

Annual Report

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Domesticated tetraploid oats. a new perspective for Morocco

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Executive summary

Oat is a relatively new crop in Morocco. Introduced by the French it is now main Morocco's forage crop. However 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 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 first year hybrids between the common oat and both wild species were grown in the field, amongst one of their parents (common oat in Morocco and *A. magna* in Israel). Over 100 backcross seeds were collected in each experiment. Chromosome counts in root tips revealed a wide range, from $2n=28$ to $2n=43$.

In spring 1998, we explored wild populations of *A. magna* and *A. murphyi* in Morocco. We discovered a number of new populations and characterized the habitats in which these species grow. Perhaps the most interesting finds during was a population in the Tangier area where the two species grow in mixed stand.

Section I

A) Research objectives

The two main objectives of the project are

- 1 To domesticate the wild tetraploid ($2n=28$) oat species *Avena magna* and *A. murphyi* by transferring to them via hybridization the domestication syndrome of the common oat (*A. sativa*, $2n=42$) The characters of the domestication syndrome in oat include seed retention at maturity, reduced awn formation and yellow and glabrous husks

The first year's objective was to grow of hybrids between the common oat and both wild tetraploids, collecting seeds from these hybrids which are the result of backcrosses to their parents, and raising plants from these seeds

- 2 To train the collaborating Moroccan scientists in the collection and utilization of oat's wild genetic resources

B) Research accomplishments

Officially, the project started on March 1st 1998, scientific activity, however, began almost a year earlier. During the spring of 1997 the principal investigator produced hybrid seeds between several oat cultivars (*A. sativa* $2n=42$) grown in Morocco and two accessions of *A. magna* ($2n=28$). At the first sign, in October 1997, of the project's potential funding, the principal investigator traveled to Morocco to discuss the initiation of the project already in autumn 1997, before official approval. The reason for this was that were the approval to be delayed, as did in fact happen, we would not lose the 1997/8 growing season. During that visit the hybrid seeds were delivered and the planting design introduced. During the 1997 visit, the principal investigator became

acquainted with the personnel that had been appointed for the project, and the local facilities were examined and plans developed for their improvement. These included the purchase of additional microscope objectives, and the erection of a small air-conditioned greenhouse.

Backcross plot (*A sativa* x *A magna*) x *A sativa*

In December 1997, *A sativa* x *A magna* F1 hybrids were planted in the Marchuch experimental farm (Morocco) and were interspaced with oat cultivars Soualem, Ghali, Tissir and Zahari, which were parents in the crosses made in 1997.

The F1 hybrids in these cross combinations are totally male sterile but have the potential to produce a few seeds when pollinated by fertile pollen from their parents. Although oat is considered as a self-pollinating plant, at flowering it releases considerable amounts of pollen. The mixed planting of F1 hybrids and oat cultivars enabled natural backcrossing to occur in the field.

In May 1998, backcross seeds were collected from the F1 plants. The few seeds (about one per panicle) were detected when the panicles became yellow. At this stage, the fertile spikelets were still green. All the empty spikelets were then removed and seeds were collected when they were mature and dry. The total number of backcross seeds obtained is as follows:

<i>(A sativa</i> (Tissir) x <i>A magna</i> 158) x Tissir	3
<i>(A sativa</i> (Ghali) x <i>A magna</i> 158) x Ghali	18
<i>(A sativa</i> (Zahari) x <i>A magna</i> 169) x Zahari	9
<i>(A sativa</i> (Soualem) x <i>A magna</i> 169) x Soualem	81

The higher number of seeds on hybrids involving Soualem reflects the higher number of hybrids in that particular combination, and probably better cross-compatibility.

The backcross seeds were germinated in October 1998 and chromosome counts were attempted in root tips of 80 seedlings (Table 1)

The seedlings were planted in pots and were raised to study chromosome pairing at meiosis, pollen stainability and seed set

(A sativa x A magna) x A magna

The backcross to the tetraploid wild parent was conducted in Israel during the 1997/98 growing season, and a parallel experiment is in progress in Morocco in 1998/99 from hybrid seeds produced in Israel. Only Soualem was used as a hexaploid parent, but of *A magna* accessions no. 158 and 169 were employed. Altogether 85 hybrid seeds were planted and interspaced with the tetraploid wild parents. In May 1998, 105 backcross seeds were collected from the F1 hybrids. The seeds were germinated in July and chromosome numbers were counted in root tips (Table 2). The plants were raised to maturity in an air-conditioned greenhouse.

In only 8 of the 95 backcross plants, pollen stainability was higher than 35% and these were the only plants that produced a few seeds by selfing. Of the 95 plants, 52 exhibited seed dispersal as the wild parents (shedding spikelets) and in 43, spikelets were retained as in the common oat. Of the latter, only four produced a few seeds.

Table 1 Distribution of chromosome numbers in (*A sativa* x *A magna*) x *A sativa* populations

2n=42

parent	Chromosome Numbers									Total
	35	36	37	38	39	40	41	42	43	
Ghali						2	5	6		13
Zahari			1		2		2	1		6
Soualem	11	11	1	9	8	7	17	14	1	61

Table 2 Distribution of chromosome numbers in (*A sativa* x *A magna*) x *A magna*

2n=28

line	Chromosome Numbers									Total
	28	29	30	31	32	33	34	35	36-40	
158	2	6	5	7	2	3	5	7	1	38
169	8	7	8	9	4	2	7	8	3	56

Collecting wild tetraploid oats

The tetraploid oats *A magna* and *A murphyi* were collected for three main purposes 1 to identify the habitats where these wild oats grow, 2 to collect more of their germplasm, and 3 to train the collaborating Moroccan scientists in the systematic procedure of detecting and collecting wild genetic resources

Since the two tetraploid oats have an affinity to heavy soil, soil maps are indispensable for targeting potential growing areas for these wild oats Unfortunately, we were unable

to obtain such a map and the excursion was based mainly on the principal investigator's and the collaborating scientists previous experience with the distribution of these wild oats. Another limiting factor was the drought which affected the wild oat populations. They were more scattered geographically and much smaller in size than in a rainy year. Moreover, the natural habitat of both *A magna* and *A murphyi* is agricultural land but the wild oats cannot tolerate cultivation and may be restricted to field borders or deserted plots. Plants of these wild oats which were probably abundant in the past have today become rare. Despite these limitations, we were able to detect some previously unreported populations, particularly of *A magna*. Perhaps most exciting was the discovery of a population in the Tangier area, presently the most northern fringe of this species, in mixed stand with *A murphyi*. Another important find was a population of weedy *A magna* which thrived in a wheat field, west of Fes. The *A magna* plants were vigorous, outstripping the wheat plants by over one meter, and produced a large number of spikelets per plant.

The distribution of *A murphyi* in Morocco is restricted mainly to the Tangier area. Rapid urbanization there has replaced some previously observed populations of *A murphyi* with houses and factories. Without appropriate measures to protect it *A murphyi*, would likely be eliminated from this area within a few years. Another small and isolated population of *A murphyi* was encountered near Ben Slimane, east of Casablanca. Soil type in that general area is sandy loam, but many small marshes are scattered there with heavy black soil. Grazing is extensive in these marshes and the natural vegetation is protected from it only when adjacent to a cultivated field.

C) Scientific impact of collaboration

The scientific impact of the collaboration has two aspects: training in cytogenetics as a tool for conducting introgression experiments, and a better approach for detecting and collecting wild genetic resources.

None of the collaborating scientists had any background in cytogenetics. It therefore became necessary to prepare a short course in cytogenetics for them, during the visits of the principal investigator to Morocco, that would enable them to understand and execute the experimental procedures. Then, under the supervision of the principal investigator and later on their own, they performed chromosome counts in the backcross population. They also conducted the field experiment and collected the rare backcross seeds.

D) Description of the project's impact

The results obtained so far are the first links in a chain that will eventually lead to the formation of domesticated tetraploid oats, and common oat cultivars with genes from the wild tetraploids. We anticipate that this new germplasm will provide Morocco with better adapted oat cultivars.

E) Strengthening of developing country institutions

Rapid progress in breeding grain crops depends, among other things, on the ability to grow two or even three generations per year, usually in controlled environment facilities. This kind of facility has been lacking in the Forage Department of the INRA in Rabat, and apparently in other INRA departments. The erection of a small air conditioned greenhouse will enable the achievement of a high rate of hybrid seeds in crosses, and the raising of two generations per year in the introgression experiments with oats and other crop plants as well.

Studying and understanding the chromosomal aspects of introgression experiments are of theoretical and practical importance. Introducing the cytogenetic dimension to the Forage Department will improve their research capacity as well as that of INRA as a whole.

F) Future work

The project is running its course though behind schedule because the delay of erecting the greenhouse in Morocco. The introgression experiments will continue for the next two years. In the $2n=28 \times 2n=42$ experiment, we hope to select a better common oat type toward the end of the project, and in the $2n=42 \times 2n=28$ experiment to select a domesticated tetraploid of *A. magna* which will be the cornerstone in developing tetraploid oat cultivars.

The work plan has undergone some revisions. Originally, a domesticated *A. magna* and *A. murphyi* were planned for crosses with Moroccan oat cultivars. However, disagreement arose with the respect of the issue of royalties should the project produce commercial cultivars. Thus, it has been agreed that the starting material will consist of wild forms of the two tetraploid species and not the domesticated type which has been developed for over 10 years in Israel.

Since the viral disease BYDV is a serious problem in Morocco, we have decided to screen the wild tetraploid accessions for BYDV tolerance, although this was not initially part of the project. It seems now that a selection of such tolerant types is almost a prerequisite for the establishment of domesticated tetraploid cultivars.

Section II

A) Managerial issues

A major managerial constraint to the project productivity has been the transfer of money to Morocco. The first transfer arrived in Morocco in August 1998, more than 3 months after the transfer order has been issued in Tel Aviv. We are experiencing the same problem with the second transfer, issued in December 1998, but not yet arrived in Morocco.

B) Budget

In the project proposal we indicated the need for an air conditioned greenhouse as a major facility to improve hybrid seed production and to raise two generations of hybrid derivatives per year. The Moroccan collaborators suggested building an air conditioned compartment in an existing greenhouse. But on the principal investigator's first visit of Morocco it was apparent that this was not a realistic option. Instead, we decided to erect a small, independent greenhouse. This is now nearing completion and it is obvious that the money allocated in the budget will not be sufficient and it will be necessary to withdraw funds (approximately \$10,000) from other items such as material, other direct costs and seasonal labor.

C) Special concerns

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

D) Collaboration

Travel the principal investigator visited Morocco twice during the first year of the project, May 16- June 24 and October 10-20. During these visits project progress in Morocco was assessed and future work was discussed with the project personnel.

Training the visit to Morocco was also used for training The trainees were Saidi Nezha, Saidi Seddik, Mouna Anchem and Souihka Allal

During the May-June visit, they were trained to detect backcross seeds following natural backcrossing of the pentaploid hybrids to their hexaploid parents They were also trained to collect wild genetic resources of oats During October the visit, the first three of them were trained in chromosome staining, chromosome counting and recognition of individual oat chromosomes

The following activities are plan for the next 6 months

Morocco

- (a) raising of (*A sativa* x *A magna*) x *A sativa* backcross plants and studying their fertility
- (b) raising of *A sativa* x *A murphyi* hybrids among their *A sativa* parents, collecting their seeds, counting chromosome numbers in their root tips and recording chromosome behavior at meiosis
- (c) raising of *A sativa* x *A murphyi* hybrids among their *A sativa* parents, collecting their seeds, counting chromosome numbers in their root tips and studying their behavior at meiosis
- (d) screening *A magna* and *A murphyi* collections for BYDV tolerance

Israel

Selection of domesticated *A magna* plants among derivatives of (*A sativa* x *A magna*) x *A magna* backcross and crossing them again to *A sativa*

E) Request for American Embassy or AID actions

As already indicated, we are facing considerable delays in fund transfers to Morocco, about 3 months from the time the order is issued until its arrival there. We would appreciate any attempts to shorten this period.