Domestication of *Avena magna* Murphy et Terrell: a wild tetraploid oat species endemic of Morocco

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Введение в культуру дикорастущего тетраплоидного вида овса Avena magna Murphy et Terrell, эндемика Марокко

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Avena magna Murphy et Terrell (2n = 4x = 28), a tetraploid oat species endemic of Morocco, has a high groat protein content (more than 20%), good resistance to diseases and good adaptability to the Mediterranean edaphic and climate conditions. Moreover, this taxon is morphologically similar to the hexaploid oat species A. sativa and promising for interspecific crosses with A. sativa in order to transfer the domestication syndrome into it. Four hybridization cycles employing four accessions of A. magna and five Moroccan hexaploid oat cultivars of A. sativa were accomplished to domesticate A. magna. Morphological characterization and cvtogenetic analysis of derivative hybrid seeds were made to determine their ploidy level and select the seeds with 2n = 28. The overall combinations yielded 81 hybrid plants with 2n varying from 28 to 29, with 58% having 2n = 28, and pollen fertility over 85%. However, 27 hybrids yielded a seed set ranging from 20 to 80%. Selected hybrid plants were subjected to pedigree selection in the field until they reached the 8th generation and assessed for agronomic performance. Three domesticated lines of tetraploid oat A. magna were selected for their good disease resistance. Analysis of groat protein content in the selected hybrids using the Kjeldahl method showed that it was 2 to 3% higher than in their parents.

Key words: oats, common oat, groat protein content, Kjeldahl analysis, human consumption.

Тетраплоидный дикий вид овса *Avena magna* Murphy et Terrell (2n = 4x = 28), эндемик Марокко, имеет высокое содержание белка в зерне (более 20%), высокую устойчивость к болезням и хорошую адаптивность к почвенноклиматическим условиям Средиземноморья. Морфологически этот вид сходен с гексаплоидным культурным видом овса *A. sativa* L., который и был использован в межвидовых скрещиваниях для одомашнивания дикого вида. Для внесения в дикий вид признаков культурного овса провели четыре цикла скрещиваний с участием четырех образцов *A. magna* и пяти марокканских гексаплоидных сортов *A. sativa*. Для отбора гибридных растений с числом хромосом 2n = 28 использовали морфологические характеристики и цитогенетический анализ.

По всем комбинациям получено 81 гибридное растение, с числом хромосом (2n) от 28 до 29. Только 58% гибридных растений имели 28 хромосом и фертильность пыльцы более 85%. Отобранные гибридные растения подвергались дальнейшему отбору в поле по агробиологическим показателям до поколения F_8 . В результате изучения по признаку устойчивости к болезням отобраны три одомашненные линии тетраплоидного овса *А. magna*. Анализ содержания белка в зерне с использованием метода Кьельдаля у отобранных гибридных линий показал, что он на 2–3% выше, чем у родительских форм.

Ключевые слова: овес посевной, содержание белка в зерне, анализ Кьельдаля, пищевое направление.

Introduction

Since the first century of our era, oats have become a major crop in Europe and in certain other Mediterranean regions. Recently, too much interest was given to this cereal since it has a caryopsis rich in protein up to 22% which is the highest value for *Avena sativa* L. (Ladizinsky, Fainstein, 1977) in addition to other components, such as oil which can reach 18.1% in some lines of *A. sativa* (Welch et al., 2000). Nutritive quality of this cereal has increased, in contrast to wheat and barley; lysine and tryptophan concentrations in oats are significantly high and adequate for a balanced nutritive diet designated for non-ruminant animals such as horse.

Oats are used in different forms: as a pure crop, green forage, hay, silage, grain and straw. In Morocco, oats are used as forage and seem to play a role for dairy cattle breeding mainly in the North-West of Morocco. It remains the most important cultivated crop for populations living in marginal lands in developing countries (Al Faiz et al., 1997). It has a high nutritive value as forage with good digestibility of dry matter exceeding 75%. Stevens et al. (2004) reported that oat hay has more digestible organic matter, more appreciated by cattle.

Oats grains are notable for high groat protein content, with a high nutritive value, and a good rate of oils, in addition to other compounds that exert good beneficial effects on health, such as antioxidants and vitamins (avenanthramides and tocols) (Mannerstedt et al., 2004; Saidi et al., 2013; Benchekroun et al., 2014). Clamot (1979) and Welch et al. (2000) have reported that oat groat protein is unique among those of other cereals grown in temperate regions, as it has a relatively good balance of amino acids, which remains stable even if the rate of groat protein is increased by genetic manipulations. Oat groat is also richer in oil, compared to other cereals. Oat groat increases the energy density, being rich in unsaturated fatty acids, and has a good potential for industrial extraction as a biofuel (Zhu et al., 2004). In addition, oat groat contains significant amounts of (1-3), (1,4)-β-D-glucan (Manzali et al., 2018). This substance is the main component responsible for decreasing plasma cholesterol which attracted the attention of processors and plant breeders (Welch et al., 2000). Recent medical reports confirm that fiber consumption prevents intestine and colon cancers. Due to its valuable components, oat producers have increased the production of this cereal since the 1990s and promoted it as a beneficial food for human health (Harlan, 1992).

Due to its beneficial effects on human health, more attention was given to this cereal, therefore the demand for oat seed is permanently increasing. This led to boosting new breeding research programs to release new cultivars for human consumption. In order to succeed in the release of new lines with high nutritive value, responding to all technological food criteria, it is necessary to employ wild oat genetic resources, since they constitute a rich reservoir of valuable genes. Among these wild oat species, *A. magna* Murphy et Terrell is a tetraploid oat species (2n = 4x = 28), discovered 28 years ago and native to Morocco. This taxon is morphologically similar to hexaploid oat species and it is difficult to distinguish it from *A. sterilis* L. According to Rajhathy and Sadasivaiah (1969), *A. magna* had been involved in the origin of hexaploid species. It has a groat protein content reaching 30%, good oil content and large seeds (1000 seed weight \approx 35 g). Since it is endemic to Morocco, *A. magna* has good resistance to diseases threatening oat cultivation, such as powdery mildew, crown rust and barley yellow dwarf virus. In addition, it has good tillering productivity (Ladizinsky 1993; Loskutov, 2001) and good adaptability to Moroccan soil and climate conditions.

Because of its impressive characteristics, this tetraploid taxon has been domesticated recently by transferring into it the domestication syndrome of the common oat *A. sativa*. The transferred characters were the non-shedding spikelets, glabrous and yellow lemma, and reduced awn formation. The domestication was achieved by crossing the common oat with the tetraploid species and then backcrossing the pentaploid hybrids with pollen of the tetraploid wild parent (Ladizinsky, 1995). Therefore, in the early 2000s, a research programme was boosted by INRA aiming to domesticate the tetraploid oat species *A. magna* by transferring the diversity of the Moroccan common oat cultivars into it, and release tetraploid oat lines developed for human consumption.

Methodology

The hybridization work involved five Moroccan common oat cultivars of *A. sativa*, registered in the national official catalogue ('Soualem', 'Amlal', 'Ghali', 'Tissir' and 'Zahri') and four accessions of *A. magna* (P1-1, P1-6, Aa2 and Aa16). *A. magna* P1-1 and P1-6 are the derivatives of the crosses accomplished previously between 'Soualem' and wild accessions of *A. magna* (158 and 169, respectively) originating from Morocco (Ladizinsky, 1995).

Seeds of both A. sativa cultivars and A. magna accessions were germinated in Petri dishes and transplanted into Jiffy pots under controlled greenhouse environmental conditions to prepare plants for hybridization crosses (mean temperature = 24°C). Backcrosses of the derivative hybrids to their tetraploid parents were performed in the greenhouse as well as in the field at the INRA experimental domain in Marchouch (60 km SE Rabat, altitude = 400 m, longitude = 6°42', latitude = 33°45', and annual rainfall = 242-650 mm). Each year we implemented two hybridization cycles, a winter generation and a summer one. Four hybridization cycles were undertaken, the first and the third were achieved manually under controlled environmental conditions in a greenhouse, aiming to backcross the pentaploid hybrids to respective common oat cultivars. The second hybridization cycle aimed to backcross the pentaploid hybrid seeds, derivatives of the crosses between A. sativa cv. 'Soualem' and A. magna P1-1 and P1-6, to their tetraploid parents, respectively. As for the fourth hybridization cycle, derivative hybrid seeds of the third cycle were pollinated by the derivative hybrid plants of the second hybridization cycle. After the second and the fourth hybridization cycles we proceeded to:

1. Chromosome counting in root tips in order to select the hybrid seeds with tetraploid chromosome number (2n = 4x = 28) (Singh, 1992).

2. Assessment of pollen stainability (Singh, 1992) and seed set for the derivative plants.

Morphological seed characterization

Derivative hybrid seeds were subjected to morphological characterization to examine lemma color, lemma hairiness,

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presence or absence of awns, and awn development. Lemma color was classified as yellow or as various degrees of coloration; lemma pubescence as glabrous or as various degree of hairiness; and awn number as presence of awns, absence of awns, one or two awns per spikelet (Ladizinsky, 1995).

Selection method

Collected seeds from each single hybrid plant having (2n = 4x = 28) issued from the fourth hybridization cycle were planted as F_2 generation at the INRA experimental station in Marchouch (68 km SE Rabat, Morocco, longitude: 6° 71 500 W, latitude: 33° 60 499 N), and subjected to pedigree selection. Selection was primarily based on resistance/tolerance to diseases, mainly crown rust, powdery mildew and BYDV.

Groat protein analysis

Seeds of the derivative lines, in addition to their respective tetraploid and hexaploid parents, were assessed for groat protein content using the conventional Kjeldahl method of chemical analysis (DIN ISO 1578, 2001).

Statistical analysis was made using SAS software version 9.1.

Results and discussion

Second hybridization cycle

The pentaploid hybrid seeds, derivative of the first crosses, achieved previously by Ladizinsky (1995), between *A. sativa* cv. 'Soualem' with each of the accessions of *A. magna* P1-1 and P1-6, were backcrossed to their tetraploid parents and yielded 18 and 59 hybrid seeds, respectively. Chromosome number of the hybrid seeds derived from the crosses involving P1-1 ranged from 2n = 28 to 41. Only two plants

had the tetraploid chromosome number (2n = 4x = 28), pollen fertility of 94% and 97%; and a seed set of 36% and 87%, respectively. Seeds derivatives of the cross involving 'Soualem' and P1-1 are scored 'B'.

The backcross involving P1-6 yielded 59 hybrid plants with 2n ranging from 28 to 41, pollen fertility ranging from 27% to 98%, and a seed set varying from 0% to 98%. Among the yielded hybrids, 17% were tetraploids with pollen fertility ranging from 43% to 96%, and a seed set from 0% to 90% (only two plants had a good seed set of 52% and 90%, respectively). Seeds derivatives of the cross involving 'Soualem' and P1-6 are scored 'C'.

Seed morphology of the tetraploid derivative seeds was similar to the one of their hexaploid parents. For the cross with P1-1, seeds had basic lemma hairiness, a yellowish lemma, and only one developed awn. As for the derivatives of the cross with P1-6, seed morphology of the hybrid seeds did not differ from the derivative seeds of the cross with P1-1.

Fourth hybridization cycle

The overall combinations of the fourth hybridization cycle yielded 236 hybrid plants, with 2n varying from 2n = 28 to 42. Crosses between different hexaploid cultivars of *A. sativa* and the derivatives of the cross which involved P1-6 yielded more tetraploid hybrid seeds (101 individuals), compared to the cross with derivatives of P1-1 (40 individuals). The most yielding cross of tetraploid seeds was the cross involving 'Amlal' (76 seeds), followed by 'Soualem' (29 seeds), 'Ghali' (24 seeds), 'Zahri' (9 seeds), and Tissir (1 seed). Crosses between 'Amlal' or 'Soualem' with the derivatives of P1-1 were the most yielding ones compared to the crosses achieved with the other cultivars (23 and 16 seeds, respectively) (Table 1).

Table 1. Frequency distribution of chromosome numbers among BC plants of the 4th hybridization cycle involving Avena magna

Таблица 1. Распределение чи	сла хромосом у гибридных раст	ений BC ₄ с участием Avena magna
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Combinations	27	28	29	30	31	32	33	34	35	36-42	Total
A × B91	1	23	2	1	1	2	1	1	0	0	31
A × C05		7	0	0	0	0	0	0	0	1	8
A × C21	0	8	0	0	0	0	0	0	0	0	8
A × C90		23	4	2	0	1	1	0	0	1	32
G × C05		24	5	0	2	0	0	0	0	2	33
S × B55		1				0	0	0	0		1
S × B91	0	16	10	4	3	1	2	1	3	0	40
S × C05	1	10	2	5	0	2	0	0	0	1	20
S × C21	1	9	2	2	1	0	0	0	1	1	16
S × C90		10	2	1	1	2	1	1	0	1	19
T × C34		1		0	0	0	2	0	0	0	3
Z × C05	1	7	1	2	1	2	3	0	0	0	16
Z × C34		2	0	1	0	0	1	0	1	0	5
Total	4	141	28	18	9	10	11	3	5	7	236

Pollen fertility ranged from 0 to 99%. Among the derivative hybrid plants, 59% were tetraploids, 75% of which had pollen fertility over 90%. Among the derivative hybrid plants, 96% had reached maturity and 50% had set seeds (1–100% seed set). Only 18% had a seed set over 50%. Hybrids with the tetraploid chromosome number represented 79%, and 20% among them set seeds over 50%.

Analysis of variance for quantitative parameters (2n, pollen fertility, seed set, and number of awns per spikelet) within and between groups of combinations revealed that no significant differences in the chromosome number (2n), pollen fertility (PF) or seed set (SS) was detected within the groups. However, a highly significant difference between groups of combinations for 2n was revealed. Derivative hybrids were significantly different in the number of awns per spikelet (Nb. Awns/Sp). As for the seed set, no significant difference within and between groups was detected (Tables 2, 3). Lemma pubescence, number of awns and awn development deviated significantly from the expected 3:1 ratio in most of the combinations (Table 4).

Analysis of segregation in spikelet characters for all hybrid derivatives within each combination revealed that lemma hairiness and lemma color were significantly and negatively correlated (P < 0.001) as for the number of awns per spikelet and awn development. No correlation was detected between lemma hairiness and awn development or between the number of awns and lemma hairiness (Table 5).

 $\frac{F_2}{Eighty}$ one hybrid plants had chromosome numbers varying from 28 to 29. Among them, 58% were tetraploids (2n = 4x = 28) with pollen fertility exceeding 85%. However, 27 hybrid plants had a set seed ranging from 20 to 80%. Selected hybrid plants were subjected to pedigree selection in the field until reaching genetic stability in the 8th genera-

Таble 2. Analysis of variance within and between groups of combinations for quantitative parametersТаблица 2. Дисперсионный анализ внутри и между группами комбинацийпо количественным параметрам

		Р					
Groups of combinations	DF	2n	Nb. Awns/Sp.	SS	PF		
А	3	0.108 n.s	0.814 n.s	0.770 n.s	0.272 n.s		
S	4	0.436 n.s	0.023*	0.854 n.s	0.486 n.s		
Ζ	1	0.828 n.s	0.934 n.s	0.358 n.s	0.486 n.s		
Global	12	0.005**	0.040*	0.369 n.s	0.040*		

Note: *** Significant at the 0.05, 0.01 probability level.

Nb. Awns/Sp.: number of awns/spikelet; SS: seed set; PF: pollen fertility.

A = (A × B91, A × C90, A × C05, A × C21); S = (S × B55, S × B91, S × C90, S × C05, S × C21); Z = (Z × C05, Z × C34); Global = (all 4th hybridization cycle combinations)

Table 3. Statistical parameters for quantitative variables within and between combinations Таблица 3. Статистические параметры количественных переменных внутри и между комбинациями

Variables	Mean + SE						
	Α	S	Z	Global			
2n	29.202 ± 0.311	29.757 ± 0.230	30.928 ± 0.610	29.676 ± 0.169			
Nb. Awns	0.910 ± 0.034	0.821 ± 0.042	0.821 ± 0.073	0.867 ± 0.023			
SS	14.910 ± 2.232	15.264 ± 2.173	11.965 ± 3.176	15.664 ± 1.363			
PF	76.815 ± 3.874	72.526 ± 3.604	55.00 ± 8.238	74.347 ± 2.296			

Note: Nb. Awn/Sp.: number of awns/spikelet; SS: seed set; PF: pollen fertility.

A = (A × B91, A × C90, A × C05, A × C21); S = (S × B55, S × B91, S × C90, S × C05, S × C21); Z = (Z × C05, Z × C34); Global = (all 4th hybridization cycle combinations)

Global analysis of the Pearson correlation coefficient for 2n, compared to the other quantitative parameters for all combinations, revealed that 2n was negatively correlated to PF (P < 0.01, r = -1877^{**}), SS (P < 0.01, r = -0.1878^{**}) and to the number of awns / Sp. (P < 0.001, P = 0.0003, r = -0.2406^{***}).

Morphological characterization of the derivative hybrid seeds revealed that 32% of the derivative seeds were hairless, and 75% had a yellowish lemma as their domesticated parents; 14% of the hybrids were awnless; the rest of the plants had mainly one awn per spikelet. No spikelet shedding was observed for the derivative hybrids. However, lemma color segregated as a single gene in all combinations.

tion. Selected lines showed good agronomic performance and therefore were assessed for groat protein content.

Groat protein content in domesticated A. magna lines

Eight selected domesticated lines of the tetraploid species *A. magna*, having good agronomic performance were grown in the field at the INRA experimental farm in Marchouch. Oat grain quality is determined by groat protein and oil contents (Welch and al., 2000). Groat protein content is more often ranked as the most important trait among grain constituents because of its high nutritional value (Doehlert et al., 2001). Therefore, domesticated lines were analyzed for this trait using the Kjeldahl method and com-

Table 4. Segregation of four spikelet morphological characters in the 4th hybridization cycle of domesticated Avena magna (KH2, 3:1)

Таблица 4. Расщепление по четырем морфологическим признакам колоска у BC4 одомашненных растений Avena magna (KH2, 3: 1)

Combinations	Lemm	a color	Lemma	pubescence	Nb. Awn	s/Spikelet	Awn development		
	Yellow	Dark	Glabrous	Pubescence	0	1	0_1	2_3	
S × B91	32	10	17	25	8	34	1	0	
KH2/ p_value	0,016	0,899	10,258	0,001	26,391	< 0.0001	_	-	
		1	1					1	
A × B91	22	6	9	19	3	25	3	25	
KH2/ p_value	0,100	0,752	10,338	0,001	10,338	0,0013	23,625	< 0.0001	
S × C90	18	1	4	15	6	13	7	12	
KH2/ p_value	2,882	0,0896	11,076	0,001	7,1956	0,0073	5,6116	0,0178	
		·					·		
A × C90	21	5	6	20	2	24	2	24	
KH2/ p_value	0,251	0,616	14,024	0,000	24,285	< 0.0001	24,285	< 0.0001	
S × C05	13	6	7	12	2	17	7	12	
KH2/ p_value	0,203	0,653	5,612	0,018	16,134	< 0.0001	5,6116	0,0178	
A × C05	5	2	2	5	0	7	0	3	
KH2/ p_value	0,023	0,880	3,022	0,082	8,4	0,0038	3,6	0,0578	
Z × C05	13	9	9	13	4	18	7	15	
KH2/ p_value	1,260	0,262	5,246	0,022	14,271	0,0002	8,2429	0,0041	
G × C05	20	9	9	20	1	28	2	27	
KH2/ p_value	0,262	0,609	11,252	0,001	31,14	< 0.0001	27,812	< 0.0001	
S × C21	11	3	3	11	2	12	7	7	
KH2/ p_value	0,050	0,823	8,046	0,005	10,441	0,0012	1,8667	0,1719	
A × C21	5	3	3	5	1	7	2	4	
KH2/ p_value	0,291	0,590	2,286	0,131	6,3492	0,0117	2,0979	0,1475	
Z × C34	4	2	2	4	1	5	1	4	
KH2/ p_value	0,101	0,751	2,098	0,148	4,1119	0,0426	3,0326	0,0816	
T × C34	5	0	1	4	1	4	1	4	
KH2/ p_value	1,429	0,232	3,033	0,082	3,0326	0,0816	3,0326	0,0816	

Note: Nb. Awn/Sp.: number of awns/spikelet; SS: seed set; PF: pollen fertility.

 $A = (A \times B91, A \times C90, A \times C05, A \times C21); S = (S \times B55, S \times B91, S \times C90, S \times C05, S \times C21); Z = (Z \times C05, Z \times C34); Global = (all 4th hybridization cycle combinations)$

Table 5. Linkage of spikelet characteristics in Avena magna derivatives of each combinationof the 4th hybridization cycle

Таблица 5. Связь признаков колоска у гибридных растений каждой комбинации ВС₄ в потомстве Avena magna

Combination	DL	К2	Р	r		
Lhairin*Awnsdev						
A × B91	3	3.896	0.273	-0.01636 ns		
A × C05	_	_	_			
A × C21	2	0.747	0.688	-0.13012 ns		
A × C90	3	5.814	0.121	-0.14249 ns		
G × C05	3	3.834	0.280	-0.11108 ns		
S × B55	-	_	_	_		
S × B91	3	3.261	0.353	0.04440 ns		
S × C05	2	0.351	0.839	0.04275 ns		
S × C21	1	0.424	0.515	0.17408 ns		
S × C90	3	6.152	0.104	0.36378 ns		
T × C34	2	0.833	0.659	0 ns		
Z × C05	3	1.776	0.620	0.00771 ns		
Z × C34	3	3.000	0.392	0		
NbAwns* Lhairin	·					
A × B91	1	1.836	0.175	0.25608 ns		
A × C05	-	-	-	-		
A × C21	1	0.686	0.408	-0.29277 ns		
A × C90	1	0.885	0.347	0.18447 ns		
G × C05	1	2.302	0.129	0.28172		
S × B55	-	-	-	-		
S × B91	1	0.982	0.322	-0.15294 ns		
S × C05	1	1.304	0.253	-0.26197 ns		
S × C21	1	0.636	0.425	−0.21320 ns		
S × C90	1	2.030	0.154	0.32686 ns		
T × C34	1	0.313	0.576	-0.25000 ns		
Z × C05	1	0.167	0.683	0.08716 ns		
Z × C34	1	0.600	0.439	−0.31623 ns		
Lhairin* Lcolor						
A × B91	1	16.1212	<.0001	-0.75879***		
A × C05	1	7.0000	0.0082	-1.00000***		
A × C21	1	8.0000	0.0047	-1.00000***		
A × C90	1	20.6349	<.0001 -0.89087***			
G × C05	1	29.0000	<.0001	-1.00000***		
S × B55	-	-	-	-		

Table 5. The end Таблица 5. Окончание

таолица	5.	ОКОН	чание	

Combination	DL	К2	Р	r	
S × B91	1	19.3015	<.0001	-0.67791***	
S × C05	1	15.0330	0.0001	-0.88950***	
S × C21	1	14.0000	0.0002	-1.00000***	
S × C90	1	5.6296	0.0177	-0.54433**	
T × C34	-	-	_		
Z × C05	1	22.0000	<.0001	-1.00000***	
Z × C34	1	6.0000	0.0143	-1.00000***	
NbAwns*Awnsdev					
A × B91	3	28.0000	<.0001	0.61758***	
A × C05	-	-	-		
A × C21	2	8.0000	0.0183	0.66667*	
A × C90	3	13.0903	0.0044	0.37550 ns	
G × C05	3	29.0000	<.0001	0.35844	
S × B55	-	-	-		
S × B91	3	15.0611	0.0018	0.53916***	
S × C05	2	3.8319	0.1472	0.40320 ns	
S × C21	_	-	-	-	
S × C90	3	8.7794	0.0324	0.64758**	
T × C34	2	0.8333	0.6592	0	
Z × C05	3	11.9167	0.0077	0.61931**	
Z × C34	3	6.0000	0.1116	0.69561 ns	
NbAwns* Lcolor					
A × B91	1	0.9164	0.3384	0.18091 ns	
A × C05	-	-	-		
A × C21	1	0.6857	0.4076	0.29277 ns	
A × C90	1	0.5159	0.4726	0.14086 ns	
G × C05	1	2.3016	0.1292	−0.28172 ns	
S × B55	-	-	_	-	
S × B91	1	0.6968	0.4039	0.12880 ns	
S × C05	1	1.0317	0.3098	0.23302 ns	
S × C21	1	0.6364	0.4250	0.21320 ns	
S × C90	1	0.4872	0.4852	0.16013 ns	
T × C34	_	-	_	-	
Z × C05	1	0.1671	0.6827	-0.08716 ns	
Z × C34	1	0.6000	0.4386	0.31623 ns	

Note: *** Significant at the 0.05, 0.01 probability level

pared to their hexaploid *A. sativa* parents: 'Soualem', 'Ghali', 'Amlal' and 'Zahri'. Some lines revealed a groat protein content exceeding by 2 to 3% that of both hexaploid and tetraploid parents (Table 6). The fact that the groat protein content was not as high as expected may have been due to the low level of protein content in the employed *A. sativa* cultivars, which are normally used for forage.

Table 6. Groat protein content in hexaploid parents and the derivative tetraploid Avena magna

Таблица 6. Содержание белка в зерне гексаплоидных родительских форм и тетраплоидных гибридов Avena magna

Species	Groat protein (%)	
A. sativa	Amlal	13
	Ghali	14
	Soualem	10
	Zahri	11
A. magna	P 1-6	11
	mag. Dom. 17	12
	mag dom. 49	12
	mag dom. 61	12
	mag. Dom. 66	13
	mag. Dom. 68	13
	mag. Dom. 76	12
	mag. Dom. 78	12
	mag. Dom. 81	13

Conclusion

Recently, oat has attracted more attention as a crop used for human consumption, and therefore, the release of new lines with a high nutritive value becomes of great importance. The exploitation of wild genetic resources is very beneficial, since this reservoir includes valuable genes. This material has served to enrich the genepool of the cultivated forms through interspecific crosses or to domesticate wild species. Therefore, the coupling of high productivity with valuable traits has been the main objective of any breeding program; it may lead to an increase in the percentage of qualitative kernel components (Loskutov, 2004). Hence, many attempts have been undertaken to domesticate some taxa in order to achieve this goal. Ladizinsky (1995) succeeded in domesticating two wild tetraploid oat species A. magna and A. murphyi Ladiz. by transferring into them the domesticated syndrome of the cultivated oat through hybridization. The objective of this study was to fully domesticate the developed lines by introducing into them more domesticated characters of Moroccan common oats cultivars. Because it is very difficult to domesticate wild species due to their wild characters, especially shattering, lemma hairiness and presence of awns, we performed many hybridization cycles, up to four (2 backcrosses with A. sativa and 2 backcrosses with tetraploid parents), in which common oat was used as the female parent. When a plant breeder contemplates hybridization, cytogenetic studies can be of great assistance in a number of ways (Love et al., 1947).

In the last hybridization cycle (4th crossing cycle), we managed to harvest the BC4 seeds, which were analyzed for their chromosome number, pollen stainability, seed set, and seed morphological characterization. Analysis of correlations between the chromosome number and the other quantitative characters confirmed the results reported by Hugh et al. (1980), that no correlation exists between 2n and PF. Morphological characterization of derivative hybrid seeds showed that lemma color segregated in all combinations as a single gene, while the other characters deviated significantly from the 3:1 ratio for most of the combinations, thus confirming what was demonstrated by Marshall and Shanner (1992), that the four spikelet characteristics are linked to one another in the common oat. This has been contradicted by Ladizinsky (1995) who reported that it does exist in *A. magna*.

According to the results of the chemical analysis undertaken by Welch et al. (2000) for different oat accessions collected from different countries, the ones originating from Morocco presented the highest groat protein content of 29,5% for A. magna. For that reason, Ladizinsky (1995) suggested that this trait may be one of the main features to domesticate A. magna. This was replied by the experiment we undertook, and hence we succeeded in domesticating 8 lines. However, our goal was to develop domesticated tetraploid lines with high nutritive value and improved groat protein content, exceeding that of their hexaploid parents. Analysis of the derived material revealed that this trait was improved by 2 to 3% in comparison to the hexaploid parent. This was due to the lower value of protein content in the common oat cultivars used in the crosses, since these cultivars were originally intended for forage use, in addition to the dilution of the responsible gene due to the large number of the achieved hybridization cycles.

The domestication goal was achieved, but the choice of the hexaploid cultivars with high groat protein content has to be considered for future crossing. Furthermore, an advanced food technology analysis should be carried out to assess the real nutritive value of the developed tetraploid lines.

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