kfardan (KFR.).

IFLLS	Herbage Yield(kg/ha)			Biologycal Yield(kg/ha)			Grain Yield (kg/ha)		
	TH	Br.	Kfr.	TH	BR.	Kfr.	TH	Br.	Kfr.
567	1423	1265	4559	3717	2359	5729	1187	684	1117
504	1487	1313	4941	3793	2518	5611	1130	756	1083
516	1366	1285	4605	3845	2629	5432	1084	624	1000
528	1486	1313	4849	4020	2533	5609	1188	616	983
531	1517	1290	4717	3820	2700	5059	1136	710	933
Mean+	1397	1272	4005	3745	2422	5050	1008	500	864
SEM	136	103	550	247	180	380	90	29	107
LSD(0.05)	386	297	1566	707	516	1090	258	110	305
CV(%)	17.00	15.00	24	12.00	20.00	14.00	16.00	31.00	21.5

(+) Mean for all 25 selections.

Table 4.1.20. erbage, biological and grain yields (kg/ha) for the top 5 lines of dwarf chickling (*Lathyrus cicera*) in advanced yield trials atTel Hadya (TH), Breda (Br) and Kfardan (Kfr.).

selection IFLLC	Herbage Yield(kg/ha)			Biologycal Yield(kg/ha)			Grain Yield (kg/ha)		
	TH	Br.	Kfr.	TH	BR.	Kfr.	TH	Br.	Kfr.
586	2263	1040	4380	4977	2088	6385	1739	606	1985
491	2511	1287	5243	5297	2801	5968	1874	1012	1831
492	2029	1141	4246	5085	2094	6410	1960	510	2145
569	2014	1070	4151	4874	2526	6462	1830	837	1925
576	2401	1112	4013	4666	1924	6389	1712	500	1948
Mean[+]	1680	1020	3838	4718	1910	5971	1700	510	1868
SEM±	186	102	431	253	187	398	115	90	147
LSD(P=0.05)	534	292	1227	719	541	1142	328	210	422
CV(%)	17	17	19	12	20	12	13	30	14

(+) Mean for all 25 lines.

Ochrus chickling

Sixteen lines of ochrus chickling were tested at Tel Hadya, Breda and Kfardan. At Tel Hadya herbage, biological and grain yields were greater than Breda and Kfardan (Table 4.1.21). This is due to the mild winter of 1995 and the late maturing lines which were unaffected by *Orobanche*, because of its resistance[2]. The low yield at Kfardan was due its susceptibility to cold[3].

Conclusions

The results of the advanced yield trials of six *Vicia* species and three *Lathyrus* species show a large diversity in herbage, grain and straw production. These species can contribute significantly to animal production in rainfed agriculture. Their use in rotation with cereals will increase yields and total biomass[4]. Furthermore, they will increase the sustainability of farming systems by acting as a disease and *Orobanche* break, and by contributing to the nitrogen nutrition of cereals. As forage legumes can be used for grazing hay, straw and grain, we can begin to see how the various species will meet the farmer's needs in the prevailing farming systems of WANA. The high grain and straw yields and early maturity of narbon vetch make it ideal feed legume for producing winter stocks of straw and grain for feeding sheep during the peak of feed demand

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Linke, K.H., A.M. Abd El Moniem and M.C. Saxena, 1993: Variation in resistance of same forage legumes species to *Orobanche Cerenata* Fask. Field Crop Research, 32,277-285.

Abd El Moniem, A.M. and P.S. Cocks, 1993: Adaptation and Yield stability of selected lines of *Lathyrus* spp. under rainfed conditions in West Asia, Euphytica 66:89-97.

Jones, M.J. and M. Singh, 1995: Yields of Crop dry matter and nitrogen in long-term barley rotation trials at two sites in northen Syria. J. Agric. Sci., Camb., 124, 389-402.

in winter[5]. It does not lose its leaves following frost, like many other feed legumes, and its seed contain around 28% crude protein. Palestine vetch can be used for hay making in spring because of its rapid spring growth, leaf-retention, and vigorous growth habit. Common vetch is a versatile feed legume crop. The rapid winter growth types can be used for early grazing, the non-shattering erect types can be used to produce grain and straw, and the leafy and rapid spring growth types can be used for hay making either in pure stand or in mixtures with barley or oats. For its long flowering period and cold tolerance, wooly-pod vetch can be used for grazing or hay making in high elevation areas[6]. For farmers who require hay or grazing dwarf chickling could be another option. The common chickling is susceptible to *Orobanche*, but in dry areas it can still be used for grain and straw.

Abd El Moniem, A.M., 1992: Narbon vetch (*Vicica narbonesis*): Apotential feed legume crop for dry areas in West Asia. J. Agronomy & Crop Science 169, 347-353.

Abd El Moniem, A.M., 1990: Growth analysis, herbage and seed yield of certain forage legume species under rainfed conditions. J. Agronomy & Crop Science 164, 34-41.

Table 4.1.21. Herbage, biological and grain yields (kg/ha) for the top 5 lines of ochrus chickling (*Lathyrus ochrus*) in advanced yield trials at Tel Hadya (TH), Breda (Br) and Kfardan (Kfr.).

selection	Herbage Yield(kg/ha)			Biologycal Yield(kg/ha)			Grain Yield (kg/ha)		
IFLLO	TH	Br.	Kfr.	TH	Br.	Kfr.	TH	Br.	Kfr.
537	2271	1504	1675	5073	2990	5035	1474	634	1004
538	2468	1876	1556	5252	2752	4941	1906	550	1089
540	2086	1914	1607	5383	2585	4697	1848	568	1050
542	2118	1816	1685	4647	2660	5006	1540	744	892
545	2305	2180	1697	5159	2592	4653	1693	673	904
Mean*	2370	1832	1574	5076	2610	4583	1177	522	837
SEM±	165	132	169	252	255	2677	136	99	758
LSD(P=0.05)	485	382	498	485	622	787	400	171	2230
CV(%)	13	17	18	12	28	12	16	270	15

Because of its resistance to *Orobanche* and susceptibility to frost spells in early winters ochrus chickling can be used for grazing or for grain and straw in areas heavely infested with *Orobanche* with mild winters. For its high nutritive value of grain and straw, earliness and erect growth habit, bitter vetch is more suitable for grain and straw for winter feeding in wet areas with more than 350 mm rainfall.

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4.1.4. Nutritional Quality.

Achieving high yield potential and adaptation to different niches and agroecological zones needs to be complemented by ensuring that the end products are acceptable by livestock. Therefore, quality parameters of hay, straw and grain are also given great consideration in the breeding program.

4.1.4.1. Hay and Straw Quality of *Vicia* spp. and *Lathyrus* spp.

The quality parameters utilized in the forage breeding program are Protein %, Neutral Detergent Fiber (NDF), Acid Detergent Fiber (ADF), and Dry Matter Organic Matter Digestibility (DOMD%).

Large differences were observed both between and within species. Hays of *V. sativa, V. ervilia* and *V. villosa spp. dasycarpa* are high in protein content, and the fiber contents of *V. ervilia* are low, resulting in high digestibility. Fiber content is high in the hay of *V. palaestina*, and its digestibility is low. This is mainly due to its tiny leaves, tall plants, and low leaf: stem ratio (Table 4.1.22a).

Hays of *L. sativus* and *L. cicera* are high in protein content, NDF% and ADF%; whereas the hay of *L. ochrus* is relatively low in protein content and fiber resulting high digestibility. This is mainly due to high in degree of leafiness and relatively thick stems. The same trend was found in the case of straws (Table 4.1.22b.).

4.1.4.2. Tanins and Protein Contents in *Vicia narbonensis* Seeds

Tanin in narbon vetch seeds has a negative effect as an anti-palatability factor and a positive effect as a protein out-flow from rumen. Tanins and protein content of eighty one new selections of narbon vetch indicate that none of the selections was tanin-free. The tanin content of the whole seed varied from 0.10 to 0.38% whereas, protein content varied from 20.3 to 28.2%.

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Table 4.1.22.a. Mean and range of Protein%, NDF%, ADF% and DOMD% for hays and straws of five *Vicia* spp. promising lines in advanced yield trials at Tel Hadya.

Species		Hay				Straw			
		Protein%	NDF%	ADF%	DOMD%	Protein%	NDF%	ADF%	DOMD%
Vicia sativa	Mean	22.00	28.60	18.50	78.00	11.70	48.00	34.00	45.00
	Range	18-25	25-32	16-22	73-82	10-14	42-54	32-37	37-52
V. ervilia	Mean	22.00	15.00	12.30	81.00	12.00	42.00	31.00	53.00
	Range	19-27	11-18	10-15	78-85	8-14	35-45	24-33	45-65
V.vil. ssp. dasycarpa	Mean	20.8	30.6	19.00	71.00	12.90	48.00	34.00	35.00
	Range	18-24	26-35	16-21	69-77	9-14	46-52	32-35	29-39
V. palaestina	Mean	18.50	35.00	21.70	63.00	7.00	58.00	37.00	21.00
	Range	14-20	33-42	20-25	57-67	2-9	54-61	34-39	13-28
V. narbonensis	Mean	-	-	-	-	14.00	43.00	32.00	47.00
	Range	-	-	-	-	9-16	36-50	30-35	41-52

Table 4.1.22.b. Mean and range of Protein%, NDF%, ADF% and DOMD% for hays and straws of three *Lathyrus* spp. promising lines in advanced yield trials at Tel Hadya.

Species		Hay				Straw			
		Protein%	NDF%	ADF%	DOMD%	Protein%	NDF%	ADF%	DOMD%
Lathyrus sativus	Mean	21.30	35.20	18.30	83.00	18.60	50.60	26.00	59.00
	Range	14-24	26-41	16-20	80-86	16-20	43-58	23-29	53-66
Lathyrus cicera	Mean	21.50	32.00	19.00	79.00	16.00	61.00	33.00	55.00
	Range	19-23	27-35	15-23	75-82	14-18	52-67	26-36	49-70
Lathyrus ochrus	Mean	17.50	26.00	17.00	86.00	14.5	43.00	26.00	62.00
	Range	14-24	19-29	14-23	82-89	12-16	33-53	22-29	56-66

4.1.4.3. Canavanine and Protein Content of Bitter Vetch (*Vicia ervilia*)

It has been reported that *Vicia* spp. differ widely in their secondary metabolite composition[7]. One of these compounds is the non-protein amino acid "canavanine", which is considered as an anti-palatability factor. This factor is a complex compound that is difficult to measure by conventional methods. Rapid analytical techniques are needed to determine the concentration of this compound in our breeding lines, such as the Capillary Zone Electrophoreses (CZE) technique developed at the Center for Legumes in Maditeranean Agriculture (CLIMA).

Seeds of twenty five promising lines, grown in advanced yield trials at Tel Hadya, were analyzed for canavanine and protein contents. The canavanine content varied from 0.05 to 0.14% with a mean of 0.09%. The protein content varied from 20.64 to 26.5% with a mean 22.4%. IFLVE 2518 was the lowest in canavanine and the highest in protein content. This indicates that there is a good scope for selection for low canavanine and high protein contents.

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4.1.4.4 Evaluation of B-ODAP and protein content of promising lines of three *Lathyrus* spp. at Tel Hadya, Breda and Kfardan

Protein and the neurotoxin B-N-Oxalyl-L- α-β-Diamino propionic Acid (B-ODAP) Syn., B-N-Oxalyl-L-Alanine (BOAA) for promising lines of *Lathyrus sativus*, *L. cicera* and *L. ochrus* grown at Tel Hadya, Breda and Kfardan, was estimated with a Near-Infrared Reflectance (NIR), Model NEOTEC 5000, with a wave length setting between 1100 and 2500 mm. Every tenth sample was verified by Macro-Kjeldahl method for crude protein and classical spectrophotometric

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Enneking, D. The Toxicity of *Vicia* spp and their utilization as grain legumes. PH.D. Thesis. University of Adelaide, Wait Agriculture Research Institute, South Australia, 1994.

analysis for B-ODAP. It was possible to develop a good calibration which permitted a good correlation (r= 0.90 and 0.93) for B-ODAP and crude protein, respectively.

Table 4.1.23 summarizes the results of crude protein % and B-ODAP% for three *Lathyrus* spp. grown at three locations. The results indicate that none of the *Lathyrus* spp. lines was B-ODAP free, although some lines were quite low showing large variation between species, between lines within species and between locations. *L. ochrus* had the highest protein and B-ODAP contents at the three locations, whereas *L. cicera* had low protein and B-ODAP content at all locations. Breda was the location with the highest protein and B-ODAP content. The presence of such variation in protein and B-ODAP suggest that there is a good potential for developing *L. sativus*, *L. cicera* and *L. ochrus* lines with low B-ODAP and high protein contents.

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4.1.5. Genetic Improvement

4.1.5.1. Improving Nutritional Quality of *Lathyrus sativus* by Breeding

In 1991/92 21 high yielding and early maturing *L. sativus* lines with a B-ODAP content ranging from 0.2 to 0.75% were crossed with late maturing low B-ODAP lines with less than 0.1%. Due to transgressive segregation toward earliness and high B-ODAP content, a large proportion of the F_2 population matured earlier than the parents. Selection in F_3 was for early maturity, small and large seed size, light cream seed colour and less than 0.1% B-ODAP. In 1994/95, 85 families with their parents were grown under rainfed at Tel Hadya and Breda to assess their yield potential and B-ODAP content.

Table 4.1.23. Mean crude protein (%) and B-ODAP(%) of three *Lathyrus* spp. at Tel Hadya, Breda and Kfardan.

Species		Crude protein			B-ODAP (%)		
		Tel Hadya	Breda	Kfardan	Tel Hadya	Breda	Kfardan
Lathyrus sativus	Mean	23.90	26.60	27.09	00.20	00.31	00.22
	±SEM	00.52	00.89	00.77	00.07	00.09	00.04
	Range	23-26	24-28	26-29	0.11-0.25	0.28-0.50	0.20-29
L. cicera	Mean	22.00	25.00	26.00	00.19	00.22	00.21
	±SEM	01.01	00.51	00.71	0-09	00.06	00.05
	Range	20-24	23-26	25-28	0.11-0.21	0.11-0.37	0.14-0.29
L. ochrus	Mean	26.07	27.60	28.00	00.53	00.61	00.56
	±SEM	01.14	01.62	00.70	00.09	00.14	00.09
	Range	24-28	23-29	26-29	0.8-1.05	0.34-0.79	0.51-0.74

Three F_4 families had low B-ODAP (0.02-0.07%) and had large white or cream-colored seeds and white flowers. The seed yield ranged from 0.6 to 1.2 tons/ha. The effect of location on B-ODAP content was not significant. These three low B-ODAP families were 10-15 days later in maturity than the lines with high B-ODAP at both locations. Days to maturity were significantly correlated with B-ODAP content (r=-0.29 and -0.36, P<0.01) at Tel Hadya and Breda, respectively. The results also showed that mean B-ODAP content was associated with seed colour: white-seeded families had a mean B-ODAP content of 0.17%, light grey-seeded families 0.29%, grey- or brown-seeded families 0.41 and black-seeded families 0.65%.

The study shows that B-ODAP content in *L. sativus* (grasspea) can be reduced by breeding. It is possible, however, that the presence of anti-nutritional factors such as B-ODAP is associated with the ability of this species to resist biotic and abiotic stresses, and that the total elimination of B-ODAP may increase the susceptibility of the plant to these stresses. This aspect is currently being investigated. Preliminary observations have revealed that low B-ODAP lines are late to flower and mature and have higher susceptibility to downy mildew (*Pernospora vicae*) and powdery mildew (*Erysiphe martii* f.sp. lathyri).

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4.1.5.2. Breeding Common Vetch for Non-shattering Pod (Seed Retention) Character

Pod shattering is common in *Vicia sativa* and it restricts the use of these legumes for producing feed. Our screening program for non-shattering characteristic indicated the existence of wide variation for this trait among common vetch germplasm collected from different places[8]. Three wild accessions with almost completely non-shattering pod habit were identified and isolated for use as genetic

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Abd El Moniem, A.M., 1993: Selection for non-shattering common vetch (*Vicia sativa*. L.). Plant Breeding 110, 168-171.

resources in the breeding program.

Genetic studies revealed that non-shattering is conditioned by single recessive gene[8]. The incorporation of non-shattering gene into agronomically promising lines was achieved by backcrossing and selection done in backcross one (BC1) to BC5, for non-shattering and early flowering and maturity. One superior selection (IFLVS-NS#715) was identified having 95-97% non-shattering pods, as compared to 40-45% in the original cultivated lines. This selection was released as a cultivar in Jordan in 1993 and is used by farmers in Jordan and it is on-farm trials in Iraq, Lebanon and Syria.

Breeding non-shattering cultivars in common vetch is continuing with the aim to incorporate the erect habit. IFLVS (NSE) #2558 was identified as erect and non-shattering and also characterized by white flower colour, which facilitates maintenance selection of the line. Seed multiplication for this line is being done at Tel Hadya and Terbol stations for distribution to national programs. The practical benefits of developing non-shattering and erect lines include increased grain yield, reduced problem of volunteers in the subsequent cereal crops, improved opportunity for mechanical harvesting and increased flexibility in the time of harvest.

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4.1.5.3. Studies of Hybrids Between *V. Sativa* ssp. *sativa* and *V. sativa* spp. *amphicarpa*

Species and subspecies hybridization is an important aspect in feed legume breeding to recombine useful genes carried by the parental species and also to widen the genetic base for selection. Our studies on amphicarpic type of *V. sativa* indicated that the ability of underground vetch (*V. sativa* spp. *amphicarpa*) to produce aerial and underground pods increases its winter hardiness, drought tolerance and persistence under heavy grazing. The disadvantages of an underground podding habit in vetch, which may limit its utilization as a pasture plant, are low rate of vegetative growth, shattering of above-ground pods and the dependence of amphicarpy to edaphic conditions. In contrast, the common vetch (*Vicia sativa* spp. *sativa*) grows well under favourable conditions but is not cold and drought tolerant and there are some lines with non-shattering pods.

To enhance the herbage production of underground vetch and to improve the drought and cold tolerance of the common vetch, crosses between the two species were initiated in 1990 to combine specific characters from the two subspecies, to study the compatibility between them, and to generate additonal variability.

Three promising lines of common vetch with non-shattering pods i.e. IFLVS #1416, 713 and 1448 were crossed with two lines of underground vetch IFLVA #2416 and 2614 to make twelve crosses (including reciprocals). High vigour was observed in the F_1 plants carrying few underground pods. The F_2 plants and F_3 progenies showed striking variability. Through multiple trait selection, the segregating F_3 progenies were classified according to the number of underground pods and the above-ground biomass production, and selections were made for progenies with 4-9 underground pods per plant, cold and drought tolerance, increased vegetative and leafiness and non-shattering above-ground pods similar to those of common vetch. Selection was carried out through F_4 for families having 6-10 underground pods, and vigorous above-ground growth. These selected families were stable in 1994/95 and the above-ground herbage yield was three times higher than the amphicarpa type parents, while maintaining 6-10 pods underground. Seeds of these new lines were multiplied and are being used to rehabilitate the marginal lands at Tel Hadya, in collaboration with PFLP.

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4.1.5.4 Genetic studies of root-knot nematode (*Meloidogyne artiellia*) resistance in common vetch

A diallel cross was used to study the inheritance of resistance to root-knot nematode (*Meloidogyne artiellia* Franklin), which is a limiting factor in introducing Common vetch in West Asia and North Africa.

Seven lines of common vetch (*V. sativa*) namely IFLVS #713, 1429, 2003, 2025, 2023, 2043 and 2052 were selected as parents of a diallel cross. These lines range from resistant (IFLVS 713 and 1249) to susceptible (IFLVS 2003) to highly susceptible (IFLVS 2025, 2023, 2040 and 2052).

The 21 F_1 were grown under artificial conditions in the green house. The infection rate was 20 second stage larvae/g soil. Seeds were sown in pots in a randomized complete block design with four replications. At flowering stage the plants were harvested, the roots washed carefully, and the density of the nematode on the roots determined. Nematodes were extracted and counted as number of nematodes per roots.

The mean number of nematodes (juveniles and larvae) extracted from the roots of the plants of different F_1 varied significantly (Table 4.1.24). In general, nematode reproduction was highest in F_1 having IFLVS 2025, 2023, 2040 and 2052 as one parent, while F_1 with IFLVS 713 and 1429 as one parent tended to have lower level of nematode reproduction. This indicates that F1 with IFLVS 713 and 1429 as one parent tended to be most resistant. The low levels of nematode reproduction exhibited by F_1 such as IFLVS 713x2003, 713x2025, 1429x2025 and 1429x2003, also indicate that development of common vetch resistant to *M. artiella* reproduction could be possible. The use of root-knot nematode resistant material in crop rotation schemes would provide common vetch growers with an alternative to nematicides for controlling the nematode. This is especially important because nematicides are cost prohibitive.

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Table 4.1.24. Numbers of nematode (Juveniles & larvae) from the roots of common vetch hybrids grown in the green house.

Hybrid			No. of nematode/ g roots	Hybrid			No.of nematode/ g roots
713	X	1429	21	2003	X	2025	368
713	X	2003	142	2003	X	2023	473
713	X	2025	96	2003	X	2040	10021
713	X	2023	133	2003	X	2025	9795
713	X	2040	320	2025	X	2023	10462
713	X	2052	142	2025	X	2040	11392
1429	X	2003	28	2025	X	2052	10781
1429	X	2025	36	2023	X	2040	13531
1429	X	2023	52	2023	X	2025	12430
1429	X	2040	167	2040	X	2052	14400
1429	X	2052	15				
Mean							4497
LSD (p=0.05)							570

4.1.6. Forage Legume Agronomy

4.1.6.1. Response of Promising Lines of *Vicia* and *Lathyrus* spp. to Simulated Grazing at Different Growth Stages at Tel Hadya (Syria) and Kfardan (Lebanon)

The objective of this trial was to study the response of *Vicia* and *Lathyrus* spp. to simulated grazing (cutting), the ability for regrowth after cutting under rainfed conditions and some forage quality parameters at various times during the growing season.

One of the questions in assessing forage productivity is at what time herbage production and quality of different *Vicia* and *Lathyrus* spp. should be measured. Our practice has been to measure herbage yield at 100% flowering stage because this is the time normally recommended for hay cuts and to measure seed yield at maturity. But if farmers are more interested in grazing, this would seem unsatisfactory. Furthermore, harvesting at a particular phenological stage can mean large difference in age and hence yield. We need to know whether growth continues after cutting at a particular stage and the changes in forage quality. Also, the regrowth rate after cutting at different growth stages may vary among different *Vicia* and *Lathyrus* spp.

The background and treatments imposed in this trial are described in the 1994 Germplasm Program Annual Report for Legumes.

Based on dry-matter production (dry herbage) of each entry from the first and second harvest, all entries were able to survive during the winter at both locations (23 frost nights at Tel Hadya, with an absolute minimum temperature -6.6^{0}C and a rainfall total of 313 mm, and 34 frost nights with an absolute minimum temperature of -6.0^{0}C and rainfall of 365 mm at Kfardan).

There were clear differences in phenology (Table 4.1.25). Palestine vetch and narbon vetch were early flowering at both locations compared to the other entries. Flowering at Tel Hadya was generally earlier than Kfardan. This confirmed the result obtained in 1994.

During the vegetative stage of growth differences in dry-matter yield at each cut were large (Table 4.1.26). Up to cut 3, bitter vetch, wooly-pod vetch, narbon vetch and orchus chickling gave the highest herbage yield. Ochrus chickling produced 0.5t/ha at first cut, but at the fifth cut (100% flowering) its production increased up to 6t/ha at Tel Hadya and 4.5t/ha at Kfardan (Table 4.1.26). Entries characterized by cold tolerance and rapid winter growth such as bitter vetch and wooly - pod vetch produced high herbage at the first cut.

Table 4.1.25. Phenological stages at different cutting times and plant ages for Vicia & Lathyrus spp. grown at Tel Hadya and Kfardan, 1994.

Location/Cut No.		Plant age (days)	Vs713+	Vs2560	Ve2517	Vp2525	Vh2548	Vd683	Vn2380	Lo542	Ls587	Lc501
Tel Hadya	1	57	V*	V	V	V	V	V	V	V	V	V
	2	73	V	V	V	V	V	V	V	V	V	V
	3	87	V	V	V	StartF	V	V	StartF	V	V	V
	4	101	StartF	V	StartF	50% F	StartF	StartF	50% F	50% F	V	StartF
	5	116	50%	StartF	50% F	100% F	100% F	25% F	100% F	100% F	StartF	50%
Kfardan	1	52	V	V	V	V	V	V	V	V	V	V
	2	70	V	V	V	V	V	V	V	V	V	V
	3	86	V	V	V	V	V	V	V	V	V	V
	4	101	V	V	V	StartF	StartF	V	StartF	StartF	V	V
	5	119	StartF	StartF	StartF	50% F	50% F	StartF	50% F	50% F	V	StartF

*V = Vegetative, F = Flowering.
+ = Vs = *Vicia sativa*, Vc = *V. ervilia*, Vp = *V. palaestina*, Vh = *V. hubrida*, Vd = *V. dasycarpa*, Vn. = *V. narbonansis*, Lo = Lathyrus orchus, Ls = *L. sativus*, L.c = *L. cicera*.

Table 4.1.26. Herbage yield (DM) (kg/ha) of different *Vicia* and *Lathyrus* spp. from cut 1 to 5 at Tel Hadya and Kfardan.

Location/Cut No.		Vs713	Vs2560	Ve2517	Vp2525	Vh2548	Vd683	Vn2380	Lo542	Ls587	Lc501	LSD (P=0.05)
Tel Hadya	1	537	550	640	310	300	720	590	501	310	301	197
	2	1215	1300	1400	1100	835	1900	1500	1450	1100	980	360
	3	2923	2500	3100	1800	1900	3500	2900	3200	1700	2000	674
	4	3700	2900	5000	3400	3200	4900	4100	4200	2600	2600	745
	5	4100	4700	6900	4900	4950	5200	5800	6100	4200	4000	1118
Kfardan	1	610	700	910	410	390	820	610	520	390	320	288
	2	1490	1610	1700	900	890	1820	1600	1400	910	890	357
	3	2800	2200	2800	1310	1600	2900	2500	3400	1500	1190	658
	4	5090	4100	4400	3100	3000	3900	3800	3800	1800	2500	1114
	5	5600	5010	5200	4200	4210	4200	5550	4500	3900	3900	859

Regrowth after cutting was recorded two weeks after each cut. *Vicia hyhrida, V. palaestina,* and *V. Villosa* ssp. *dasycarpa*, showed rapid regrowth after cutting up to cut 2. Most of the entries did not regrow after cut 3 at Tel Hadya, but *V. sativa 713, V. palaestina* and *V. hybrida* regrew poorly after cut 4 at Kfardan (Table 4.1.27). The regrown materials were left until maturity for seed yield estimation at both locations. After cut one, all entries produced seeds from the regrown materials which varied from 150 kg/ha for wooly-pod vetch to 610 kg/ha for bitter vetch at Tel Hadya and from 90 kg/ha for narbon vetch to 490 kg/ha for common vetch # 713 at Kfardan (Table 4.1.28). Wooly-pod vetch and narbon vetch have no ability to produce seed after cut two at both locations.

Results of Table 4.1.26 and 4.1.28 reveal that most of the entries regrew after early cutting (cut 1 and 2) providing more than one ton/ha dry herbage yield and enough seeds for resowing. If farmers desperately need a forage crop for early grazing (late February - begining of March), common vetch, bitter vetch, wooly-pod vetch and ochrus chickling are recommended. Grazing at this stage should be moderate to give ability to plants to regrow for seed production. Palaestine vetch and common vetch are suitable for late grazing.

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4.1.6.2. Studies on underground vetch (*V. sativa ssp. amphicarpa*)

4.1.6.2.1. The effect of plant density on the hardseededness of underground vetch.

In November 1993, underground vetch (selection 2647) was seeded at 10 rates (50, 120, 200, 250, 320, 400, 500, 800, 1260 & 2000 seeds per/m^2) in 0.5m x 6m plots at Tel Hadya. The experimental procedure and results are detailed in the 1994 annual report.

After maturity (July 1994) approximately 200 underground seeds from each plot were put in cloth bags and placed in the field on the soil surface.

Table 4.1.27. Regrowth after cutting, on 1-9 scale (where 1 is poor, 9 excellent) from cuts 1 to 5 for different *Vicia* and *Lathyrus* spp., grown at Tel Hadya and Kfardan.

Location/Cut No.		Vs713	Vs2560	Ve2517	Vp2525	Vh2548	Vd683	Vn2380	Lo542	Ls587	Lc501	LSD (P=0.05)
Tel Hadya	1	6.9	7.0	7.5	8.5	8.0	9.0	5.0	6.0	7.0	8.0	1.2
	2	6.0	6.9	6.5	8.5	9.0	8.0	4.0	4.0	6.0	6.0	1.6
	3	3.0	3.2	2.0	6.0	4.0	5.0	3.0	2.0	4.0	3.0	0.98
	4	-	-	-	-	-	-	-	-	-	-	-
	5	-	-	-	-	-	-	-	-	-	-	-
Kfardan	1	5.9	6.0	5.9	7.0	6.0	7.0	4.5	6.0	5.0	7.0	1.9
	2	7.0	7.0	4.6	7.0	5.0	6.0	3.0	4.3	3.0	5.9	1.7
	3	4.5	4.9	1.0	5.6	5.0	3.0	2.0	3.0	3.0	2.6	1.1
	4	2.1	-	-	4.3	2.0	-	-	-	-	-	1.2
	5	-	-	-	-	-	-	-	-	-	-	-

Table 4.1.28. Seed yield (kg/ha) from the regrowth after cuttings 1,2 and 3 at full maturity of different *Vicia* and *Lathyrus* spp. at Tel Hadya and Kfardan.

	Tel Hadya				Kfardan			
	Cut 1	Cut 2	Cut 3	Maturity	Cut 1	Cut 2	Cut 3	Maturity
Vicia sativa 713	510	413	200	1394	490	196	140	1977
V. sativa 2560	490	407	210	1828	416	146	137	2100
V. ervillia 2517	610	600	-	2186	462	346	-	2500
V. palaestina 2525	450	440	180	1653	246	183	146	1400
V. hybrida 2548	210	150	50	1519	241	173	30	1700
*V.v.spp.dasycarpa*683	150	50	-	1084	110	30	-	990
V. narbonensis 2380	180	70	-	2864	90	20	-	2700
Lathyrus ochrus 542	410	390	60	1881	320	120	-	1400
L. sativus 587	550	490	70	1413	410	320	50	1630
L.cicera 501	390	430	90	1298	299	310	40	1440
Mean	395	344	86	1712	308	184	54	1783
SEM±	51	40	14	126	49	21	7.5	189
LSD(P=0.05)	110	89	39	347	135	89	22	490

Hardseed determinations were made at regular intervals through to the beginning of 1995. For this period there was a rapid reduction in hardseed during the late autumn winter months until only 20-40% of the original seed remained hard. Following harvest the next season hardseed determinations were continued from July 1995 on a fortnightly basis until mid January 1996. At present (15/12/95), for all treatments, %90 of the remaining seed is still hard.

4.1.6.2.2. Barley-underground vetch rotation trial.

The background and treatments imposed in this trial are described in the 1993 Legume Program annual report. In the 1993/94 season all plots were sown to Barley (Mari/Aths*2//M-Att-73-337-1) and underground vetch was allowed to volunteer as a weed. Barley yields were measured and the size of the underground vetch seedbank was assessed at the end of the season. The results are detailed in the 1994 Germplasm Program annual report.

In the 1994/1995 season underground vetch was allowed to regenerate. The size of the seed bank was determined at the end of the season. Hardseed determinations are ongoing.

The seedbank status of the rotation trial from June 1990 to June 1995 is described in Table 4.1.29. Early grazing caused a reduction in seed yield by approximately 75%. During February underground vetch is still at an early seedling stage. Presumably, the damage caused by grazing was severe enough to retard seed production. Grazing in March and April had no effect on seed production. This would indicate that the root and underground stem system are well developed by March and that the majority of the seed bank is contributed by underground seed.

During the 1990/91 and 1993/94 seasons each plot was sown to barley. This caused, on average, 80% reduction in the seed bank size compared to the previous year. Between 80-90% of the underground vetch seedbank germinates during any given season, yielding plant densities of up to 2700 seedlings/m^2. This results in intense intra-specific competition made even more severe by the competition from a rapidly developing barley canopy. The seedlings which survive to maturity would be under low light intensities not conducive to seed production. The differences between grazing treatment plots in 1991 would be due to the seed bank sizes of the previous year.

Table 4.1.29. Seed bank of underground vetch (seeds/m2) at the end of each season for the duration of the barley-vetch rotation trial. The grazing treatments were imposed in the year of establishment (1989). Barley was planted during the 1990/91 and 1993/94 season.

Main plot grazing treatments (1989)	89/90	90/91 (Barley)	92/93	93/94 (Barley)	94/95
Grazed February	633	160	1461	409	620
Grazed March	2320	312	2628	462	696
Grazed April	2991	492	221	755	952
No Grazing	3009	662	2912	649	487
LSD (P=0.05)	1089	317	1494	487	525

LSD(P=0.05) = 799 for between year compariso

From 1993 onwards the seedbank sizes were the same within each year irrespective of the 1989 grazing treatments. During the 1992/1993 season underground vetch recovered from the barley phase to yield a seed bank which did not differ from the year of establishment. Of more significance perhaps, is the lack of recovery during the 1994/1995 season after the 1993/1994 barley phase. There was no difference between the seed yield under the barley in 1994 and that of the following ley season. This could be due to more intense competition from faster growing weed species which increased in number as the trial progressed. It is probable that the seed bank would be exhausted entirely if subjected to a barley rotation during the 1995/1996 season.

At flowering in 1993 half of each main plot was cut to the ground to asses the impact of defoliation. (Table 4.1.30) shows the effect of the cutting treatments. In general cutting during flowering made no difference to the size of the seed bank. The exception was the no grazing mainplot treatment where cutting did apparently cause a reduction in yield. It is not clear why this should be the case since there was no difference between the yield of the non-cut sub treatments indicating similar plant densities. The same can be said for the difference between the sub treatments within the grazed in February treatment for the 1994/1995 season.

Table 4.1.30. Comparison of underground vetch seed yield (seeds/m2) for noncut and cut at flowering sub treatments imposed in 1992/93.

Main plot Grazing treatments (1989)	92/93		93/94 (Barley)		94/95	
	noncut	cut	noncut	cut	noncut	cut
Grazed Feb.	1461	1398	409	651	620	114**
Grazed March	2628	2176	462	498	696	636
Grazed April	2218	2785	755	524	952	741
No Grazing	2912	1335**	649	540	487	531

(**) denote a significant difference between cutting treatmts (P=0.01).

Table 4.1.31 details barley grain yield for the 2 barley phases. Barley yielded more in rotation with underground vetch than after fallow. During the 1991/1992 season the barley-fallow treatment yielded about half a ton less than the barley-vetch rotation. Whilst in 1993/1994 it yielded almost a ton less. This indicates that underground vetch has value as a nitrogen providing ley species.

Table 4.1.31. Barley grain yield (t/ha) for main plot grazing treatments for both barley rotation years.

Main Plot Grazing Treatments (1989)	90/91	93/94
Grazed Feb.	2.09	1.87
Grazed March	2.11	1.68
Grazed April	2.05	1.92
No Grazing	2.01	1.87
Barley-fallow	1.54	0.74
LSDs (P=0.05)	0.25	0.65

LSD = (P=0.05) 0.66 between years comparisons

4.1.6.2.3. Comparison of underground vetch germplasm originating from different environments.

The aim of this trial was to assess the variation within the available underground vetch germplasm and attempt to correlate the variation to the environmental conditions from which the germplasm originated.

A collection of 72 accessions of underground vetch originating from Algeria, Cyprus, Jordan, Syria and Turkey was assembled. 15 seeds per accession were planted into jiffy pots which were kept in a greenhouse until the seedlings had reached a height of approximately 5 cm. When the worst of the winter frosts had abated they were hardened outside of the greenhouse and then planted in rows in the field at Tel Hadya.

In addition to the above germplasm, further material was collected from Jebal Abdul Aziz in the Hasake province located in north east Syria. The collection was done in a systematic way so as to assess the relative distribution of underground vetch in a grazing protected area and in an adjacent grazed area. 25 samples of 5 cores per sample were taken at 20 meter intervals along a transect. Four transects were taken per habitat. The elevation of each sample site was recorded. Vetch seeds were extracted from the samples, counted, weighed and then planted in the field as described above. The soil was retained for nutrient analysis.

52% of the grazing protected sites contained vetch seed. In total, 295 vetch seeds were collected of which approximately 80% were underground vetch. By contrast, only 10% of the grazed sites contained vetch seed (29 seeds) 98% of which were underground vetch.

The seed taken from the transects were planted in the field along with the other accessions. Nineteen morphological and agronomic characteristics were recorded.

To investigate whether the hardseed breakdown patterns differ between accessions, seed from each accession was collected at maturity (May 1995) and placed in cloth bags which were left on the soil surface in the field at Tel Hadya. Regular hardseed determinations were made to January 1996.

At this stage, only a preliminary analysis of the data has been undertaken, the summary of which is presented below.

Days to first flower were normally distributed about a mean of 120 days. The earliest accession flowered at 53 days while the latest flowered at day 140. Over 50% of accessions flowered between 117 and 122 days.

Days to pod maturity were narrowly spread around a mean of 152 days. The earliest accession finished at 148 days the latest at 172 days. Over 90% of accessions matured between days 155 and 160.

In general, for both flowering and podding the earliest accessions originated from Jordan and southern Syria. The exception were those accessions collected from Jebal Abdul Aziz which fell into the early pod maturity group.

Plot means for the yield parameters were ranked using 2 methods: (i) nearest neighbour analysis and (ii) means adjusted against check plots which were spaced at regular intervals throughout the trial. The two different methods yielded ranks which corresponded well.

Plot means for single plant dry weight were normally distributed about a mean of 24.2 g/plant. The minimum was 4.7 g/plant while the maximum was 126.5 g/plant.

Of the top 10 accessions, by far the highest yielding variety was V.*sativa* ssp.*nigra* which originated from Mersin, Turkey, three were Vicia *sativa* accessions (2 of which were *originally received as* V.*sativa* spp. *amphicarpa*), while four of the highest yielding underground vetch accessions were collected from Jebal Abdul Aziz (44 g/plant).

Plot means for the total number of seeds per plant were normally distributed about a mean of 284 seeds/plant. The maximum was 659 seeds/plant while the minimum was 38 seeds/plant. The top ten accessions were underground vetch. Three of them originated from the collection at Jebal Abdul Aziz (Hasake province north east Syria). The other accessions were Acc# 3175, 5155, 5162, 5150, 4241, 2614 and 3358.

It is interesting to note here that earliness of flowering and pod maturity did not necessarily correspond with higher seed yields.

The collection in Jebal Abdul Aziz demonstrates underground vetch's adaptation to both grazed and marginal environments. That is, the majority of legumes found were underground vetch. The ability to set seed underground and its adaptation to marginal environments and to grazing, suggests the use of underground vetch in a self regenerating pasture system and or in a cereal-legume rotation. The grazing treatments in the rotation trial show that it can still set a sizeable seed bank if it is not grazed too early.

However, a robust self-regenerating pasture plant needs to maintain a durable seedbank. This relies upon either production of large amounts of seed and or seed which will lose its hardseededness slowly over 4-5 years. The hardseed breakdown data presented demonstrates that 45%-90% of new seed becomes permeable within the first year. The optimal density for biomass production was shown to be at a seeding rate of 320 seeds/m^2. At this density it produced approximately 5000 seeds/m^2. If 70% of these seeds become permeable in the first year, around 3500 seeds/m^2 will germinate. This is roughly 10 fold more than the optimum density and would probably result in a high mortality rate and reduced yields in those that survived.

In other words, a lot of the seed that is produced would be wasted. This would explain why the seedbank in the rotation trial crashes during the cereal phase and is almost exhausted after the 5th season even though it has been grazed only once and been subjected to only 2 cereal phases.

However, there is some degree of variability for both seed yield and rate of hardseed breakdown, even in the limited amount of material worked with. For instance 1 out of 4 of the selections tested show a different pattern of hardseed breakdown (A larger range of accessions are being tested this year).

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4.1.7. Forage Legume Pathology

Pathology research on forage legume was confined to the screening for resistance to the three major diseases, ascochyta leaf spot and stem blight (*Ascochyta* spp.), botrytis blight (*Botrytis cinerea*) and downy mildew (*Perenospora* spp.). The screening was done at Tel Hadya on a total od 152 entries including *Vicia narbonensis* (36), *V. sativa* (25), *V. ervilia* (25), *Lathyrus sativus* (25), *L. cicera* (25) and *L. ochrus* (16). This material represents the advanced yield trials of the forage legume breeding program. Each of the six species was tested in 3 replications, in a randomized complete block design and in plots of 3 rows, each 2.5 x 0.3m.

Creation of artificial epiphytotics was done by using infected stubble from previous season (previous planting) and also by 3 times inoculation (after emergence and development) with a suspension containing the pathogen propagation units. In addition to disease data taken in the screening field, notes were taken on the same species and entries planted in the "Demonstration Field" at Tel Hadya under natural infection with downey mildew. Selection criteria were <2.8 score on a 1-5 scale.

Among the *Vicia* spp. for Ascochyta, the highest percentage of resistant lines (84%) was from the species *V. ervilia*. For Botrytis, *V. sativa* had the highest % resistant lines (56%); and for downy midlew *V. narbonensis (28%)* and *V. sativa* (16%) were resistant to the disease. One line (# 2498-94) of *V. sativa* was resistant to all 3 diseases, whereas 5 lines (# 2561-94, 2377-94, 2382-94, 2463-94, 2470-94) of *V. narbonensis* have resistance to the 3 diseases.

Among *Lathyrus* spp., all 16 entries of *L. ochrus* were resistant to all thre diseases. In *L. cicera* and *L. sativus* there were 92 and 52% resistant lines respectively for ascochyta respectively. For downy mildew, 60% of the lines in *L. cicera* and 24% in *L. sativus* were resistant to the disease and for botrytis, both species have had a good percentage of resistant lines, 64 and 40% respectively. One line (# 553-94) of *L. sativus* showed resistant to all three diseases and 10 (# 486-94, 487-94, 489-94, 490-94, 491-94, 492-94, 493-94, 496-94, 498-94, 500-94) of *L. cicera* were resistant to all diseases.

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4.1.8. Use of Forage Legumes by NARS

In Morocco, two varieties of narbon vetch, one variety of wooly-pod vetch and one variety of Common vetch were released. The Jordanian National Program in collaboration with the Mashreq project, released one variety of non-shattering Common vetch (Vicia sativa # 715) and recommended for release another three lines of Common vetch and one line of Common chickling (*Lathyrus sativus*).

The Mashreq/Maghreb project in response to farmer's demand and to enhance farmer adoption, expanded its activities in the demonstration of *Vicia sativa* (Common vetch) in the three Mashreq countries (Syria, Jordan and Iraq). The main purpos is to use it for direct grazing by sheep, and therefore, farmers who own sheep or goats were the target group for such activities. However, farmers were given the option to harvest part of their fields for seed production.

In Jordan, twenty demonstrations were conducted with a total area of 37.8 ha. A total of 2578 small rumimant which consisted of 20 flocks grazed Common vetch for a period ranged from 9 to 30 days depending on flock size. In Syria, 7 ha were planted in 9 locations, a total number of 801 head, consisted of 9 flocks of sheep and goats, grazed the area for between 6 and 60 days depending on flock size. In northern Iraq, 4 on-farm studies were conducted with an area of each location ranged from 1 to 1.5 ha, sheep grazed Common vetch at stocking rates of 10, 15 and 20 ewes per ha.

As a result Iraq and Jordan started a seed increase program for Bekia to make seed available for farmers.

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4.1.9. Annual Forage Legumes for Turkish Highlands

The Central Highlands of Turkey (CHT) are characterized by cold winters and dry summers. Under these extreme air temperatures, forage crops are still underexploited as a part of crop rotations. Although half of the total vetches in Turkey is grown in the CHT, farmers still use the local cultivars that are spring-planted and have low productivity. This generally results in shortage of good quality feed especially during the winter supplementary feeding time. Considering this, a collaborative Turkey/ICARDA project to identify annual forage legumes for both autumn and spring planting in the harsh environment of the CHT was initiated in the Center Institute of field crops (in Ankara) in the 1992/93 crop season. Three-year results of this project have shown that autumn-planted annual forage legumes performed significantly better than the spring-planted ones. Among vetches, hungarian vetch (*Vicia pannonica*) and wooly-pod vetch (*V. villosa* ssp. *dasycarpa*) were more promising than others for autumn planting and for grazing and/or hay. Narbon vetch (*V. narbonensis*) also performed better in autumn planting over the spring planting. Chicklings (*Lathyrus* spp.) also proved promising additional alternatives to spring-planted vetches. Of a number of accessions evaluated, acc. 751 of hungarian vetch and 694 of wooly-pod vetch for autumn planting, and acc. 793/A of narbon vetch and 794 of common chichling (*L. sativus*) performed better than others. On-farm evaluation of the promising species and accessions has been initiated in the CHT.

4.1.10. International Testing Program

The international testing program on lathyrus and vetch is a vehicle for the dissemination of genetic materials and improved production practices, in the form of international nurseries and trials, to the national programs. The genetic materials comprise elite lines with wide or specific adaptation, special morphological or quality traits, and resistance to common biotic and abiotic stresses. Nurseries are only sent on request and often include germplasm specifically developed for a particular region or a national program. A list of trials suppplied in the 1995/96 season is given in Table 4.1.32.

The testing program helps in identification of genotypes with specific and wide adaptation. The performance data permit assessment of genotype x environment interaction and help in targeting breeding efforts for specific agro-ecological conditions.

The salient features of the 1993/94 international nursery results, received from cooperators until 31 October 1995, are presented here. The stability analyses of some of the trials were done using Eberhart and Russell (1966) model.

Table 4.1.32. Distribution of Legume International Nurseries to cooperators for the 1995/96 season.

International Trial/Nursery	No.of sets
Lathyrus Adaptation Trial (ILAT)	
- *Lathyrus sativus* (ILAT-LS-96)	41
- *Lathyrus cicera* (ILAT-LC-96)	31
- Lathyrus ochrus (ILAT-LO-96)	29
Vetch Adaptation Trial (IVAT)	
- *Vicia sativa* (IVAT-VS-96)	51
- *Vicia narbonensis* (IVAT-VN-96)	42
- *Vicia ervilia* (IVAT-VE-96)	35
- *Vicia villosa* ssp *dasycarpa* (IVAT-VD-96)	36
Total	265

For the first time three separate International Lathyrus Adaptation Trials (ILAT) namely, *Lathyrus sativus* (ILAT-LS), *Lathyrus cicera* (ILAT-LC), and *Lathyrus ochrus* (ILAT-LO); and four separate International Vetch Adaptation Trials (IVAT) namely, *Vicia sativa* (IVAT-VS), and *Vicia narbonensis* (IVAT-VN), *Vicia ervilia* (IVAT-VE), and *Vicia villosa* ssp *dasycarpa* (IVAT-VD) were supplied to cooperators during 1993/94. In each of these trials there were 15 test entries and one local check. The results of these trials are discussed separately as below:

In ILAT-LS, the mean seed yield of selections across locations varied between 778 and 1255 kg/ha. The stability analysis indicated that the selections Sel -531, -533, -510, -522, -529, -504, -520, -528, and -508 with above average mean seed yield and non-significant deviations, were relatively stable and predictable across environments.

Reselts from ILAT-LC were returned from 6 locations. The mean seed yield of entries across locations ranged between 2113 and 2675 kg/ha. Eight selections (Sel -493, -491, -500, -486, -495, -492, -490 and -494) gave higher seed yield than the overall mean but only two of these selections, namely Sel -486 and -492 showed predictable behavior across environments.

The results for ILAT-LO were reported from 8 locations from 7 countries. The mean seed yield of selections across locations ranged between 1389 and 1730 kg/ha. The five heaviest yielding entries across locations included Sel -540, -538, -547, -550, and -545 with seed yield of 1730, 1688, 1655, 1568, and 1541 kg/ha respectively. The stability analysis of seed yield of the entries revealed that two selections namely, Sel -540 and -550 were adaptable across environments.

The results of IVAT-VS, IVAT-VN, IVAT-VE and IVAT-VD were reported from 10, 10, 9, and 11 locations, respectively. In IVAT-VS, the mean seed yield for entries across locations varied between 876 and 1500 kg/ha. Nine entries namely, Sel -2640, -2560, -2497, -2637, -2483, -2505, -2556, -2504, and -2642 exceeded the overall mean yield but only two selections -2497 and -2483 were stable and predictable.

In IVAT-VN, the mean seed yield of selections across environments varied between 1668 to 2004 kg/ha. Eight entries exceeded the overall mean. Among these Sel -2461, -2385, -2388, -2468, -2390, -2467, and -2464 were stable with predictable behavior.

In IVAT-VE, the mean seed yield of selections varied between 731 and 1028 kg/ha. Nine selections exceeded the overall mean seed yield and seven of these namely, Sel -2521, -2519, -2512, -2520, -2517, -2513, and -2515 were stable and predictable across environments.

In IVAT-VD, the mean seed yield of selections across locations varied from 1040 to 1326 kg/ha. Six selections exceeded the overall mean in seed yield and three of these namely, Sel -2455, -2431, and -2456 were stable and predictable across environments.

5. DRY PEA IMPROVEMENT

Although peas have been cultivated in the ICARDA region for millennia, yields are low because of lack of high yielding and stable genotypes, and poor crop management. To improve and stabilize yield of peas, an integrated approach to pea improvement was initiated at ICARDA in 1986/87 following the receipt of grant from the Ministry for Economic Cooperation, Germany (BMZ). The strategy is to capitalize on existing research done at a number of institutions in the developed and some developing countries, and restrict activities to the identification of dry pea varieties adapted to the farming systems of WANA. The work on pea improvement is therefore concentrated in the following area:

I. Assemble enhanced germplasm/cultivars from institutes working on dry pea in developed and developing countries and test them at ICARDA sites to identify superior lines for evaluation by the national programs in WANA.

II. Develop suitable production technology and transfer it to national programs.

5.1. Germplasm Collection and Evaluation

Fifty seven germplasm and improved cultivars were assembled during 1994/95. Out of these were from Australia 41 (Acc Nos.- 601 to - 641) and 16 from India (Acc Nos. - 644 to -659). These were evaluated for yield performance and cold tolerance under Tel Hadya conditions.

5.1.1. Pea Genetic Evaluation Trial

The new accessions obtained from various institutions were evaluated at Tel Hadya in Genetic Evaluation Trial with 64 entries. The data were recorded on various phenological and morphological characters. Days to flower ranged from 115 days (for Acc No. 603) to 145 days (for Acc No. 641); days to maturity ranged from 135 days (for Acc Nos. -615, -636, -642, -643, -646, and -224) to 154 days (for Acc No. 641); harvest index ranged from 9% (for Acc No. 641) to

45% (for Acc No. 603) and seed yield ranged from 252 (for Acc No. 641) to 2616 kg/ha (for Acc No. 622). None of the test entries exceeded the improved check cultivar, Acc No.225, by a significant margin. The five highest seed yielding entries included Acc Nos. -622, -639, -225, -617 and -603 with seed yields of 2616, 2569, 2459, 2329 and 2224 kg/ha, respectively (Table 5.1.1).

5.1.2. Evaluation for Cold Tolerance

In the cold tolerance nursery 208 dry pea accessions were evaluated during 1994/95 season. Visual cold tolerance ratings on a 1-9 scale (where 1 = free from damage, 9 = killed) were assigned after the susceptible check was killed. Fourteen accessions were tolerant to cold having a rating of 3 (Table 5.1.2).

5.2. Yield Trials

5.2.1. Preliminary Yield Trial (PYT)

Sixty four entries selected from the genetic evaluation and preliminary yield trials of the previous season (1993/94) were evaluated for yield performance in an 8 x 8 lattice design during 1994/95 at Tel Hadya and Terbol. Adjusted seed yield for the entries varied from 610 to 2452 kg/ha at Tel Hadya; and from 308 to 2521 kg/ha at Terbol. Location means at Tel Hadya and Terbol were 1379 and 1569 kg/ha, respectively. None of the test entries exceeded the improved check cultivar Acc No.225. Based on the mean yield over locations the five heaviest yielding lines included Acc Nos. -552, -501, -101, -557 and -553 with seed yields of 2487, 2145, 2109, 2087 and 2069 kg/ha respectively (Table 5.2.1).

Table 5.1.1. Adjusted seed yield (YD=kg/ha) and rank (R), biological yield (BALD=kg/ha), days to flower (DFLR), days to maturity (DMAT) and harvest index (HI) of some of the high yielding entries in Pea Genetic Evaluation Trial at Tel Hadya during 1994/95.

Acc. No.	Name	YD	R	BALD	R	DFLR	DMAT	HI
601	88P007-2-31	1721	31	4297	40	125	137	40
602	88P022-3-15	1924	19	4485	37	125	137	43
603	88P034-3-13	2224	5	4925	19	115	136	45
604	88P034-3-15	2067	8	4819	23	116	136	43
605	88P034-3-24	1882	21	4731	29	115	139	40
612	88P050-6-7	1929	16	5084	16	123	138	38
613	88P050-6-9	1766	24	4618	34	120	137	38
614	88P050-6-11	1749	26	4897	20	122	137	36
616	88P090-5-21	1926	18	4755	28	126	136	41
617	88P090-5-23	2329	4	6072	4	128	138	38
618	88P090-5-26	2014	10	5186	14	126	138	39
619	88P101-10-1	1927	17	4652	32	126	138	41
620	88P106-2-1	1728	30	5233	12	125	138	33
621	88P106-2-5	1850	23	5111	15	126	136	36
622	88P106-3-1	2616	1	6825	2	126	637	38
623	88PX00-11-3	1760	25	4562	35	121	137	39
624	88PX00-11-12	1635	35	4123	45	123	138	40
625	88P007-3-20	1736	29	4711	30	123	139	37
626	88P022-3-22	2183	6	4972	17	119	138	44
630	88P101-7-9	1963	15	5519	9	128	137	36
633	88P022-6-21	1967	12	5423	10	125	138	36
634	88P022-6-22	1966	13	5777	6	125	139	34
638	Spring pea 3	1916	20	4840	22	123	138	40
639	Spring pea 4	2569	2	6538	3	126	140	39
640	Spring pea 5	1697	32	5636	8	123	139	30
643	Pusa-10	1964	14	4967	18	112	135	40
647	DDR-11	1688	33	4389	38	125	136	38
650	DDR-14	1740	28	4003	48	123	136	43
651	DDR-15	1856	22	4785	26	122	137	39
653	DMR-7	2015	9	6005	5	126	138	34
654	DMR-11	1744	27	4779	27	123	137	36
655	DMR-20	1643	34	5233	13	125	139	31
656	DMR-24	1984	11	5705	7	125	138	35
658	DMR-26	2090	7	5296	11	120	137	39
659	DMR-27	1625	36	4250	43	128	136	38
225	Aleppo Local	2459	3	6959	1	128	138	35
Grand Mean		1616		4518				
S.E. Of Mean		313.5		698.4				
LSD at P=0.05		890.2		1984.1				
C.V.		27.4		21.9				

Table 5.1.2. Accessions with tolerance to cold.

Acc	Name	Origin	Acc	Name	Origin
12	JM2570	Afghanistan	197	D200-4-3	USA
17	MG100616	Greece	199	D166-1-15-1W	USA
77	K-129	Greece	205	D166-1-24-6W	USA
85	506-V2	Afghanistan	206	D166-1-4-2W	USA
86	643-V2	Afghanistan	346	PIMOS12069	USA
186	D166-3-1	USA	354	PIMOS12077	USA
190	D166-1-14-2	USA	470	WIR-1878	CIS

5.2.2. Pea International Adaptation Trial (PIAT)

Twenty three entries selected from PYT and PIAT conducted during the previous season and a local check were tested in PIAT at Tel Hadya and Terbol during 1994/95. Eight test entries at Terbol and 5 test entries at Tel Hadya yielded significantly better than the local check. The five highest yielding entries at Tel Hadya included Acc Nos. -372 (DMR-3), -448 (TARA), -526 (88P038-10), -504 (Local Ethiopian Cultivar) and -548 (WA932), and at Terbol included Acc Nos. 21 (Local Selection 1690), 447 (G22763-2C), 498 (P301), 546 (WA930), and 545 (88PX0034). On the basis of mean seed yield over locations, Acc No. 21 (Local Selection 1690) ranked first and was followed by Acc No. 372 (DMR-3), with seed yields of 2000, 1933, 1908, 1822 and 1813 kg/ha, respectively (Table 5.2.2). The Acc No. 284 (PS210158) was the earliest to flower and to mature.

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Table 5.2.1. Adjusted seed yield (YLD=kg/ha) and rank (R), days to flower (DFLR) and days to maturity (DMAT) of some of the entries in Preliminary Yield Trial at Tel Hadya, and Terbol during 1994/95.

Acc. No.	Entry name	Tel Hadya YLD	Tel Hadya R	Terbol YLD	Terbol R	Mean over locations YLD	Mean over locations R	DFLR	DMT
101	JI 231	2200	3	2018	8	2109	3	105	160
496	M 152-8	1844	8	1910	14	1877	11	114	161
497	P 94-1	1682	13	1858	17	1770	14	95	159
501	P 397-4	1869	5	2420	3	2145	2	100	159
520	88P 037-5	1860	7	1884	16	1872	12	106	161
549	WA933	1372	29	1997	9	1685	15	99	158
552	88P 109-11	2452	1	2521	1	2487	1	100	161
553	89P 109-12	2155	4	1982	11	2069	5	97	160
554	89P 109-13	1551	20	2489	2	2020	6	99	160
555	89P 110-1	1663	14	1942	12	1803	13	93	159
556	89P 110-6	1273	38	1809	23	1541	28	99	158
557	89P 111-1	2363	2	1811	22	2087	4	100	161
558	89P 123-3	1556	19	1520	36	1538	30	94	160
559	89P 124-4	1643	16	1669	32	1656	17	94	159
560	89P 127-2	1868	6	1996	10	1932	9	100	160
561	89P 129-4	1426	26	1768	26	1597	24	103	162
562	89P 134-2	1395	27	1515	38	1455	38	99	160
563	89P 142-6	1369	31	1683	30	1526	31	100	163
565	89P 150-18	1076	51	1911	13	1494	33	103	158
566	89P 150-20	1335	33	1905	15	1620	20	101	159
568	89P 156-5	1171	45	1681	31	1426	39	110	160
569	89P 166-12	1771	10	2221	4	1996	7	108	161
570	89P 170-7	1533	23	1760	27	1647	18	105	160
573	PS 010603	1325	34	1785	25	1555	26	101	159
574	PS 010735	1601	18	1367	42	1484	36	97	162
576	PS 010840	1480	25	1835	21	1658	16	105	160
577	PS 010843	1549	21	1063	57	1306	46	100	162
581	PS 110010	1394	28	1708	29	1551	27	101	160
584	PS 110038	1517	24	1733	28	1625	19	100	160
590	PS 110462	1243	40	1575	34	1409	40	92	161
591	PS 010497	1622	17	1595	33	1609	22	102	160
592	PS 110594	1369	30	1848	20	1609	22	98	160
593	PS 010598	1120	50	1854	18	1487	34	100	160
594	PS 110638	1812	9	1269	47	1541	29	97	161
403	ACC 1670	1541	22	1376	41	1459	37	102	152
448	TARA	1129	49	1573	35	1351	41	110	154
452	Praire N.11	1184	42	042	7	1613	21	111	155
498	P 301	1691	12	2073	6	1882	10	96	151
499	P 350-1	1269	39	1848	19	1559	25	103	152
504	Eth. Local	1176	44	1795	24	1486	35	110	152
517	88P 022-6	1644	15	1385	40	1515	32	101	150
225	Aleppo Loc	1769	11	2180	5	1975	8	113	163
Grand Mean		1379		1569					
S.E Of Mean		207.9		179.8					
LSD AT P=0.05		582.6		503.7					
C.V %		26.1		19.9					

Table 5.2.2. Mean seed yield (YLD=kg/ha) and rank (R), days to flower (DFLR), and days to maturity (DMAT), and plant height (PTHT) of entries at Tel Hadya and Terbol in PIAT-95.

Acc. No.	Entry Name	Terbol YLD	Terbol R	Tel Hadya YLD	Tel Hadya R	Mean over locations YLD	Mean over locations R	DFLR	DMAT
164	MG102521	1296	19	1903	10	1599	13	124	158
284	PS210158	1176	20	1958	7	1567	17	96	151
296	A149	1667	6	1519	21	1593	14	118	156
321	AMAC	963	24	1375	22	1169	22	107	151
372	DMR-3	1463	11	2403	1	1933	2	109	153
380	DMR-8	1083	21	1926	8	1505	19	106	152
403	ACC 1670	1611	8	1528	19	1570	16	109	153
445	305PS210572	1389	15	1560	18	1475	21	102	153
447	G22763-2C	2000	2	1644	14	1822	4	119	156
448	TARA	1352	18	2208	2	1780	7	119	157
452	Praire No. 11	1593	9	1588	16	1591	15	125	158
498	P301	1889	3	1926	9	1908	3	105	153
499	P350-1	1435	13	1579	17	1507	18	115	155
504	Ethiopian Loc	1361	17	2074	4	1718	9	121	158
507	SV88269	1028	23	1056	24	1042	24	130	159
517	88P022-6	1454	12	1523	20	1489	20	112	154
526	88P038-10	1481	10	2144	3	1813	5	110	154
545	88PX0034	1704	5	1870	11	1787	6	106	153
546	WA 930	1843	4	1630	15	1767	8	108	153
548	WA 932	1370	16	2051	5	1711	11	104	153
8	Aleppo Local-1	1398	14	2000	6	1699	12	117	156
21	SY Local 1690	2259	1	1741	13	2000	1	117	156
225	Aleppo Local	1611	7	1819	12	1715	10	117	156
Location Mean		1478		1758					
S.E. of Mean		195.8		289.8					
LSD at P=0.05		557.3		825.0					
C.V. %		23.0		28.6					

5.3. International Testing Program

The international testing program on dry pea is a vehicle for the dissemination of genetic materials and improved production practices, in the form of international nurseries and trials, to the national programs in and outside the WANA region. The genetic materials comprise elite lines with wide or specific adaptation, and special morphological or quality traits. Nurseries are only sent on request and the pea germplasm has been assembled from other centers and then tested for adaptation at ICARDA. A total of 64 sets of the Pea International Adaptation Trials (PIAT) were dispatched for the 1995/96 season.
The testing program helps in identification of genotypes with specific and wide adaptation. The performance data

permit assessment of genotype x environment interaction and help in targeting breeding efforts for specific agro-ecological conditions.

The salient features of the 1993/94 international nursery results, received from cooperators until 31 October 1995, are presented here. The stability analyses of some of the trials were done using Eberhart and Russell model.

The results of Pea International Adaptation Trial (PIAT) were reported from 19 locations. Some of the test entries exceeded the local check in seed yield by a significant ($P \leq 0.05$) margin. The mean seed yield across environments varied from 2176 to 3018 kg/ha. The stability analysis for seed yield revealed that 12 entries namely, Syrian Local Aleppo, G22763-2C, MG 102029, Local Sel. 1690, 305 PS 210572, MG 100446, MG 102256, A0149, MG 101831, Collegian, DMR-3, and Le 25 with above average yield and non-significant deviations from of environments.

6. COLLABORATIVE RESEARCH

6.1. West Asia Regional Program

6.1.1 Expansion in forage legumes plantation in the Mashreq region

In response to farmer's demand and to enhance farmer adoption, the Mashreq/Maghreb project, expanded its activities in the demonstration of "Bekia" *Vicia sativa* in the three Mashreq countries Syria, Jordan and Iraq. The main purpose is to use Bekia for direct grazing by sheep, and therefore, farmers who own sheep or goats were the target group for such activities. However, farmers were given the option to harvest part of their fields for seed production.

In Jordan, twenty demonstrations were conducted with a total area of 37.8 ha. A total of 2578 small ruminants which consisted of 20 flocks, grazed Bekia for a period ranging from 9 to 30 days depending on flock size. In Syria, 7 ha were planted in 9 locations, a total number of 801 head, consisting of 9 flocks of sheep and goats, grazed the area for a period ranging from 6 to 60 days depending on flock size. In northern Iraq, 4 on-farm studies were conducted on 1 to 1.5 ha in each location. Sheep grazed the Bekia at stocking rates of 10, 15 and 20 ewes per ha.

As a result Iraq and Jordan started a seed increase program for Bekia to make seed available to farmers.

6.2. North Africa Regional Program

6.2.1 Tunisia

6.2.1.1. Legume Disease

There was little disease pressure during this dry season, yet, viral diseases were severe. In particular the filiform virus disease was observed for the first time on chickpea; while on fababean, botrytis and mildew were widespread.

6.2.1.1.1. Ascochyta sp.

Good resistance to *A. fabae* found was confirmed in 29 H and few BPL's. More sources of tolerance to *A. Rabiei* were identified: 64% of tested varieties were rated 4.5-5.0 (1-9 scale).

6.2.1.1.2. Wilt

In the screening of the international ILC's collection, 2173 ILCs were tested and 13% were found resistant. Over the past few years, a total of 5300 lines were tested and about 10% were found resistant.

6.2.1.1.3. *Orobanche foetida*

Integrated control has been made available to farmers. Moreover, genetic tolerance was identified in the international collection of the BPLs:

- 16 BPLs have a good level of tolerance
- Spanish and Egyptian genetic material showed tolerance to *Orobanche foetida* and *Orobanche crenata* (8/9-128 and 402/29/84).

6.2.1.2. Chickpea Varietal Development

Progress has been made against the most frequent diseases, *Ascochyta* and Wilt. Three lines were particularly interesting for possessing acceptable levels of resistance and good seed size. Their performance is shown in Table 6.1.

Table 6.1. Reaction to Ascochyta and Wilt seed size and yield of three promising chickpea lines.

Chickpea	84-92C	T17W1	INRAT92
Ascochyta	4.6	6.5	4.5-5.0
Wilt	S	S	R
100 SW	33g	48 g	30-37 g
Yield	Good	Average	Fair

6.2.2 Algeria

6.2.2.1. Lentil and Chickpea Development

Research on food legumes, mainly on lentils and chickpea is very important at ITGC, Algeria. These crops are very much present in the diet and the dominant rotations throughout the country. A large number of good lines have been selected from nurseries distributed by ICARDA>

The most promising selections were:

Lentil

1.	36-14	FLIP 90-13L, 90-8L
2.	17-22	FLIP 88-6L, 90-41L, 87-48L
3.	7-9	FLIP 88-71L

Chickpea

1.	15-23	FLIP 90-105C, 90-58C, 86-50C
2.	6-13	FLIP 90-76C, 90-77C, 89-93C
3.	9-113	FLIP 87-96C, ILC6043, 89-62C, 90-96C

6.2.3. Libya

6.2.3.1. Food and Feed Legume Programme

Cooperation with ICARDA's food and feed legume projects in 94/95 season was limited to receiving regular international nurseries, as well as continuation of joint research work, planned previously with other national programmes within the region.

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7.1. Journal Articles

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8. STAFF LIST 1995 - Legumes

1. Dr Mohan C. Saxena	Leader Germplasm Program
2. Dr William Erskine	Lentil Breeder
3. Dr Khaled Makkouk	Virologist
4. Dr Michael Baum	Molecular Biologist
5. Dr Ali Abdel Moneim	Forage Legume Breeder
6. Dr Habib Ketata	Training Scientist
7. Dr K.B. Singh	Chickpea Breeder (ICRISAT)
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29. Mr Khaled El-Dibl	Senior Research Technician
30. Mr Omar Labban	Senior Research Technician
31. Mr Nidal Kadah	Senior Research Technician
32. Mr Ra'afat Azzo	Research Technician
33. Mr Mohamed K.Issa	Research Technician
34. Mr Bounian Abdel Karim	Research Technician

35. Mr Diab Ali Raya	Research Technician
36. Mr Mohamed El-Jasem	Research Technician
37. Ms Setta Ungi	Research Technician
38. Ms Nouran Attar	Research Technician
39. Mr Joseph Karaki	Assistant Research Technician (Terbol)
40. Mr Ghazi Khatib	Assistant Research Technician (Terbol)
41. Ms Aida Naimeh	Research Technician (Terbol)
42. Mr Noaman Ajanji	Driver/Store Keeper
43. Ms Mary Bogharian	Senior Secretary
44. Mrs Hasna Boustani	Senior Secretary

Consultant

45. Dr B. Bayaa	Pathologist