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Domesticated tetraploid oats: a new perspective for Morocco

Principal Investigator: Gideon Ladizinsky

Grantee Institution: The Hebrew University of Jerusalem

Collaborator: Chauki Al Faiz

Institution: INRA Rabat

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Executive Summary

Oat is a relatively new crop in Morocco, but the popular oat cultivars in Morocco are selections from varieties of US origin and are not properly adapted to the local climatic conditions. Particularly in dry years, crop failure has a disastrous effect because farmers are forced to sell their goats and sheep at sacrificial prices. Morocco is the home to many wild oat species which are well adapted to the local ecological conditions. Among them are the tetraploid (2n=28) species *Avena magna* and *A. murphyi*. Both have large grains with high protein content (about 30% more than in the cultivated oat), and exhibit tolerance to several oat diseases. The main purpose of the project is to domesticate these two tetraploid oats by transferring to them, via hybridization, the domestication syndrome of the common oat. The second objective is to train Moroccan scientists in the handling and utilization of oat's wild genetic resources.

During the second year of the project we have selected domesticated tetraoploids resulting from crosses between hexaploid oat cultivars growing in Morocco and the wild species *A. magna and A. murphyi*. With *A. magna* we are already in the second hybridization cycle and hybrids (2n=35) between the domesticated form and hexaploid oat cultivars are grown now in Morocco in the field surrounding by the domesticated tetraploid form for natural backcross. Domesticated forms of *A. murphyi* are being crossed now in Israel with hexaploid oat cultivars and will be planted next year in Morocco in a backcross experimental design.

In another series of experiments we have selected hexaploid derivatives from 2n=35 hybrids between hexaploid oat cultivars and the wild tetraploids following backcross to the hexaploid parents. These derivatives will be grown up to F_4 and their performance will be compared with their hexaploid parents.

Section I

A) Research objectives

The objectives in the second year of the project were:

1. To initiate the second hybridization cycle with domesticated *A. magna*.
2. To produce hybrid seeds between domesticated *A. murphyi* and common oat cultivars grown in Morocco.
3. To grow in the field *A. sativa* x *A. murphyi* hybrids within *A. sativa* stand for obtaining backcross seeds from which hexaploid derivatives will be selected.
4. To initiate selection of hexaplolid types in the first segregating generation of the backcross (*A. sativa x A. magna*) x *A. sativa*.
5. To explore further the distribution of wild *A. magna and A. murphyi* in Morocco and to collect seeds.

B) Research accomplishments

<u>Backcross to common oat</u>

1. (*A. sativa x A. magna) x A. sativa*: backcross plants of this combination were grown in a net house in Rabat. Despite regular treatments against aphids (vectors of the viral disease BYDV) most plants showed symptoms of the disease such as red leaves, small number of tillers per plant, reduced vigor and blasting of florets. At maturity all the plants retained their seeds. The partial sterility due to the unstable chromosome numbers of the backcross plants became more pronounced following the BYDV attack. Nevertheless, sufficient number of seeds were collected for the growing of the F_2 generation. Altogether, 76 F_2 families were planted in Marchouch experimental

farm in the form of a row per family to distinguish between homozygous and heterozygous families for seed non-shattering as explained below:

A. sativa- seed non-shattering, genotype AA

A magna- seed-shattering, genotype aa

A. sativa x A. magna seed non-shattering, genotype Aa

(A. sativa x A. magna) x A. sativa-seed non-shattering, genotypes AA, Aa

F_2 *(A. sativa x A. magna) x A. sativa*- Families AA bred through. Aa segregate

2. *(A. sativa x A. murphyi) x A. sativa*: hybrid seeds between three oat cultivars of Morocco, Soualem, Tissir and Amlal and two *A. murphyi* genotypes, 45-55 and 50-52 were planted among their cultivated parents as pollen donors in Marchouch Experimental Farm. Plant development was good and so the overlapping flowering time of parents and hybrids. As a result a large number of seeds were collected on the hybrids as indicated in Table 1 (the hybrids are self-sterile):

Table 1. Backcross seeds collected on various *(A. sativa x A. murphyi) x A. sativa* combinations

Hybrids	no. seeds collected
Soualemn x 45-55	4665
Soualem x 50-52	3546
Tissir x 45-55	4261
Tissir x 50-52	1070
Amlal x 45-55	116
Amlal x 50-52	309

The range of chromosome numbers was estimated by counting 2n in samples of the various combinations. Counts of chromosome numbers were made in root tips following

Feulgen staining and are summarized in Table 2. By deducting 21 (chromosome number in *A. sativa* gamete), from these numbers, chromosome number in the viable female gametes of the F_1 hybrids is revealed. It appears that the most viable gametes had 21 chromosomes, and adding to them those with one more or one less chromosome they together constitute more than 50% of the viable gametes.

The resulting plants were grown in an air-conditioned greenhouse for further study of their pollen and seed fertility. A large number of seeds (150-1000) of each hybrid combination were planted in the field in Rharb Experimental Station.

Table 2. Frequency distribution of chromosome numbers in backcross plants (*A. sative x A. murphyi) x A. sativa.*

	2n											
combination	34	35	36	37	38	39	40	41	42	43	45	Total
Soualem x 45-55	1	1	1	0	5	6	6	4	9	3	0	36
Soualem x 50-52	1	2	0	1	3	4	4	4	10	6	2	35
Tissir x 45-55	0	1	1	3	3	3	5	4	19	4	0	43
Tissir x 50-52	0	1	0	2	3	3	5	2	13	2	0	31
Amlal x 45-55	0	1	0	0	4	4	2	5	8	2	4	30
Amlal x 50-52	0	1	1	0	3	2	9	6	7	12	1	42
Total	2	7	3	6	21	22	31	23	66	29	7	217

Backcross to the tetraploid species

1. Production of *A. sativa* x *A. magna* (domesticated) hybrid seeds. Domesticated *A. magna* plants were selected in autumn 1998 in Israel and were crossed as male parents to *A. sativa* in spring 1999. The hybrid seeds were planted in Morocco in autumn 1999 in Marchouch Experimental Farm, surrounded by their tetraploid parents and tetraploid lines selected in BARD project in 1993. This experiment constitutes the beginning of the second hybridization cycle.
2. (*A. sativa* x *A. magna* wild) x *A. magna* wild: although this combination was grown in Israel in the process of selecting domesticated *A. magna*, we thought that it would be useful to run a similar experiment in Morocco to show the Moroccan collaborators how domesticated forms can emerge from this cross. The hybrid seeds were planted in Marchouch Experimental Farm surrounded by their wild parents. Unfortunately, the hybrids terminated flowering before the wild tetraploid plants have not even started. As a result, no seeds were formed on the hybrids.
3. Selection of domesticated *A. murphyi*: Accessions no. 45-55 and 50-52 of *A. murphyi* were crossed as male parents with *A. sativa* cultivar Soualem. The hybrids were planted in Israel in 1999 growing season surrounded by plants of the tetraploid parents as pollen donors (the hybrids are self-sterile). One hundred and five backcross seeds were collected on the hybrids, of which 95 were germinated in summer 1999 and chromosome counts in root tips revealed a range of 2n from 28 to 36. Fifty four plants exhibited the seed-shattering habit of the wild parent. This kind of segregation is expected in a backcross of a heterozygous to its recessive parent. Severe sterility was typical among the backcross plants. Only two plants with non-shattering seeds,

2n=29 and 2n=30, produced some seeds in autumn 1999. Among their progeny plants with 2n=28 were selected and they crossed in spring 2000 with *A. sativa* to initiate the second hybridization cycle.

Collecting wild tetraploid species

The year 1999 was relatively dry in Morocco. Rainfall was below average with patchy distribution and it terminated, in most places, at the end of January. The consequence for wild stands was catastrophic because of heavy pressure of grazing and cutting. Despite these, we conducted a collection trip to the north western part of Morocco to detect populations of *A magna* and *A. murphyi*. We collected seeds in 13 populations of which two of *A. murphyi* and four of *A. magna* were firstly detected in this trip. Seeds from about 100 plants were collected in each population and were kept separately for multiplication and later for evaluation. The seeds and the details of the source populations are kept in the Forage Department of INRA. Rabat.

C) Scientific impact of collaboration

During the second year of the project the impact of the scientific collaboration is become more and more apparent both in specific fields and general issues. The collaborator scientists and their technicians have acquired the cytological techniques for handling mitotic chromosomes and counting chromosome numbers in root tips is a routine now. What is still needed is more experience with meiotic chromosomes. Inadequate growing conditioned and particularly the delay in erecting the air-conditioned greenhouse have been the major reasons for that. Now when the greenhouse is operational we anticipate a similar progress in this matter as well.

The collaborators have gained confidence in exploration of wild oat species and have learnt the importance of ecological factors in the process of identifying the habitats where the various species use to grow.

A more general aspect, the process of exploiting wild genetic resources is much clearer for the Moroccan collaborators. Seed collecting in the wild is only the first and a relatively easy step. Selecting useful genes in the wild material and transferring them to a domedsticated background is by far more laborious but rewarding.

E) Strengthening of developing country institutions

The completion of the air-conditioned greenhouse marks a new era for the Forage Department of INRA in Rabat. It appears that this is the first greenhouse of that kind for annual crops in INRA. It open the way to speed up breeding programs by growing two generations per year instead of only one at the moment. The establishment of a small cytogenetic unit in the Forage Department within the frame work of the project, has instantly provoked the interest of scientists inside and outside the Department of applying chromosome studies in their material.

F) Future work

During the last year of the project we hope to complete the second hybridization round with domesticated *A. magna*, and if lucky, to initiate the third round. With domesticated *A. murphyi*, the second hybridization cycle will be completed shortly after the end of the project. Unfortunately, winter 1999/2000 has been extremely dry. Rainfall was plentiful in November-December, but since then not a drop of rain. This is an alarming situation for Morocco as a whole with immediate consequences for the project. Water for

irrigation in the experimental station are scarce and the few green patches attract all kind of pests which are rare in a normal year.

Section II

A) Managerial issues

The project runs its course with no managerial problems.

B) Budget

In spite of spending excessive fund on the air-conditioned greenhouse we secured enough fund for running costs in the third year of the project.

C) Special concerns

No change has occurred with this item during the second year of the project.

D) Collaboration

Travel: During the second year of the project the Principal Investigator visited Morocco three times. The first one, March 16- March 22, was devoted for inspecting the experimental plots, the state of the greenhouse, training the Moroccan collaborators in collecting immature anthers for meiotic studies and in making interspecific crosses. The second one, May 23-June 5 was for harvesting backcross seeds and collecting wild *A. magna* and *A. murphyi*. In the last visit, October 10-17, hybrid seeds of *A. sativa x A. magna* (domesticated) were brought for planting in the autumn. The results of the previous season were evaluated and the goals for the next season were discussed resulting in a clear work plan.

Training: The visit of the principal Investigator to Morocco have been used also for training as indicated above This method of Visit and Teach appears successful and

ensures proper transmission of knowledge and a regular follow up of progress and drawback.

Anticipated activities in the next 6 months

Morocco

1. Selecting fertile F_2 families of *(A. sativa x A. magna) x A.sativa* which retain their seeds at maturity.
2. Seed harvest of F_1 plants of the *(A. sativa x A. murphyi) x A. sativa* cross.
3. Harvest of backcross seeds (*A. sativa x A. magna*- domesticated) *x A. magna*-domesticated, counting chromosome numbers in root tips and raising the F_1 plants in the greenhouse.

Israel

1. Production of hybrid seeds *A. sativa x A. murphyi*-domesticated.

E)Request for American Embassy Tel-Aviv or AID action

None