

IDENTIFICATION OF THE DIVERSITY OF CULTIVATED PLANTS AND THEIR WILD RELATIVES FOR SOLVING FUNDAMENTAL AND APPLIED PROBLEMS

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Release of two hexaploid oat (*Avena sativa* L.) cultivars with high nutritive value of groat for human consumption: an innovation at the Moroccan national level

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Moroccan oat breeding has traditionally targeted forage yield, yet the wild tetraploid oat species *Avena murphyi* Ladiz. possesses protein- and β -glucan-enhancing alleles largely absent from cultivated germplasm.

The two decades of research, exploiting the wild tetraploid oat species *A. murphyi* through backcross introgression led to the release of the first Moroccan improved hexaploid oats cultivars, 'Al Fawze' and 'Abtah', developed for human consumption.

Peer-reviewed papers, institutional reports and breeder datasets were screened for local *Avena* L. diversity and conservation, interspecific hybridization procedures, multi-environment performance under rainfed Mediterranean conditions, and nutritional and techno-functional traits relevant to food, feed and industrial use.

Introgressed lines achieved 17% groat protein content, 5% groat β -glucan content and 38 g thousand-kernel weight while sustaining 34–43 q ha⁻¹ grain yield in addition to broad resistance to crown rust, powdery mildew, and BYDV. A 20% substitution of bread-wheat flour with 'Al Fawze' flour increased loaf protein by 14% without altering dough rheology, signaling immediate value for bakery and cereal products.

Leveraging endemic wild germplasm fast-tracked the creation of climate-adapted, nutrient-dense oats for human consumption. Next steps include certified-seed scale-up, processor partnerships, and consumer-focused validation of sensory quality, sustainability, and health benefits.

Keywords: forage crop, *Avena murphyi*, hybridization, bioactive compounds, β -glucans, groat proteins, indigenous genetic resources, wild species

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ИДЕНТИФИКАЦИЯ ГЕНЕТИЧЕСКОГО РАЗНООБРАЗИЯ КУЛЬТУРНЫХ РАСТЕНИЙ И ИХ ДИКИХ РОДИЧЕЙ ДЛЯ РЕШЕНИЯ ФУНДАМЕНТАЛЬНЫХ И ПРИКЛАДНЫХ ПРОБЛЕМ

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Создание двух гексаплоидных сортов овса (*Avena sativa* L.) пищевого назначения с высокой питательной ценностью крупы: национальная инновация Марокко

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Селекция марокканского овса традиционно нацелена на урожайность кормового зерна, однако дикий тетраплоидный вид овса *Avena murphyi* Ladiz. обладает аллелями, повышающими содержание белка и β-глюканов в зерне, которое значительно превосходит показатели возделываемых сортов.

Два десятилетия изучения интрогрессии возвратного скрещивания с использованием популяций дикого тетраплоидного вида овса *A. murphyi* привели к созданию первых марокканских улучшенных гексаплоидных сортов овса 'Al Fawze' и 'Abtah' пищевого назначения.

Рецензируемые статьи, научные отчеты и базы данных селекционеров проанализировали на предмет местного разнообразия и сохранения генетических ресурсов рода *Avena* L. Провели межвидовые скрещивания и полевые испытания гибридов в нескольких почвенно-климатических зонах Средиземноморья с богатым земледелием, изучили биохимические и технологические признаки, значимые для пищевого, кормового и технического направлений использования культуры.

Крупа интрогрессивных линий овса содержала до 17% белка и до 5% β-глюканов, масса 1000 зерновок достигала 38 г, урожайность зерна сохранялась на уровне 34–43 ц/га, выявлена комплексная устойчивость к корончатой ржавчине, мучнистой росе и вирусу желтой карликовости ячменя. При замещении 20% муки мягкой пшеницы мукой сорта овса 'Al Fawze' содержание белка в полученном хлебе составляло 14% без изменения реологии теста, что свидетельствует о повышении качества хлебобулочных и других пищевых изделий из этой муки.

Использование эндемичной дикой зародышевой плазмы ускорило создание хорошо адаптированных, богатых питательными веществами новых сортов овса для пищевого использования. Следующими шагами станут масштабное увеличение производства сертифицированных семян, заключение партнерских соглашений с переработчиками, проверка вкусовых качеств продукции, ее экологической чистоты и пользы для здоровья человека.

Ключевые слова: кормовая культура, *Avena murphyi*, гибридизация, биоактивные вещества, β-глюканы, содержание белка, местные генетические ресурсы, дикие виды

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Introduction

Global and national production context

Agriculture remains central to livelihoods and development in many regions, and cereals underpin human and animal nutrition because of their energy density and versatility. Within this group, oats have attracted renewed attention for both resilience and food-grade value. According to FAOSTAT, oat ranks as the seventh most economically important cereal worldwide, after maize, rice, wheat, barley, sorghum, and millet. In 2023, global oat production was projected to decline markedly to ~23.1 million tons, the lowest in 11 years, mainly due to reduced planted areas and yields in Canada, the European Union, and the United States. Over the same horizon, total coarse grains were expected at ~1,511 million tons, with maize at a record ~1,215 million tons; total cereals near ~2,815 million tons (<https://www.fao.org/worldfoodsituation/csd/en>).

In Morocco, FAOSTAT time series also show fluctuations: ~15,009 ha and ~5,551 t in 2020; 17,845 ha and 6,390 t in 2021; 13,040 ha and 3,740 t in 2022, reflecting the rainfall dependence of rainfed systems (<https://www.fao.org/faostat/en/#data/QCL>).

Importance and utilization of oats

Oats have long been important as livestock feed (grain, green fodder, silage, and hay) and are valued for their high fodder nutritional value and dry matter digestibility exceeding 75%. Oat straw also exhibits more digestible organic matter than other cereal straws, which is appreciated by livestock (Stevens et al., 2004). In parallel, oats remain an important grain for populations in marginal areas in emerging countries. Oat seeds combine proteins of high nutritional value, oils, and health-promoting constituents such as avenanthramides and tocopherols (Peterson, Mannerstedt Fogelfors, 2004; Alrahmany, Tsopmo, 2012). Classic work highlighted that oat groat proteins have a favorable amino acid balance that remains stable even when total protein increases through genetic progress (Clamot, 1979; Welch et al., 2000). Higher groat oil (unsaturated fatty acids) increases energy density and has potential for industrial extraction (e.g., biofuel). (1→3),(1→4)- β -D-glucans are a major component implicated in lowering plasma cholesterol; groat protein and oil remain key grain quality traits (Zhu et al., 2004). Medical studies favoring whole-grain fiber intake to reduce blood cholesterol and intestinal/colon cancer risk underpinned the 1990s resurgence of interest in oats for human health (Harlan, 1992). Bran and caryopsis residues also enable furfural production for solvent and pharmaceutical applications (Harlan, 1992; Saidi, 2015).

Oat cultivation in Morocco

Oats are grown as a winter crop in favorable and intermediate rainfed regions, either as a pure stand or mixed with vetch (notably in the Saïs), for hay, green fodder, and grain. Oats contribute significantly to dairy systems in northwestern Morocco, while barley dominates in the middle and lower semi-arid zones; fodder yield depends on the cutting stage and cutting $\times$ genotype $\times$ environment interactions (Al Faiz et al., 1997). Cultivated area has often exceeded 100,000 ha (~70,000 ha pure, and 50,000 ha vetch-oat), making oats the third fodder crop after alfalfa and barley and contributing ~36% of total fodder production (Al Faiz et al., 2004; Saidi, Al Faiz, 2008). The 2020–2022 FAOSTAT figures cited above confirm the recent volatility in area and output (<https://www.fao.org/faostat/en/#data/QCL>).

Oat genetic resources in Morocco

Morocco is a center of diversification for *Avena*, with ~30 wild species (Saidi, Ladizinsky, 2005). Endemic taxa include *A. atlantica* B.R. Baum. & Fedak. ($2n = 2x = 14$), *A. agadiriana* B.R. Baum. & Fedak. and *A. magna* Murphy & Terrell ($2n = 4x = 28$), while *A. murphyi* Ladiz. ($2n = 4x = 28$) and *A. prostrata* Ladiz. ($2n = 2x = 14$) are found in limited northern regions (Leggett, 1992). Although hexaploid cultivated oat (*A. sativa* L.) is important for feed and food, limited efforts have historically targeted conservation and use of wild relatives to enrich the cultivated gene pool narrowed by selection (Leggett, 1992). From the mid-1960s, breeders undertook introgression from wild oats to improve groat nutritive value, leading to protein-rich cultivars (Moser, Frey, 1994). Foundational cytogenetic studies clarified affinities among wild and cultivated *Avena*, suggesting tetraploids (e.g., *A. murphyi*) underpinned hexaploid evolution (Ladizinsky, 1969; Thomas, Bhatti, 1975). Tetraploids with AADD genomes share morphological traits with AACCCD hexaploids, facilitating hybridization owing to chromosome similarity (Rajhathy, Sadasivaiah, 1969; Thomas et al., 1980). This rationale supported interspecific crosses to transfer desirable alleles from wild to cultivated backgrounds (Ladizinsky, Johnson, 1972; Ladizinsky, Fainstein, 1977a; Loskutov, 2001b; Loskutov, Rines, 2011; Manzali et al., 2023, 2025).

Why broadening the genetic base of *A. sativa* matters now?

Modern hexaploid oats have a relatively narrow genetic base, constraining progress in agronomic performance (yield stability, disease resistance) and techno-functional quality (kernel geometry, groat recovery, protein, oil, β -glucans) under rainfed Mediterranean conditions. In contrast, the tetraploids *A. magna* and *A. murphyi* endemic and edapho-climatically adapted to Morocco carry very high groat protein (up to ~30%), elevated oil, large kernels (1000-kernel weight $\approx$ 35 g), strong tillering, and resistance to powdery mildew and crown rust (Ladizinsky, Fainstein, 1977a; Leggett, 1992; Loskutov, 2001a). Their strong meiotic affinity with cultivated oats (AADD $\leftrightarrow$ AACCCD) enables efficient allele transfer; the homoeology of major loci for groat protein further favors backcrossing (Rajhathy, Sadasivaiah, 1969; Thomas, Bhatti, 1975; Marshall et al., 1992; Welch et al., 2000; Zhu et al., 2004).

Moroccan breeding program redirected toward food-grade oats

Since the early 1980s, INRA-Morocco has released hexaploid cultivars adapted to contrasting agroclimates (Al Faiz, 1990). The first-wave 21 hulled and two naked oats, derived from temperate North-American introductions, focused on forage biomass and rust tolerance. However, shared pedigrees eroded resistance durability (crown rust, BYDV, smut) and limited physiological plasticity under recurrent drought. Anticipated climate-change pressures in North Africa reinforce the need for novel alleles to stabilize yield while raising grain quality endpoints. Morocco's secondary diversification center provides such variability, notably tetraploids *A. murphyi* and *A. magna* (Leggett, 1992; Saidi, Ladizinsky, 2005). Building on this evidence, INRA redirected its program in the 2000s toward food-grade oats. One pipeline implemented interspecific introgression via repeated backcrosses between the Moroccan hexaploid cultivar 'Amlal' and a wild *A. murphyi* accession from northern Morocco, followed by pedigree selection to genetic stability (Saidi et al., 2016). Subsequent multilocation trials under rainfed and irrigated conditions

have evaluated advanced lines for agronomy and food-relevant grain quality (Saidi et al., 2020, 2025).

Study gap, hypothesis, and objectives

Despite strong biological and historical reasons to leverage Moroccan tetraploids for broadening *A. sativa*, there is still a shortage of integrated evidence linking introgression to the combined response of (i) agronomic performance (yield, disease resistance) and (ii) techno-functional grain traits (thousand-kernel weight, groat recovery, groat protein, β -glucans) under rainfed Mediterranean conditions. This study reports the development and multi-environment testing of two hexaploid oat lines derived from repeated backcrosses between 'Amlal' and *A. murphyi*, documents ploidy stabilization and phenotypic segregation through eight generations of pedigree selection, and evaluates agronomic performance and grain quality gains relative to commercial checks, with an emphasis on food-grade applications. We hypothesized that introgression from *A. murphyi* would increase the mean and variance for targeted quality and processing traits while maintaining or improving yield and disease resistance, thereby broadening the effective genetic base of *A. sativa* for Mediterranean breeding programs (Ladizinsky, Fainstein, 1977a; Loskutov, 2001a; Welch et al., 2000; Zhu et al., 2004; Saidi et al., 2016, 2020, 2025).

Material and methods

Plant material

The study used two parental materials. The wild tetraploid donor was *Avena murphyi* accession P45-55 ($2n = 4x = 28$) collected in the Tangier region, described with 23.7 g 100 g⁻¹ DM of groat protein and tolerance to BYDV. The recurrent parent was *A. sativa* cv. 'Amlal' ($2n = 6x = 42$), registered in 1996 in the Moroccan national catalogue; semi-early, adapted to rainfed zones of northwestern Morocco, with typical yield/quality traits as stated in the original manuscript. Program descendants used as benchmarks in advanced trials: 'Amlal' (conventional control) and the released introgression descendants 'Al Fawze' and 'Abtah' from the same pipeline.

Methods

Interspecific crosses and backcrossing

The first crossing cycle was realized under greenhouse conditions in the early 2000s. The hexaploid parent *A. sativa* cv. 'Amlal' ($2n = 6x = 42$), used as a female parent after manual emasculation, was manually pollinated by the pollen collected from the tetraploid parent plants of *A. murphyi* (P45-55) (Fig. 1). Collected hybrid seeds were backcrossed to their hexaploid parent in the field, at the INRA-Morocco Experiment Station in Marchouch, located 65 km southeast of Rabat (longitude 6°71' 600 W, latitude 33°60' 499 N, 410 masl).

Ploidy determination

Ploidy of the hybrid seeds derivative from the undertaken crosses was determined using the conventional method in order to select hybrids having the hexaploid ploidy level ($2n = 6x = 42$). The determination of chromosome number was checked in root tips of the pregerminated hybrid seeds in Petri dishes after being treated according to R. J. Singh's methodology (2016). Chromosomes were visualized and counted using an optical microscope.

Morphological characterization

In order to analyze the segregation of the spikelet phenotypic characters of the hexaploid hybrid seeds, the progeny was characterized for wild morphological characters such as absence/presence of awns, awn vigor, seed color, and lemma hairiness.

Pedigree selection

F₂ progenies were grown at INRA-Marchouch (33° 47' N, 06° 71' W) and advanced to F₈ by pedigree selection to reach genetic stability prior to quantitative evaluation.

Crop establishment and site conditions

At the Marchouch Experiment Station, average annual precipitation during the seed-increase period was ≈ 374 mm (mean of 571, 296 and 255 mm reported by the station), with a mean annual temperature $\approx 23.79^\circ\text{C}$; soils are described as rich black, crumbling and well-drained. Plots at the station were managed in RCB, 4 rows $\times$ 1 m (4 m²), 3 replications, with manual weeding and no chemical herbicides or N fertilizer applied.

Field establishment for the performance trials followed the national guidance for oats: late October to late November sowing, row spacing 15–20 cm, depth 2–5 cm, and post-sowing rolling when needed (rate adjusted to thousand-kernel weight), with trials conducted under rainfed conditions.

Assessment of agronomic performance

Preyield (behavior) trials

Location and design. Eighth-generation, true-breeding progenies were grown for two consecutive seasons (2020/21 and 2021/22) at the INRA-Marchouch Experiment Station (33°47' N, 06°71' W, 410 masl) in a randomized complete-block design (RCBD) with four replications, following the layout principles of K. A. Gomez and A. A. Gomez (1984). Each plot contained six 5 m rows spaced 0.20 m apart (6 m²).

Traits recorded. Days to heading and physiological maturity were noted when 50 % of spikes reached the target stage; early vigor was rated 1–9 at tillering (Oelke et al., 1992). Plant height and lodging (%) were measured at the milk stage. Grain yield was obtained by combine-harvesting the central four rows, cleaning, and adjusting to 120 g kg⁻¹ seed moisture (International Seed Testing Association, 2018). Aboveground biomass was cut at the milk stage, oven-dried (65°C, 48 h), and expressed as dry-matter yield (t ha⁻¹).

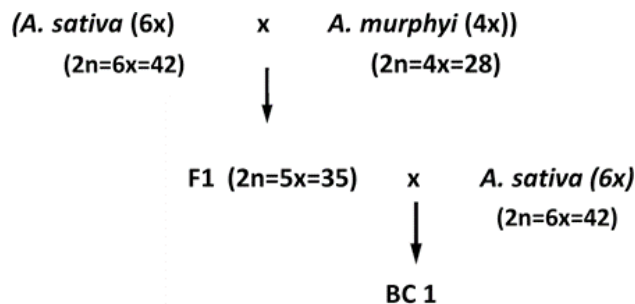


Fig. 1. Crossing pattern

Рис. 2. Схема скрещивания

Advanced multilocation trials

Sites and seasons. The three best preyield performers plus two commercial checks ('Amlal' and 'Al Fawze') were evaluated for three seasons (2022/23–2024/25) at five rainfed sites: Marchouch, Meknes, Tassaout, Tangier, and Sidi Allal Tazi (300–650 mm of annual rainfall) representing Morocco's principal oat zones (Al Faiz et al., 1997).

Experimental layout. Trials followed an RCBD with four replications; plot size and management matched the preyield stage. Basal fertilizer (60 kg N + 60 kg P₂O₅ ha⁻¹) was applied according to the national oat guidelines (ONSSA, 2020); weeds were hoed manually to avoid herbicide × genotype interactions.

Disease and pest assessment.

Crown rust (*Puccinia coronata* f. sp. *avenae* P. Syd. & Syd.) and powdery mildew (*Blumeria graminis* (DC) Speer) were rated twice (booting and soft-dough) on the modified Saari-Prescott 0–100 severity scale; on each date, severity (%) was estimated on 20 tillers per plot taken from the central rows (edge rows excluded) to avoid border effects. No foliar fungicides or insecticides were applied, and infection relied on natural field pressure (Saari, Prescott, 1975).

Barley yellow dwarf virus (BYDV) symptoms were scored at late tillering on a 0–5 ordinal scale where 0 = no reddening, and 5 = severe stunting with leaf reddening; scores were recorded on 20 tillers per plot from central rows (Comeau, Haber, 1986).

Insect pests. Natural infestations of oat aphid (*Rhopalosiphum padi* L.) and armyworm (*Mythimna unipuncta* Haworth) were monitored weekly from stem elongation to early grain fill on 10 randomly selected tillers per plot (central rows). Resistance was indexed as the area under the pest progress curve (AUPPC), computed by the trapezoidal method from consecutive weekly counts as described by M. J. Jeger and S. L. H. Viljanen-Rollinson (2001); higher AUPPC indicates greater pest pressure. (Jeger, Viljanen-Rollinson, 2001)

Assessment of grain quality parameters

Weight of one thousand seeds (WTS)

The weight of one thousand seeds was determined according to the ISTA rules (International Seed Testing Association, 1985). A sample of 1000 seeds per line in three replications was weighted. The WTS is the average of the weights of the three samples expressed in grams.

Determination of groat and husk proportions

Groat and husk proportions were calculated according to the methodology described by (Doehlert et al., 1999) and (Hall et al., 2003). This method consists of counting and weighing 100 grains in three replications per line and carrying out the husking and weighing of groat and husks separately. Proportions (%) are calculated by:

$$(\text{Groat or husk weight} / 100\text{-seed weight}) / 100$$

Determination of groat protein content

Whole oat groats were first ground in an IKA MF-10 basic impact mill equipped with a 0.5 mm stainless-steel screen to obtain a uniform flour. Crude protein was then measured by the Kjeldahl procedure (AOAC 979.09): after digestion, distillation and capture of the liberated ammonia in boric acid, the distillate was back-titrated with 0.1 N HCl. Protein content on a dry-matter basis was calculated as:

$$\text{Protein (\%DM)} = \frac{(V - V_0) \times 1000 \times N \times 14 \times F \times 100}{P \times \text{DM\%}},$$

where V and V₀ are the titrant volumes for sample and blank (mL); N is acid normality (0.1); 14 is the atomic mass of

nitrogen; F is the nitrogen-to-protein factor (6.25 for oat flour, or 5.7 for wheat flour); P is the test portion (g); DM % is the sample dry-matter content.

Determination of groat β-glucan content

The content of soluble β-glucans in groat was measured through an enzymatic method according to AACC (32-23), AOAC (995-16), and ICC (168), using a kit of mixed β-glucans (Megazyme International Ltd., Wicklow, Ireland) suggested by B. V. McCleary et al. (2006).

Analysis of the nutritional quality and preliminary tests of flour mixtures' rheological behavior

Mixtures of flours were made from bread wheat cv. 'Kenz' and different proportions of hexaploid oat flour from cv. 'Al Fawze', selected from the hybrid lines resulting from the interspecific crosses realized between hexaploid oat cv. 'Amlal' and the wild tetraploid oat accession of *A. murphyi* P45-55, selected for its high groat nutritional value, registered in the INRA–Morocco official catalogue, and used here as an initiative test for industrial uses. All flour mixtures were analyzed for the ability to form bread with good technological and nutritional characteristics. Mixtures with different oat incorporation proportions of flour in wheat flour at the rates of 10%, 15%, 20%, 25% and 30% were baked and subjected to gastronomic tests.

Dough formulation (baker's % on total flour): 1% of salt, 1.5% of baker's yeast, and 53% of water.

The sedimentation index of wheat-oat composite flours was measured using the Zeleny sedimentation test following AACCI Method 56-61.02 and ICC Standard 116/1; results were expressed in milliliters, with readings corrected for flour moisture as specified in the method. Triplicate determinations were performed per formulation.

Loaf volume was determined within 30 min of baking by the rapeseed displacement method (AACCI 10-05.01); each value is the mean of at least three loaves per treatment. Specific volume was calculated as loaf volume divided by loaf weight (cm³ g⁻¹), and crumb density as loaf weight divided by loaf volume (g cm⁻³).

Registration in the Moroccan official catalogue

The registration in the INRA–Morocco official catalogue of the selected lines as new cultivars was done according to UPOV standards of distinction, homogeneity and stability (DHS), and agronomic and technological value tests (ATV).

Chemicals and reagents

All chemicals and reagents (acids, bases, salts, buffers, and organic solvents used for sample preparation, Kjeldahl digestion/distillation, and β-glucan extraction) were analytical grade and purchased from Sigma-Aldrich/Merck (Merck KGaA, Darmstadt, Germany; distributed by Sigma-Aldrich, St. Louis, MO, USA). The mixed-linkage β-glucan assay kit was obtained from Megazyme (Wicklow, Ireland).

Statistical analysis

The obtained results were statistically processed with SAS 9.3 (SAS Institute, Cary NC), using ANOVA analysis of variance (Proc Glimprocedure) and a posteriori multiple comparison of means by Duncan's test. *P* < 0.05 values were considered as significant.

Results and discussion

Interspecific crosses

The first crossing cycle generated pentaploid hybrids (2n = 5x = 35) with a very low fertility rate. In order to restore their fertility, pentaploid hybrids were backcrossed to their hexaploid parent and 30 hybrids were thus obtained.

Ploidy analysis of hybrid seeds

Ploidy examination of the backcrossed hybrid seeds revealed that their chromosome number varied from 34 to 45; with 72% of the hybrids having a hexaploid ploidy level. This confirms that most fertile female gametes of the first hybridization cycle had 20 to 21 chromosomes. In addition, analysis of pollen fertility showed that 28% hybrid plants had pollen fertility above 50%. We noticed that even plants with an aneuploid chromosome number had good fertility, which translated into good grain yield (Fig. 2). The study of the correlation of pollen fertility to the other parameters, using the Pearson coefficient, showed that the correlation of pollen fertility with chromosome number was not significant ($r = 0.19$, $P < 0.05$), but on the other hand it was highly significant with grain yield ($r = 0.61^{***}$, $P < 0.001$). According to G. Ladizinsky and R. Fainstein (1977a), the number of chromosomes does not have a very influencing effect on the viability of the gametes, and it is rather their constitution and their compatibility that are the most important.

other and this correlation is influenced by the effect of the genotype. Thus, each morphological character is controlled by a single gene, but these genes are closely linked to each other (Ladizinsky, Fainstein, 1977a).

Selection

Derivative hybrids from the interspecific cross (*A. sativa* × *A. murphyi*) × *A. sativa* were subjected to field pedigree selection during eight years, starting from the F_2 generation to the F_8 generation. Among the thirty tested lines, ten achieved genetic stability and showed good resistance/tolerance to diseases, such as powdery mildew, *Helminthosporium*, crown rust, and barley yellow dwarf virus.

Assessment of agronomic performance

The ten selected lines were tested for agronomic performance during two years in the field (Fig. 4). The annual rainfall for the two growing seasons was 530 and 570 mm, respectively. Plant vigor of the assessed lines was similar or even superior to that of the hexaploid parent 'Amlal'. The height of hybrid plants exceeded that of their parent 'Amlal' (145 cm) by

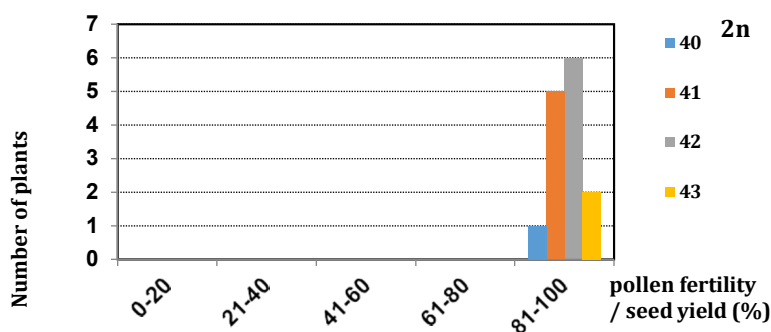


Fig. 2. Distribution of BC_1 plants from [*A. sativa* (cv. 'Amlal') × *A. murphyi* P45-55] × *A. sativa* by chromosome number and by classes of pollen fertility and seed yield per plant

Рис. 2. Распределение растений BC_1 от [*A. sativa* (cv. 'Amlal') × *A. murphyi* P45-55] × *A. sativa* по числу хромосом и по классам фертильности пыльцы и урожайности семян на растение

Morphological characterization of hybrid seeds

Morphological characterization of the backcrossed seeds showed the existence of great morphological variability within the hybrid seeds, due to the segregation of their morphological characters (Fig. 3). Analysis of the resulting quantitative variables using the independence χ^2 test highlighted the existence of a highly significant effect of the genotype on lemma hairiness ($\chi^2 = 73.75^{**}$) and awn vigor ($\chi^2 = 92.01^{**}$). As for the number of awns per spikelet, the genotype had a non-significant effect on this character ($\chi^2 = 12.55$ ns). In general, morphological characters are correlated with each

2 to 6 cm. The average grain yields of the hybrid lines over the two years were 34.8 q ha^{-1} and 35.10 q ha^{-1} , respectively. On average, the lines' grain yield by 3 to 12 q ha^{-1} exceeded that of the hexaploid parent 'Amlal' (31 q ha^{-1}). Therefore, the hybridization work allowed the derivative lines to have a grain yield gain of 9.68% to 38.7% (see Fig. 5). The average dry matter yields of the hybrid lines were 9.8 t DM/ha and 10.2 t DM/ha respectively, exceeding that of cv. 'Amlal' (10 t DM/ha) (Fig. 6). According to the obtained results, four hybrid lines, F09-01 (genotype A07-13), F09-02 (genotype A07-20), F09-03 (genotype A07-32), and F09-04 (genotype



Fig. 3. Morphological characterization of hybrid seeds' spikelets

Рис. 3. Морфологическая характеристика колосков гибридных зерен



Fig. 4. Assessment of agronomic performance of derivative hybrid lines in the field
Рис. 4. Оценка агрономических показателей гибридных линий в полевых условиях

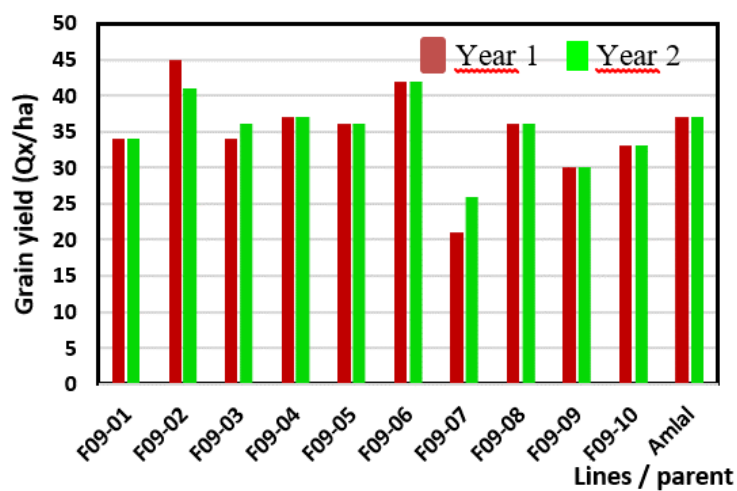


Fig. 5. Hybrid line grain yield assessed over two years at the Marchouch Experiment Station
Рис. 5. Урожайность зерна гибридных линий, оцененная за два года на Маршущской опытной станции

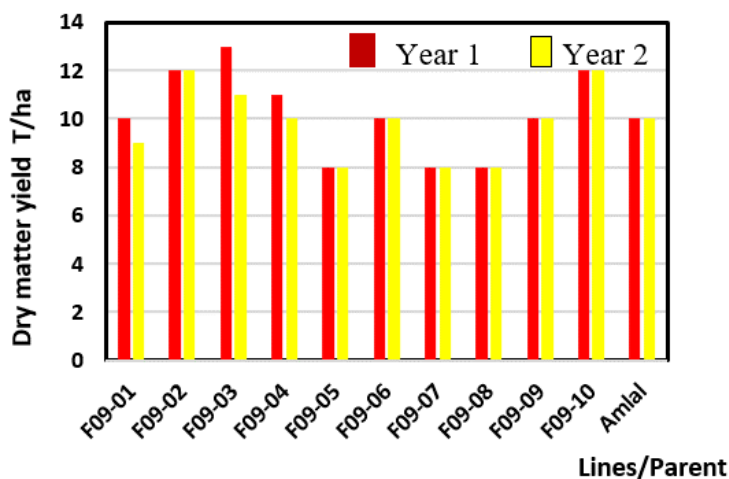


Fig. 6. Hybrid line dry matter yield assessed over two years at the Marchouch Experiment Station
Рис. 6. Урожайность сухой массы гибридных линий, оцененная за два года на Маршущской опытной станции

A07-36), were selected for further assessment for nutritional and technological quality traits.

Assessment of the hybrid lines' technological quality

Weight of one thousand seeds (WTS) of hybrid lines

The hybrid lines varied in WTS from 36 to 39 g, exceeding their hexaploid parent *A. sativa* cv. 'Amlal' (33.33 g) by 2 to 5 g. Thus, this trait was improved through interspecific crosses to the tetraploid parent *A. murphyi* (P45-55).

Groat and husk proportions in hybrid lines

Analysis of groat proportions for the hybrid lines revealed that it ranged from 68% to 74%, higher than that of both the tetraploid parent *A. murphyi* P45-55 (58%) and the hexaploid parent 'Amlal' (66%) (see Fig. 7). Husk proportions of the derived lines varied between 26% and 32%, lower than in both the tetraploid parent *A. murphyi* P45-55 (42%) and the hexaploid parent *A. sativa* cv. 'Amlal' (34%) (Fig. 7).

Line A07-13 delivered the most compelling improvement, raising groat recovery to 74%, about 12 percentage points above the cultivated parent 'Amlal' and 16 points above the wild donor *A. murphyi* P45-55, while A07-36 (70%) and A07-20 (68%) also outperformed the check at $P \leq 0.05$. Because groat percentage is a reliable proxy for milling yield (Doehlert et al., 1999; Hall et al., 2003), these gains translate directly into higher flour output and lower dehulling waste, showing that the backcross strategy retained the high-protein alleles of *A. murphyi*, yet eliminated its thick-hull penalty. The PPSD results show a clear milling-quality gain after one backcross: all BC₁ lines carry < 33% husk, while A07-13 combines the lowest husk (25.96%) with the highest groat (74.04%), outperforming both the donor P45-55 (42.39% husk; 57.61% groat) and the recurrent parent 'Amlal' (33.75%; 66.25%), with differences supported by $P = 0.0001$, $CV \leq 5.24\%$, and $LSD = 3.177$ (Table 1). Higher groat percentage is a direct

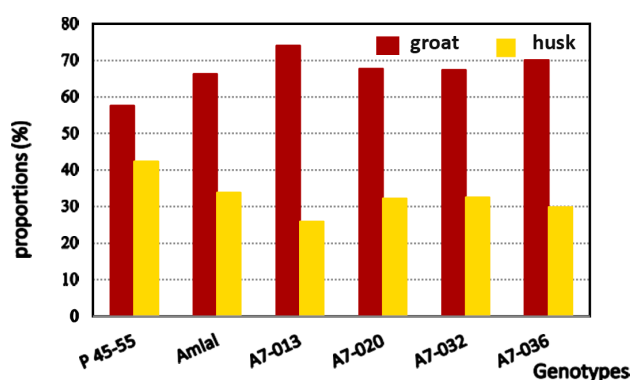


Fig. 7. Groat and husk proportions in the derivative lines of [*A. sativa* ('Amlal') × *A. murphyi* (P45-55)] × *A. sativa* ('Amlal')

Рис. 7. Соотношение зерна и пленок линий [*A. sativa* ('Amlal') × *A. murphyi* (P45-55)] × *A. sativa* ('Amlal')

Table 1. PPSD analysis for groat and husk proportions in the progeny of the cross [*A. sativa* ('Amlal') × *A. murphyi* (P45-55)] × *A. sativa* ('Amlal')

Таблица 1. Анализ PPSD для соотношения зерна и пленок у потомства скрещивания [*A. sativa* ('Amlal') × *A. murphyi* (P45-55)] × *A. sativa* ('Amlal')

Lines/parents	Groat %	Husk % /
A07-13	74.04 a	25.96 d
A07-20	67.80 bc	32.20 bc
A07-32	67.50 bc	32.50 bc
A07-36	70.08 b	29.92 c
Amlal	66.25 c	33.75 b
P45-55	57.61 d	42.39 a
ns	0.0001**	0.0001**
C.V (%)	2.71	5.24
LSD	3.177	3.177

Note: the means in a column sharing the same letter do not differ at $P \leq 0.05$ (Fisher's protected LSD); ** indicates significance at $P < 0.01$; CV is the coefficient of variation of the ANOVA residual

Примечание: средние значения в столбце, имеющие одну и ту же букву, не различаются при $P \leq 0,05$ (НСР по критерию Фишера); ** указывает на значимость при $P < 0,01$; CV – коэффициент вариации остатка

proxy for milling/dehulling yield and an explicit breeding target in modern oat programs: recent work using multitrait genomic selection treats groat % as a core quality trait, because it predicts recoverable edible kernel and lowers processing losses (Ardayfio et al., 2025).

In parallel, genome-scale studies now map the loci for kernel and nutritional traits underpinning food-grade oats, supporting the feasibility of decoupling hull burden from end-use composition – consistent with the pattern observed here (Dhakal et al., 2024).

Groat protein content in hybrid lines

Groat protein content for the selected lines exceeded that of the hexaploid parent 'Amlal' (10.46%) by 1 to 7%. The highest protein content of 17.29% was recorded for line (F09-01) A07-13 (Fig. 8). Multiple comparison of PPSD means revealed the existence of a highly significant difference between the studied lines and their parents (0.0001**, $P < 0.05$). Thus, the hybridization work made possible a genetic gain from 12% to 65% in the progeny for this trait.

Combining all the above results, line A07-13 was selected for its good agronomic performance in addition to its nutritional features. This line was registered in the INRA–Morocco national catalogue under the name of cv. 'Al Fawze'. This cultivar was subjected to further nutritional quality tests and preliminary tests of rheological behavior. Adding different proportions of 'Al Fawze' flour to that of the widely used Moroccan cultivar of bread wheat 'Kenz' improved the nutritional quality of the produced bread. Protein content in the mixture of 30% of oat and 70% of bread wheat flours was increased by 14% and the dry matter content by 16.02%, compared to that of bread wheat flour from cv. 'Kenz'.

Incorporation of 30% of oat flour into 70% of bread wheat flour improved the β -glucan content of the mixture by up to 1.20%. On the other hand, both color and rheological behavior of the flour mixture were affected, acting particularly on the water absorption rate and paste stability (Fig. 10). Across wheat–oat blends with the use of 'Al Fawze', protein rose steadily with substitution (14.20% at 0% to 16.02% at

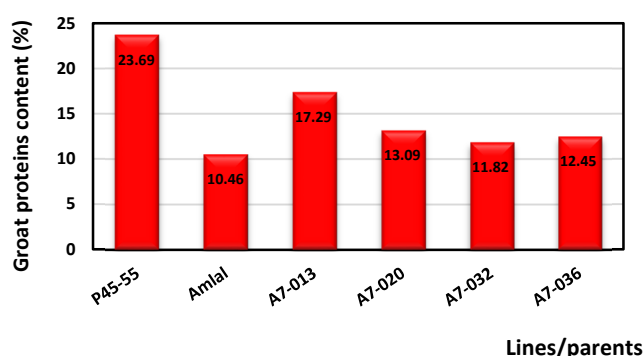


Fig. 8. Groat protein content in the derivative lines of [*A. sativa* ('Amlal') × *A. murphyi* (P45-55)] × *A. sativa* ('Amlal')

Рис. 8. Содержание белка в крупе линий от скрещивания [*A. sativa* ('Amlal') × *A. murphyi* (P45-55)] × *A. sativa* ('Amlal')

Groat β -glucan content in hybrid lines

Analysis of the β -glucan content in the groat of the selected lines showed that it ranged from 2.94 to 4.29%, realizing an improvement of this trait by 0.18 to 1.53% in comparison to the hexaploid parent 'Amlal' (2.76%). The highest rates were scored for lines A07-13 and A07-32: 4.29% and 4.1%, respectively (Fig. 9). Hence, through hybridization work, we were able to achieve a genetic gain of 2.44 to 54.28% in the derivative lines for groat β -glucan content.

Nutritional quality and preliminary rheological behavior tests of flour mixtures

30% → 17.14% at 100%), and β -glucans increased sharply from 15 to 30% of the oat share (0.18 to 1.20 g/100 g), as expected for oat fortification. Gluten dilution lowered the sedimentation index with higher oat levels (34.00 at 0% to 17.67 at 30%), a well-documented effect in wheat–oat composites. Technologically, the best balance occurred at 20% of oat, where specific volume peaked (13.36 cm³ g⁻¹) and crumb density was minimal (0.075 g cm⁻³); at 25–30%, nutritional gains continued but loaf structure started to decline. This pattern agrees with recent studies showing that moderate oat inclusion ($\approx$ 10–20%), with proper water/yeast optimization,

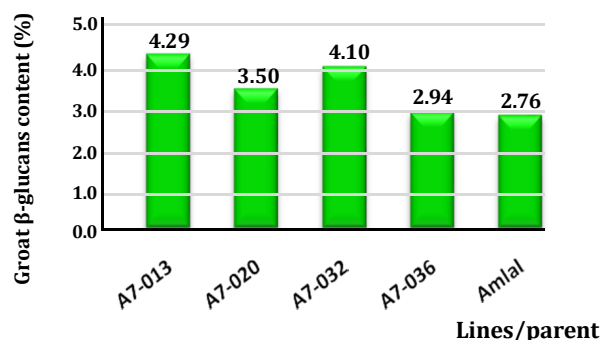


Fig. 9. Groat β -glucan content in the derivative lines of [*A. sativa* ('Amlal') × *A. murphyi* (P45-55)] × *A. sativa* ('Amlal')

Рис. 9. Содержание β -глюканов в крупе линий от скрещивания [*A. sativa* ('Amlal') × *A. murphyi* (P45-55)] × *A. sativa* ('Amlal')

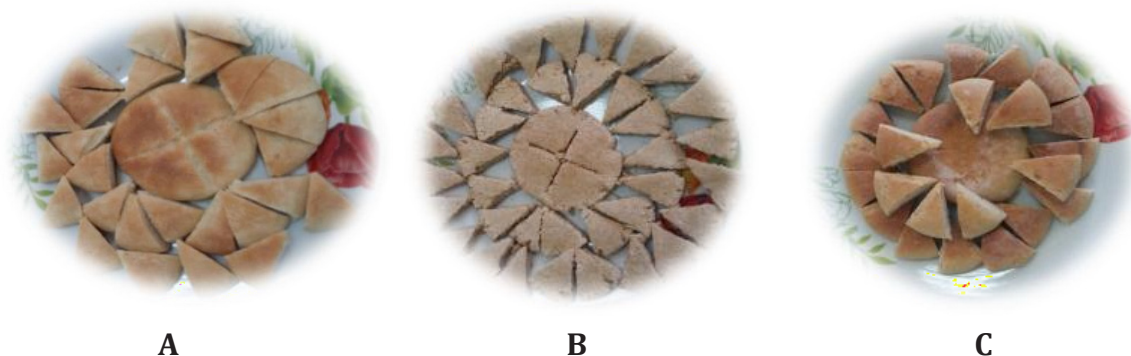


Fig. 10. Bread samples: A – baked from 100% ‘Kenz’ bread wheat flour; B – baked from 100% ‘Al Fawze’ oat flour; C – baked from a mixture of ‘Kenz’ (80%) and ‘Al Fawze’ (20%) flours

Рис. 10. Образцы хлеба, изготовленного из: А – 100% пшеничной муки сорта ‘Kenz’; В – 100% овсяной муки сорта ‘Al Fawze’; С – мучной смеси сортов ‘Kenz’ (80%) и ‘Al Fawze’ (20%)

can retain or maximize loaf volume while boosting fiber, whereas higher levels typically reduce volume and increase firmness due to gluten dilution and β -glucan viscosity effects (Rashed et al., 2024). It markedly improved the nutritional quality without influencing too much the technological quality required by the bakery industries and recommended bread bakeries (Table 2).

(Fig. 11), meaning more saleable flour per ton of grain. Even more striking are the compositional gains: protein rises from 10% to 17% and 13%, while β -glucan content increases from 3% to 5% and 4%. These levels meet or exceed the thresholds associated with cholesterol-lowering and low-glycemic properties in human diets (Welch et al., 2000; Zhu et al., 2004) (see Table 3, 4).

Table 2. Physicochemical composition and bread-quality attributes of wheat breads incorporating different levels of ‘Al Fawze’ oat flour

Таблица 2. Физико-химический состав и качественные показатели пшеничного хлеба с различным содержанием овсяной муки сорта ‘Al Fawze’

‘Al Fawze’ oat flour, %	Wheat flour, %	Protein, %	β -glucans, g/100 g	Sedimentation index	Specific volume, cm ³ /g	Density, g/cm ³
0	100	14.20 ^d	0.12 ^d	34.00 ^a	–	–
10	90	14.85 ^c	0.10 ^c	20.67 ^c	–	–
15	85	15.19 ^c	0.18 ^c	22.33 ^b	11.04 ^d	0.090 ^{ab}
20	80	15.27 ^c	0.99 ^{cb}	20.67 ^c	13.36 ^b	0.075 ^d
25	75	15.93 ^b	1.19 ^b	19.33 ^c	–	–
30	70	16.02 ^b	1.20 ^b	17.67 ^d	11.72 ^{dc}	0.085 ^{abc}
100	0	17.14 ^a	4.04 ^a	12.33 ^e	12.08 ^{bcd}	0.082 ^{bcd}

Note: the means with different letters a, b, c, etc. indicate significant differences according to Duncan’s test, $P \leq 0.05$

Примечание: средние значения с разными буквами a, b, c и др. указывают на значимые различия согласно тесту Дункана, $P \leq 0,05$

Cultivar registration in the official national register

‘Al Fawze’ and ‘Abtah’ outperform their parent ‘Amlal’ in every key trait shown in Tables 3 and 4. Grain yield rises by $\approx 10\%$ in ‘Al Fawze’ and by $> 35\%$ in ‘Abtah’, confirming that the introgressed germplasm confers a genuine productivity advantage rather than a mere quality trade-off. Both retain full field resistance to crown rust and powdery mildew and show only moderate barley-yellow-dwarf symptoms, a disease-control profile that typically safeguards 15–20% of yield otherwise lost to infection (Saari, Prescott, 1975).

Technologically, thousand-kernel weight climbs from 32 g in ‘Amlal’ to 38 g and 34 g, a range associated with superior milling efficiency and reduced broken groats during dehulling (Doehlert et al., 1999; Hall et al., 2003). Groat recovery itself improves from 66% to 74% (‘Al Fawze’) and 68% (‘Abtah’)

Conclusion

Interspecific introgression from the endemic tetraploid *Avena murphyi* (P45-55) into hexaploid *A. sativa* (‘Amlal’) produced fertile backcross derivatives (72% hexaploid), in which pollen fertility correlated with grain yield ($P < 0.001$). After F_2 – F_8 pedigree selection, ten stable lines combined higher field performance (mean 34.8–35.1 q ha⁻¹ vs. 31 q ha⁻¹ for ‘Amlal’) with robust disease profiles. Milling traits improved markedly: $< 33\%$ of husk and 68–74% of groat in all BC₁ lines, with A07-13 at 74.04% of groat / 25.96% of husk and TKW 36–39 g. Composition also advanced (groat protein up to 17.29%, β -glucans up to 4.29%). In bread prototypes, 20% of ‘Al Fawze’ flour maximized the specific volume (13.36 cm³ g⁻¹) and minimized the crumb density (0.075 g cm⁻³), while in-



Fig. 11. Kernels of new released hexaploid *Avena sativa* cvs. 'Al Fawze' and 'Abtah'

Рис. 11. Зерновки новых выведенных гексаплоидных сортов *Avena sativa* 'Al Fawze' и 'Abtah'

Table 3. Agronomic traits of the registered cvs. 'Al Fawze' and 'Abtah' compared to their hexaploid parent 'Amlal'

Таблица 3. Хозяйственно ценные признаки зарегистрированных сортов 'Al Fawze' и 'Abtah' в сравнении с их гексаплоидным родителем – сортом 'Amlal'

Cultivars	Plant cycle	Disease resistance assessment				Grain yield, q ha ⁻¹
		Crown rust	Powdery mildew	Helminthosporium	Barley yellow dwarf virus (BYDV)	
'Amlal'	Semi-early	Resistant	Moderately sensitive	Resistant	Moderately sensitive	31
'Al Fawze'	Semi-early	Resistant	Resistant	Moderately resistant	Resistant	34
'Abtah'	Semi-early	Resistant	Resistant	Moderately resistant	Resistant	43

Table 4. Selected technological parameters of the registered cvs. 'Al Fawze' and 'Abtah' compared to their hexaploid parent 'Amlal'

Таблица 4. Отдельные технологические параметры зарегистрированных сортов 'Al Fawze' и 'Abtah' в сравнении с их гексаплоидным родителем – сортом 'Amlal'

Cultivars	Selected technological parameters			
	Weight of 1000 seeds, g	Husk proportion, %	Groat protein content, %	Groat β -glucan content, %
'Amlal'	32.33 $\pm$ 1.35 ^c	66.25 $\pm$ 1.04 ^c	10.46 $\pm$ 0.03 ^c	2.76 $\pm$ 3.03 ^c
'Al Fawze'	38.20 $\pm$ 1.52 ^a	74.03 $\pm$ 0.15 ^a	17.29 $\pm$ 0.29 ^a	4.29 $\pm$ 0.03 ^a
'Abtah'	34.40 $\pm$ 0.03 ^b	67.80 $\pm$ 0.96 ^b	13.09 $\pm$ 0.40 ^b	3.50 $\pm$ 1.04 ^b

Note: the means with different letters a, b, c, etc. indicate significant differences according to Duncan's test, $P \leq 0.05$

Примечание: средние значения с разными буквами a, b, c и др. указывают на значимые различия согласно тесту Дункана, $P \leq 0,05$

creasing protein and β -glucans. On this basis, A07-13 ('Al Fawze') and A07-36 ('Abtah') were registered, delivering higher yield, groat recovery and nutritional value than 'Amlal'. The results broaden the genetic base of Moroccan oats and provide a practical route to food-grade, climate-adapted cultivars. Industrial scale-up of milling and baking trials is the next step.

Recommended fields of application

Oats have a rich and diverse composition of proteins, unsaturated fatty acids, fibers and antioxidants, such as polyphenols and tocopherols. This particular composition supplies this crop with beneficial properties that could be converted into antioxidant effects, preventing against diabetes, obesity, cancer, and cardiovascular diseases. In Morocco, however, oat

cultivation for human consumption has not yet been developed compared to that for animal feed. This research supported the release of new and original cultivars from Moroccan wild oat germplasm. The exploitation of new cultivars as beneficial food for human health opens up new prospects for both farmers and industrials (flourmills, biscuit factories, breakfast cereals, etc.).

High levels of quality parameters of these cultivars, derived from interspecific crosses, provide an optimistic tool for improving groat nutritional value, especially in regions of the world where deficiency of minerals and other major nutritional compounds affects populations. The existence of essential nutrients in the grain of the newly released cultivars can remedy the problems of malnutrition by acting as supplements in the daily diet, thus making it possible to cover our daily needs in proteins, soluble fibers, essential minerals, etc.

The results of this research showed for the first time the relevance and usefulness of combining both breeding work and technological assessment. It should be noted in this regard that the evaluation of essentially nutritional parameters, namely proteins and β -glucans in groat, was carried out for the first time in Morocco. The obtained results made it possible not only to distinguish oats from other cereals, but also to differentiate the new hexaloid cultivars from those commonly produced in the world, as reported in the literature.

Furthermore, the results of this research could further encourage the use of oats in industrial preparations, where wheat holds exclusive rights. Moreover, the flour from the selected lines could be a source of valuable nutrients for human consumption and can therefore be used in the formulation of typical foods, nutritious and beneficial to health, of those commonly elaborated with wheat flour.

Thus, the development of new cultivars with high nutritional value, conceived for human consumption, can contribute to increasing the socio-economic impact of oat cultivation, mainly in North Africa where oats are still mainly used as fodder.

Estimation of socio-economic benefits

The demand for healthy and multifunctional products is increasing day after day with the societies' evolution and the awareness of everyone's well-being. In this context, we offer two new oat cultivars, already recognized for their beneficial effects on health, which also have even higher nutritional qualities. All oat trade value chain branches would benefit from this new developed material. Farmers could ensure good commercialization of their production due to the good grain yield and the excellent performance of the cultivars. The manufacturer could add value to its products by including oats in the recommended proportions. The consumer would gain from a quality product beneficial for his health, especially that the number of diabetics requiring a healthy and safe diet is rapidly increasing worldwide.

The adoption of these cultivars with good productivity and excellent agronomic, technological and nutritional performance by farmers will certainly ensure a good margin of profitability. Seed growers will also benefit from these cultivars, due to their grain quality and the guaranteed good yield levels, which will make up for the lack of certified seeds produced by the formal sector.

Morocco could even consider exporting seeds of these two cultivars to major oat consumer countries, because of their exceptionally good groat protein content, compared to that of other cultivars grown elsewhere and released without using wild germplasm available in our rich flora.

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