

RENSEQ IN POTATO: COVERING THE SPECTRUM OF APPLIED AND FUNDAMENTAL RESEARCH



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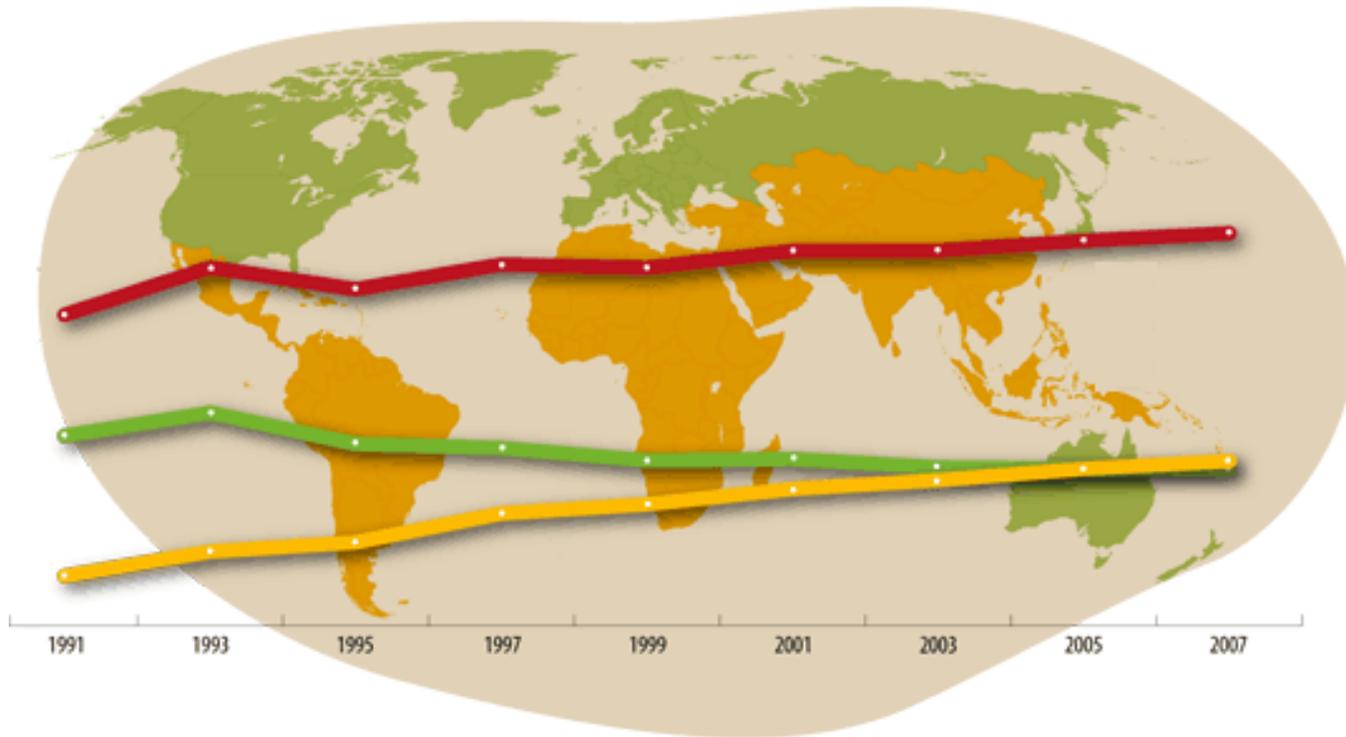
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Arbor RenSeq Webinar; March 10th 2022



University
of Dundee

POTATO IS THE MOST IMPORTANT NON-CEREAL FOOD CROP



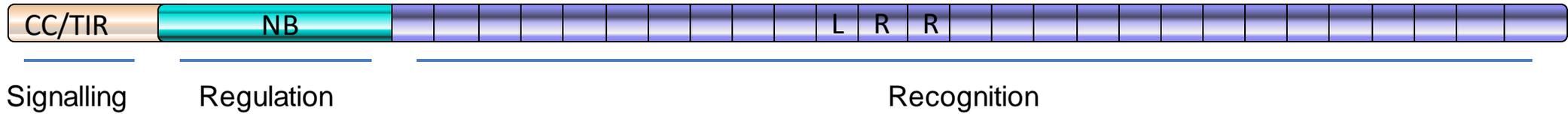
THE FAMINE IN IRELAND.— FUNERAL AT SKIBBEREEN.— FROM A SKETCH BY MR. H. SMITH, CORK.



BEGGAR-WOMAN AND CHILDREN.

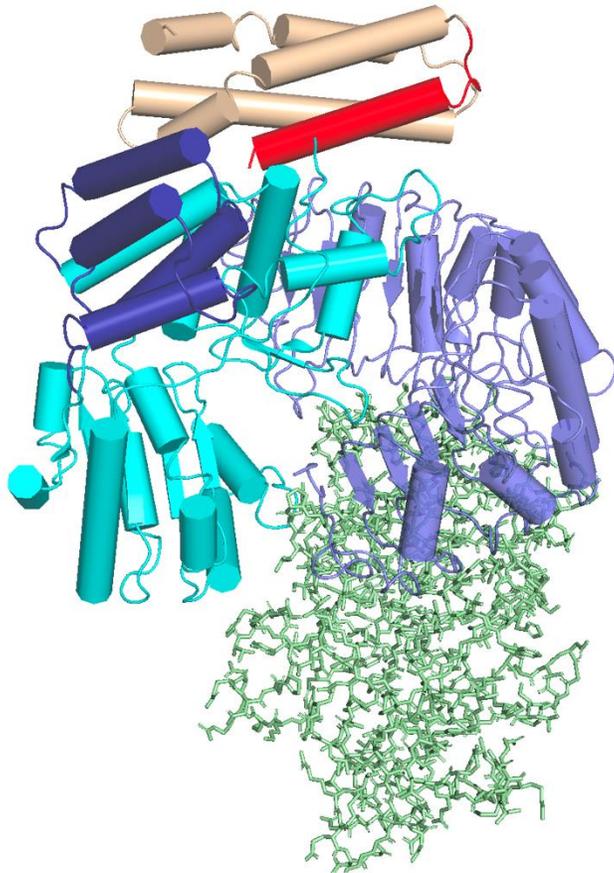
- Consumed by a billion people worldwide
- Global potato production exceeds 300 million metric tons
- Diseases can have major global impact: Late blight - Famine in the late 1840s
- PCN: threatens the seed potato industry
- More than 25% of the potato crop is lost to disease

FUNCTIONAL NB-LRR RESISTANCE GENES CONTROL DIVERSE PATHOGENS

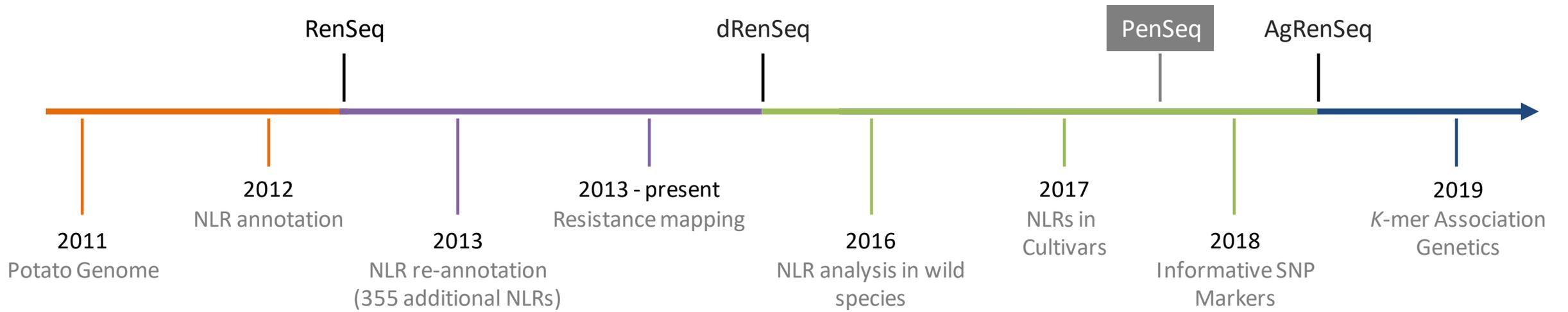


Effective against diverse and taxonomically unrelated pathogens

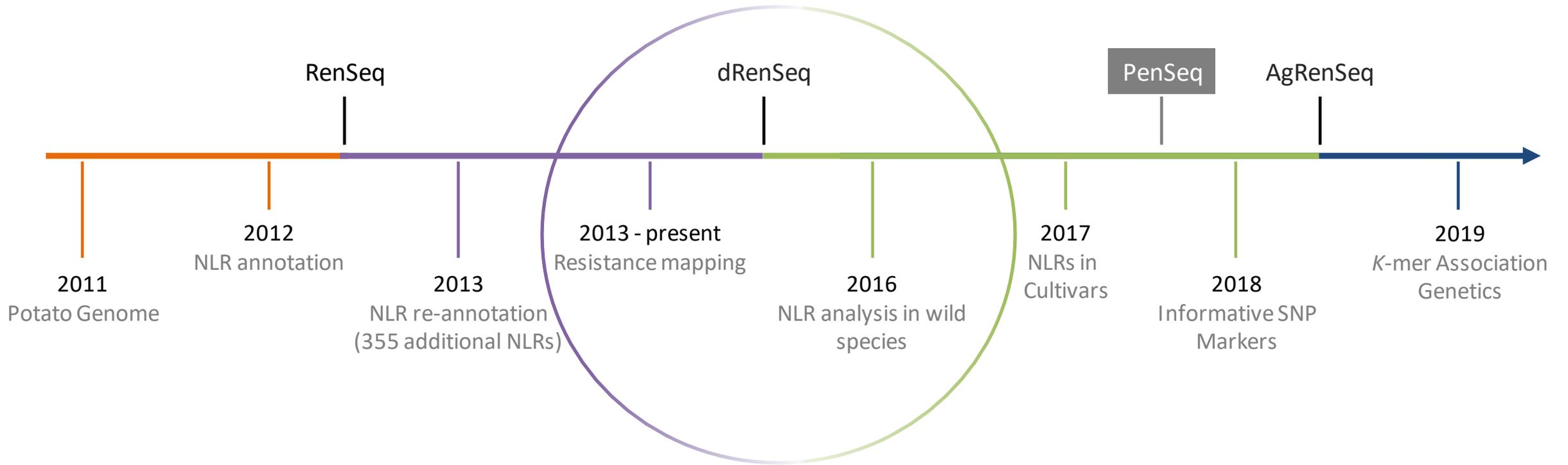
- Bacteria
- **Viruses**
- **Nematodes**
- Insects
- **Filamentous fungi**
- **Oomycetes** (including field resistances; Rui *et al.*, 2018 JXB)



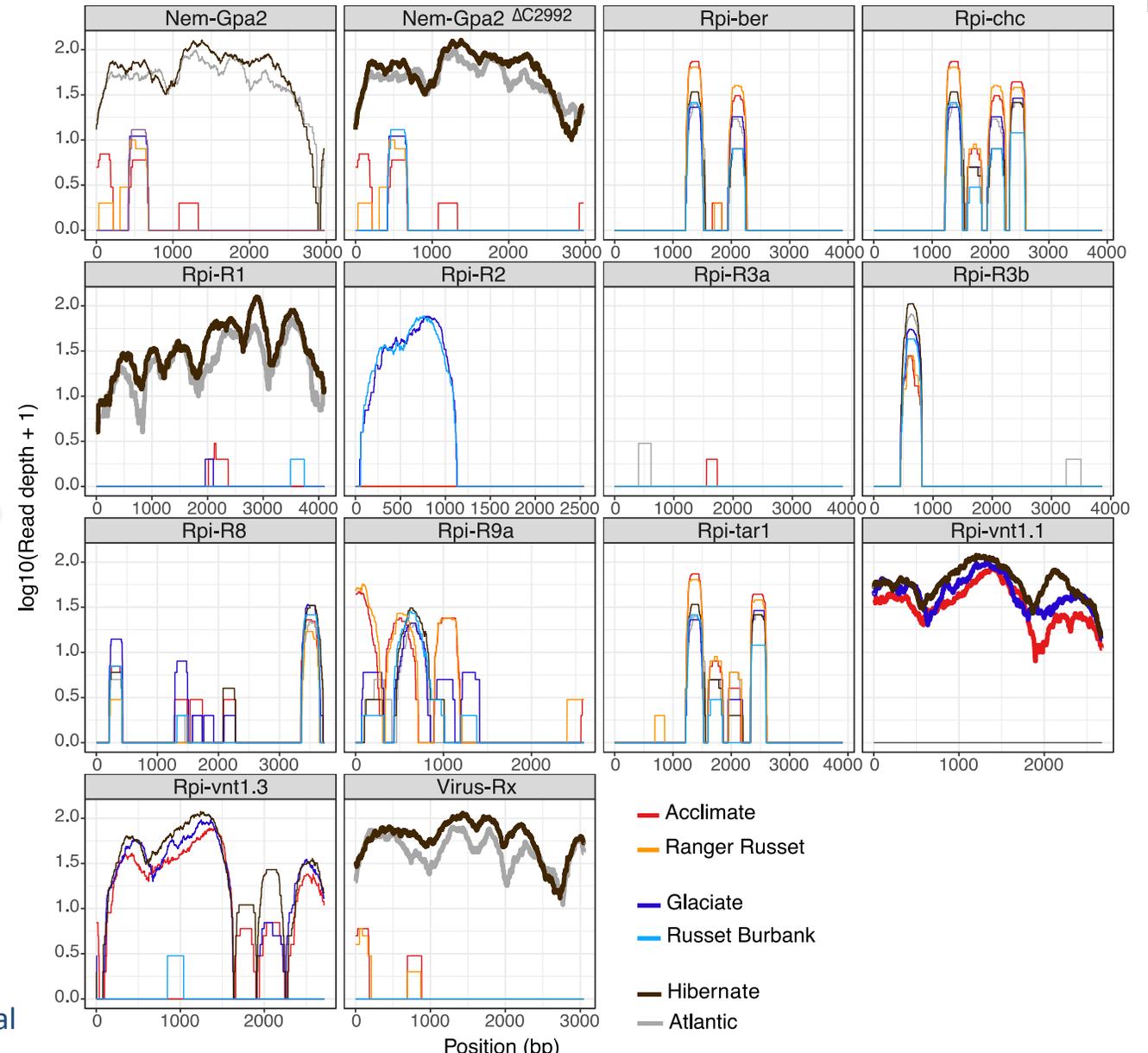
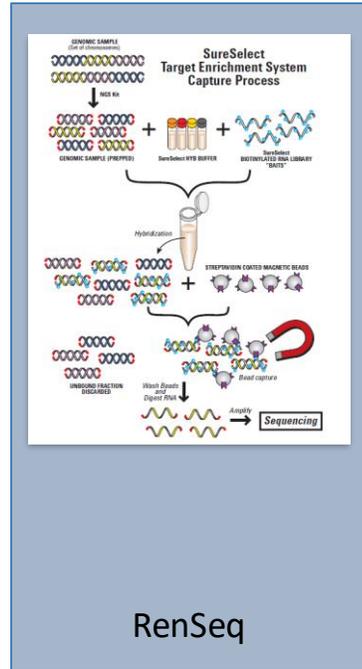
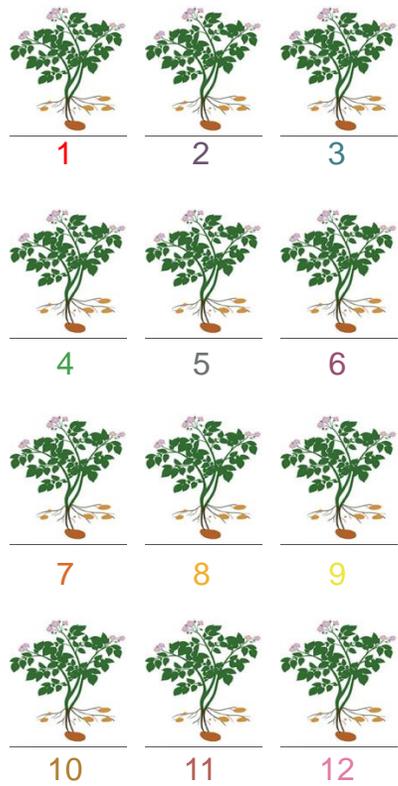
RENSEQ : RESEARCH AND BREEDING TOOL



dRENSeq AND RENSeq-BASED ANALYSIS OF WILD SPECIES



DIAGNOSTIC RENSEQ (dRENSEQ) TO TRACK NLRs IN VARIETIES AND WILD SPECIES



IDENTIFYING NOVEL SOURCES OF RESISTANCES

- Commonwealth Potato Collection (CPC)
- started in 1938 & 1939 with collecting expeditions to Mexico and South America
- now ~1500 accessions, >80 species (1/3 cultivated landraces)
- Maintained as true seeds on a 20 year (+) rotation
- Annually 60-80 accessions are rejuvenated



RENSEQ DATA - WILD SPECIES



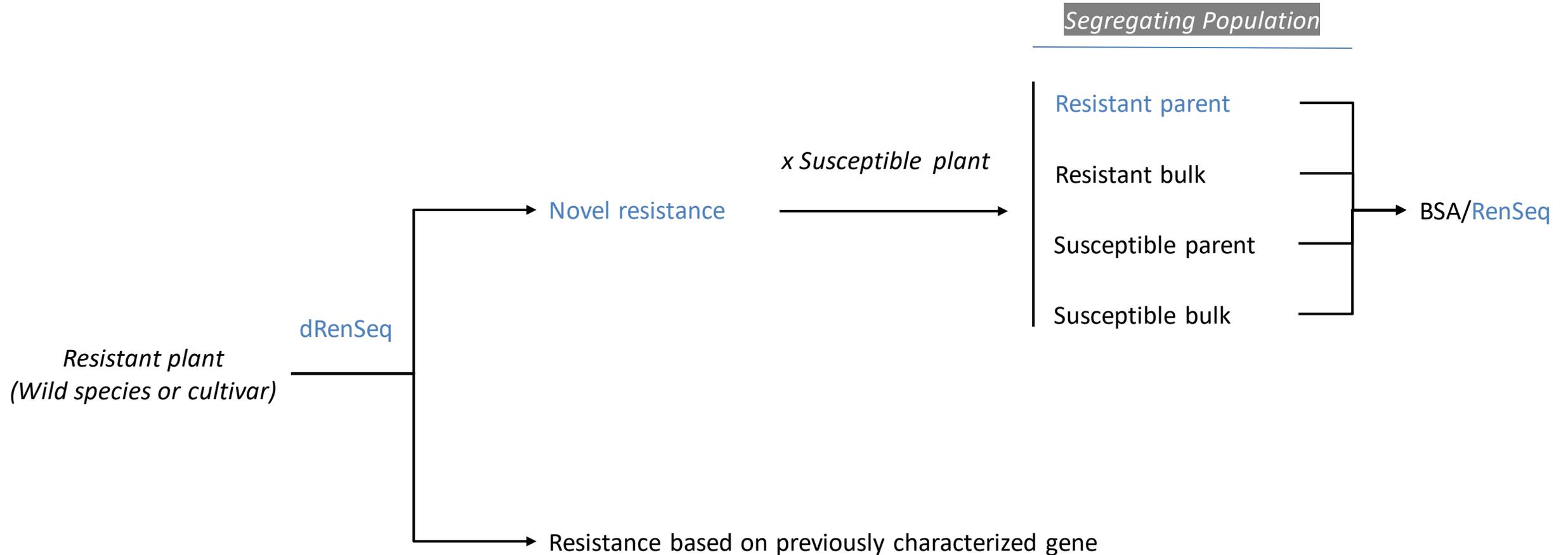
Illumina RenSeq:

- ~ 150 diploid accession from the CPC
- ~ 80 accession from the International Potato Centre (CIP)

PacBio RenSeq:

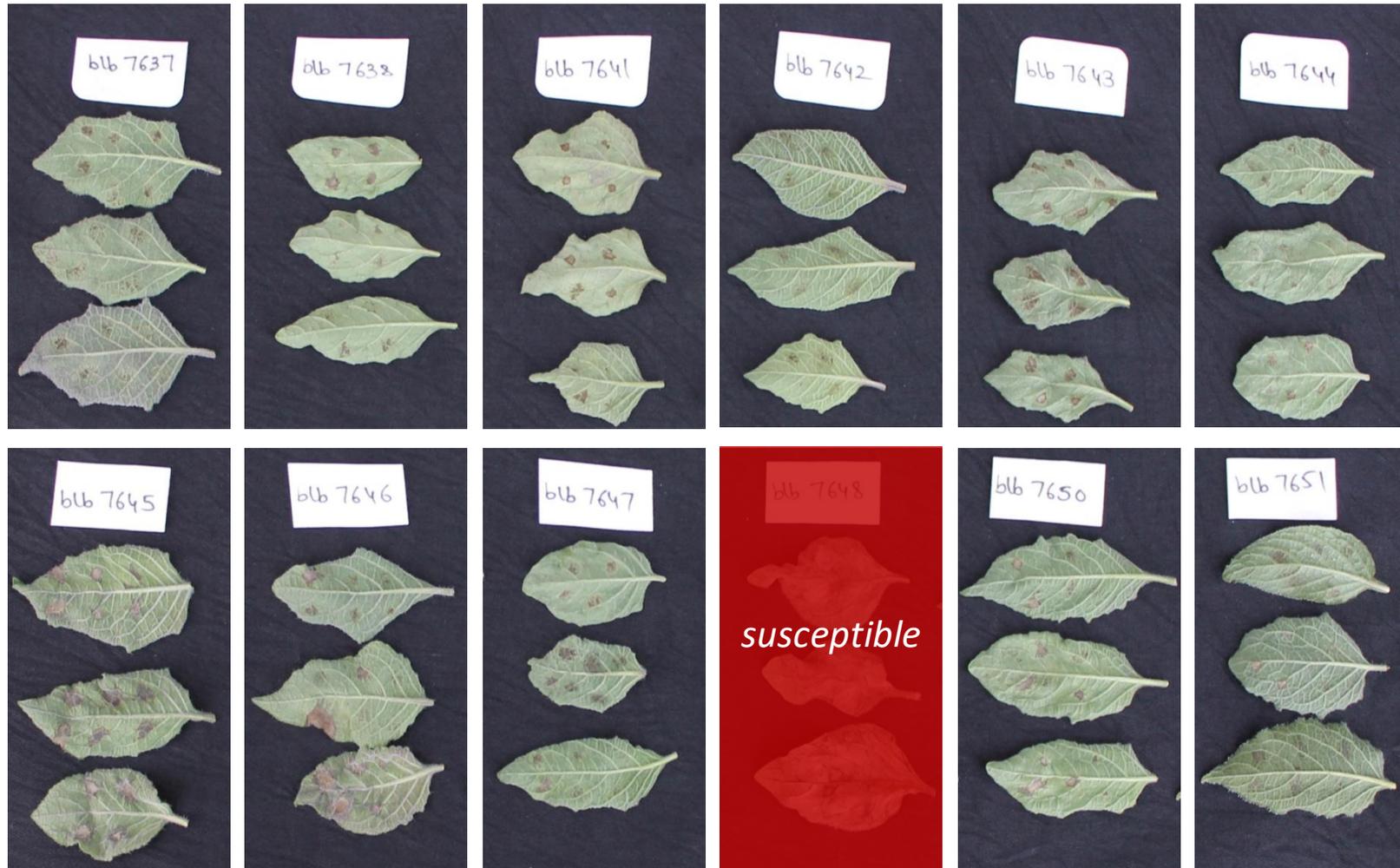
- ~ 96 diploid accession from the CPC
 - Phenotypic evaluation for blight in 2021
 - Phenotypic evaluation for nematode and PVY resistance in 2022
- Can we identify new resistances?

DRENSAQ WORKFLOW TO PRIORITISE NOVEL RESISTANCES



IDENTIFYING NOVEL SOURCES OF RESISTANCES

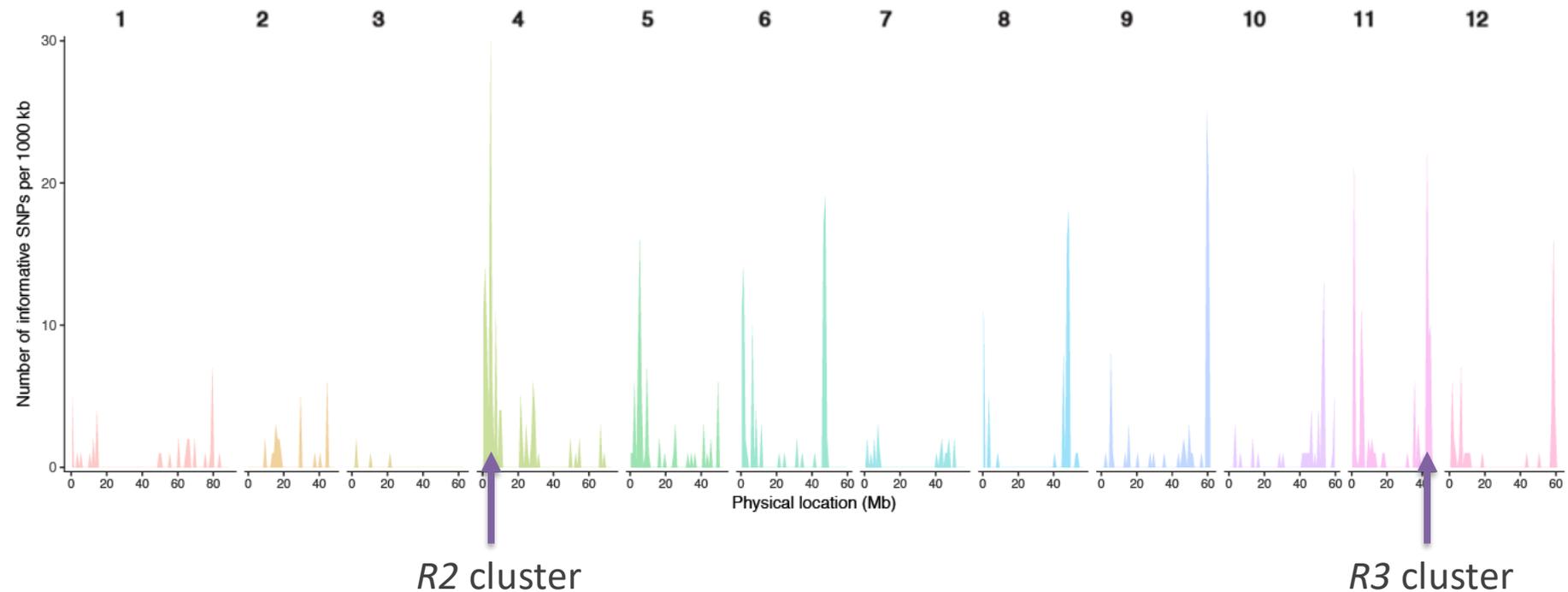
12 *Solanum bulbocastanum* accessions from the Commonwealth Potato Collection



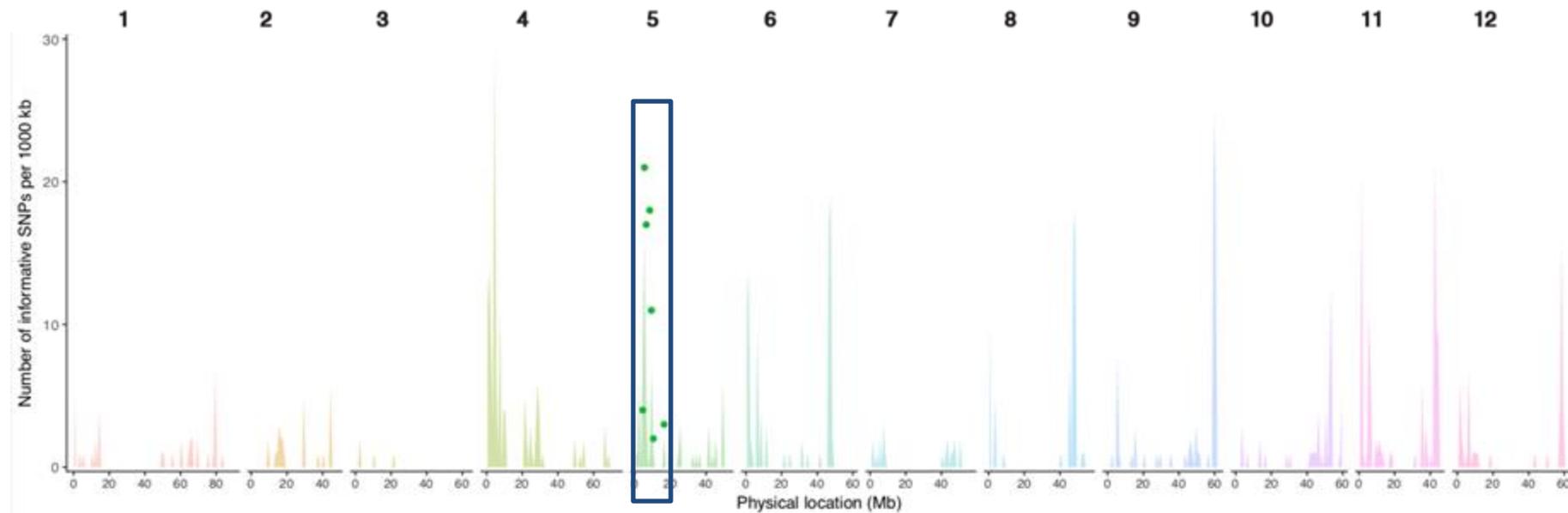
dRENSEQ IDENTIFIES TWO NOVEL RESISTANCES IN 7641 AND 7650



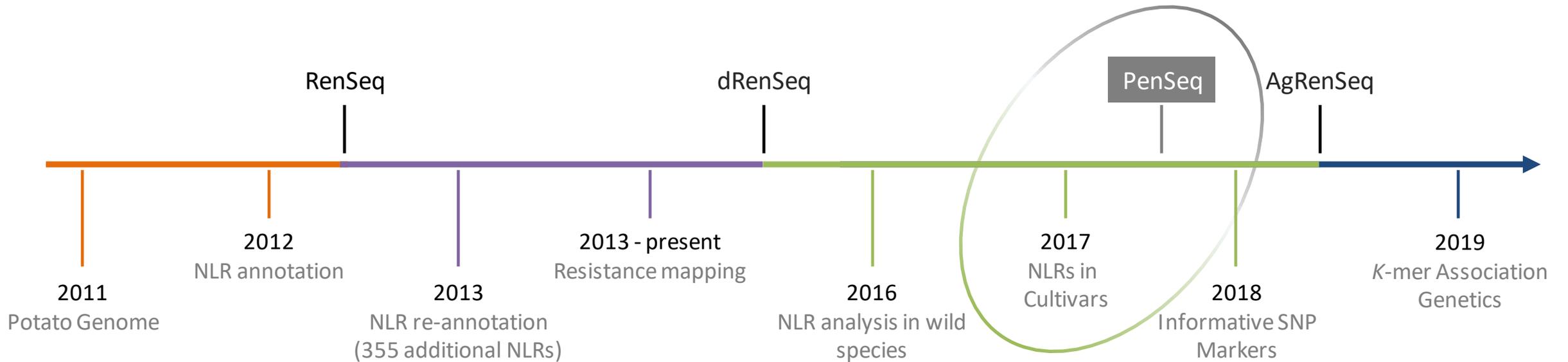
BULKED SEGREGANT ANALYSIS TO MAP RESISTANCE: *S. BULBOCASTANUM* 7650 x *S. MICHOCANUM*



BULKED SEGREGANT ANALYSIS TO MAP RESISTANCE: *S. BULBOCASTANUM* 7650 x *S. MICHOCANUM*



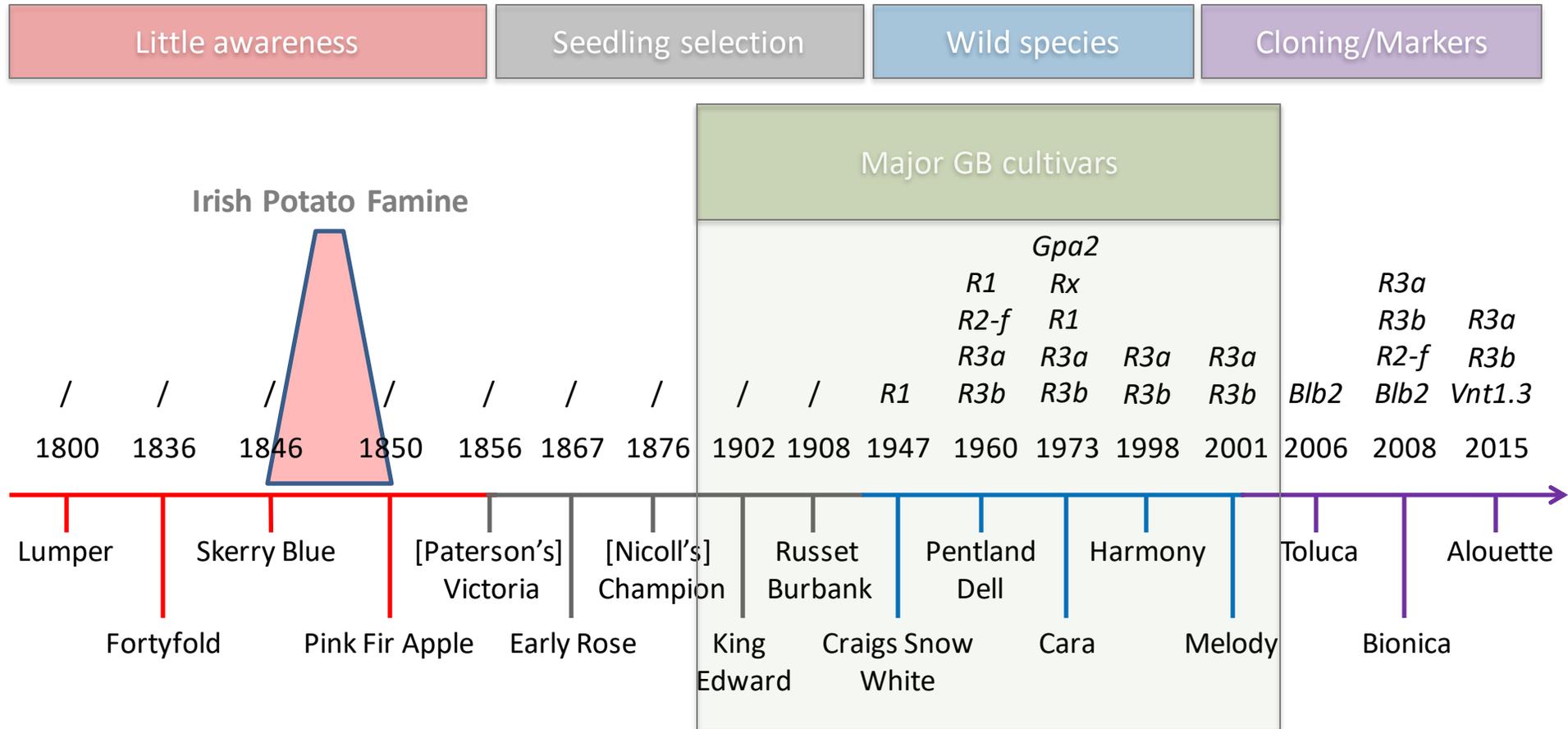
RENSEQ: STUDYING NLR DEPLOYMENT IN CULTIVARS



- RenSeq of 275 varieties (Illumina)
- RenSeq of 97 breeding clones (Illumina)
- RenSeq of 10 varieties (PacBio)

HISTORICAL PATTERN OF NLR DEPLOYMENT IN UK CULTIVARS

Breeding for disease resistance

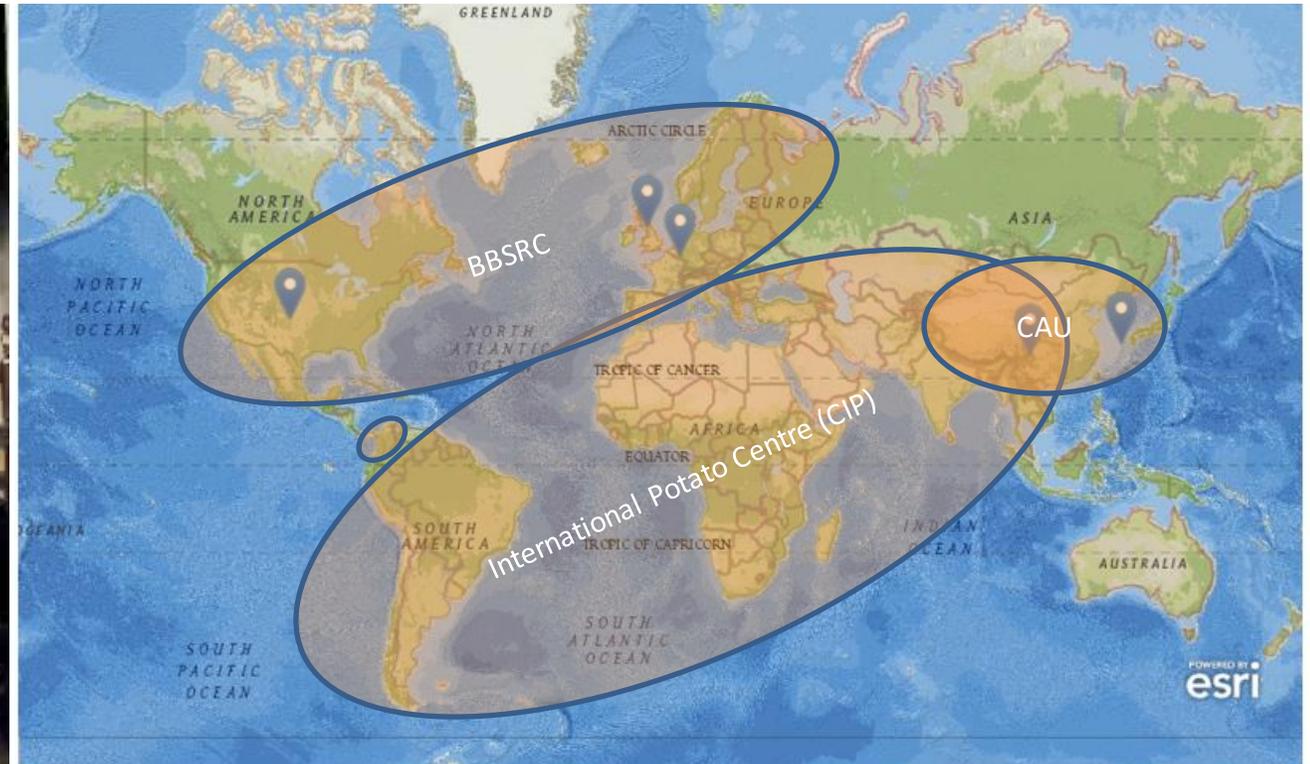


RENSEQ DATA FOR VARIETIES/BREEDING CLONES

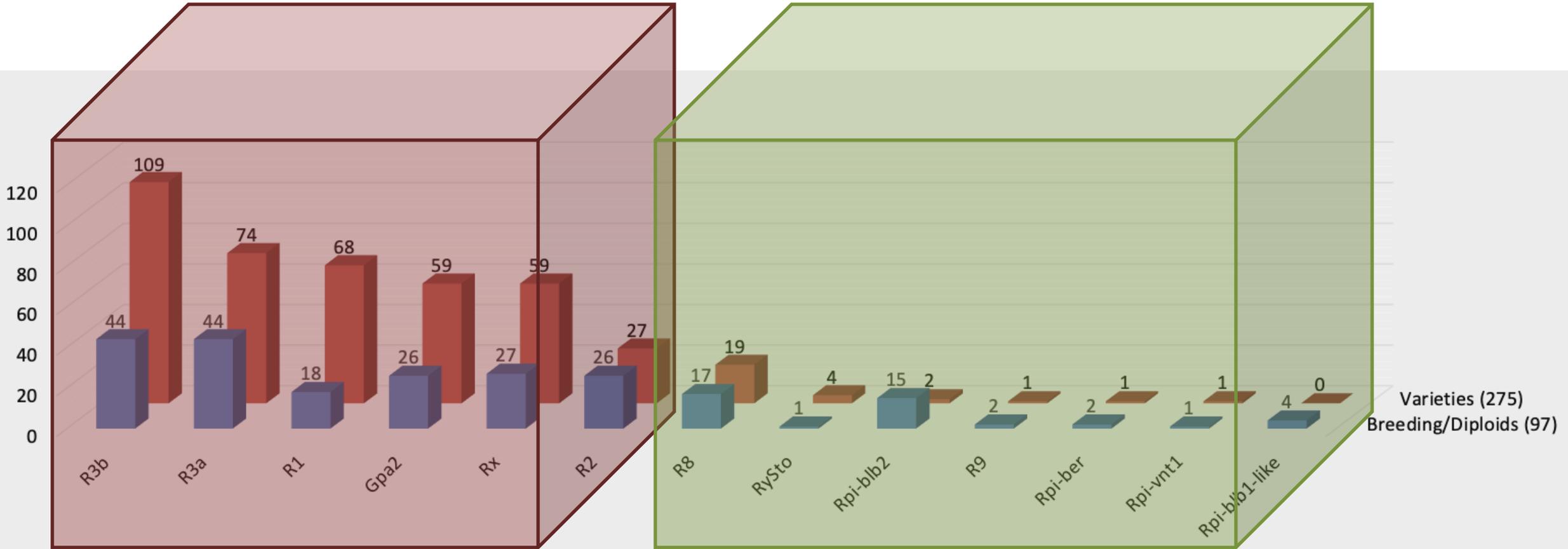
Historical Deployment of NLRs



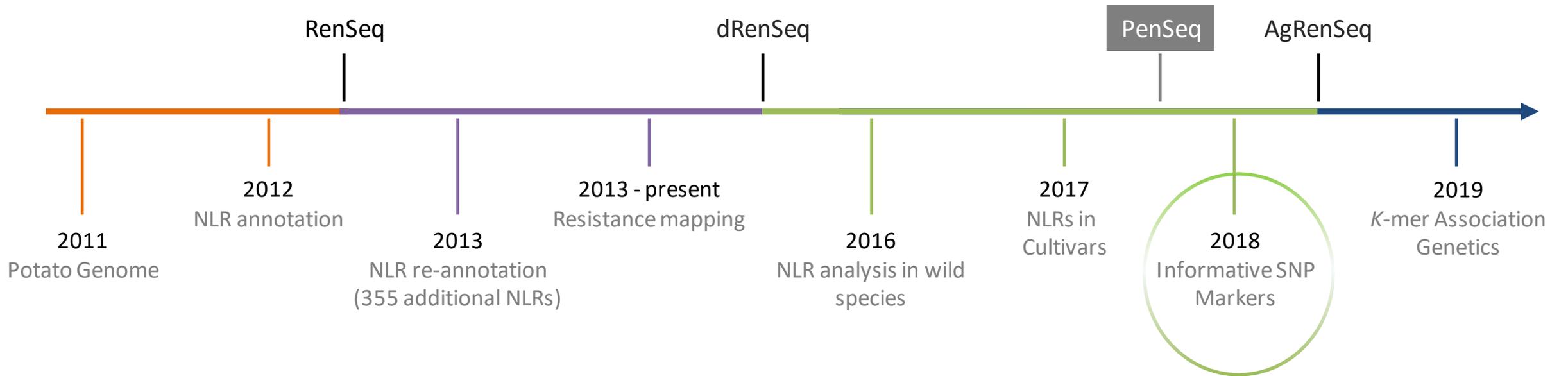
Geographical Deployment of NLRs



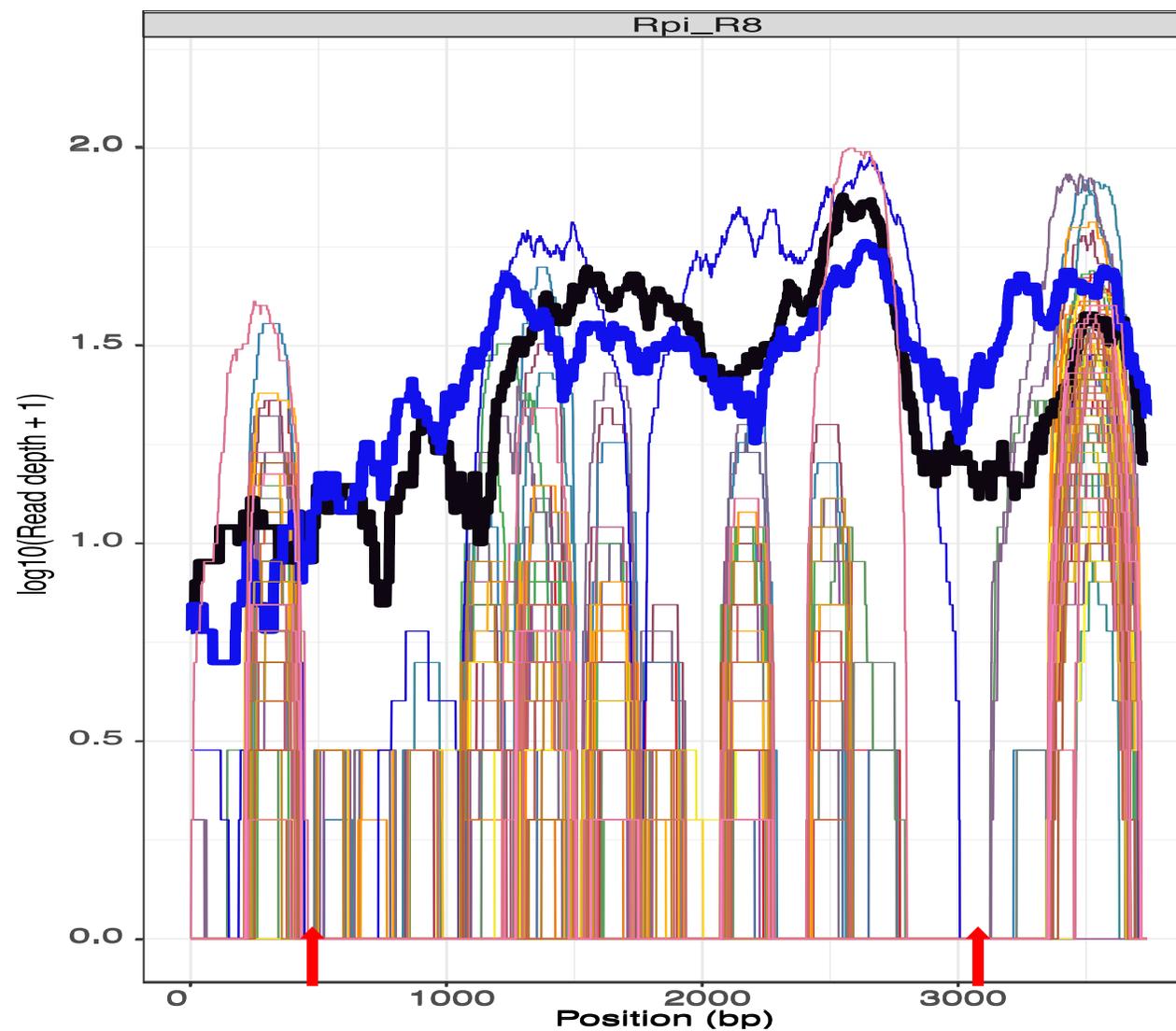
DISTRIBUTION OF FUNCTIONAL NLRs IN VARIETIES AND BREEDING CLONES



RENSEQ: INFORMATIVE MARKERS FOR BREEDING

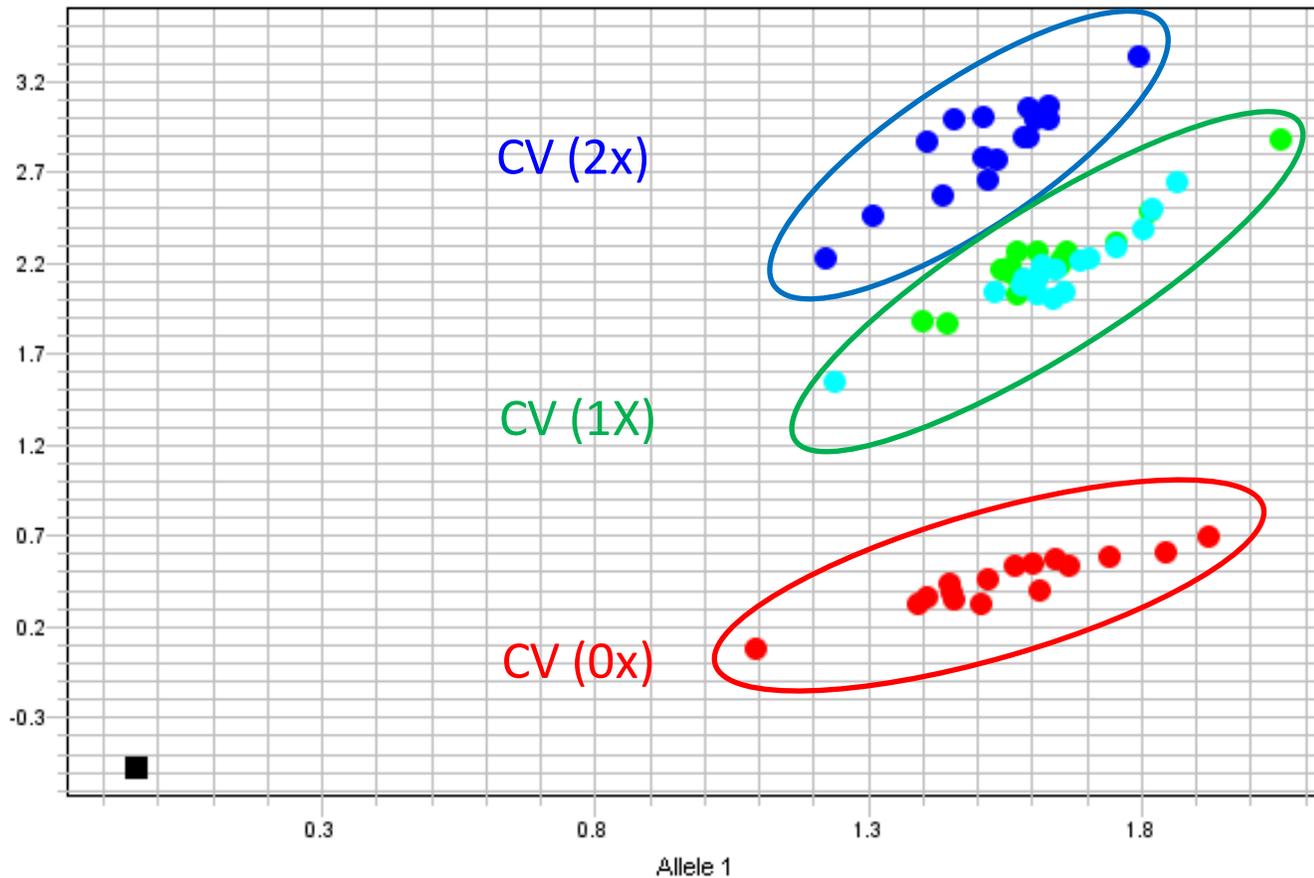


CONVERTING RENSEQ-READS INTO INFORMATIVE AND HIGHLY TRANSFERABLE MARKERS



INFORMATIVE AND HIGHLY TRANSFERABLE MARKERS INFORM COPY NUMBERS

Allelic Discrimination Plot



Markers:

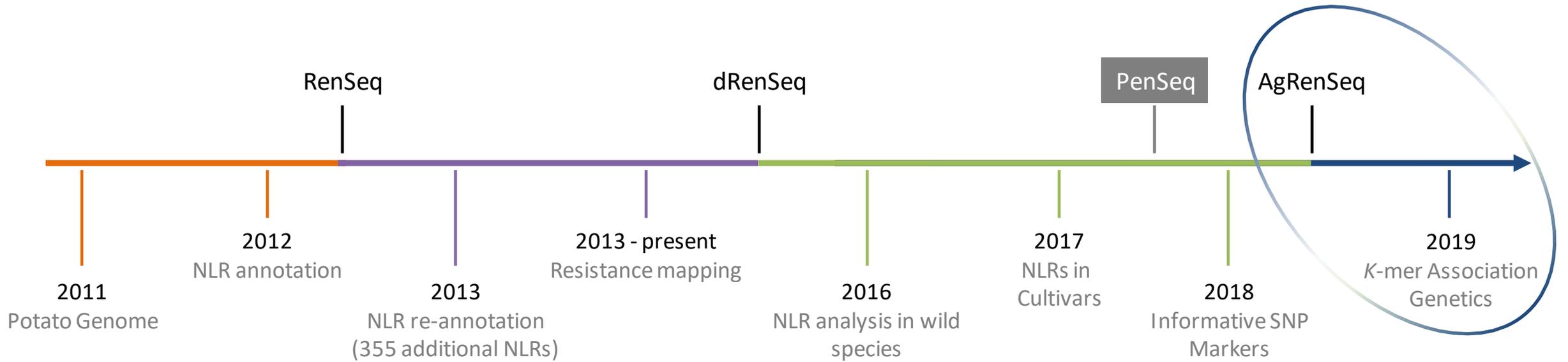
- *R8*
- *R9a*
- *Rpi-blb2*
- *Rpi-vnt1*

- *Gpa2*
- *Rx*
- *R2*
- *R3a*
- *R3b*

New markers:

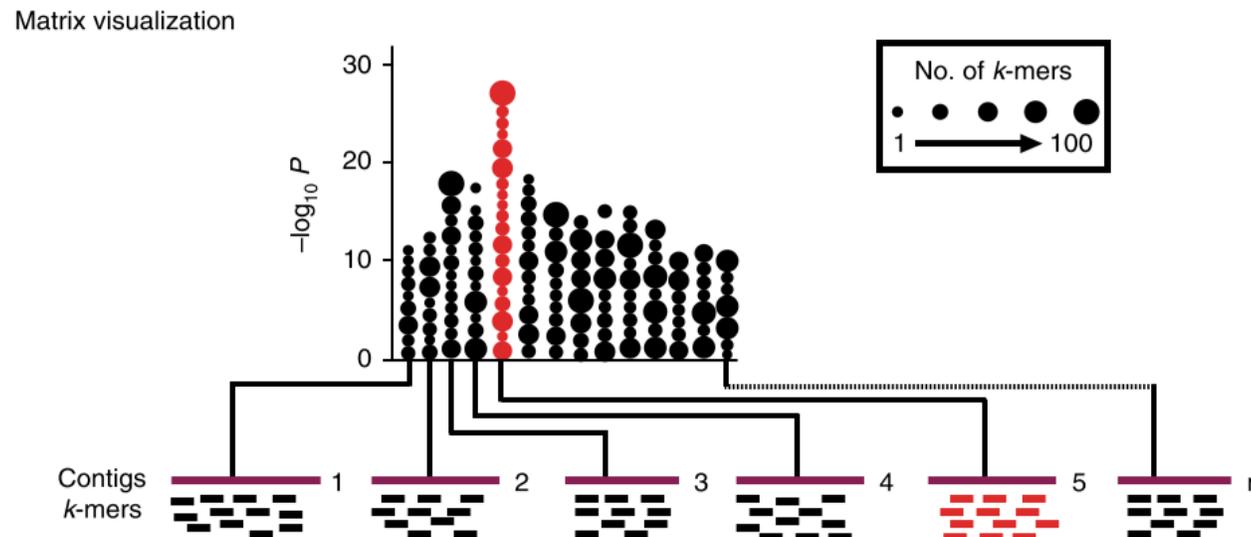
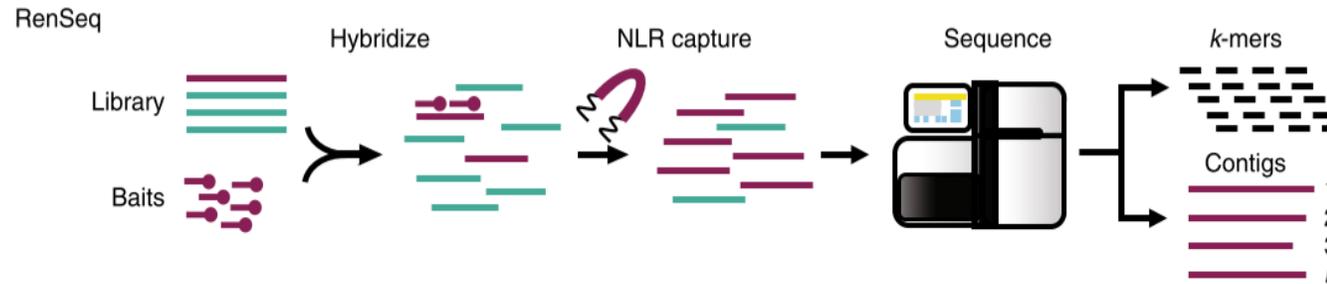
- *Ry(sto)*
- *H1*
- *Gpa5 (dev)*

AGREnSEQ : ASSOCIATION STUDIES FOR MULTIPLE RESISTANCES AND DIFFERENT PATHOGENS



AGRESEQ: K-MER BASED ASSOCIATION GENETICS

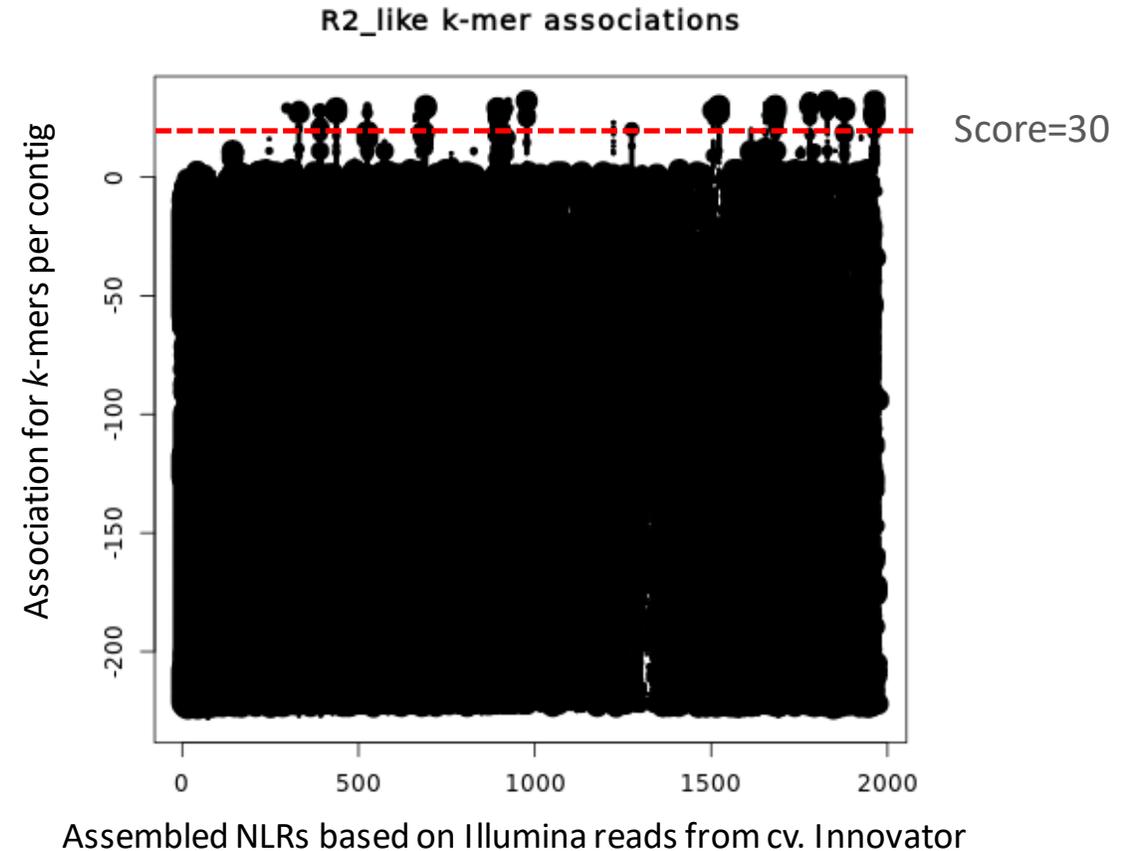
Takes advantage of our existing RenSeq data and the recombination during breeding



(Arora, S., Steuernagel, B., ... & Wulff, B.B.H.; 2019)

AGRENSAQ : R2-LIKE PROOF OF CONCEPT WITH ILLUMINA-BASED REFERENCE FAILS TO IDENTIFY FULL-LENGTH R2-LIKE GENE

- Cultivar Innovator Illumina-based NLR representation
- Total number of varieties/breeding clones: 334
- Varieties containing *R2-like*: 43
- Varieties without *R2-like*: 291
- Associated contigs: 16

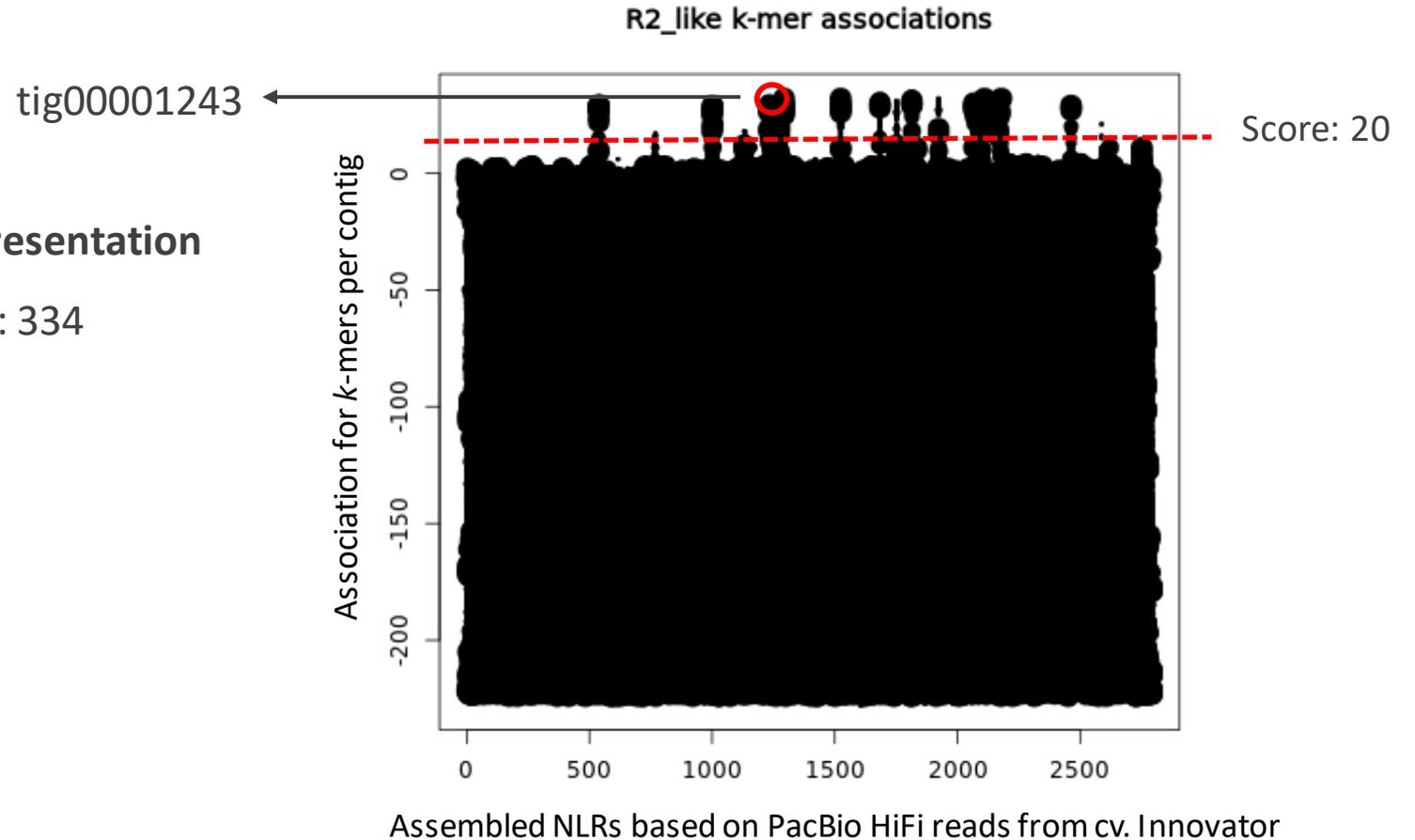


BG	Contig	Identity	Mismatch	Gap	E	Matched length	Length of BG	% Coverage of BG
R2-like	scf7180000022945	100	0	0	0	1306	2545	51

BG: Benchmark gene

SMRT-AGRESEQ : R2-LIKE PROOF OF CONCEPT WITH PACBIO-BASED REFERENCE IDENTIFIES FULL-LENGTH R2-LIKE GENE

- Cultivar Innovator PacBio-based NLR representation
- Total number of varieties/breeding clones: 334
- Varieties containing *R2-like*: 43
- Varieties without *R2-like*: 291
- Associated contigs: 18



BG	Contig	Identity	Mismatch	Gap	E	Matched length	Length of BG	% Coverage of BG
R2-like	tig00001243	100	0	0	0	2545	2545	100

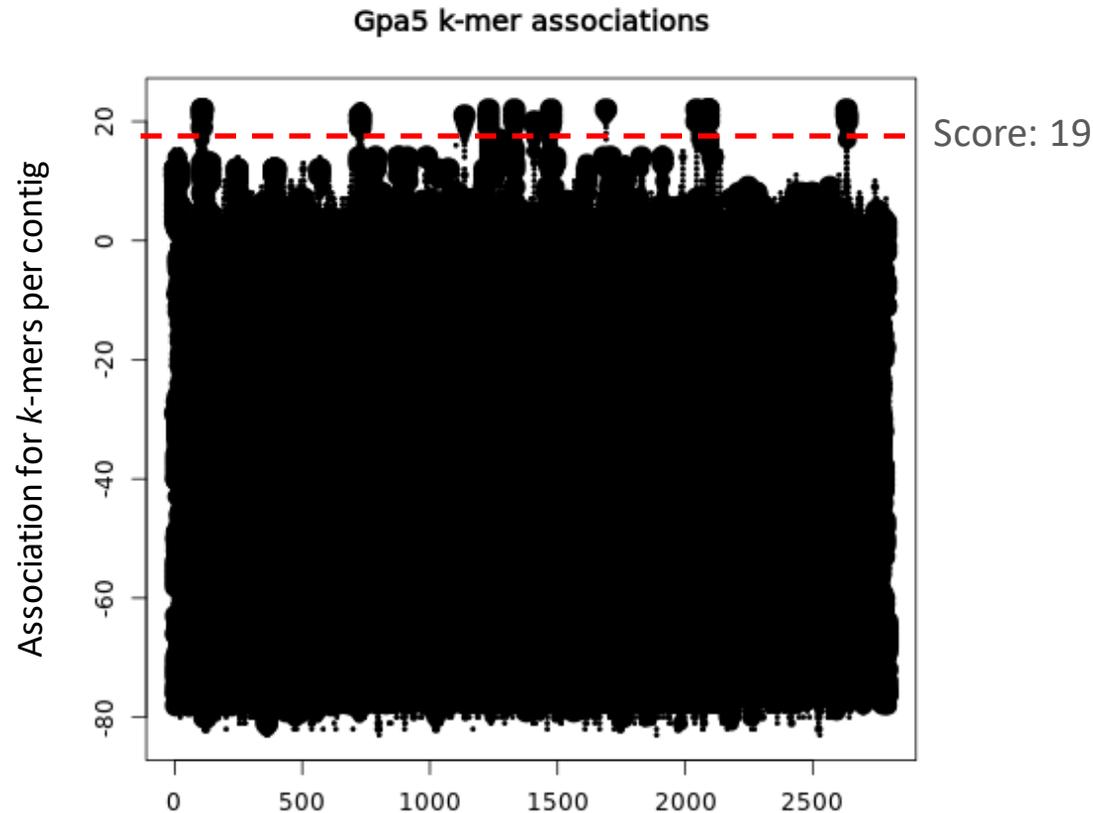
GPA5 – NEMATODE RESISTANCE

- Potato Cyst Nematodes (PCN) arrived after the famine
- *Globodera rostochiensis*
 - Pathotype Ro1 is controlled by *H1*
 - Resistance derived from *Solanum tuberosum* ssp. *Andigena*; CPC accession 1673
- *Globodera pallida*
 - Pathotypes Pa2/3, 1 are controlled by *Gpa5*
 - Resistance derived from *Solanum vernei* 'Innovator-resistance'



Photo by Ulrich Zunke; University of Hamburg;

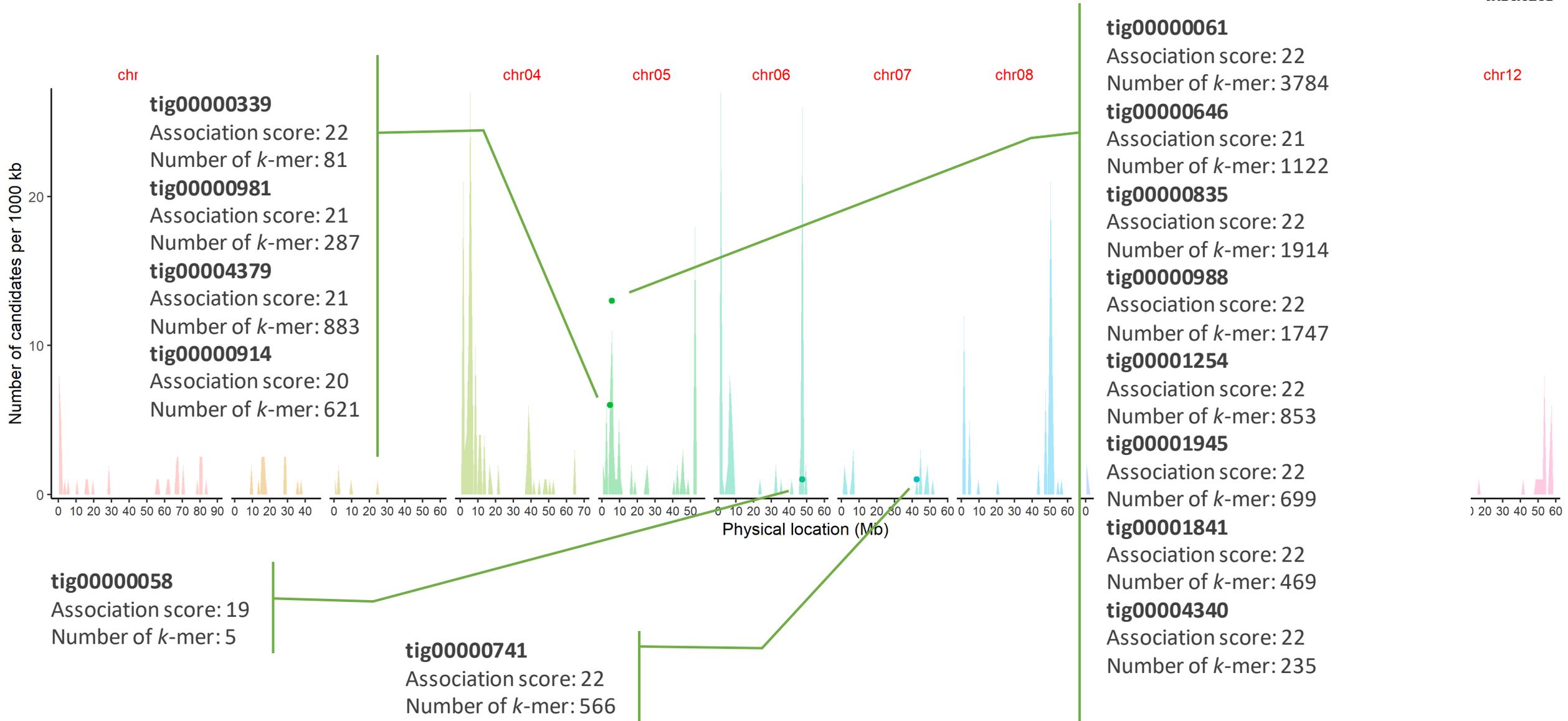
SMRT-AGRESEQ IDENTIFIES CANDIDATE CONTIGS FOR THE ELUSIVE GENE *GPA5*



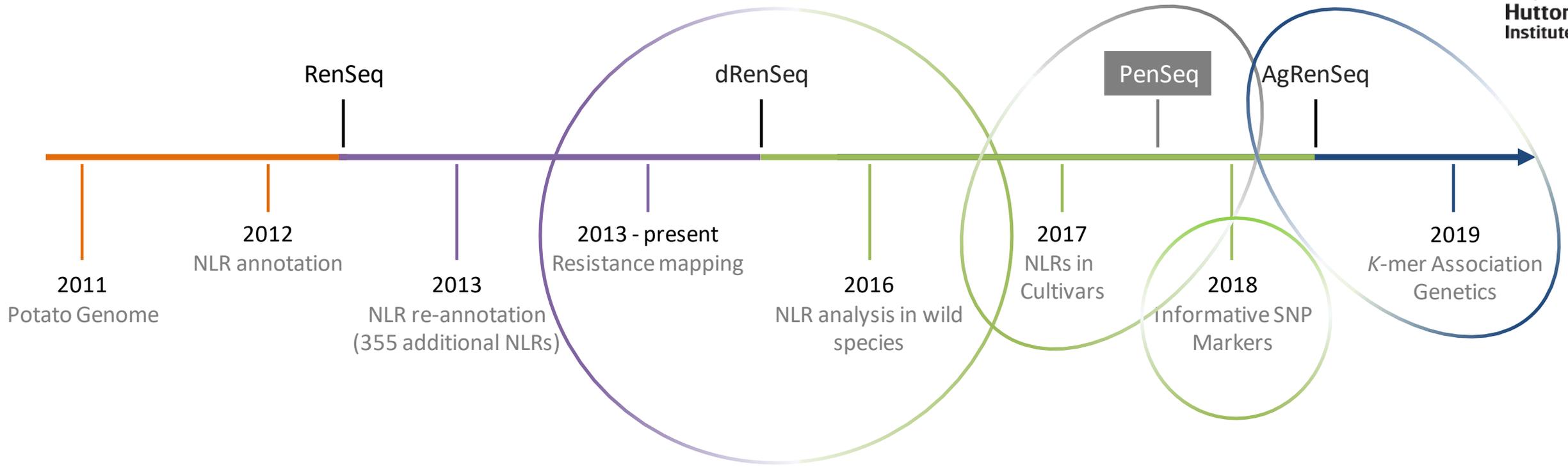
Assembled NLRs based on PacBio HiFi reads from cv. Innovator

- Cultivar Innovator PacBio-based NLR representation
- Total number of varieties: 128
- Varieties with *Gpa5*: 24
- Varieties without *Gpa5*: 104
- Associated contigs: 14
- Candidate NLRs: 9

GPA5 ASSOCIATED CONTIGS MAP TO POTATO LG 5



SUMMARY



RenSeq enables us to:

- Identifying new resistances from the CPC and other germplasm collections
- Track functional resistances in established cultivars and breeding clones
- Inform breeders about parental material for crosses
- Develop markers for important resistances
- Identify candidate genes for elusive resistances deployed in cultivars



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