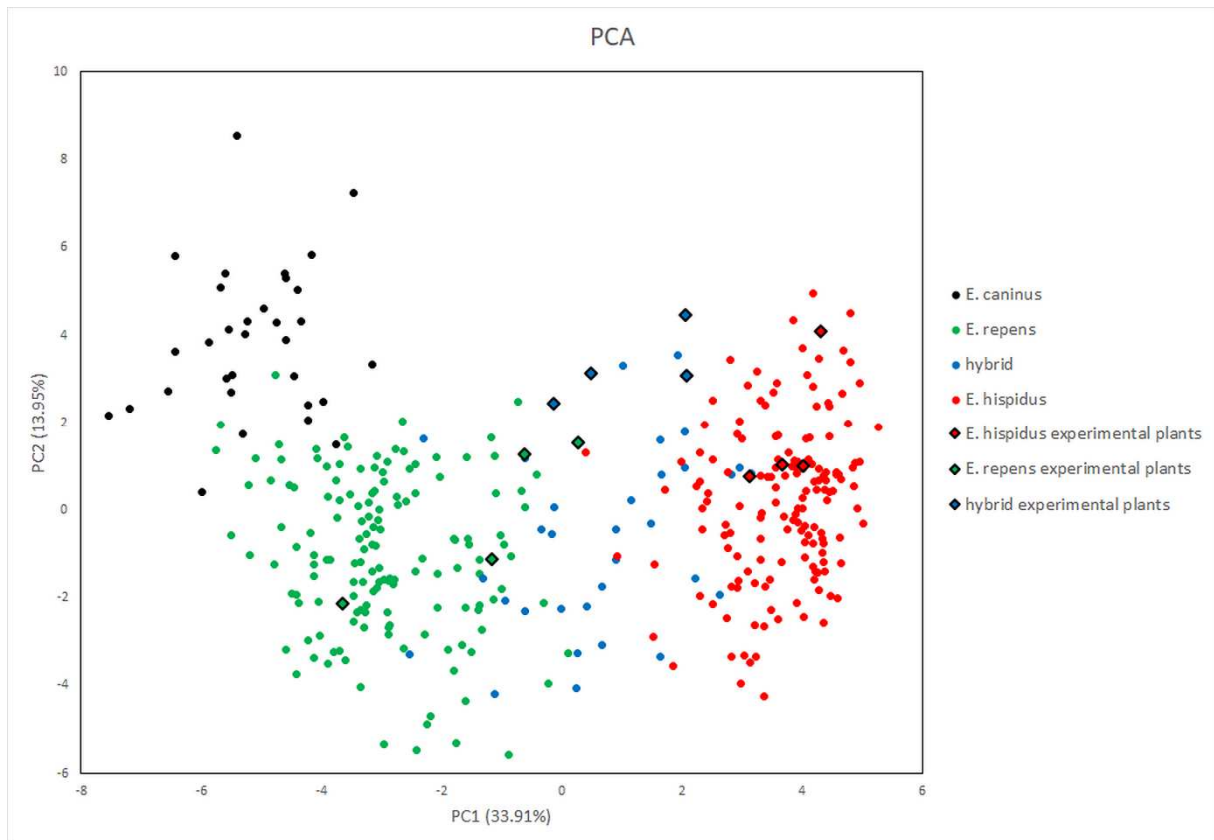


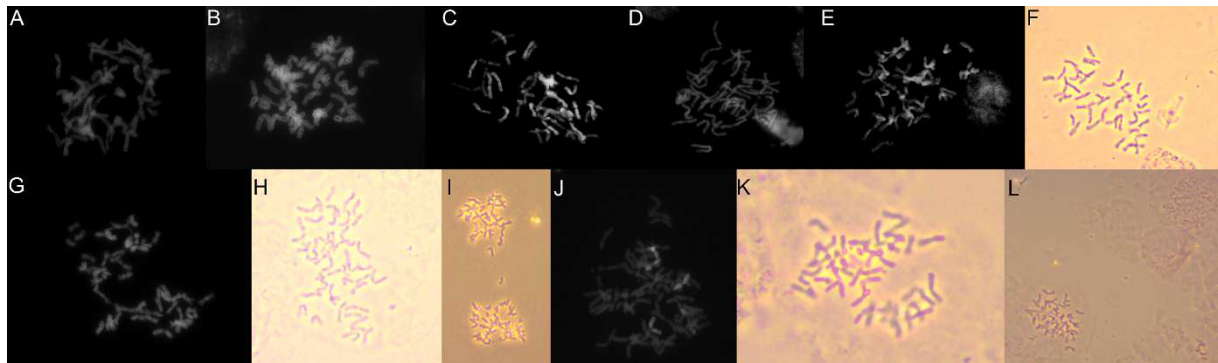
Urfusová R., Mahelka V., Krahulec F., Veškrna O. & Urfus T. (2022) The mentor effect increases the rate of selfing in couchgrasses. – *Preslia* 94: 377–397.

**Electronic Appendix 1.** – Parental plants in pollination experiments, their absolute genome sizes (GS; in pg) and ploidy levels. Ploidy of *Elymus* plants were verified by chromosome counts, ploidy levels of crops were verified by their producers – the breeding stations Selgen and Danko. *Elymus* taxa were morphologically determined and their identity was confirmed by morphometrics (following Urfusová et al. 2021).

Parental plant	Species	GS (pg)	ploidy level
MO24c	<i>E. repens</i>	25.02	6x
CS22a	<i>E. repens</i>	24.50	6x
T14P4	<i>E. hispidus</i>	27.77	6x
CS1f	<i>E. hispidus</i>	27.85	6x
MO5b	hybrid	26.74	6x
T13P12	hybrid	28.77	6x
T10P7	<i>E. repens</i>	30.81	7x
T14P8	<i>E. repens</i>	30.22	7x
T9P1	hybrid	30.50	7x
T1P1	hybrid	35.44	8x
T4P4	<i>E. hispidus</i>	33.18	8x
T4P5	<i>E. hispidus</i>	32.71	8x
Secale Gadello	<i>Secale cereale</i>	17.55	2x
JJ 6R 610 A	<i>Hordeum vulgare</i>	10.84	2x
Lancelot	<i>Hordeum vulgare</i>	10.84	2x
JJ cross 43	<i>Hordeum vulgare</i>	11.21	2x
Triticum monococcum	<i>Triticum monococcum</i>	12.61	2x
K1 Bohemia	<i>Triticum aestivum</i>	37.74	6x
SG-U2078-18	<i>Triticum aestivum</i>	36.48	6x
K2 Illusion	<i>Triticum aestivum</i>	35.71	6x
(SG-S1500-14) Lorien	<i>Triticum aestivum</i>	36.49	6x
SG-U5003-16	<i>Triticum aestivum</i>	35.76	6x
Viriato	<i>Triticum aestivum</i>	38.12	6x
Triticale MAMUT	<i>Triticale</i>	43.40	6x
Triticale PORTO	<i>Triticale</i>	42.08	6x



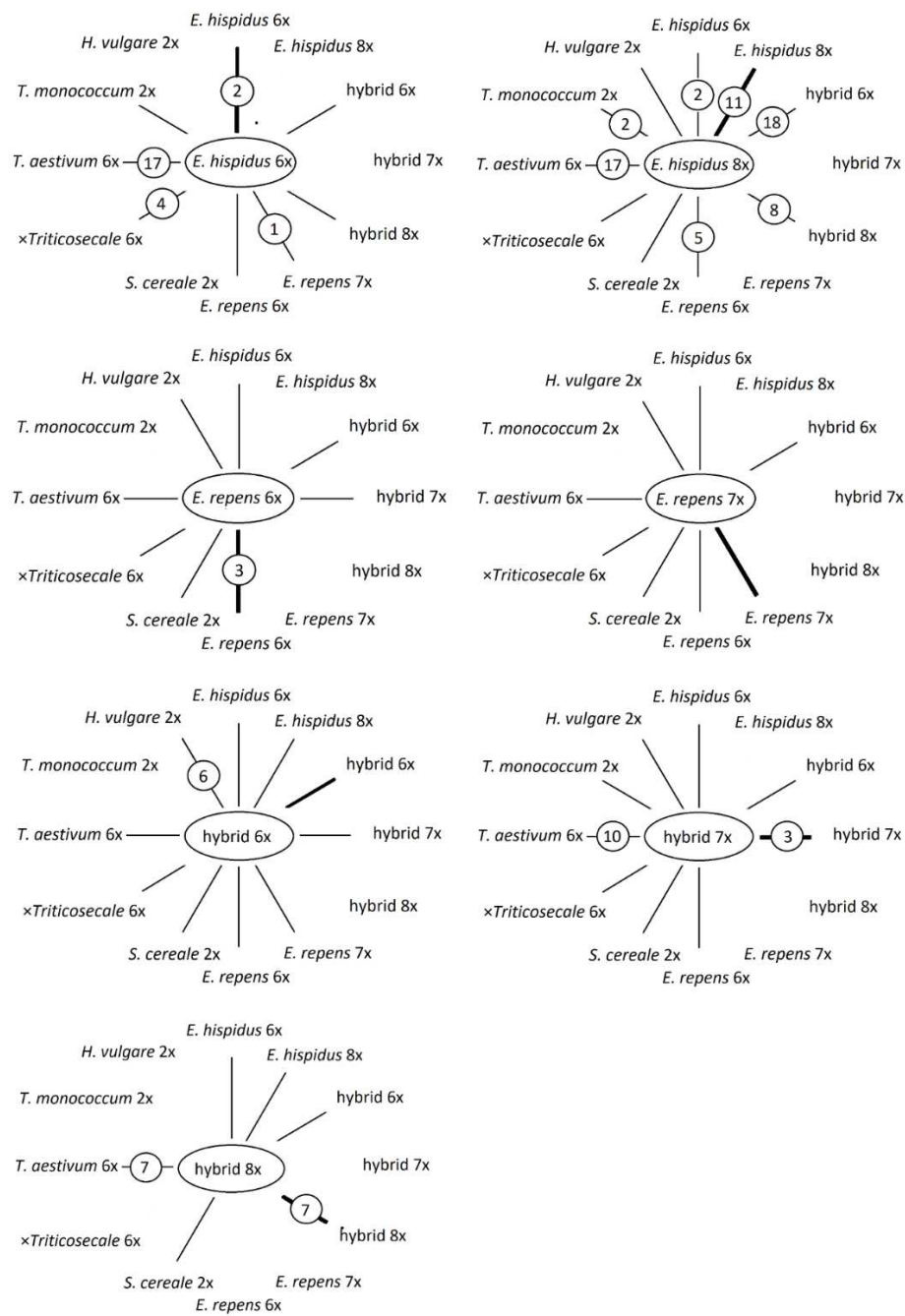
**Electronic Appendix 2.** – Ordination diagram of principal component analysis (PCA) of individuals from the study of Urfusová et al. (2021), with added *Elymus* plants used in this study – encircled in black.



**Electronic Appendix 3.** – Chromosomes of parental plants. A, B – hexaploid *E. repens*; C, D – hexaploid hybrid; E, F – hexaploid *E. hispidus*; G, H – heptaploid *E. repens*; I – heptaploid hybrid; J – octoploid hybrid; K, L – octoploid *E. hispidus*. Black-and-white images – magnification 600×, colour images – 400×.

**Electronic Appendix 4.** – *Elymus* species used in the open-pollination control experiment. The ploidy levels of part of these plants were confirmed by chromosome counts.

<b>sample</b>	<b>species</b>	<b>GS (pg)</b>	<b>ploidy</b>
Rum17-3c	<i>E. repens</i>	24.77	6x
MO24c	<i>E. repens</i>	25.02	6x
Mol17-3b	<i>E. repens</i>	25.09	6x
CS1f	<i>E. hispidus</i>	27.85	6x
SR17-1a	<i>E. hispidus</i>	28.11	6x
Ukr17-3c	<i>E. hispidus</i>	30.01	6x
Ukr17-7b	<i>E. hispidus</i>	29.55	6x
Ukr17-3ch	<i>E. hispidus</i>	29.12	6x
MO5b	hybrid	26.74	6x



**Electronic Appendix 5.** – Schematically expressed pollination experiment based on maternal plants (marked by ellipse) and realized crossings linked to pollen donor taxa/ploidal cytotypes. The thin line corresponds to induced selfing and the thick line to core selfing. Successful pollinations are marked by numbers of developed seeds in a circle.

**Electronic Appendix 6.** – Results of particular generalized linear mixed-effects models (GLMM) testing the effect of ploidy level and species of maternal plant and paternal plant and type of selfing on seed set with Tukey Contrasts. Seed set refers to number of seeds/number of florets ratio of each single pollinated spike. Maternal plant individual was used as random effect.

## GLMM

### Seed set: entire

maternal plant ploidy level (regardless of species):  $\chi^2 (2) = 14.884$ , Df resid = 257, p = 0.0005862

Multiple Comparisons of Means: Tukey Contrasts:

	Estimate	Std. Error	z value	Pr(> z )
7x - 6x == 0	0.8363	0.9745	0.858	0.66326
8x - 6x == 0	-2.5969	0.8476	-3.064	0.00620
8x - 7x == 0	-3.4331	1.1140	-3.082	0.00601

maternal plant species (regardless of ploidy level):  $\chi^2 (2) = 17.474$  Df resid = 257, p = 0.0001605

Multiple Comparisons of Means: Tukey Contrasts:

	Estimate	Std. Error	z value	Pr(> z )
<i>E. repens</i> - <i>E. hispidus</i> == 0	3.875	1.050	3.692	< 0.001
hybrid - <i>E. hispidus</i> == 0	2.027	0.681	2.976	0.00789
hybrid - <i>E. repens</i> == 0	-1.848	1.032	-1.790	0.16802

maternal plant species combined with ploidy level:  $\chi^2 (6) = 30.193$ , Df resid = 253, p = 3.613e<sup>-05</sup>

Multiple Comparisons of Means: Tukey Contrasts:

	Estimate	Std. Error	z value	Pr(> z )
<i>E. hispidus</i> 8x - <i>E. hispidus</i> 6x == 0	-1.6108	0.8830	-1.824	0.48151
<i>E. repens</i> 6x - <i>E. hispidus</i> 6x == 0	2.0220	1.2307	1.643	0.60732
<i>E. repens</i> 7x - <i>E. hispidus</i> 6x == 0	21.4800	256.0000	0.084	1.00000
hybrid 6x - <i>E. hispidus</i> 6x == 0	2.6343	1.0456	2.519	0.12578
hybrid 7x - <i>E. hispidus</i> 6x == 0	0.6897	1.0368	0.665	0.99271
hybrid 8x - <i>E. hispidus</i> 6x == 0	-0.0712	0.9711	-0.073	1.00000

<i>E. repens</i> 6x - <i>E. hispidus</i> 8x == 0	3.6328	1.2579	2.888	0.04713
<i>E. repens</i> 7x - <i>E. hispidus</i> 8x == 0	23.0908	256.0014	0.090	1.00000
hybrid 6x - <i>E. hispidus</i> 8x == 0	4.2451	1.1118	3.818	0.00199
hybrid 7x - <i>E. hispidus</i> 8x == 0	2.3005	1.0811	2.128	0.29182
hybrid 8x - <i>E. hispidus</i> 8x == 0	1.5396	0.9884	1.558	0.66597
<i>E. repens</i> 7x - <i>E. repens</i> 6x == 0	19.4580	256.0028	0.076	1.00000
hybrid 6x - <i>E. repens</i> 6x == 0	0.6123	1.3761	0.445	0.99922
hybrid 7x - <i>E. repens</i> 6x == 0	-1.3323	1.3697	-0.973	0.94967
hybrid 8x - <i>E. repens</i> 6x == 0	-2.0932	1.3211	-1.584	0.64770
hybrid 6x - <i>E. repens</i> 7x == 0	-18.8457	256.0020	-0.074	1.00000
hybrid 7x - <i>E. repens</i> 7x == 0	-20.7903	256.0019	-0.081	1.00000
hybrid 8x - <i>E. repens</i> 7x == 0	-21.5512	256.0018	-0.084	1.00000
hybrid 7x - hybrid 6x == 0	-1.9446	1.1874	-1.638	0.61135
hybrid 8x - hybrid 6x == 0	-2.7055	1.1746	-2.303	0.20547
hybrid 8x - hybrid 7x == 0	-0.7609	1.1511	-0.661	0.99295

autonomous vs. induced selfing:  $\chi^2$  (1) = 8.3748, Df resid = 258, p = 0.003805

Multiple Comparisons of Means: Tukey Contrasts:

	Estimate	Std. Error	z value	Pr(> z )
autonomous selfing - induced selfing == 0	0.7188	0.2576	2.79	0.00527

### **Seed set: hexaploid maternal plant**

maternal plant species:  $\chi^2$  (2) = 9.7565, Df resid = 126, p = 0.00761

Multiple Comparisons of Means: Tukey Contrasts:

	Estimate	Std. Error	z value	Pr(> z )
<i>E. repens</i> - <i>E. hispidus</i> == 0	2.0225	1.2586	1.607	0.2395
hybrid - <i>E. hispidus</i> == 0	2.6777	1.1345	2.360	0.0469
hybrid - <i>E. repens</i> == 0	0.6551	1.4544	0.450	0.8930

autonomous vs. induced selfing:  $\chi^2$  (1) = 2.9208, Df resid = 128, p = 0.08745

**Seed set: maternal plant *E. hispidus***

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maternal plant ploidy level:  $\chi^2 (1) = 4.407, Df \text{ resid} = 79, p = 0.03579$

Multiple Comparisons of Means: Tukey Contrasts:

	Estimate	Std. Error	z value	Pr(> z )
8x - 6x == 0	-1.3904	0.6583	-2.112	0.0347

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autonomous vs. induced selfing:

$\chi^2 (1) = 0.0006874, Df \text{ resid} = 79, p = 0.00176$

Multiple Comparisons of Means: Tukey Contrasts:

	Estimate	Std. Error	z value	Pr(> z )
selfing - induced == 0	1.0201	0.3261	3.128	0.00176

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**Seed set: maternal plant *E. repens***

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maternal plant ploidy level:  $\chi^2 (1) = 4.1199, Df \text{ resid} = 64, p = 0.04238$

autonomous vs. induced selfing:  $\chi^2 (1) = 3.9436, Df \text{ resid} = 64, p = 0.04705$

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**Seed set: maternal plant hybrid**

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maternal plant ploidy level:  $\chi^2 (2) = 6.0933, Df \text{ resid} = 108, p = 0.04752$

Multiple Comparisons of Means: Tukey Contrasts:

	Estimate	Std. Error	z value	Pr(> z )
7x - 6x == 0	-2.333	1.745	-1.337	0.3744
8x - 6x == 0	-3.761	1.826	-2.060	0.0983
8x - 7x == 0	-1.428	1.833	-0.779	0.7159

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autonomous vs. induced selfing:  $\chi^2 (1) = 0.61782, Df \text{ resid} = 109, p = 0.4319$

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**Seed set: autonomous selfing**

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maternal plants species (regardless on ploidy level):  $\chi^2 (2) = 6.407, Df \text{ resid} = 112, p = 0.04062$

Multiple Comparisons of Means: Tukey Contrasts:

	Estimate	Std. Error	z value	Pr(> z )
<i>E. repens</i> - <i>E. hispidus</i> == 0	1.7988	0.7542	2.385	0.0442
hybrid - <i>E. hispidus</i> == 0	0.9651	0.5575	1.731	0.1906



hybrid - <i>E. repens</i> == 0	-0.8337	0.7445	-1.120	0.4979
<hr/>				
<u>ploidy level of maternal plants (regardless on species):</u>	$\chi^2 (2) = 7.2812, Df \text{ resid} = 112, p = 0.02624$			
Multiple Comparisons of Means: Tukey Contrasts:				
	Estimate	Std. Error	z value	Pr(> z )
7x - 6x == 0	0.3314	0.8441	0.393	0.9178
8x - 6x == 0	-1.3140	0.6528	-2.013	0.1076
8x - 7x == 0	-1.6454	0.7852	-2.095	0.0894

**Seed set: induced selfing**

diploid vs. polyploid paternal plant	$\chi^2 (1) = 1.941, Df \text{ resid} = 142, p = 0.1636$			
<hr/>				
<u>ploidy level of maternal plants (regardless on species):</u>	$\chi^2 (2) = 8.2165, Df \text{ resid} = 141, p = 0.01644$			
Multiple Comparisons of Means: Tukey Contrasts:				
	Estimate	Std. Error	z value	Pr(> z )
7x - 6x == 0	1.157	1.431	0.809	0.6935
8x - 6x == 0	-2.995	1.209	-2.478	0.0344
8x - 7x == 0	-4.152	1.653	-2.511	0.0315
<hr/>				
<u>maternal plants species (regardless on ploidy level)</u>	$\chi^2 (2) = 17.269, Df \text{ resid} = 141, p = 0.0001779$			
Multiple Comparisons of Means: Tukey Contrasts:				
	Estimate	Std. Error	z value	Pr(> z )
<i>E. repens</i> - <i>E. hispidus</i> == 0	20.341	6311.326	0.003	0.99999
hybrid - <i>E. hispidus</i> == 0	3.087	1.085	2.846	0.00883
hybrid - <i>E. repens</i> == 0	-17.253	6311.326	-0.003	1.00000
<hr/>				
diploid vs. polyploid paternal plant	$\chi^2 (1) = 1.941, Df \text{ resid} = 142, p = 0.1636$			
<hr/>				
<u>ploidy level of paternal plants (regardless on species):</u>	$\chi^2 (3) = 15.142, Df \text{ resid} = 140, p = 0.0017$			
Multiple Comparisons of Means: Tukey Contrasts:				
	Estimate	Std. Error	z value	Pr(> z )

6x - 2x == 0	0.9742	0.3805	2.560	0.0434
7x - 2x == 0	1.4377	1.1397	1.261	0.5545
8x - 2x == 0	-0.9016	0.8214	-1.098	0.6617
7x - 6x == 0	0.4635	1.0894	0.425	0.9704
8x - 6x == 0	-1.8758	0.7328	-2.560	0.0434
8x - 7x == 0	-2.3394	1.3094	-1.787	0.2511

$\chi^2 (7) = 29.389$ , Df resid = 136, p =

paternal plants species (regardless on ploidy level): 0.0001229

Multiple Comparisons of Means: Tukey Contrasts:

	Estimate	Std. Error	z value	Pr(> z )
<i>E. repens</i> - <i>E. hispidus</i> == 0	0.35335	0.98805	0.358	1.0000
<i>Hordeum vulgare</i> - <i>E. hispidus</i> == 0	-1.12641	0.99041	-1.137	0.9398
hybrid - <i>E. hispidus</i> == 0	0.01617	0.91425	0.018	1.0000
<i>Secale cereale</i> - <i>E. hispidus</i> == 0	-0.03220	1.05058	-0.031	1.0000
<i>T. aestivum</i> - <i>E. hispidus</i> == 0	1.11796	0.94695	1.181	0.9271
<i>T. monococcum</i> - <i>E. hispidus</i> == 0	0.84444	1.18872	0.710	0.9960
× <i>Triticosecale</i> - <i>E. hispidus</i> == 0	0.92341	1.05152	0.878	0.9855
<i>Hordeum vulgare</i> - <i>E. repens</i> == 0	-1.47975	0.57292	-2.583	0.1436
hybrid - <i>E. repens</i> == 0	-0.33717	0.52468	-0.643	0.9979
<i>Secale cereale</i> - <i>E. repens</i> == 0	-0.38555	0.68130	-0.566	0.9991
<i>T. aestivum</i> - <i>E. repens</i> == 0	0.76462	0.55956	1.366	0.8545
<i>T. monococcum</i> - <i>E. repens</i> == 0	0.49110	0.86670	0.567	0.9990
× <i>Triticosecale</i> - <i>E. repens</i> == 0	0.57007	0.70786	0.805	0.9913
hybrid - <i>Hordeum vulgare</i> == 0	1.14258	0.45951	2.487	0.1796
<i>Secale cereale</i> - <i>Hordeum vulgare</i> == 0	1.09420	0.53618	2.041	0.4211
<i>T. aestivum</i> - <i>Hordeum vulgare</i> == 0	2.24437	0.48971	4.583	<0.001
<i>T. monococcum</i> - <i>Hordeum vulgare</i> == 0	1.97085	0.76714	2.569	0.1487
× <i>Triticosecale</i> - <i>Hordeum vulgare</i> == 0	2.04982	0.69743	2.939	0.0567
<i>Secale cereale</i> - hybrid == 0	-0.04837	0.58803	-0.082	1.0000
<i>T. aestivum</i> - hybrid == 0	1.10179	0.36763	2.997	0.0479

<i>T. monococcum</i> - hybrid == 0	0.82827	0.80150	1.033	0.9637
× <i>Triticosecale</i> - hybrid == 0	0.90724	0.57101	1.589	0.7299
<i>T. aestivum</i> - <i>Secale cereale</i> == 0	1.15016	0.61234	1.878	0.5319
<i>T. monococcum</i> - <i>Secale cereale</i> == 0	0.87664	0.85208	1.029	0.9646
× <i>Triticosecale</i> - <i>Secale cereale</i> == 0	0.95561	0.78691	1.214	0.9162
<i>T. monococcum</i> - <i>T. aestivum</i> == 0	-0.27352	0.82062	-0.333	1.0000
× <i>Triticosecale</i> - <i>T. aestivum</i> == 0	-0.19455	0.62448	-0.312	1.0000
× <i>Triticosecale</i> - <i>T. monococcum</i> == 0	0.07897	0.95564	0.083	1.0000

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**Electronic Appendix 7.** – Results of generalized linear mixed-effects models (GLMM) testing the effect of ploidy level and species of maternal plant and species of paternal plant on seed set and germination rate. Seed set refers to number of seeds/number of florets ratio of each single pollinated spike.

Model used	GLMM	GLMM
dataset	seed set: entire	germination rate: entire
<u>Maternal plant:</u>		
maternal plant species	$\chi^2 (2) = 12.247$ , Df resid = 246, p = 0.002191	$\chi^2 (2) = 0.0631$ , Df resid = 37, p = 0.9689
maternal plant ploidy	$\chi^2 (2) = 5.3194$ , Df resid = 246, p = 0.06997	$\chi^2 (2) = 0.0851$ , Df resid = 37, p = 0.9584
<u>Paternal plant:</u>		
paternal plant species	$\chi^2 (6) = 26.31$ , Df resid = 250, p = 0.0001949	$\chi^2 (6) = 0.2$ , Df resid = 41, p = 0.9998
paternal plant ploidy	$\chi^2 (2) = 0.8474$ , Df resid = 246, p = 0.6546	$\chi^2 (2) = 0.2279$ , Df resid = 37, p = 0.8923
autonomous vs. induced selfing	$\chi^2 (1) = 0.4382$ , Df resid = 245, p = 0.508	$\chi^2 (1) = 0.0011$ , Df resid = 36, p = 0.9733
ploidy of pollination (homo x heteroploid)	$\chi^2 (1) = 0.2986$ , Df resid = 245, p = 0.5848	$\chi^2 (1) = 0.0544$ , Df resid = 36, p = 0.8156

Maternal plant individual was treated as a random factor in GLMM models. Numbers are  $\chi^2$  values obtained from LRT tests (GLMM).

**Electronic Appendix 8.** – Results of GLMM analysis of entire dataset only with presence or absence of evolved seeds (successful vs. unsuccessful crosses) and GLMM analysis of the successful crosses only.

### GLMM

#### Seedset: entire seedset; 0/1 seeds

maternal plant species	$\chi^2 (2) = 19.401$ , Df resid = 246, p = 6.127e-05
maternal plant ploidy	$\chi^2 (2) = 4.7052$ , Df resid = 246, p = 0.09512
paternal plant species	$\chi^2 (6) = 9.7404$ , Df resid = 250, p = 0.136
paternal plant ploidy	$\chi^2 (2) = 0.9692$ , Df resid = 246, p = 0.616
autonomous vs. induced selfing	$\chi^2 (1) = 2.4503$ , Df resid = 245, p = 0.1175
ploidy of pollination (homo x heteroploid)	$\chi^2 (1) = 0.008$ , Df resid = 245, p = 0.9287

#### Seedset: only successful pollinated spikes; number of seeds combined with number of florets

maternal plant species	$\chi^2 (2) = 0.5825$ , Df resid = 37, p = 0.7473
maternal plant ploidy	$\chi^2 (2) = 5.0465$ , Df resid = 37, p = 0.0802
paternal plant species	$\chi^2 (6) = 16.055$ , Df resid = 41, p = 0.01346

paternal plant ploidy	$\chi^2 (2) = 1.2398, \text{Df resid} = 37, p = 0.538$
autonomous vs. induced selfing	$\chi^2 (1) = 7.6176, \text{Df resid} = 36, p = 0.00578$
ploidy of pollination (homo x heteroploid)	$\chi^2 (1) = 1.2693, \text{Df resid} = 36, p = 0.2599$

Maternal plant individual was treated as a random factor in GLMM models. Numbers are  $\chi^2$  values obtained from LRT tests (GLMM).