

Applied biotechnology to combat the leaf rust caused by *Puccinia tritricina* and powdery mildew caused by *Blumeria graminis* in polish wheat cultivars



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INTRODUCTION AND GOAL

The main purpose of crop production is to achieve the highest possible yield while minimizing use of pesticides. Growing varieties with beneficial traits, also with a high potential of yield is closely connected with their resistance to fungal and viral diseases. The important role is playing breeding for resistance. At present, many tools of molecular genetics and biotechnology exist which can be successfully used to obtain plant disease resistance.

Leaf rust and powdery mildew are occurring in all wheat growing regions of Poland and every year contribute to losses in cereal production. The main reason for these losses is the cultivation of varieties that are susceptible to these diseases.

The aim of presented research is:

1. Pyramiding of leaf rust and powdery mildew resistance genes in a one genotype;
2. To clarify the location Lr55 gene in wheat genome;
3. Screening for resistance leaf rust and powdery mildew.



STAGE 1. Pyramiding of leaf rust and powdery mildew resistance genes in a one genotype

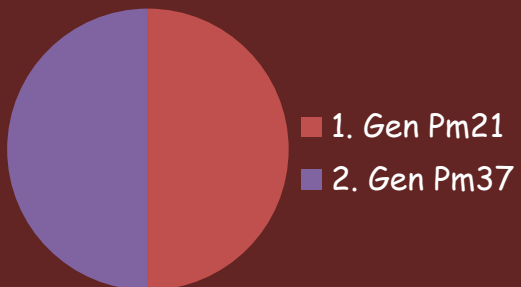
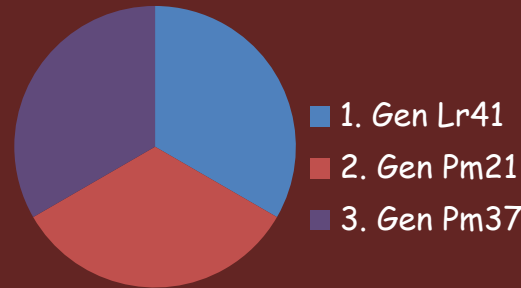
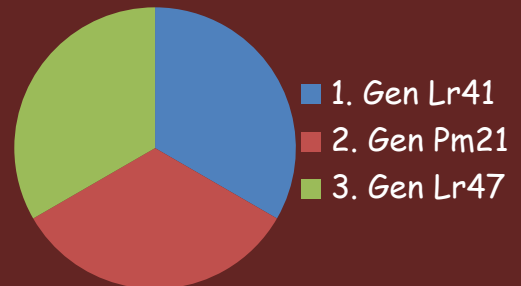
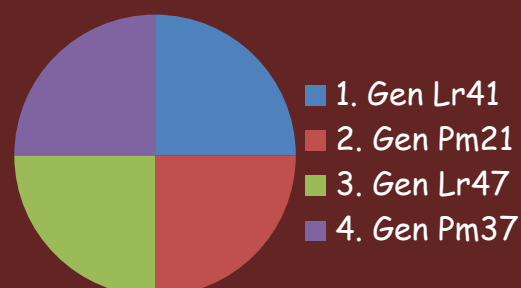
PARENTAL PLANT

1. Recurrent Parents (RP) - 5 varieties of winter wheat: Nadobna, Bogatka, Meteor, Lexus, RAH979.
2. Donor lines:
 - line KS90WGR10 = Lr39 (=Lr41) gene, as the sources of resistance to leaf rust derived from *T. tauschii*,
 - line UC996610010 = Lr47 gene, as the sources of resistance to leaf rust derived from *T. speltoides*,
 - line KS04WGR45 = Lr55 gene, as the sources of resistance to leaf rust derived from *Elymus trachycaulis*,
 - line Yangmai 5 = Pm21 gene, as the sources of resistance to powdery mildew derived from *Dasyphyrum villosum*,
 - line NC99BGTA11 = Pm37 gene, as the sources of resistance to powdery mildew derived from *T. timopheevii*.



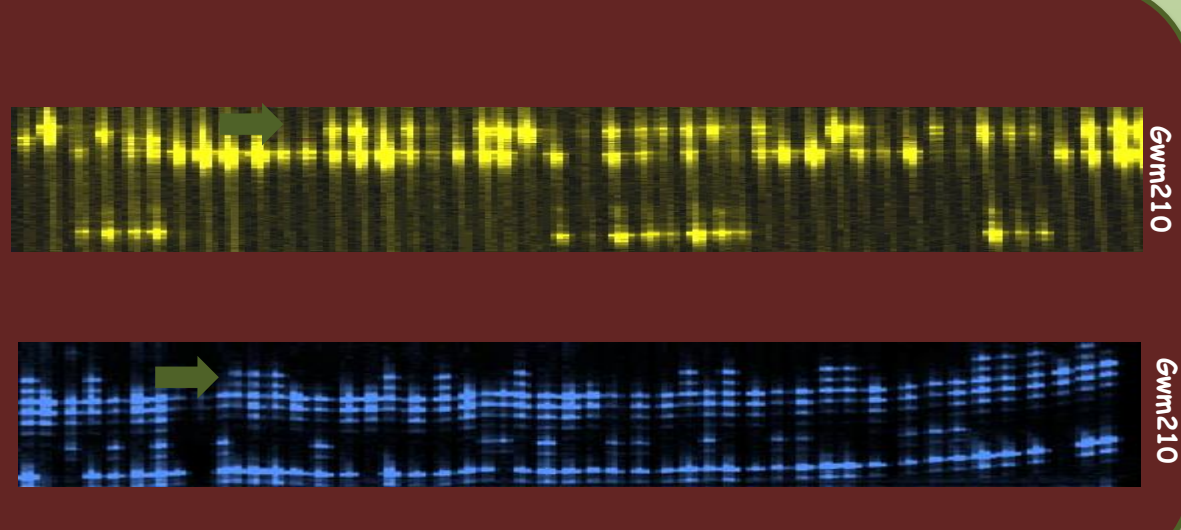
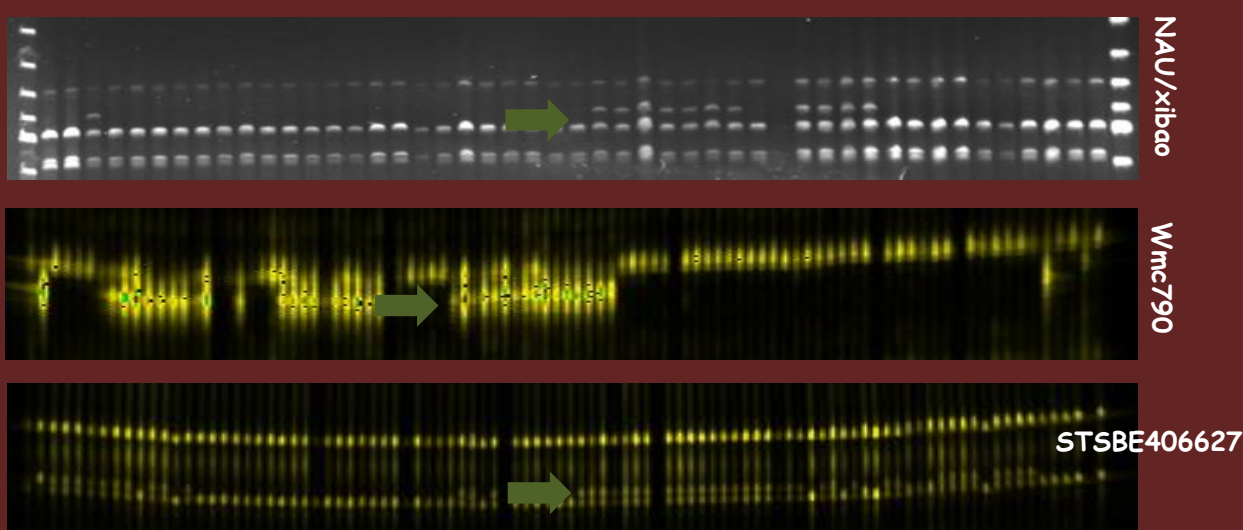
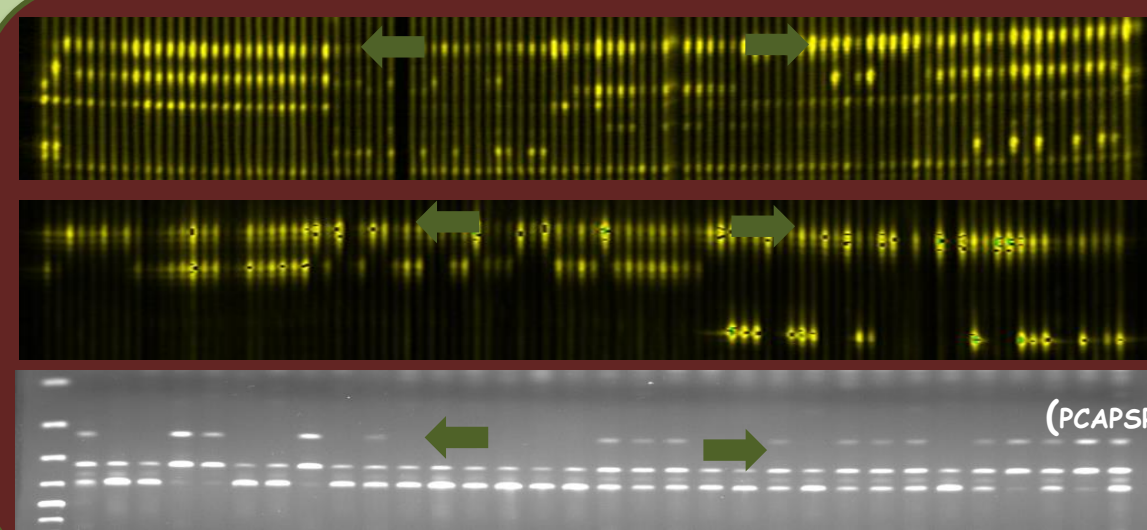
UP TO NOW, THE FOLLOWING COMBINATIONS OF RESISTANCE GENES WERE OBTAINED

No	Population	Gene Pyramiding using molecular markers	Goals for 2016
1.	BIO4_2a	(Lr41+Pm21+Lr47) x Bogatka x RAH979 x Lr47	x Bogatka
2.	BIO4_2b_1	(Lr41+Pm21+Lr47) x Bogatka x RAH979 x Nadobna x Lr47	x Bogatka
3.	BIO4_2b_2	(Lr41+Pm21+Lr47) x Bogatka x RAH979 x Nadobna x Nadobna	in progress x Pm37
4.	BIO2_2a	(Lr41+Pm21+Lr47) x Bogatka x Nadobna x Lr47	in progress x Bogatka v Nadobna
5.	BIO2_2b_1	(Lr41+Pm21+Lr47) x Nadobna x Bogatka x Lr47	x Nadobna
6.	BIO2_2b_2	(Lr41+Pm21+Lr47) x Nadobna x Bogatka x Bogatka	in progress x Pm37
7.	BIO8_1_1	(Lr41+Pm21+Pm37) x Bogatka x Lexus x Meteor x Pm37	x Bogatka
8.	BIO8_1_2	(Lr41+Pm21+Pm37) x Bogatka x Lexus x Meteor x Bogatka	in progress x Lr47
9.	BIO8_3	(Lr41+Pm21+Pm37) x Bogatka x Lexus x Meteor x Nadobna x Pm37	x Nadobna
10.	BIO8_2a	(Lr41+Pm21+Pm37) x Bogatka x Lexus x Meteor x Pm37	x Bogatka
11.	BIO8_2b	(Lr41+Pm21+Pm37) x Bogatka x Lexus x Meteor x Pm37	x Bogatka
12.	BIO8_4b	(Lr41+Pm21+Pm37+Lr47) x Bogatka x Lexus x Meteor x Nadobna x Pm37	x Bogatka
13.	BIO8_4a	(Lr41+Pm21+Pm37+Lr47) x Bogatka x Lexus x Meteor x Pm37	x Bogatka
14.	BIO8_4c	(Lr41+Pm21+Pm37+Lr47) x Bogatka x Lexus x Meteor x (Pm36 un)	in progress



MOLECULAR ANALYSIS - FOREGROUND SELECTION

1. Screening Lr39(= gene: Gdm35, Barc124, Gwm210, Gwm261, Gwm296.
2. Screening Lr47 gene: (PCAPSR+PS10L+PS10L2), Gwm60.
3. Screening Lr55 gene: DArT (molecular mapping in progress)
4. Screening Pm21 gene: NAU/xibao.
5. Screening Pm37 gene: Gwm332, Wmc790, STSBE406627.



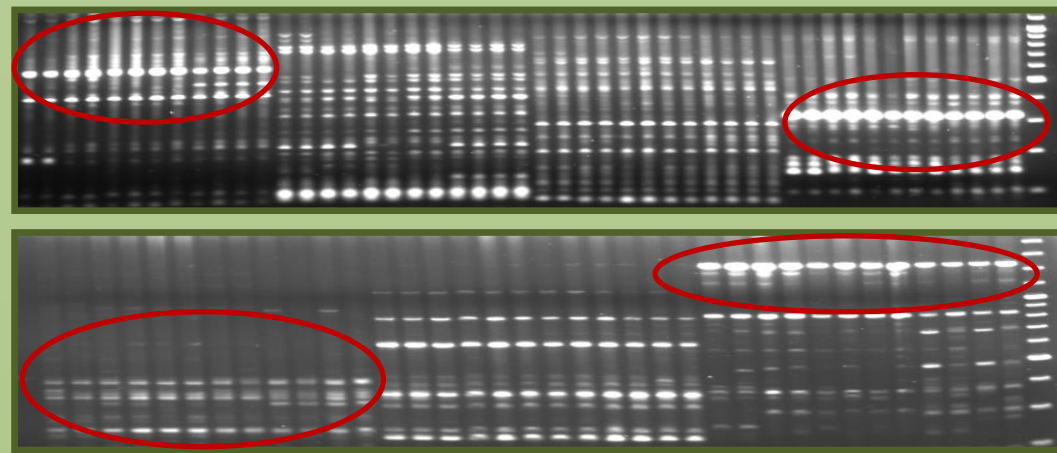
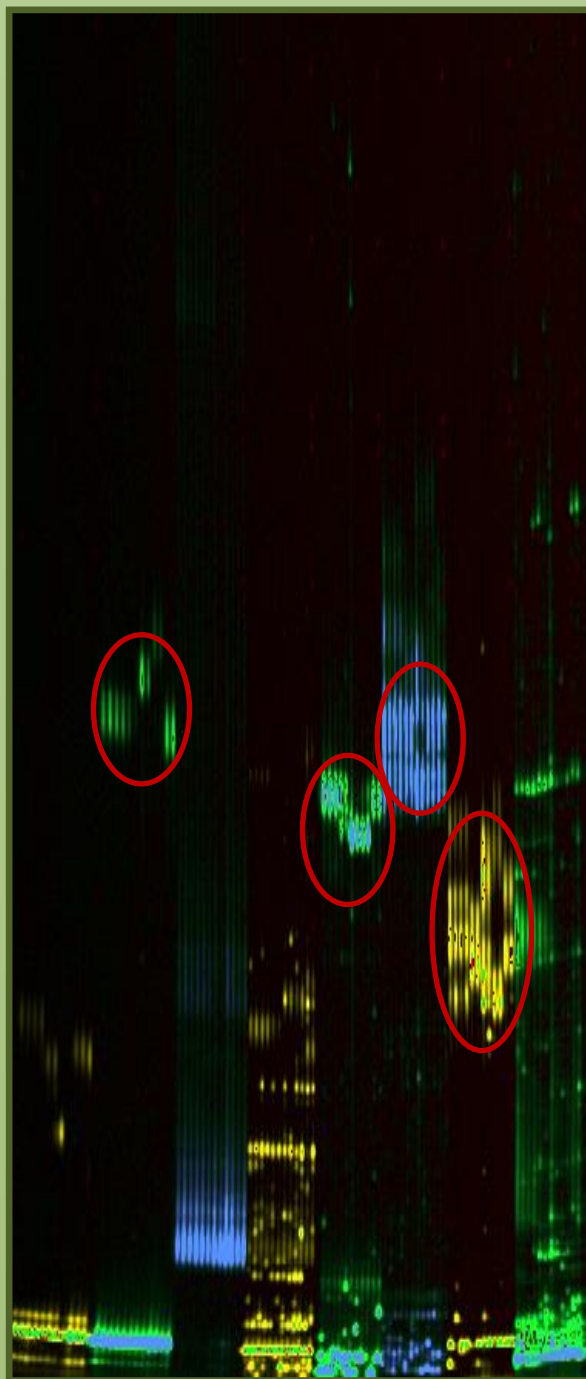
STAGE 2. To clarify the location Lr55 gene in wheat genome

MOLECULAR MAPPING

1. Four mapping populations: F₂ (Lr55 x Bogatka); (Lr55 x Nadobna); (Lr55 x Kampana); (Lr55 x Muszelka).
2. SSRs: 65 polymorphic markers.
3. DArTs: 18 markers and 15 restriction enzymes.

UP TO NOW, THE FOLLOWING COMBINATIONS OF RESTRICTION ENZYMES WERE DONE

DArTs	BamHI	MboI	MseI /TruI	RsaI	AluI	HinfI	BsuRI/ HaeIII	EcoRI	PstI
wpt6427_3	+	-	-	-	-	-	-	-	-
wpt8986_1	-	+	-	+	-	-	+	+	-
wpt5678_1	-	-	-	-	-	-	-	-	-
wpt5678_3	-	+	+	+	+	-	+	+	-
wpt6777_1	+	+	+	+	+	+	+	+	+
wpt6777_3	-	-	-	-	-	-	-	-	-
wpt5801_1	-	+	-	-	-	-	+	-	-
wpt5801_2	-	+	+	+	+	-	+	+	+



SUMMARY

In summary, pyramiding of several resistance genes using marker assisted proved that this strategy is more efficient and speed up recurrent genome recovery. However, this approach can not completely replace the traditional methods, especially for verification of the results in pathology tests. The resistance tests using differential set of host and pathogen are essential in MAS, especially when the foreground selection results can be sometimes problematic to interpret. The practical outcome of presented study are, the newly produced homozygous wheat lines which can be used as the source of effective resistance for the leaf rust and powdery mildew in breeding programs.

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STAGE 3. Screening for resistance leaf rust and powdery mildew

REACTION OF BLUMERIA GRAMINIS FF. SSP ISOLATES COLLECTED FROM TRITICALE

Cultivar / lines

Isolates	Axiolator Pm1	Avalon Pm2	Assuan Pm3a	Chul Pm3b	Sonara Pm3c	Kallari Pm3d	Weihenst Pm4	Kormoran Pm5	Pm6	Transeer Pm7	Disponent Pm8	Maris Unt Pm2+6	Kudatt Pm3a+4b	Boxer Pm4b+5	Sorbas Pm4b+8	Kronjuwel Pm	Granada Pm5+8	Apollo Pm2+4b+8	Soppo Pm1+2+4b+9	Amigo Pm17	Pm21	Pm22	Pm23	Pm29	Pm35	Pm37	Pm38	Pm39	Pm39(L+U46)	Nimbus	Maris Templer 2013Pm	XX186 Pm19/2013	Drabber Pm12,3,4,6,9
1476	0	0	0	0	0	0	4	4	4	2	0	0	4	0	0	0	4	0	0	4	4	0	0	2	4	4	4	4	4	4	2	2	0
1477	0	4	4	0	0	4	4	4	4	4	4	4	4	0	0	0	4	0	0	4	4	0	0	2	4	4	4	4	4	4	2	0	
1478	0	4	0	4	4	4	4	4	4	4	4	4	4	0	0	0	4	0	0	4	4	0	0	4	4	4	4	4	4	4	4	0	
1479	4	0	0	0	4	0	4	4	4	4	2	4	0	4	4	4	4	0	0	4	4	0	4	2	0	0	4	4	4	4	4	4	
1480	0	4	0	0	0	0	4	4	2	0	4	0	0	0	0	0	4	0	0	4	4	0	0	4	0	0	4	4	4	4	4	4	
1481	0	4	2	0	4	0	4	4	4	4	0	4	4	0	0	0	4	0	0	4	4	0	0	4	0	0	4	4	4	4	4	4	