

**Program  
Abstracts  
List of participants**

**International Conference on  
Breeding for Stress Tolerance in  
Cool Season Food Legumes**

**Organized by**

**University of Naples  
Faculty of Agriculture  
Portici, Italy**

**International Center  
for Agricultural Research  
in the Dry Areas (ICARDA)  
Aleppo, Syria**

**10 - 12 September 1990  
"La Cappella", Villa Rufolo  
Ravello, Italy**

## OBJECTIVES OF THE CONFERENCE

The objectives of the conference are:

1. To review the past work on resistance to biotic stresses (diseases, insect-pests, and parasites) and abiotic stresses (temperature, moisture, and salt) in cool season food legumes (chickpea, faba bean, lentil, and pea).
2. To plan out the future strategy to solve problems related with resistance breeding to stabilize food legume production.
3. To publish the proceedings to serve as a reference book on resistance breeding.

## INTERNATIONAL ORGANIZING COMMITTEE

Dr. M.C. Saxena  
Prof. F. Saccardo  
Dr. K.B. Singh

## LOCAL ORGANIZING COMMITTEE

Prof. F. Saccardo  
Dr. A. Leone  
Dr. P. Crino  
Dr. A. Porta-Puglia  
Dr. F. Calcagno

## EDITORIAL COMMITTEE

Dr. K.B. Singh  
Dr. M.C. Saxena  
Prof. F. Saccardo  
Dr. A. Porta-Puglia

**PROGRAM  
ABSTRACTS  
LIST OF PARTICIPANTS**

**INTERNATIONAL CONFERENCE  
ON  
BREEDING FOR STRESS TOLERANCE  
IN COOL SEASON FOOD LEGUMES**

**ORGANIZED BY**

**University of Naples  
Faculty of Agriculture  
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**International Center  
For Agricultural Research  
in the Dry Areas (ICARDA)  
Aleppo, Syria**

**SEPTEMBER 10-12, 1990  
"LA CAPPELLA", VILLA RUFOLIO  
RAVELLO, ITALY**

**Program**

**Sunday, 9 September**

17:00 - 18:00 Registration at the Conference Hall

**Monday, 10 September**

**Session 1:      Opening session**  
**Chairman: Prof. E. Porceddu**

17:00 - 18:00 Registration at the Conference Hall

08:00 - 08:05 Welcome Prof. L. Postiglione

08:05 - 08:10 Conference objectives Dr. K.B. Singh

08:10 - 08:20 Inauguration

08:20 - 08:25 Chairman's remarks Prof. E. Porceddu

08:25 - 08:40 The challenge of biotic and  
 abiotic stress resistance in  
 cool season food legumes Dr. M.C. Saxena

08:40 - 08:45 Vote of thanks Prof. F. Saccardo

**Session 2:      Stresses resistance breeding**  
**Chairman: Prof. E. Porceddu**

08:45 - 09:30 Problems and prospects of stress  
 resistance breeding in chickpea K.B. Singh

09:30 - 10:15 Problems and prospects of stress  
 resistance breeding in faba bean L.D. Robertson

10:15 - 10:45 Coffee break

10:45 - 11:30 Problems and prospects of stress  
 resistance breeding in lentil W. Erskine,  
 M.C. Saxena

11:30 - 12:15 Problems and prospects of stress  
 resistance breeding in pea L. Monti,  
 L. Frusciante,  
 R. Romano

12:15 - 13:30 L U N C H

**Session 3:**      Disease resistance  
Chairman: Dr. R. Johnson

- |               |   |  |
|---------------|---|--|
| 13:30 - 14:15 | Screening for disease resistance in chickpea          | R.M. Jimenez-Diaz,<br>P. Crino, H. Halila,<br>C. Mosconi |
| 14:15 - 15:00 | Screening for disease resistance in faba bean         | S.B. Hanounik,<br>G.J. Jellis,<br>M.M. Hussein           |
| 15:00 - 15:30 | Coffee break  |  |
| 15:30 - 16:15 | Methods for selection of disease resistance in lentil | M.P. Khare, B. Bayya,<br>S.P.S. Beniwal                  |
| 16:15 - 17:00 | Screening peas for disease resistance                 | J.M. Kraft,<br>W.J. Kaiser                               |

Tuesday, 11 September

**Session 4:**      Insect, parasite, and virus resistance  
Chairman: Dr. A.v. Schoonhoven

- |               |  |   |
|---------------|--|---|
| 08:00 - 08:45 | Screening and selection criteria for insect resistance in cool season food legumes | S. Weigand,<br>M. Pimbert   |
| 08:45 - 09:30 | Selection for nematode resistance in cool season food legumes                      | N. Greco,<br>M. Di Vito   |
| 09:30 - 10:15 | Problems and prospects of selecting Orobanche resistance                           | J.I. Cubero,<br>S. Khalil   |
| 10:15 - 10:45 | Coffee break   |   |
| 10:45 - 11:30 | Screening for virus resistance in cool season food legumes                         | K. Makkouk,<br>L. Bos, N. Horn,<br>B. Sreenivasa Rao                |
| 11:30 - 12:15 | Mechanisms of resistance to fungal pathogens in cool season food legume            | W. Barz <u>et al.</u><br><br>D. Meier, R. Tenhaken,<br>R. Vogelsang |

**Session 5: Abiotic stress resistance**  
**Chairman: Prof. L. Monti**

- |               |  |  |
|---------------|--|--|
| 13:30 - 14:15 | Screening for cold and heat tolerance in cool-season food legumes  | R.S. Malhotra,<br>M.C. Saxena                              |
| 14:15 - 15:00 | Selection for drought and salinity tolerance in cool season food legumes   | N.P. Saxena,<br>C. Johansen,<br>M.C. Saxena,<br>S.N. Silim |
| 15:00 - 15:30 | Coffee break   |  |
| 15:30 - 16:15 | Mechanisms of resistance to drought, cold and heat in cool season food legumes with emphasis on pea and chickpea | J. Wery, O. Turc,<br>J. Lecoeur                            |
| 16:15 - 17:00 | Physiological and morphological basis for stress-resistance in pulse crops with special emphasis on chickpea     | F. Calcagno,<br>G. Gallo                                   |

Wednesday, 12 September

**Session 6: New strategies for stress resistance**  
**Chairman: Dr. M.C. Saxena**

- |               |  |   |
|---------------|--|---|
| 08:00 - 08:45 | Use of molecular genetics for stress breeding in cool-season food legumes            | J.K. Key, A. Leone  |
| 08:45 - 09:30 | Mutagenesis and chromosome manipulation for stress resistance in cool season legumes | F. Saccardo,<br>A. Enrico, P. Crino<br>B. Ocampo, G. Venora |
| 09:30 - 10:15 | Use of wild species as a source of resistance in pulse crops                         | F.J. Muehlbauer   |
| 10:15 - 10:45 | Coffee break   |   |
| 10:45 - 11:30 | Durable resistance to pathogens and how to breed for it                              | J.E. Parlevliet   |
| 11:30 - 12:15 | Strategies for multiple stress resistance breeding in cool season food legumes       | A. Porta-Puglia,<br>K.B. Singh,<br>A. Infantino             |
| 12:15 - 13:30 | L U N C H  |   |

**Session 7:**      **Breeding methods for stress resistance**  
**Chairman: Dr. K.J. Frey**

- |               |   |               |
|---------------|---|---------------|
| 13:30 - 14:15 | Utilization of germplasm resources of cool season food legumes in breeding for stress tolerance | S. Jana       |
| 14:15 - 15:00 | Breeding methods for stress resistance in self-pollinated crops                                 | A.E. Slinkard |
| 15:00 - 15:30 | Coffee break  |               |
| 15:30 - 16:15 | Population improvement for stress resistance in cross-pollinated crops                          | J.I. Cubero   |

**Session 8:**      **Closing session**  
**Chairman: Prof. Scarascia Mugnozza**

- |               |  |                            |
|---------------|--|----------------------------|
| 16:15 - 16:30 | Strategies for breeding disease resistant crop varieties | K.J. Frey                  |
| 16:30 - 16:50 | Summation of some of the issues raised in the workshop   | R. Johnson                 |
| 16:50 - 17:00 | Vote of thanks   | F. Saccardo<br>M.C. Saxena |

THE CHALLENGE OF DEVELOPING ABIOTIC AND BIOTIC STRESS RESISTANCE  
IN COOL-SEASON FOOD LEGUMES

M.C. Saxena

International Center for Agricultural Research in the Dry Areas  
ICARDA, P.O. Box 5466, Aleppo, Syria

Cool-season food legumes - chickpea, faba bean, lentil, and peas - account for nearly 35% of the area and 55% of pulse production in the world. In the Mediterranean rainfed farming systems, they are the most dominant pulse crops. Their productivity, however, is low because of a wide range of abiotic and biotic stresses. The major abiotic stress factors are cold, heat, drought, nutrient deficiency, nutrient toxicity, and lodging. Water-logging can also be a constraint in some areas. The major biotic stresses are those caused by fungal, bacterial and viral diseases, insect pests, nematodes, and parasitic broomrape (*Orobanche crenata*). The relative importance of different stress factors differs not only from crop to crop but also for the same crop in different regions.

Host-plant resistance, either alone or as a component of an integrated control strategy, appears to be the most practical and economic approach for reducing the effects of abiotic and biotic stresses. Research on host-plant resistance/tolerance has been undertaken by several national and international programs in the major production areas and notable progress made. However, the stresses often occur together in an interacting fashion, necessitating the incorporation of multiple stress resistance in suitable agronomic backgrounds. Also, the variations in pathotypes and the existence of biotypes in insect pests, make durable resistance breeding a difficult task. Conventional breeding strategies have shown good success but their effectiveness can be considerably increased with the application of suitable biotechnologies. Improvement in screening techniques, use of molecular markers for identification and utilization of economically important genes, use of *in vitro* culture for transferring desirable genes from wild to cultivated species, and genetic engineering using alien genes all offer opportunities for improving the resistance of future cultivars of cool-season food legumes to different stresses.



PROBLEMS AND PROSPECTS OF STRESS RESISTANCE BREEDING IN CHICKPEA<sup>1</sup>

K.B. Singh

International Center for Agricultural Research in the Dry Areas  
(ICARDA), P.O. Box 5466, Aleppo, Syria

Chickpea suffers from a number of biotic and abiotic stresses which reduce the yield and make production unstable. Sources of resistance have been identified for several important biotic stresses such as diseases (Ascochyta blight, Fusarium wilt, dry root rot, and Botrytis gray mold), pea leaf roll virus, cyst nematode, insect pests (pod borer and leaf miner), and Orobanche spp. Also, sources of resistance have been identified for a few abiotic stresses, such as cold, drought, iron deficiency chlorosis and lodging. But effort to identify sources of resistance to heat, salinity and water logging have not met with success. Utilizing these sources of resistance, cultivars with resistance to Ascochyta blight, Fusarium wilt and cold have been released in several countries. However, success in insect resistance breeding has been limited. Breeders have developed lodging resistant lines which may soon be released. For other stresses, where sources of resistance have been found, efforts are underway to breed resistant cultivars.

These stress resistant cultivars have, however, never been popular with growers with possibly two exceptions. These were Fusarium wilt-resistant cultivars in Mexico and Ascochyta blight and cold resistant cultivars in the Mediterranean region. A major reason for low popularity of resistant cultivars has been the lack of multiple stress resistance. In most areas more than one stress is important, hence the need for multiple stress resistant cultivars arises.

Cultivars with multiple stress resistance are of paramount importance if chickpea is to be introduced in new areas or conditions. Ascochyta blight and cold resistant cultivars for winter sowing in the Mediterranean region; Phytophthora root rot and root lesion nematode resistant cultivars for Australia; heat, collar rot and Colletotrichum blight resistant cultivars for early sowing in Peninsular India; and terminal heat and drought tolerant chickpeas for sowing as a catch crop in rice-based farming systems of south Asia are a few well demonstrated examples. Chickpea production has remained static for the last three decades, but it can be increased substantially if appropriate resistant cultivars are available for introduction in new areas and/or conditions.

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<sup>1</sup> Joint contribution from ICARDA, P.O. Box 5466, Aleppo, Syria and ICARISAT (International Crops Research Institute for the Semi-Arid Tropics), Patancheru P.O., A.P. 502 324, India.

**PROBLEMS AND PROSPECTS OF STRESS RESISTANCE BREEDING IN LENTIL****W. Erskine and M.C. Saxena****International Center for Agricultural Research in the Dry Areas  
(ICARDA), Aleppo, Syria**

Lentil production is limited by lack of moisture and unfavourable temperatures throughout its distribution. Salinity and waterlogging are of local importance only. Progress has been made in breeding for tolerance to drought through selection for an appropriate phenology and increased water use efficiency and in breeding for winter hardiness through selection for cold tolerance.

The diseases rust, vascular wilt and Ascochyta blight, caused by Uromyces viciae fabae, Fusarium oxysporum f.sp. lentis and Ascochyta lentis, respectively, are the major fungal pathogens of lentil. Cultivars with resistance to rust and Ascochyta blight has been released to growers and resistant sources to vascular wilt are being exploited. Sources of resistance to several other fungal and viral diseases of regional importance are known. In contrast, however, the pea leaf weevil (Sitona spp.) and the parasitic weed broomrape (Orobanche spp.) are major yield reducers of lentil, and to a lesser extent the cyst nematode (Heterodera ciceri), but efforts to find sources of resistance to these biotic stresses have proved fruitless.

Avenues for possible future research in lentil on both biotic and abiotic stresses are discussed.

## SCREENING FOR DISEASE RESISTANCE IN CHICKPEA

R.M. Jimenez-Diaz<sup>1,2</sup>, P. Crino<sup>3</sup>, M.H. Halila<sup>4</sup>,  
C. Mosconi<sup>3</sup> and A. Trapero-Casas<sup>1</sup>

- 1,2. Catedra de Patologia Vegetal, Depto. de Agronomia, Universidad de Cordoba, Apdo. 3048, 14080 Cordoba, Spain. Instituto de Agronomia y Proteccion Vegetal, CSIC, Apdo. 3048, 14080 Cordoba, Spain.
3. ENEA C.R.E. Casaccia, Dept. of Agroindustrial Research and Development, S.P. Anguillarese 301, 00060 Roma, Italy.
4. Food Legume, INRAT, 2080 Ariana, Tunisia.

Ascochyta blight (AB, Ascochyta rabiei) and Fusarium wilt (FW, Fusarium oxysporum f.sp. ciceri) are the most important chickpea diseases throughout the world. The use of resistant cultivars is the most practical and cost-efficient method for control of both diseases. Methods to identify resistance to AB and FW in chickpea germplasm lines and segregating populations are discussed. The influence of the epidemiology of diseases, pathogenic variability in the pathogens and inheritance of resistance in the host is considered. Better understanding of the epidemiology of AB and FW is needed. The effects of interaction among environmental factors on epidemics of AB and FW, and the role of the teleomorph of A. rabiei in the pathogen variability and dispersal should be further investigated. High pathogenic variability exists in both pathogens. The identification of pathotypes of A. rabiei and F. oxysporum f.sp. ciceri should be carried out with common standardized methods and differentials. More information on occurrence and distribution of pathotypes is needed.

**SCREENING FOR DISEASE RESISTANCE IN FABA BEAN****S.B. Hanounik<sup>1</sup>, G.J. Jellis<sup>2</sup> and M.M. Hussein<sup>3</sup>**

1. ICARDA, Douyet Research Station, BP 2335, Fes, Morocco.
2. P.B.I. Trumpington, Cambridge, CB22 1Q, UK.
3. P.O. Box 30, Khartoum, North, Sudan.

Faba bean is attacked by a wide range of diseases. Resistance to major diseases has been identified and confirmed through ICARDA's international disease screening nurseries. A few sources of resistance have been found stable over the past decade at different locations, while others showed variable reactions. These advances stimulated much of the recent breeding activities, and cultivars with resistance to major faba bean diseases are being released for the first time. The evidence of physiological variation in certain pathogens warrants continued search for new sources of resistance. Approaches to improve present methods of screening for resistance are discussed.

## SELECTION OF METHODS OF DISEASE RESISTANCE IN LENTIL

M.N. Khare<sup>1</sup>, B. Bayaa<sup>2</sup> AND S.P.S. Beniwal<sup>3</sup>

1. Jawaharlal Nehru Agricultural University, College of Agriculture, Rewa, M.P. 486001, India.
2. University of Aleppo, Faculty of Agriculture, Aleppo, Syria.
3. International Center for Agricultural Research in the Dry Areas, P.O. Box 5466, Aleppo, Syria.

Diseases are among the major stresses affecting growth and productivity of lentil. Among of the diseases affecting lentil, vascular wilt (Fusarium oxysprum f.sp. lentis), root rots (F. roseum, F. solani, Rhizoctonia bataticola, R. solani, Pythium spp.) collar rot (Sclerotium rolfsii), sclerotinia rot (Sclerotinia sclerotiorum), stem and pod rot (Botrytis cinerea), Ascochyta blight (Ascochyta lentis), rust (Uromyces fabae), downy mildew (Peronospora lentis), powdery mildew (Erysiphe poligoni), root-knot (Meloidogyne sp.), and viruses (pea seed-borne mosaic virus, bean yellow mosaic virus, and pea enation mosaic virus) diseases are important. The selection methods for identifying resistant genotypes depend upon the nature of the pathogen involved and its mode of transmission. Specific methods have been developed to screen for different host-pathogen interactions either under natural field conditions or artificially created epidemics. Soil-borne pathogens are grown in an appropriate medium, then mixed with the soil so as to produce an evenly distributed inoculum. In the case of air-borne pathogens, such as A. lentis, plants are inoculated by spraying a standardized spore suspension. The obligate parasites causing rust, downy and powdery mildews are maintained on the host plant. The screening is usually done in hot spots. Screening against seed-borne pathogens is carried out in laboratory or field as per need. Specific methods are used for viruses and nematodes. Sources of resistance have been identified among cultivated species as well as in wild Lens species. However, the genetics of resistance for fungal diseases have not been fully investigated.

## SCREENING PEAS FOR DISEASE RESISTANCE

J.M. Kraft<sup>1</sup> and W.J. Kaiser<sup>2</sup>

1. USDA-Agricultural Research Service IAREC, RR 2 Box 2953A, Prosser, WA 99350-9687.
2. USDA-Agricultural Research Service, Washington State University, Pullman, WA 99164.

Peas for both seed and green harvest are susceptible to a number of serious fungal, bacterial, and viral diseases that affect seedlings, roots, and above ground portions of the crop. Seedling fungal diseases are caused primarily by Rhizoctonia solani and Pythium spp. The most important root diseases caused by fungi include Fusarium solani f.sp. pisi and Aphanomyces euteiches. Above ground fungal diseases discussed in this paper include Ascochyta spp., downy and powdery mildew, caused by Peronospora pisi and Erysiphe pisi, respectively. Over 20 viruses are reported to naturally infect peas. This paper will focus on four of the most important viruses affecting this crop worldwide; namely bean (pea) leafroll virus (BLRV), pea enation mosaic virus (PEMV), pea seedborne mosaic virus (PSbMV), and pea streak virus (PSV). Resistance to the fungal and viral diseases discussed in this paper, along with screening procedures, have been identified and are discussed. Breeding for virus resistance in pea is an area where molecular genetic techniques and isozyme analysis can play an important role in the near future.

SCREENING AND SELECTION CRITERIA FOR INSECT RESISTANCE  
IN COOL SEASON FOOD LEGUMES

S. Weigand<sup>1</sup> and M. Pimbert<sup>2</sup>

1. ICARDA, P.O. Box 5466, Aleppo, Syria
2. ICRISAT, Patancheru P.O., Andhra Pradesh 502 324, India

The cool season food legumes suffer yield losses from several insect pests in field and store. In chickpea, leaf miner (Liriomyza cicerina), pod borers (Helicoverpa armigera and Heliothis virescens), and seed beetles (Callosobruchus chinensis and C. maculatus) are the main pests. In lentil, Sitona crinitus is most important but aphids (Aphis craccivora and Acyrtosiphon pisum) also cause yield losses. The main insect pests of faba bean are aphids (Aphis fabae and Aphis craccivora) and leaf weevil (Sitona lineatus) in the field, and different species of Bruchus and Callosobruchus in storage. Aphids (Acyrtosiphon pisum), leaf weevil (S. lineatus), and pod borer (Laspeyresia nigricana) are the most damaging insect pests in pea.

The identification of host-plant resistance requires practical and reliable methods to measure resistance. Depending on the insect pest and the crop, different field and laboratory screening techniques have been developed.

Screening for chickpea leaf miner is done under natural infestation using a visual damage score based on per cent leaf mining and plant defoliation. Screening for pod borers is also done in field using natural populations which are occasionally supplemented by laboratory reared insects. Chickpea lines resistant to these pests have been identified and studies on the mechanism of their resistance initiated. The latter will help in the development of more efficient selection criteria and screening techniques for use in breeding programs. As field screening of lentil for resistance to Sitona did not give conclusive results, a laboratory technique needs to be developed. Screening for aphids resistance in faba bean and lentil is done in the laboratory using standardized artificial infestation and promising lines are further tested in the field under natural infestation. Screening for multivoltine species of storage insect pests is conducted in the laboratory but the technique has been extensively used only in chickpea. Faba bean lines have been screened under natural infestation in the field as well as under artificial infestation in the laboratory for resistance to univoltine species of storage insect pests. The selection criteria used in the screening are discussed.

## SELECTION FOR NEMATODE RESISTANCE IN COOL SEASON FOOD LEGUMES

N. Greco and M. Di Vito

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Investigations on selection of cool season food legumes for resistance to nematodes are limited. No commercial cultivar with resistance to nematodes is so far available. Moderate resistance to Heterodera goettingiana was identified in accessions of Pisum abyssinicum governed by a single recessive gene. Extensive screening has been made of Cicer spp. and resistance to H. ciceri has been found in C. bijugum, to Meloidogyne artiellia in several species of Cicer, and to M. incognita and Rotylenchulus reniformis in C. arietinum. Resistance to Ditylenchus dipsaci occur in Vicia faba. No resistance has been found in lentil. Information on inheritance of resistance is unknown. Screening undertaken by independent investigators requires the use of standard methods. Preliminary screening is usually based on visual observation of the nematode infestation or of its symptom. Assessment of the nematode reproduction and studies on histology and metabolic changes of the infested plants could provide further confirmation of the resistance. Identifying resistant sources, understanding of resistance and nematode pathotypes are necessary to undertake a breeding program. Biotechnology and other new methods could profitably be employed to study nematode resistance.



## SCREENING FOR VIRUS RESISTANCE IN COOL SEASON LEGUMES

K.M. Makkouk<sup>1</sup>, L. Bos<sup>2</sup>, N. Horn<sup>3</sup>, and B. Srinivasa Rao<sup>3</sup>

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Chickpea, faba bean, lentil and pea are reported to be naturally attacked by 9, 32, 9, and 28 viruses, respectively. Only few are of economic importance at any one location. Host-plant resistance is considered to be the method of choice for their control. Therefore, screening for virus resistance by evaluating cultivars or breeding populations is the first step towards progress. This is best accomplished by using well defined virus inoculum and artificial inoculation methodology to achieve 100% infection of susceptible germplasm. Visual observation of inoculated plants, and measurements of virus multiplication levels of ELISA are used to assess the host reaction to the virus. In chickpea, accessions exhibiting low incidence of chickpea stunt virus have been identified. Chickpea genotypes with resistance to cucumber mosaic (CMV) and bean yellow mosaic (BYMV) viruses were reported. In faba bean, genotypes with resistance to BYMV were identified, and two recessive genes were found to confer this resistance. In lentil, accessions with resistance to pea seed-borne mosaic virus (PSbMV) were identified and this resistance was found to be conditioned by a single recessive gene. In pea, breeding lines with resistance to PSbMV, pea mosaic virus, pea top yellow virus (= BLRV), clover yellow vein virus, pea enation mosaic virus, plantago mottle virus and watermelon mosaic virus were identified. Success has been reported for screening for pea enation mosaic virus and bean yellow mosaic virus resistance in peas, indirectly by screening for the isozymes alcohol dehydrogenase and phosphoglucosmutase, respectively.

**MECHANISMS OF RESISTANCE TO FUNGAL PATHOGENS IN COOL SEASON FOOD LEGUMES**

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Plants including legumes respond to fungal infection with the expression of inducibly formed defence reactions. These may comprise: (1) cell wall reinforcement by deposition of callose, lignin, ester-bound cinnamic acids/polyphenols, and hydroxyproline - rich glycoproteins; (2) accumulation of acidic lytic enzymes (chitinases, glucanases), production of polygalacturonase inhibitors and various pathogenesis - related proteins of still unknown functions, and (3) formation of phytoalexins. In resistant genotypes, the induction of defence reactions is associated with the hypersensitive response.

The legumes will be reviewed with regard to the expression of the alternative pathways and to the mechanisms pathogenic fungi use to penetrate the plant tissues with the aid of cutinase, pectinase, polygalacturonase, and other hydrolases.

The fungus Ascochyta rabiei induces in resistant genotypes of chickpea (Cicer arietinum L.) high levels of pterocarpan phytoalexins medicarpin and maackiain together with their biosynthetic enzymes and lytic enzymes in form of various isoforms together with other PR-proteins.

In susceptible genotypes of chickpea a fungal glycoprotein suppressor strongly inhibits phytoalexin and isoflavone formation. The effect of the solanapyrone phytotoxins recently isolated from spore-forming A. rabiei isolates on chickpea leaf tissue will be described.

Financial support: Deutsche Forschungsgemeinschaft and Fonds der Chemischen Industrie.

**SCREENING FOR COLD AND HEAT TOLERANCE IN COOL SEASON FOOD LEGUMES****R.S. Malhotra and M.C. Saxena****International Center for Agricultural Research in the Dry Areas  
(ICARDA), P.O. Box 5466, Aleppo, Syria**

Cold and heat stresses are important among the abiotic stresses to which the cool season food legumes are subjected. Both these stresses can occur any time from emergence to the reproductive phase of the crop development and can cause heavy yield losses to all these crops. For any effective breeding program to improve upon the tolerance to these stresses the reliable screening techniques are the pre-requisites. Percent plant survival has been used as a parameter for cold tolerance in faba bean, pea and lentil. But in chickpea, in addition to percent plant survival, visual scores for individual plant or line have been developed. Although some laboratory screening techniques are available for cold tolerance in some of these crops, there is a need to refine and standardize them for all food legumes. With respect to tolerance to heat, little work has been done in cool season food legumes. It is suggested that screening methods used in other crops can be applied with little modifications.

**SELECTION FOR DROUGHT AND SALINITY TOLERANCE  
IN COOL SEASON FOOD LEGUMES**

N.P. Saxena<sup>1</sup>, C. Johansen<sup>1</sup>, M.C. Saxena<sup>2</sup>, and S.N. Silim<sup>1</sup>

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2. ICARDA, P.O. Box 5466, Aleppo, Syria.

Drought and salinity are two major stresses to food legume crops, including chickpea, lentil, faba bean, and pea. Genetic improvement for tolerance to these stresses, although a cheaper option compared to alleviating these stresses through management, has its own limitations. Effective field and laboratory methods of screening germplasm for drought and salinity tolerance have been described. Useful variability and traits associated with terminal drought tolerance have been identified in chickpea and lentil. Methodology of screening progenies for drought tolerance is described. An example in chickpea is the selection for increased rooting capacity in early generations.

The low levels of salinity tolerance so far found in these legumes do not justify initiating a genetic enhancement program on salinity tolerance at this stage. However, possible screening methods and breeding procedures are presented for future use.

It is proposed that a greater understanding of the physiological mechanisms conferring tolerance to defined types of drought and salinity stress would enhance the germplasm screening process, and the ability to select progeny in segregating populations. It is concluded that a greater emphasis is required on breeding for drought tolerance in these legumes. Identifying new sources of tolerance, by systematic screening of germplasm collected from stress prone habitats, should receive a high priority. If genetic improvement of salinity tolerance in these legumes is contemplated, as a supplement to management alternative, then all effort should be devoted to the germplasm screening phase.

**MECHANISMS OF RESISTANCE TO DROUGHT, COLD AND HEAT IN COOL  
SEASON FOOD LEGUMES WITH EMPHASIS ON PEA AND CHICKPEA**

**J.Wery, O. Turc and J. Lecoœur**

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Cool season food legumes experience abiotic stresses linked with temperature (cold or heat) or water (drought), which are frequently among the most limiting factors for yield and biological nitrogen fixation, particularly in the Mediterranean region. Nevertheless, progress in breeding for resistance to these stresses has been very weak compared to biotic stresses. On the basis of the literature and our own experiments we will present the mechanisms involved in resistance to these abiotic stresses and analyse the opportunity to use them in a breeding program.

With regard to the lack of work on food legumes, we will use knowledge primarily obtained from other crops, in particular cereals, and confront it with scattered observations made on cool season food legumes, mainly pea and chickpea. The comparison between these two species in a Mediterranean environment will be used to identify the mechanisms of resistance when knowledge on intraspecific variability is unavailable.

For each type of stress (chilling, freezing, heat or drought) we will review: (1) the characterization of the stress (intensity and position in the plant cycle) most frequently experienced by cool season food legumes; (2) its effects on processes involved in plant production (at cell, organ or plant level), and the evidence of genotypic variability for these effects; and (3) the mechanisms of escape, avoidance or tolerance sufficiently established and easy to measure to be used in a breeding program.

Mechanisms such as escape by accelerated phenology osmotic adjustment or membrane stability deserve particular attention. They could be involved both in resistance to water and temperature stress.

PHYSIOLOGICAL AND MORPHOLOGICAL BASIS FOR STRESS RESISTANCE  
WITH SPECIAL EMPHASIS ON CHICKPEA

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Studies were conducted to assess the effect of moisture, temperature and photoperiod on three phenological phases (germination, growth and reproduction) of chickpea by staggering of sowing date from autumn to early spring in a Mediterranean environment of Sicily in Italy. On the basis of physiological knowledge of stress tolerance in higher plants, the traits have been determined which allow, directly or indirectly, chickpea plant to overcome the detrimental effects caused by water and temperature variations. Furthermore, associations between morphological and structural traits and physiological reactions of chickpea plants have been examined. It is believed that the characters identified through these studies might help breeders to select genotypes with improved tolerance to abiotic stresses such as moisture and temperature.

## USE OF WILD SPECIES AS A SOURCE OF RESISTANCE IN PULSE CROPS

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Stresses from heat, drought, poor soil and pests are becoming increasingly important as production areas for cool season food legumes, such as lentil, chickpea, pea and faba bean, are shifted to marginal areas. Resistance to those stresses may be found in the wild relatives of these crops, but evaluations of wild accessions are needed to identify useful genes. The wild species of Lens, Cicer and Pisum are often found in the Middle East in dry stony habitats where heat and drought stresses prevail and where stress resistance genes are needed for survival. A sizeable collection of wild species of these crop plants is available for breeding purposes. The wild species have been placed in either primary, secondary or tertiary gene pools. Wild species in the primary gene pool can be hybridized with the cultivated species but some partial sterility may limit genetic recombinations. Introgression of genes from Lens orientalis to L. culinaris and from Cicer reticulatum to C. arietinum is currently underway. Hybridizations with species of the secondary gene pool encounter greater sterility as a result of chromosomal structural rearrangements. Hybridization with species of the tertiary gene pool are generally not successful, but techniques such as embryo rescue can be used to obtain hybrids. Molecular marker assisted introgression based on close genetic linkages can be used to transfer genes from a wild source to a cultivated background.

**DURABLE RESISTANCE TO PATHOGENS AND HOW TO BREED FOR IT****J.E. Parlevliet****Plant Breeding Department, Agricultural University,  
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Resistance to pathogens may be durable for several reasons. 1. The farming system prevents the build up of inoculum reducing the changes of adaptation in the pathogen population. 2. Pathogens differ in their versatility; especially biotrophic and hemibiotrophic airborne fungi adapt easily to introduced resistance. 3. Non-durable, race-specific genes may last longer if certain strategies were applied, such as multilines or cultivar mixtures, gene deployment and multiple gene barriers. Neither of these possibilities are considered to be of practical value. 4. Resistance genes themselves are durable; the pathogen can not adapt to these genes at least not within short time span.

The genetically durable resistance is often of a quantitative nature. To discern small differences in resistance good screening and assessment methods are necessary. Provided relatively small differences in resistance can be measured free from confounding other effects (interplot, interference, earliness, tallness, irregular distribution of inoculum) one should select against susceptibility and against complete resistance, i.e. removal of the most susceptible entries and those which are virtually free of the pathogen (assumed to carry a major gene). Only against pathogens belonging to groups not known to form races easily any resistance observed can be selected and used



STRATEGIES FOR MULTIPLE STRESS RESISTANCE  
BREEDING IN COOL SEASON FOOD LEGUMES

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Cool season food legumes including chickpea, faba bean, lentil, and pea suffer yield losses from a number of biotic and abiotic stresses. Sources of resistance for many stresses have been identified. Utilizing these sources, cultivars with resistance to a single stress (disease, insect, Orobanche spp., cold, heat, or drought) have been bred. Some progress has also been made in developing cultivars with two to three stresses in some crops. Due to lack of multiple stress resistant cultivars, food legumes continue to suffer heavy yield losses and most importantly these stresses make production unstable.

Several strategies could be adopted to develop cultivars with multiple stress resistance. Program for germplasm enhancement for combined resistance to several important stresses can be initiated. Germplasm with multiple stress resistance would be useful in resistance breeding. Stepwise breeding for resistance could be another approach to follow. Shuttle breeding, that is, screening segregating material in one generation for one stress and for the second stress in the following generation. A minimum of two cycles would be required to ensure resistance to both stresses. A few wild species possess resistance to two to four stresses. For example, some accessions of Cicer bijugum possess resistance to Ascochyta blight, Fusarium wilt, leaf miner, and cold. If genes for resistance from these species could be transferred to cultigen with the help of biotechnology, pace of development of cultivars with multiple stresses in chickpea could be accelerated.

The implementation of breeding programs for multiple resistance requires a multidisciplinary collaboration. International cooperation plays a major role in integrating experiences and providing sources of germplasm resistance. Integrated crop management in respect of tolerant cultivars can help reduce yield losses.

UTILISATION OF GERMLASM RESOURCES OF COOL SEASON FOOD LEGUMES  
IN BREEDING FOR STRESS TOLERANCE

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An estimated 70,000 accessions of cool season food legumes and their wild evolutionary progenitors are stored in the worldwide network of gene banks and genetic resource centers. This has led to the general belief that natural diversity in these food legumes is safely preserved. However, information on the utilization of these rich sources of diversity for increasing crop tolerance of abiotic and biotic stresses is not easily available. A survey of recent publications on the use of crop genetic resources indicates that the preserved diversity is relatively under-used, particularly for genetic improvement of abiotic-stress tolerance in cool season food legumes. The causes of this limited utilization of genetic resources are examined. It is argued that the important reasons for the under-utilization of germplasm are (a) non-availability of techniques for rapid and reliable screening of large number of accessions for stress tolerance, (b) lack of appreciation of the critical role of evaluation biologists in genetic resources conservation programs, and (c) unrealistic expectation that plant breeders will evaluate large collections of germplasm and identify potentially useful genetic materials.

Systematic evaluation and documentation of germplasm resources are central to the efficient exploitation of the preserved diversity. For three important cool season food legumes, significant progress towards comprehensive evaluation of available accessions has been made in the past decade by the International Center for Agricultural Research in the Dry Areas (ICARDA). Recent (1987-88) evaluation data for one of these crops, chickpea (*Cicer arietinum* L.), are analyzed to demonstrate the usefulness of well-planned germplasm evaluation in identifying genotypes for stress-tolerance. Shannon-Weaver information index was used to estimate phenotypic diversity for 24 characters. Average diversity for response to stresses is only about 50% of that for other morpho-physiological traits. There are substantial regional differences in diversity, as well as frequency of desirable accessions for stress-related characters. Discrete log-linear multivariate techniques are used to elucidate associations among stress related characters. The analyses indicate that these associations differ among different chickpea growing regions of the world. Implications of these findings for utilising germplasm collections of cool season food legumes are discussed in relation to increasing tolerance of abiotic and abiotic stresses.

BREEDING METHODS FOR STRESS TOLERANCE  
IN SELF-POLLINATED FOOD LEGUMES

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The primary objective of a breeding program in a self-pollinated crop is to isolate the best homogeneous line from a series of crosses and do it in as short a time as possible. All breeding methods applicable to self-pollinated crops are effective, provided genetic variation is present for the trait(s) under consideration and the breeder can successfully separate (select) this genetic variability from the environmental variability for that trait(s). The standard breeding methods for self-pollinated crops are the 1) pedigree method, 2) bulk method, and 3) various modified bulk methods, such as the mass pedigree and the single-seed descent methods. These methods are effective, but they are inefficient in that selection for traits of low heritability, such as yield and stress tolerance, is delayed several generations. Efficiency can be increased (a new cultivar developed sooner) by maximizing the opportunity to progeny test selections, using the  $F_2$  derived family method. Thus, replicated progeny tests in the  $F_3$ ,  $F_4$ ,  $F_5$  and  $F_6$  and selection between and within crosses can effectively eliminate all  $F_2$ -derived families except for a couple of families from a couple of crosses. These can then be tested nationally and the best one released following concurrent selection and progeny testing of the pre-breeder seed for homogeneity.

BREEDING OF PEAS FOR WINTER SOWING\*

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Results are reported on a breeding program for obtaining pea varieties adapted for winter sowing. Cold tolerance was the main objective with a procedure foreseeing an evaluation of the material in controlling conditions in laboratory (at 7°C) and a selection in open field.

A parallel program was also carried out for resistance to powdery mildew with selection in both greenhouses and open fields. Several advanced lines was selected with different agronomic traits according to the different disciplines of the pea production: fresh market, canning, freezing, protein and fodder.

Confirmations of the efficiency of the selection procedures were obtained in open fields in 1987 for cold tolerance when a severe cold stress occurred and in 1990 for powdery mildew resistance when a heavy fungus attack was present.

\* Confirmation No. 61 from Centro di Studio per il Miglioramento Genetico degli Ortaggi, CNR, Portici (Napoli), Italy.

## MOLECULAR GENETICS FOR STRESS BREEDING IN COOL SEASON FOOD LEGUMES\*

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Advances in molecular genetics of higher plants make it timely to identify and to dissect simple metabolic traits that confer stress tolerance. In this paper, we try to summarize the recent work concerning the molecular basis of plant response and resistance to environmental stresses. Emphasis is modeled on systems other than cool season legume crops, where extensive literature is not available, as preliminary concepts to outline possible ways of identifying and isolating genes related to stress resistance or susceptibility. Our review is limited to low and high temperature stress, drought and NaCl excess, since these are the main constraints limiting yield performance of the main cool season legume crops cultivated in the Mediterranean areas.

\* Contribution No. 62 from Centro di Studio per il Miglioramento Genetico degli Ortaggi, CNR, Portici (Napoli), Italy

**MUTAGENESIS AND CHROMOSOME MANIPULATION FOR STRESS RESISTANCE IN  
COOL SEASON LEGUMES**

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The capability to withstand various kinds of stresses represents a prerequisite for the improvement of grain legume varieties under low input conditions of marginal lands. When winter sowing is adopted, considerable benefits can derive particularly from cold tolerance, disease resistance and adjustment of crop duration to the growing season available. In order to obtain these desired traits, mutation induction and chromosome manipulation can play an important role as methodologies complementary to breeding activities.

The use of mutagenic agents on different ontogenetic stages of the plant has been reported. Much attention has also been paid to M<sub>1</sub> chimerism and to some chromosome rearrangements detected respectively after seed and gamete irradiation. Besides, the strategy for the selection of resistance to biotic and abiotic stresses has been described. From a practical point of view, mutant resistant cultivars of pea, faba bean, chickpea have been released and several lines have been used in cross breeding programs.

In order to transfer valuable resistances from wild species, cytogenetics techniques have to be taken into account by legume breeders. In this context, rearrangements (deletions, duplications, traslocation) and trasfer of chromosomes provided a great deal for increasing the genetic variation. Preliminary knowledge of the kariotype is also useful in order to establish the relationship among the species and to finalize the interspecific hybridizations.

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