



**ADER 116**  
**2015-2018**

Use of biotechnological methods to increase breeding material genetic variability and accelerate genetic progress in crop yields and stability in major crops under climate changes

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# ADER 116 2015-2018

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**Main objective:** Improving farm economic performance by increasing the efficiency of using natural resources and technological inputs for sustainable agriculture in the context of climate change.

**Specific objectives:**

- Increase the efficiency of biotechnological methods used for the development of new DH lines;
- Assessment of mutant/recombinant DH lines (with/without wheat-rye translocation 1AL:1ARS) for diseases resistance, abiotic stress and other interested traits, in different geographic areas;
- Testing and characterization of the introgression lines obtained by hybridization among wheat lines/cultivars and related species regarding disease resistance, abiotic stress factors in different geographic areas in order to increase genetic variability;
- Development of new improved wheat, barley and triticale DH lines, more adapted to climate changes and more performants than existing genotypes.



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## Expected result 1:

- Creating at least 2-3 genotypes with new superior introgressions that open up new perspectives of genetic progress for the crops level and stability under the conditions of climate change.

## Results 1:

- ✓ 11 synthetic lines obtained by hybridization among synthetic amphiploids (*Triticum durum* x *Aegilops tauschii*) with common wheat varieties (E5A/Faur-3 lines, E16A/Faur- 1 line, E23A/Faur - 1 line, E25A/Faur-1 line, E7A/Glosa-3 lines și E18A/Glosa - 2 lines) selected and used in the wheat breeding program as sources due to their characteristics for quality, grain size, TKW, tolerance to biotic and abiotic stresses (NARDI Fundulea).

Nr.crt	Line code	Combination	Protein %	Sedimentation	Dough tenacity (W)	Hardness
1	25F1101P2	E25A x Faur	15.6	57.2	330	56.1
2	16F352	E16A x Faur	14.9	50.4	258	26.5
3	5F622	E5A x Faur	14.9	48.6	304	24.4
4	5F635	E5A x Faur	15.4	53.7	309	31
5	5F1073	E5A x Faur	15.6	59.7	321	33.5
6	23FS1-5	E23 x Faur	18.1	66.2	390	52.8

## Results 1:

- ✓ 19 new hybrids of synthetic amphiploids, between 6 genotypes - modern lines of *Triticum durum* (Agedur, Condur, Pandur, Grandur, DDU-8, DDU-156, DDU-2-24, GCO-3-25) and 7 biotypes of the species *Aegilops tauschii* (*Aegilops squarrosa*)/ *Aegilops tauschii* sq. *strangulate* (F1 generation at the end of the project) (NARDI Fundulea) were created.

Nr.crt.	♀ line	♂ line
1	Agedur	<i>Ae. tauschii squarrosa</i> 2468
2	Condur	<i>Ae. tauschii squarrosa</i> 2448
3	Condur	<i>Ae. tauschii squarrosa</i> 2453
4	Condur	<i>Ae. tauschii squarrosa</i> 2568
5	Grandur	<i>Ae. tauschii squarrosa</i> 2453
6	Grandur	<i>Ae. tauschii squarrosa</i> 2468
7	Pandur	<i>Ae. tauschii</i> sq. <i>strangulata</i>
8	Pandur	<i>Ae. tauschii squarrosa</i> 2569
9	DDU-8	<i>Ae. tauschii squarrosa</i> 2448
10	DDU-8	<i>Ae. tauschii squarrosa</i> 2468
11	DDU-8	<i>Ae. tauschii squarrosa</i> 2453
12	DDU-8	<i>Ae. tauschii squarrosa</i> 2530
13	DDU-2-24	<i>Ae. tauschii squarrosa</i> 2448
14	DDU-2-24	<i>Ae. tauschii squarrosa</i> 2453
15	DDU-2-24	<i>Ae. tauschii squarrosa</i> 2468
16	DDU-156	<i>Ae. tauschii squarrosa</i> 2448
17	DDU-156	<i>Ae. tauschii squarrosa</i> 2468
18	GCO-3-25	<i>Ae. tauschii squarrosa</i> 2453
19	GCO-3-25	<i>Ae. tauschii squarrosa</i> 2468

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## Results 1:

- ✓ 7 new combinations between elite lines from interspecific hybrids and modern common wheat cultivars (NARDI Fundulea) were created.

Code	♀ parent genealogy	♂ parent genealogy
H.53- b	DDU - 217 / <i>T. timopheevi</i> (pop)	Miranda
H.54	DDU - 8 / <i>T. dicoccoides</i> 4-3 (pop)	Pitar
H.58	DDU-2-24 / <i>T.dicoccoides</i> 4-10 (pop)	Pitar
H.64	Glosa / <i>T.dicoccoides</i> 4-10 (pop)	Miranda
H72	DDU- 8 / <i>T. charlicum</i> 6-3 (pop)	Pitar
H75	DDU - 156 / <i>T. charlicum</i> 6-2 (pop)	Litera
H.85- a	Gliadine (397-2-83) / Glosa (pop)	Pitar

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### Results 1: New combinations for germplasm diversification were obtained

- ✓ 3 new combinations between synthetic amphiploids and common wheat modern lines E35-A /A38-04, E32-A /11248G2-1 (F2 generation at the end of the project) and another new combination A35-14/E32-A in the F1 generation at the end of the project (SCDA Pitești).
- ✓ A hybrid combination of synthetic amphiploid x modern wheat variety - AS5/PAJURA (F2 generation) (SCDA Șimnic).
- ✓ 10 DH lines from the hybridization between Izvor x F000628 are introduced, as sources, in the wheat breeding program at NARDI Fundulea, another 6 lines (Al -II- 20, Al -II- 45, Al -II- 78, Al -II- 236, Al -II- 27A și Al -II- 268) in breeding program from SCDA Turda, 11 DH lines were used as sources at SCDA Pitești, 16 DH lines were used as sources at Craiova University- SCDA CARACAL, and 6 DH lines were used as sources at SCDA Șimnic
- ✓ DH line of triticales 09498TD2-111 was selected as source for the triticales breeding program.



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### Expected result2:

- The introduction into the official system of testing (ISTIS) at least 2 DH genotypes obtained by the biotechnology methods and ensuring the seeds requirement for breeders in order to rapid multiplication and expansion in farms.

### Results 2:

- ✓ **Zamfira line (F13347GP1)** is a wheat double haploid line (DH line) obtained by *Zea* biotechnological method, selected from the hybridization F06659G1-1/2\*OTILIA, and it is under official testing system ISTIS, second year (NARDI Fundulea).
- ✓ **The barley DH line, *DH406-3***, obtained by the *Bulbosum* biotechnological method, was introduced into the official testing system ISTIS in 2018 (NARDI Fundulea).



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## Results 2: Perspective lines

- ✓ Introducing into the official testing system ISTIS the line **LM 16** and providing seeds for breeders for rapid multiplication and expansion in farms, will be available in the Autumn 2019. At this line, **LM 16**, all the quality indicators were at the same level as the reference cultivar, Izvor (SCDA Șimnic).
- ✓ DH wheat lines Ai- II 193, Ai- II 223, Bi- I 3, Bi- I 40, Bi- II 40, Bi- II 58 și Bi- II 69 are recommended for being cultivated in the South of the country and seeds have been provided for two farmers from this area (Craiova University-SCDA Caracal).
- ✓ DH lines of wheat: Al -II- 183, Al -II- 193, Al -II- 55, Bl -I- 3, Bl -I- 40, Bl -I- 39, Bl -II- 65 și Bl -II- 68 were selected for future testing (2018-2019) in comparative plots and possible for future testing in the official network ISTIS (SCDA Turda).
- ✓ DH lines of wheat: Ai- II-201, Bi-I-47, Bi-II-82 were selected and used as sources in the wheat breeding program at SCDA Pitești.

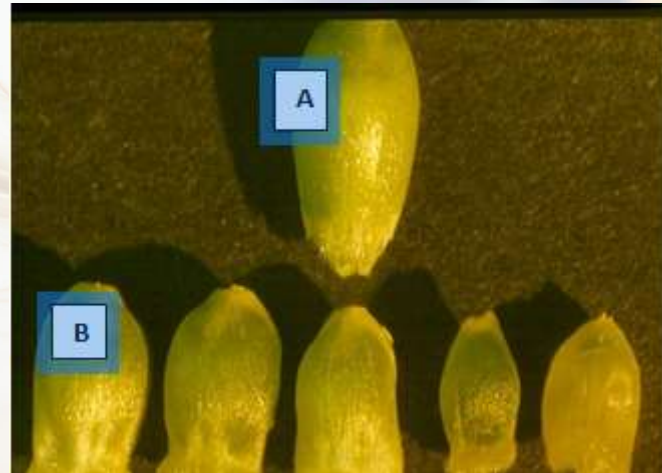
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### Expected result3:

- Increasing the efficiency of biotechnological methods used, by optimizing operating parameters in order to obtain better results in the future research.

### Results 3:

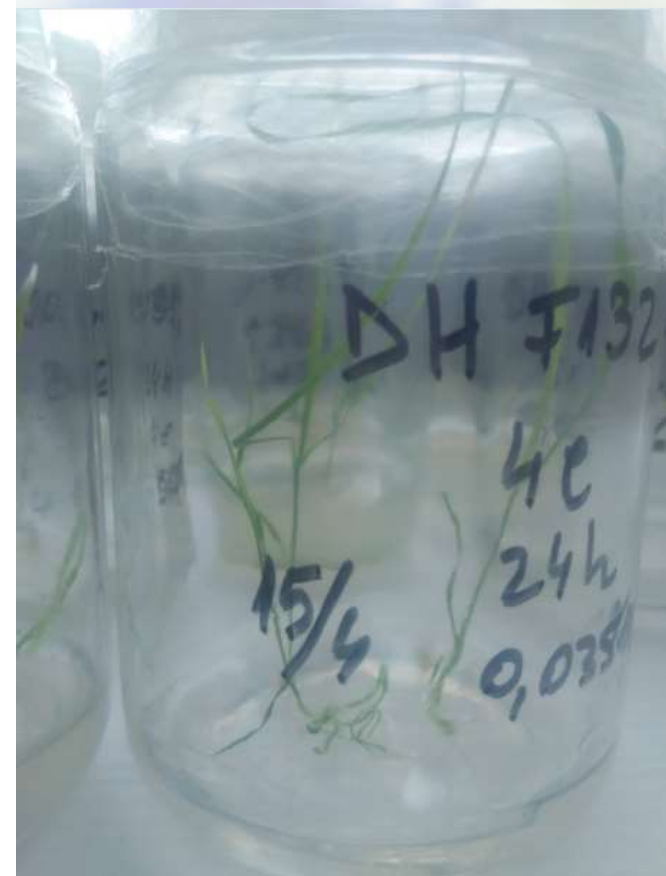
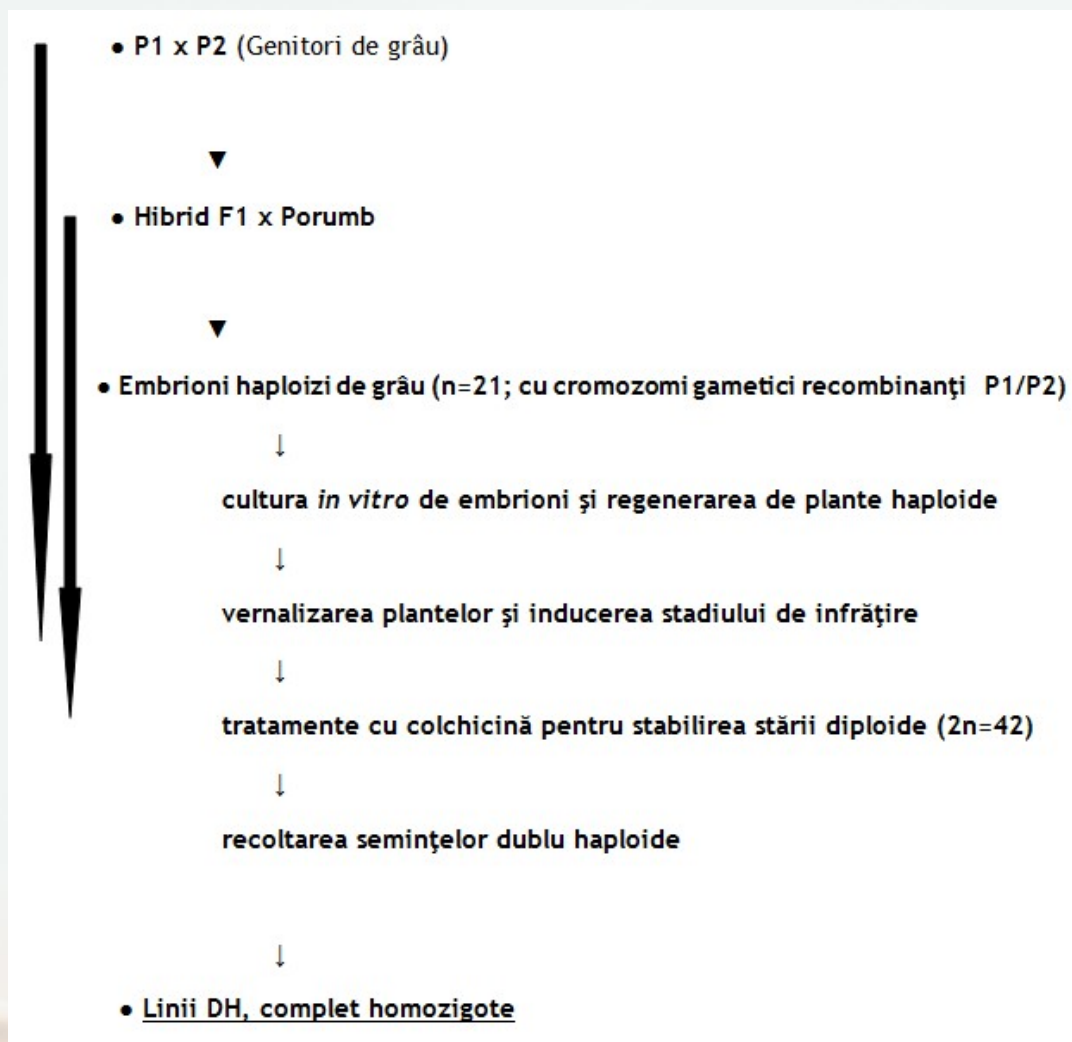
- ✓ Haploid chromosomes duplication protocol by “*in vitro*” treatment;



Caryopses formed by self-pollination (A) and by cross-pollination with maize (B)

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## The Simplified Scheme for the Production of DH Wheat Lines



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## Results 3:

- ✓ DNA isolation method from a single dry seed available for three cereals (wheat, barley and triticale).
- ✓ The highlighting of new allelic variants using molecular markers (DNA) as well as combinations of positive alleles with a positive impact on thousand kernel weight (TKW).

Molecular markers located on the 1RS chromosome associated with TKW

Marker	Dif MMB	p
wms1223	3,0	0,000254
<b>TSM592</b>	<b>3,2</b>	<b>0,000285</b>
PSC200	3,0	0,002512
TSM120	2,6	0,003774

Combinations of favorable alleles and their association with TKW

Combinatie	P	F+F	F+I	I+F	I+I
TaFLO D1+TaTPP-6AL1	0,000149	45,8	43,7	<b>50,2</b>	47,1
Secară+TaFLO D1	0,000319	45,8	<b>49,6</b>	43,3	46,4
Secară+TaTPP-6AL1	0,000991	<b>48,6</b>	47,0	46,7	43,0
Secară+TaSST-D1	0,0023	46,8	<b>49,7</b>	43,9	45,7
TaSST-D1+TaFLO-D1	0,028519	43,7	48,5	47,0	<b>49,0</b>



# Molecular marker analysis

DNA isolation



DNA amplification  
(PCR)



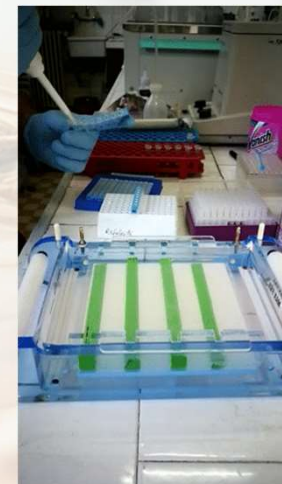
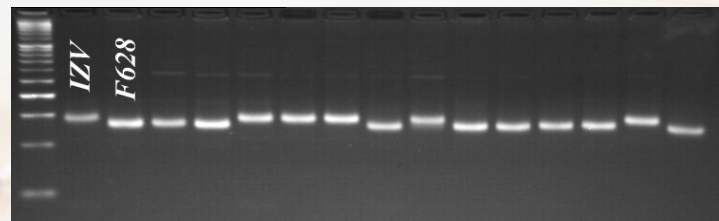
Digestion with  
restriction enzymes



Electrophoresis

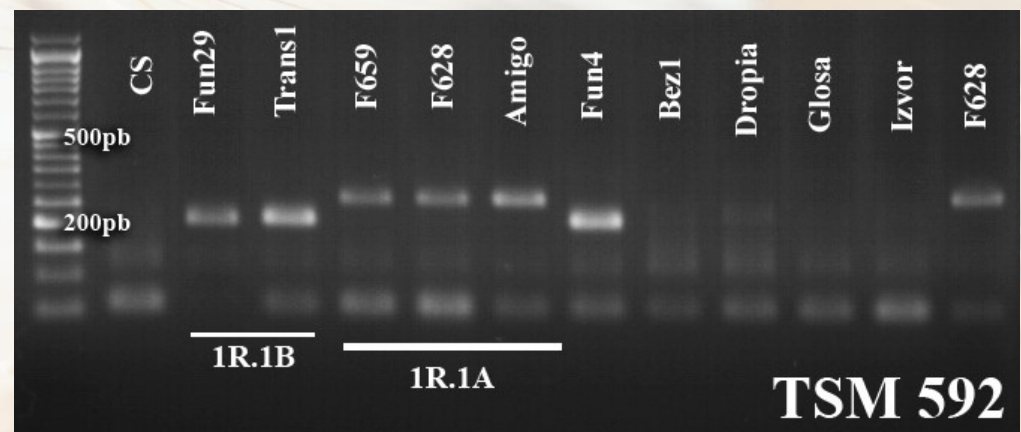
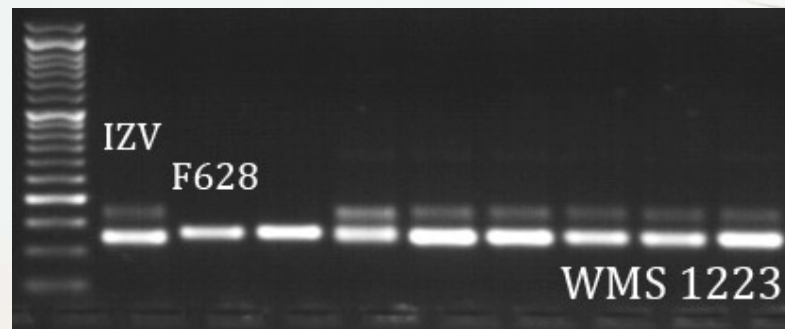
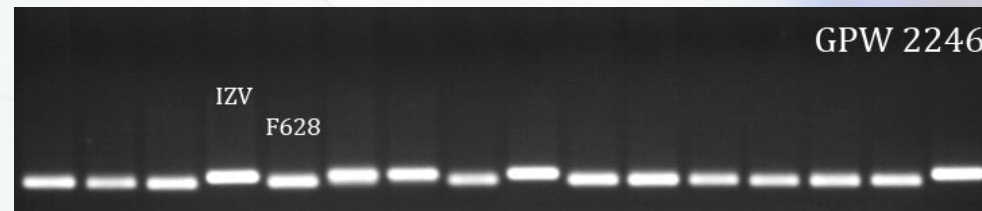
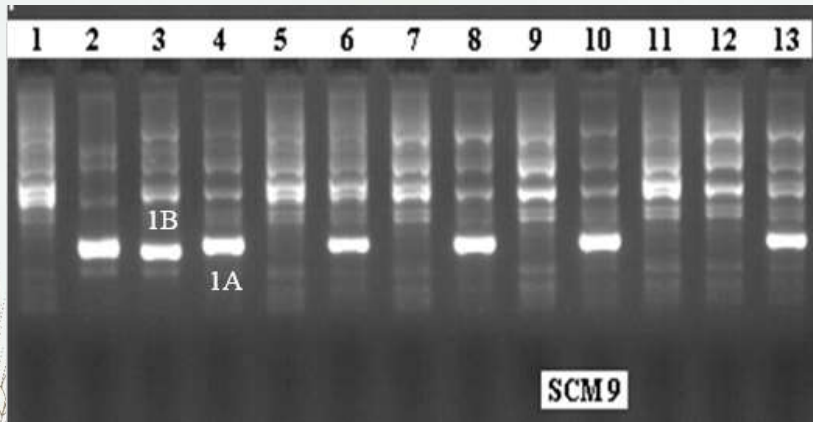


Results analysis



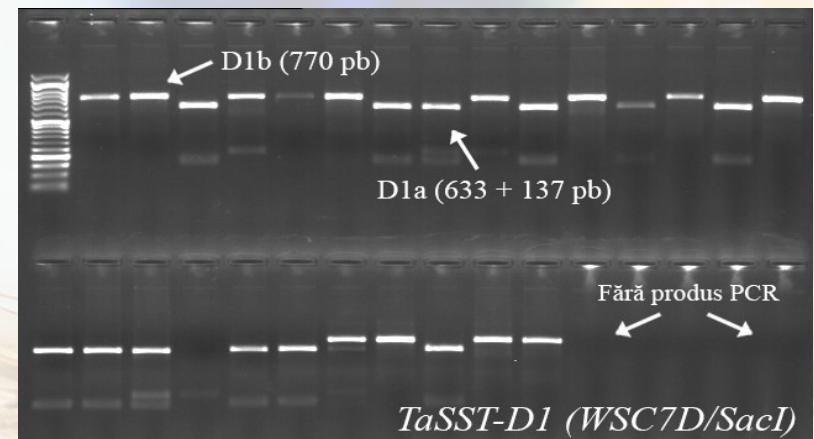
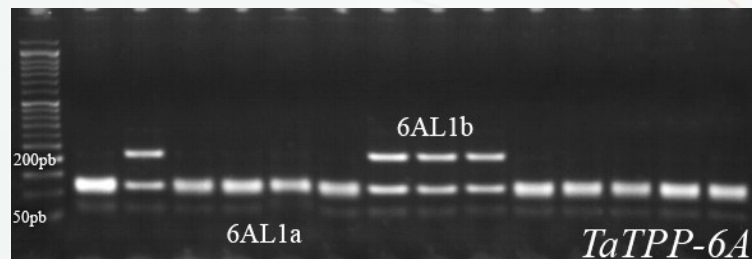
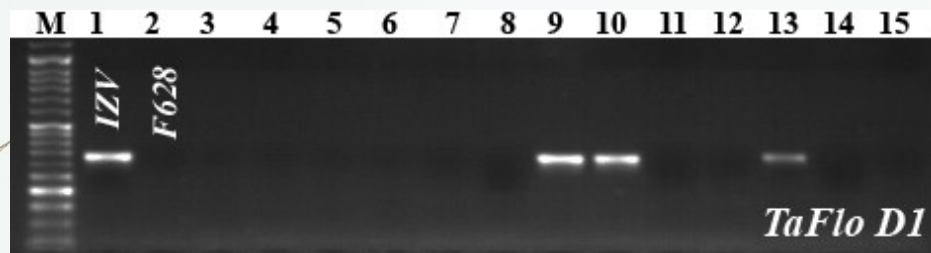
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Molecular markers for the detection of rye chromatin in a wheat background



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Genetic variability at some loci involved in the control of grain size and weight in wheat





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### Expected result 4:

- Dissemination of the results obtained by ADER116 project.

### Results 4:

- ✓ Organization of the International Conference : 17th EWAC - The European Cereals Genetics Cooperative EUCARPIA Cereals Section International Conference, 03-08 June 2018

Place: Bucharest (Capital Plaza Hotel), 54 participants;

- From abroad - 26 researchers and students from 11 countries (Algeria, Bulgaria, Canada, Russia, Ukraine, Hungary, Serbia, Germany, Mexico, United Kingdom, Poland)
- From Romania - 28 (14,8% participation from the university and 85,2% from the research field)

Papers presented - 23 papers and 14 posters;

Activities: visiting the experimental fields.



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17th EWAC Eucarpia International Conference  
June 3-8, Bucharest, Romania



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## Results 4:

### ✓ Participation at the scientific events :

- National : Annual Scientific Session of NARDI Fundulea (2016/2017/2018); Session of papers from SCDA Secuieni (2016/2017/2018); Prospects of Millennium Agriculture III”- USAMV Cluj-Napoca (2017); Session of papers of USAMV Timișoara (2016/2017/2018), Agriculture for Life, Life for Agriculture, USAMV Bucuresti (2016, 2017, 2018).
- International: 13th International Wheat Genetics Symposium (2017); „Fifth international conference on radiation and applications in various fields of research (2017); International Multidisciplinary Scientific GeoConferences SGEM (2016/2018).

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### Results 4:

#### ✓ Publications:

- articles published in journals indexed in ISI with impact factor - 4;
- articles published in journals indexed in ISI without impact factor - 1;
- articles published in journals indexed in BDI - 12.