



RUSTWATCH



*This project has received funding from
the European Union's Horizon 2020 research and innovation programme under
grant agreement No 773311.*

Grant Agreement: 773311

Call: Research and approaches for emerging diseases and pests
in plants and terrestrial livestock

Type of action: Research and Innovation action

Date: 16-03-2020

DELIVERABLE NUMBER	D1.2
DELIVERABLE TITLE	Report on race typing and genotyping of wheat rust fungi in Europe, 2019
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GRANT AGREEMENT N.	773311
DOCUMENT TYPE	Report
WORKPACKAGE N. TITLE	WP1 Understanding pathogen biology and drivers
LEAD CONTRACTOR	Aarhus University
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PLANNED DELIVERY DATE	February 2020, M22
ACTUAL DELIVERY DATE	March 2020, M23
DISSEMINATION LEVEL	Public
STATUS	Complete
VERSION	V1
REVIEWED BY	Contributing partners



EXECUTIVE SUMMARY

WP1 in the RustWatch project aims to bring together expertise and results from cereal rust diagnostic laboratories in Europe. The present report presents a summary of the 2019 rust populations for the partner countries and countries without national rust diagnostic lab facilities with respect to *Puccinia striiformis* (Pst, wheat yellow rust), *Puccinia triticina* (Pt, wheat brown rust) and *Puccinia graminis* f.sp. *tritici* (Pgt, wheat stem rust). The core activities are based on genotyping of incoming samples of rust infected wheat (occasionally other cereals) from project partners and associated networks, e.g., trial sites for Value for Cultivation and Use (VCU) and agricultural advisory services. Alignment of race- and genotyping results from the national rust diagnostic labs is another core activity, which takes place during the 1st quarter of the year, Aarhus University taking care of yellow- and stem rust and INRA taking care of brown/leaf rust samples.

Key highlights in 2019

Yellow rust:

- At least two new races were detected in PstS10, the most prevalent genetic group in Europe. The new races were adapted to local, widely grown wheat varieties, and only detectable by including these varieties in the race testing.
- A new race with *Yr10*-virulence was detected in PstS13. Isolates of this genetic group has severely affected triticale (northern Europe), durum wheat (southern Europe) and bread wheat (South America).
- Genetic group PstS7 with the original Warrior race was less prevalent, but spreading to new areas, whereas PstS8 (with the Kranich race) was observed in few cases only.
- Additional groups were detected in several countries, e.g. PstS15 consisting of a single genotype and race, which was first observed in France and this year also in Denmark.

Stem rust:

- Clade III-B (TTRTF, the “Sicily race”), which has been widespread in Sicily since 2016, was detected in Spain in 2019, following the detection in main land Italy and Croatia in 2017-2018.
- Clade IV-F, containing race TKKTF, was widespread in Europe, Northern Africa and the Middle East (11 countries).
- Clade IV-B, containing the races TKTTF and TTTTF, was detected in southern Europe. Only the race TKTTF was detected in Europe
- A new genetic group containing the race RFCNC was detected in Slovakia and in Czech Republic. A closely related variant of this group containing the race RFCPC was detected in Hungary.
- Multiple races with unusual virulence combinations were observed on cereals in local areas in Sweden and Spain, where the alternate host, *Berberis vulgaris*, was present. High diversity was confirmed by genotyping.

Leaf rust:

- Fam 166, a well-known leaf rust family which has become widely spread, was found in every participating country in 2019 (except for a single sample from Denmark), following a tendency already observed in 2018.
- Fam 106 314 2, the second known largest leaf rust family, was found in most samples from Germany and Belgium, and in some samples from Spain, Austria, Poland, Slovakia.
- Samples collected on durum wheat in Italy were part of the single genetic family, which is referred to as «Durum», which has already been observed in previous years.
- Less prevalent families were found in Austria, Poland, Italy, Norway and Denmark. Results do not yet allow us to conclude whether they are increasing in frequency.
- Three new genotypes were found in Austria, Poland, and Slovakia. Due to only few observations it is too early to assign them to new families.

The genotyping provided high-resolution results for genetic grouping of isolates, which to a large extent corresponded with race groups based on virulence phenotype. Global summary of SSR genotyping and race phenotyping results from WP1 are available online (<http://www.wheatrust.org/>), including an updated table showing the [relationship between races and genetic groups](#).

In 2019, a total of 148 (stem rust), 295 (yellow rust), and 164 (leaf rust) samples and derived isolates were handled (Table 1, 2 and 3). Each sample generally consisted of multiple rust infected leaves or stems. Representative leaves/stems were selected for alive spore recovery, and parallel genotyping based on the incoming sample, resulting in alive recovery of 143 (stem rust), 228 (yellow rust) and 135 (leaf rust) based on submissions in 2019.

The genotyping was generally based on Simple Sequence Repeat (SSR) analyses of incoming samples of rust infected plant material without prior recovery, whereas the race phenotyping was based on bioassays of differential wheat lines using spore samples of alive, pure isolates grown under strict experimental conditions (Hovmøller et al. 2017).

Table 1. Number of stem rust samples and derived isolates (*P. graminis* f.sp. *tritici*) handled in 2018-2019.

Geographic area	Country	2018			2019			Grand Total
		Dead	Recovered	Total	Dead	Recovered	Total	
Europe	Czech Republic		5	5				5
	Denmark	1		1		9	9	10
	France					1	1	1
	Hungary					1	1	1
	Italy	4	60	64	1	55	56	120
	Poland		14	14				14
	Slovakia		14	14				14
	Spain				2	74	76	76
	Sweden	8	29	37	2	5	7	44
	Total		13	122	135	5	143	148

Table 2. Number of yellow rust samples and derived isolates (*P. striiformis*) handled in 2019.

Geographic area	Country	2019		Grand Total
		Dead	Recovered	
Europe	Belgium		16	16
	Czech Repu	3		3
	Denmark	8	49	57
	France	2	16	18
	Germany	6	16	22
	Hungary	2	6	8
	Italy	7	53	60
	Latvia	19	2	21
	Netherlands		4	4
	Norway		5	5
	Poland		5	5
	Slovakia	3	3	6
	Spain	6	38	44
	Sweden	11	15	26
Total		67	228	295

Table 3. Number of leaf rust samples and derived isolates (*P. triticina*) handled in 2019.

Country	Sample state			Grand total
	Dead	Not considered	Recovered	
Austria	3		15	18
Belgium	1		7	8
Czech Rep.	9			9
Denmark			1	1
Germany			4	4
Hungary			2	2
Italy	7	4	62	73
Latvia	3			3
Norway			4	4
Poland	2		24	26
Slovakia			10	10
Spain			5	5
Sweden			1	1
Grand total	25	4	135	164

Nomenclature of races and genetic groups

Genetic groups of yellow rust are named Pst followed by a digit. Race variants are designated by the additional virulence observed or (-) in case a new variant had fewer virulences than the first defined race within the considered lineage. Race names already adopted by the farming community in Europe are maintained, e.g., “Warrior” and “Kranich”, which are named according to the wheat variety where they caused the first confirmed epidemic outbreak. A comprehensive justification and rationale for the naming of significant *P. striiformis* races and genetic groups has been published (Ali et al. 2017) and an updated [summary](#) is available on the GRRC website. The genotypic grouping of stem rust is according to Olivera Firpo et al. (2017) and [Olivera et al. \(2019\)](#), and is based on detailed alignment of the present SSR genotyping results and published SNP results; the race grouping is based on 20 internationally agreed wheat lines differentiating virulence in stem rust (Jin et al., 2008).

In yellow rust, we observed a strong correlation between genetic groups and race phenotypes for many areas, similarly, most races of wheat stem rust were associated with specific genetic groups (‘clades’). In few cases, a race phenotype of stem rust was assigned to more than one genetic group, reflecting the need for additional wheat lines differentiating races of stem rust.

A more comprehensive documentation of the nomenclature of stem rust races and genotypes, and the underlying experimental procedures are available on the [GRRC website](#), where new mapping tools were implemented recently. Geographical location of races/genotypes is shown on maps in case such information has been provided.

For leaf rust, genetic groups are named “families”, noted “Fam”, followed by a 3 to 7 digits number coding the

virulence phenotype. Thus, a family gathers individuals sharing the same genotype according to the 19 SSR markers used at INRAE, and a similar combination of virulence.

As an example, the following table defines the different pathotypes possibly found in the genetic group Fam 166 (Table 4). All of them share the exact same SSR genotype, and have a common virulence combination on resistance genes *Lr1*, *Lr3*, *Lr3bg*, *Lr10*, *Lr13*, *Lr14a*, *Lr15*, *Lr17*, *Lr17b*, *Lr37*. A variation for virulence is possibly found on *Lr20*, *Lr23* and *Lr26*. Thus up to $2^3=8$ close pathotypes can be found in this family.

Table 4. Pathotypes found in the genetic group Fam166.

Genetic group	Pathotype Code	Virulence phenotype																			
		1	-	-	-	3	3bg	-	10	13	14a	15	-	17	-	-	-	17b	37	-	-
Fam 166	166 316	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	-	-	-	17b	37	-	-
	166 317	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	-	-	26	17b	37	-	-
	166 336	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	20	-	-	17b	37	-	-
	166 337	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	20	-	26	17b	37	-	-
	166 357	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	-	23	26	17b	37	-	-
	166 376	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	20	23	-	17b	37	-	-
	166 377	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	20	23	26	17b	37	-	-

2018-2019 results – wheat stem rust

A total of 156 samples from 9 countries 2018-2019 were successfully genotyped (Table 5); 84 of these were also race typed using a standard set of 20 wheat lines used to differentiate races of stem rust.

Geographic group	Country	Genetic group	2018	2019	Total	
Europe	Czech Republic	Other	4		4	
	Czech Republic Total		4		4	
	Denmark	Clade IV-F		6	6	
	Denmark Total			6	6	
	France	Clade IV-F		1	1	
	France Total			1	1	
	Hungary	Other		1	1	
	Hungary Total			1	1	
	Italy		Clade III-B	20	16	36
			Clade IV-B	6		6
			Clade IV-F	12	23	35
	Italy Total			38	39	77
	Poland		Clade IV-E.2	3		3
			Clade IV-F	2		2
	Poland Total			5		5
	Slovakia		Other	8		8
	Slovakia Total			8		8
	Spain		Clade III-B		4	4
			Clade IV-B		2	2
			Clade IV-F		4	4
Other				32	32	
Spain Total				42	42	
Sweden		Other	12		12	
Sweden Total			12		12	
Europe Total			67	89	156	

Table 5. SSR genotyping of samples of *P. graminis* f.sp. *tritici* collected in 2018 and 2019. Results are shown as number of isolates within genetic group (clade). Significant races within clades are shown in Table 6 and virulences in Table 7. Graphical presentation of results available on www.wheatrust.org.

Genetic group	Race_name	Number of observations
Clade III-B	TTRTF	18
	- ^{a)}	22
Clade III-B Total		40
Clade IV-B	TKTTF	4
	-	4
Clade IV-B Total		8
Clade IV-E.2	TKKTF	2
	-	1
Clade IV-E.2 Total		3
Clade IV-F	TKKTF	24
	-	25
Clade IV-F Total		49
Other	LKMNC	2
	RFCNC	8
	RFCPC	1
	Other	6
	-	39
Other Total		58
Grand Total		156
^{a)} not tested		

Table 6. Relationship between genetic groups and prevalent races detected within these in Europe, 2018-2019

Table 7. Correspondence between race name and virulence on common stem rust differential lines

Race_name	Virulence corresponding to NA differentials 1-20 (main R-gene indicated)																			
	Sr5	Sr21	Sr9e	Sr7b	Sr11	Sr6	Sr8a	Sr9g	Sr36	Sr9b	Sr30	Sr17	Sr9a	Sr9d	Sr10	SrTmp	Sr24	Sr31	Sr38	SrMcN
LKMNC	5	-	-	-	-	6	8a	9g	36	-	-	17	9a	-	10	-	-	-	-	McN
RFCNC	5	21	-	7b	-	-	8a	9g	-	-	-	17	9a	-	10	-	-	-	-	McN
RFCPC	5	21	-	7b	-	-	8a	9g	-	-	-	17	9a	-	10	Tmp	-	-	-	McN
TKKTF	5	21	9e	7b	-	6	8a	9g	-	9b	30	17	9a	9d	10	Tmp	-	-	38	McN
TKTTF	5	21	9e	7b	-	6	8a	9g	36	9b	30	17	9a	9d	10	Tmp	-	-	38	McN
TTKSK	5	21	9e	7b	11	6	8a	9g	-	9b	30	17	9a	9d	10	-	-	31	38	McN
TTKST	5	21	9e	7b	11	6	8a	9g	-	9b	30	17	9a	9d	10	-	24	31	38	McN
TTKTK	5	21	9e	7b	11	6	8a	9g	-	9b	30	17	9a	9d	10	Tmp	-	31	38	McN
TTKTT	5	21	9e	7b	11	6	8a	9g	-	9b	30	17	9a	9d	10	Tmp	24	31	38	McN
TTRTF	5	21	9e	7b	11	6	8a	9g	36	9b	-	17	9a	9d	10	Tmp	-	-	38	McN
TTTTF	5	21	9e	7b	11	6	8a	9g	36	9b	30	17	9a	9d	10	Tmp	-	-	38	McN

Clade III-B, consisting of a single race (TTRTF), a.k.a. the “Sicily race”, was detected for the first time in Spain (Fig. 2). In addition to the virulences shown in Table 7, this race is virulent on several durum wheat varieties (Sr13b) and additional virulence to Sr50, i.e., higher infection type than usual according to Patpour et al. 2019.

The genetic group Clade IV-F, was quite widespread in Europe, i.e., Denmark, France, Italy, Poland, and Spain. Only a single race was detected in this group, TKKTF (Fig. 1, Table6). Clade IV-F was reported by Olivera et al. (2019) and is present in Africa, Middle East and West Asia.

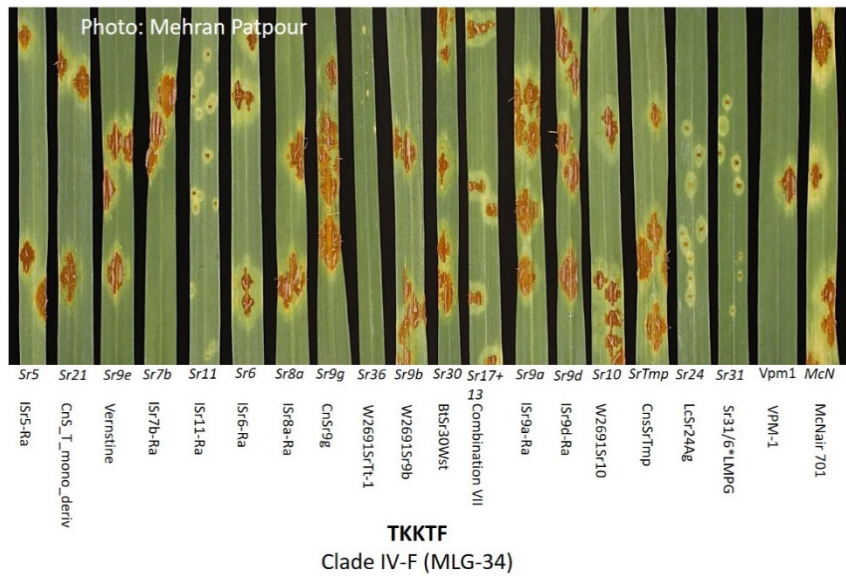


Figure 1. Virulence phenotype of race TKKTF on standard differential set of 20 lines

Clade IV-B was detected in southern Europe and northern Africa, in Europe only race TKKTF was detected. Clade I (Ug99) was not detected in Europe.

Race RFCNC belonging to another genetic group was detected in Slovakia in 2018 and 2019 and in Czech Republic in 2019. A closely related variant of this group containing the race RFCPC was detected in Hungary in 2019. The race LKMNC belonging to another genetic group (not previously defined) was also detected in Czech Republic in 2019.

Multiple races were derived from wheat, barley and rye in Sweden and Spain, in both cases sampled in local areas in proximity to the alternate host, *Berberis vulgaris*. Many of the races were characterized by avirulence on multiple differential lines, and many of the isolates were relatively poorly adapted to wheat. The unusual virulence patterns of selected isolates are currently being investigated in more detail, in combination with rust susceptibility profiles of commercially important wheat varieties in Europe and elsewhere. The high genetic diversity was confirmed by a high number of unique and diverse genotypes.

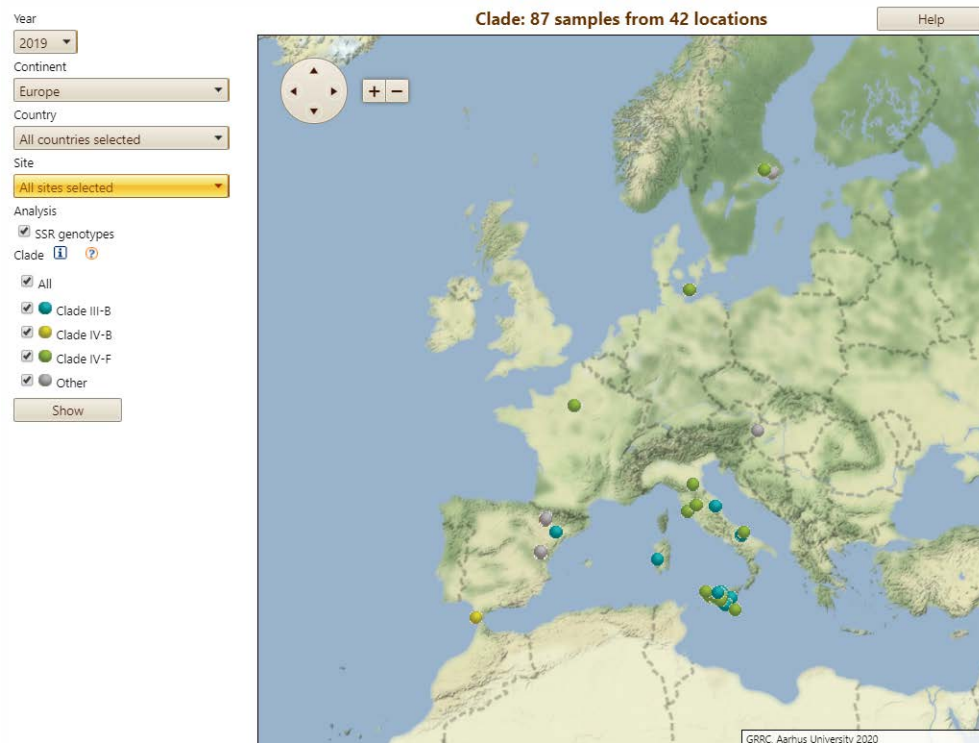


Figure 2. Distribution of genetic groups of stem rust based on samples submitted to Aarhus University, 2019 (samples with GPS information only). Detected races within groups shown in Table 6

2019 results – wheat yellow rust

A total of 412 samples collected in 17 countries in 2018 and 2019 were successfully genotyped (Fig. 3; Table 8). Race typing was done for 65 isolates for alignment and confirmation of genetic grouping and emergence of new races within genetic groups.

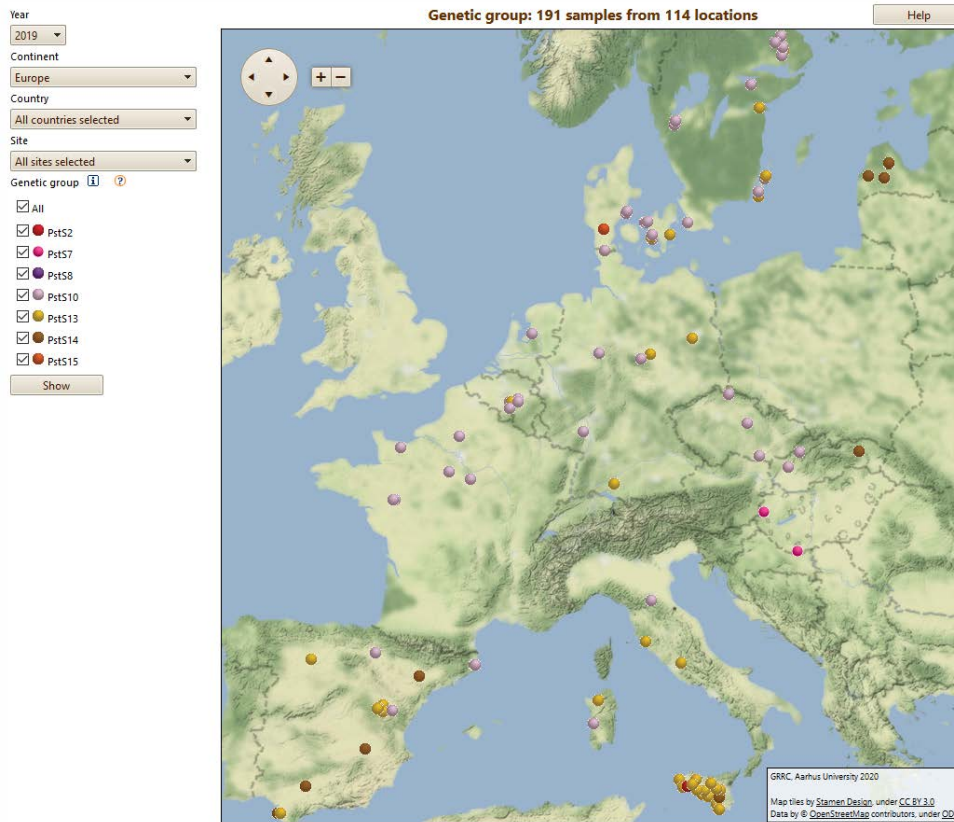


Figure 3. Distribution of genetic groups of yellow rust based on samples submitted to Aarhus University, 2019 (samples with GPS information only). Detected races within groups shown in Table 9.

PstS10 was the most prevalent group on bread wheat in Europe, so far dominated by a single race (virulence phenotype: 1,2,3,4,-,6,7,-,9,-,-,17,-,25,-,32,Sp,AvS,-) (Table 9). However, in recent years, new races adapted to local, widely grown wheat varieties have emerged within PstS10, resulting in quantitative shifts in rust susceptibility of such varieties, e.g., from ‘resistant’ to ‘susceptible’ or from ‘susceptible’ to ‘extreme susceptible’. It has not been possible to diagnose such new races within PstS10 by molecular techniques nor by standard wheat differential lines, so far only by including the varieties in question into the differential set, e.g., varieties Kalmar and/or Benchmark. However, correct diagnosis of races based on such quantitative differences requires strict and well-defined experimental conditions, which may not always be available.

In terms of virulences corresponding to commonly used Yr-genes, the races within PstS10 are similar to the only race detected so far in PstS7 (“Warrior”), except for the resistance specificity in the varieties Warrior (UK) and Ambition (DK). The “Warrior” race is still present, but often in low frequency, in many areas in Europe. Likewise, the PstS8 group (containing the “Kranich” race) has become less frequent in most areas of Europe. A new race in a new unique genetic group, termed PstS15, was detected in France in 2017, 2018 and 2019 and in 2019 also in Denmark. The significance of this race in terms of potential shifts in rust susceptibility is being investigated via ongoing RustWatch project activities.

PstS13 and PstS14 were detected in multiple countries and continents in recent years, and in 2019 a new Yr10-virulent variant was detected in Poland. PstS13 isolates are highly aggressive on several crop types, e.g., multiple triticale varieties (northern Europe and South America), durum wheat (southern Europe) and multiple spring wheat varieties (South America) resulting in significant yield losses (Carmona et al., 2019). PstS14 is highly dominant in Morocco, and otherwise present in multiple European countries, but often in low frequencies.

Table 8. SSR genotyping of samples of *P. striiformis* 2018-2019. Results shown as number of isolates within genetic groups in each country, significant races and virulences within each group shown in Table 9.

Geographic group	Country	SSR_name	2018	2019	Grand Total
Europe	Austria	PstS7	2		2
		PstS10	10	15	25
	Belgium	PstS13		1	1
		PstS7	1		1
		PstS10	2	3	5
	Czech Republic	PstS7	8		8
		PstS7	1		1
		PstS10	34	32	66
	Denmark	PstS13	2	12	14
		PstS14	4		4
		PstS15		4	4
		PstS7	2		2
	Estonia	PstS8		1	1
		PstS7	2		2
		PstS10	4	12	16
	France	PstS13	1		1
		PstS15	1	1	2
		PstS7	4	3	7
	Germany	PstS10	2	4	6
		PstS13	6	3	9
		PstS8		1	1
	Hungary	PstS7		4	4
		PstS10		3	3
	Italy	PstS13	8	24	32
		PstS13		1	1
		PstS14	1	1	2
		PstS2		1	1
		PstS4	16		16
		PstS7		2	2
		PstS10		1	1
	Latvia	PstS14	9	18	27
		PstS7	3	1	4
		PstS10	3	3	6
	Netherlands	PstS7		1	1
		PstS10	1	5	6
	Norway	PstS7	1		1
		PstS10	1		1
	Poland	Other	1		1
		PstS10	2		2
		PstS13	4		4
Slovakia	PstS10	1	2	3	
	PstS14	1	1	2	
	PstS7	2	1	3	
Spain	PstS10	10	10	20	
	PstS13	4	11	15	
	PstS14	6	6	12	
	PstS14		1	1	
	PstS7	4		4	
Sweden	PstS10	23	16	39	
	PstS13	4	5	9	
	PstS7	3		3	
	PstS8	4		4	
Ukraine	PstS13	4		4	
	PstS13	4		4	
Europe Total		202	210	412	

Table 9. Correspondence between genetic groups and prevalent races of *P. striiformis* sampled from epidemic sites since 2000, Global Rust Reference Center, January 2020.

Common names of prevalent races and genetic groups in yellow rust - GRRC, January 2020			
Genetic group	Race	Virulence phenotype*	Prevalence in geographical region
PstS0	Brigadier	1,2,3,-,-,-,-,9,-,-,17,-,25,-,-,-,AvS,-	Europe
	Brigadier,v4	1,2,3,4,-,-,-,-,9,-,-,17,-,25,-,-,-,AvS,-	Europe
	Madrigal_Lynx	1,2,3,-,-,6,-,-,9,-,-,17,-,25,-,-,-,AvS,-	Europe
	Madrigal_Lynx,v4	1,2,3,4,-,-,6,-,-,9,-,-,17,-,25,-,-,-,AvS,-	Europe
	Robigus	1,2,3,4,-,-,-,-,9,-,-,17,-,25,-,-,32,-,AvS,-	Europe
	Solstice_Oakley	1,2,3,4,-,-,6,-,-,9,-,-,17,-,25,-,-,32,-,AvS,-	Europe
	Tulsa	-,-,3,4,-,-,6,-,-,-,-,-,-,25,-,-,32,-,AvS,-	Europe
PstS1	PstS1	-,2,-,-,-,6,7,8,9,-,-,-,-,25,-,-,-,AvS,-	North America, Australia
	PstS1,v1	1,2,-,-,-,6,7,8,9,-,-,-,-,25,-,-,-,AvS,-	East Africa
	PstS1,v1,v27	1,2,-,-,-,6,7,8,9,-,-,-,-,25,27,-,-,AvS,-	East Africa
PstS2	PstS2	-,2,-,-,-,6,7,8,9,-,-,-,-,25,-,-,-,AvS,-	East Africa, West Asia, South Asia
	PstS2,v1	1,2,-,-,-,6,7,8,9,-,-,-,-,25,-,-,-,AvS,-	East Africa
	PstS2,v3	-,2,3,-,-,6,7,8,9,-,-,-,-,25,-,-,-,AvS,-	East Africa
	PstS2,v27	-,2,-,-,-,6,7,8,9,-,-,-,-,25,27,-,-,AvS,-	East Africa, West Asia, North Africa
	Pst2,v1,v27	1,2,-,-,-,6,7,8,9,-,-,-,-,25,27,-,-,AvS,-	East Africa, West Asia
	PstS2,v3,v27	-,2,3,-,-,6,7,8,9,-,-,-,-,25,27,-,-,AvS,-	East Africa
	PstS2,v10,v24	-,2,-,-,-,6,7,8,9,10,-,-,-,24,25,-,-,-,AvS,-	East Africa, West Asia
	PstS2,v10,v24,v27	-,2,-,-,-,6,7,8,9,10,-,-,-,24,25,27,-,-,-,AvS,-	West Asia
	PstS2,v3,v10,v24,v27	-,2,3,-,-,6,7,8,9,10,-,-,-,24,25,27,-,-,-,AvS,-	East Africa
PstS3	PstS3	-(2),-,-,-,6,7,8,-,-,-,-,-,-,-,AvS,-	Europe, North Africa, West Asia
	PstS3,v25	-(2),-,-,-,6,7,8,-,-,-,-,25,-,-,-,AvS,-	Europe, East Africa
PstS4	Triticale2006	-,2,-,-,-,6,7,8,-,10,-,-,24,-,-,-,-,-	Europe
PstS5	PstS5	1,2,3,4,-,-,6,-,-,9,-,-,-,-,25,-,-,32,-,AvS,Amb	Central Asia
	PstS5,v17	1,2,3,4,-,-,6,-,-,9,-,-,17,-,25,-,-,32,-,AvS,Amb	Central Asia, South Asia
PstS6	PstS6	1,2,-,-,-,6,7,-,9,-,-,17,-,-,27,-,-,AvS,-	East Africa, Central Asia, South Asia
PstS7	Warrior	1,2,3,4,-,-,6,7,-,9,-,-,17,-,25,-,-,32,Sp,AvS,Amb	Europe
PstS8	Kranich	1,2,3,-,-,6,7,8,9,-,-,17,-,25,-,-,32,-,AvS,Amb	Europe
PstS9	PstS9	1,2,3,4,-,-,6,-,-,9,-,-,-,-,25,27,32,-,AvS,Amb	Central Asia, South Asia
	PstS9,v17	1,2,3,4,-,-,6,-,-,9,-,-,17,-,25,27,32,-,AvS,Amb	Central Asia
PstS10	Warrior(-)	1,2,3,4,-,-,6,7,-,9,-,-,17,-,-,25,-,-,32,Sp,AvS,-	Europe, North Africa
	Kalmar	1,2,3,4,-,-,6,7,-,9,-,-,17,-,-,25,-,-,32,Sp,AvS,-	Europe
	Benchmark	1,2,3,4,-,-,6,7,-,9,-,-,17,-,-,25,-,-,32,Sp,AvS,-	Europe
PstS11	PstS11	-,2,-,-,(4),-,-,6,7,8,-,-,-,17,-,-,27,32,-,-,AvS,-	Central and West Asia, East Africa
PstS12	Hereford	-,2,3,-,-,6,7,8,-,-,-,17,-,-,25,-,-,32,-,AvS,-	Europe
PstS13	Triticale2015	-,2,-,-,-,6,7,8,9,-,-,-,-,-,-,-,AvS,-	Europe, South America, West Asia
PstS14	PstS14	-,2,3,-,-,6,7,8,9,-,-,17,-,25,-,-,32,(Sp),AvS,-	Europe, North Africa
PstS15	PstS15	1,2,3,-,-,6,7,-,9,-,-,17,-,-,25,-,-,32,-,AvS,Amb	Europe

* Figures and symbols designate virulence and avirulence (-) corresponding to yellow rust resistance genes: Yr1, Yr2, Yr3, Yr4, Yr5, Yr6, Yr7, Yr8, Yr9, Yr10, Yr15, Yr17, Yr24, Yr25, Yr27, Yr32, and the resistance specificity of Spalding Prolific (Sp), Avocet S (AvS) and Ambition (Amb), respectively. Kalmar and Benchmark are new races in PstS10, which can only be distinguished by these varieties.

Leaf rust

In 2019, a total of 164 samples were received. From the gathered information, they were collected on at least 63 different cultivars (21 of which were durum wheat cultivars). Four of them were not considered, as they displayed only yellow or stem rust symptoms. Then 25 samples were not recovered and were probably dead upon arrival. They are currently waiting for genotyping.

The 54 live samples collected from durum wheat are to be genotyped, before discussing any need for pathotyping. Currently, previous work conducted on recent durum wheat leaf rust populations in France revealed no diversity, neither on the durum differential set (proposed by Goyeau et al., 2012), nor on the set of 19 SSR used for bread wheat. Thus, we are to check first if some genotype diversity is detectable in these European samples. If so, we will phenotype the isolates on the available durum differential set, improving it as needed by adding new differential lines. Genotyping of the durum leaf rust samples is ongoing, with 12 results available up to now.

A single-pustule isolation was made for the 135 recovered samples, among which the first 85 were then genotyped. Among those 85 samples, 73 bread wheat samples were also phenotyped (Table 10).

Table 10. Number of leaf rust samples and derived single pustule isolates processed

	Dead	Single-pustule isolated	Total	Genotyped	Phenotyped
Durum wheat	4	54	58	12	0
Bread wheat	21	81	102	73	81
Total	25	135	160	85	81

This allowed to identify seven families, of which two main families stand out: Fam 106 314 2 and Fam 166, already observed in France and across Europe. Fam 106 314 2 is virulent on gene *Lr28*, introduced in British cultivars in 2006, and then used in the European germplasm, with cultivars registered from 2011 for example in France.

Fam 166 combines 12 virulences against the 23 *Lr* genes included in the differential set. Families such as Triticale and Durum underline the low diversity of known leaf rust races collected on those species. Durum wheat being an important part of the wheat crops in Italy, it appears as expected that most of the samples from Italy are revealed as being part of the Durum family. As expected, at this stage no genetic diversity was observed in the samples collected from durum wheat.

Three unknown genotypes appeared, from a Polish an Austrian, and a Slovakian sample. However, it is too early to confirm if they do represent new families, especially because no other isolate was ever observed having the same genotype. They also present original pathotypes, detailed in the table below (Table 11).

Table 11. Virulence phenotypes of newly detected European leaf rust genotypes in 2019

Country	Pathotype Code	Virulence phenotype																			
		1	-	-	-	3	3bg	3ka	-	13	14a	15	-	17	-	-	-	-	37	-	-
Austria	165314	1	-	-	-	3	3bg	3ka	-	13	14a	15	-	17	-	-	-	-	37	-	-
Poland	165334	1	-	-	-	3	3bg	3ka	-	13	14a	15	-	17	20	-	-	-	37	-	-
Slovakia	164754	1	-	-	-	3	3bg	-	-	13	14a	15	16	17	-	23	-	-	37	-	-

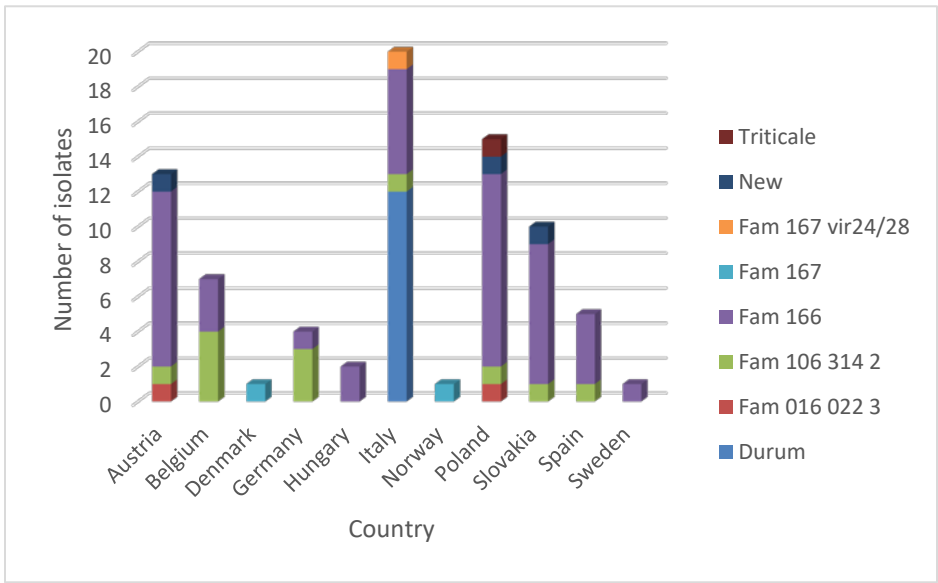


Figure 4. Chart of leaf rust families distribution across Europe in 2019. Only 79 out of the 85 genotyped isolates appear on the chart, as 6 of them still have uncertain results. This is possibly due to a mix, and could lead to a new purification of those samples to further investigate them.

Table 12. Correspondence between leaf rust genetic groups and virulence phenotypes

Genetic group	Pathotype Code	Virulence phenotype																			
Fam 106 314 2	106 314 2	1	-	-	-	-	-	-	10	13	14a	15	-	17	-	-	-	-	37	-	28
	106 315 2	1	-	-	-	-	-	-	10	13	14a	15	-	17	-	-	26	-	37	-	28
	106 334 2	1	-	-	-	-	-	-	10	13	14a	15	-	17	20	-	-	-	37	-	28
	106 335 2	1	-	-	-	-	-	-	10	13	14a	15	-	17	20	-	26	-	37	-	28
	166 314	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	-	-	-	-	37	-	-
	166 334	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	20	-	-	-	37	-	-
Fam 166	166 316	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	-	-	-	17b	37	-	-
	166 317	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	-	-	26	17b	37	-	-
	166 336	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	20	-	-	17b	37	-	-
	166 337	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	20	-	26	17b	37	-	-
	166 357	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	-	23	26	17b	37	-	-
	166 376	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	20	23	-	17b	37	-	-
	166 377	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	20	23	26	17b	37	-	-
	Fam 167	167 315	1	-	-	-	3	3bg	3ka	10	13	14a	15	-	17	-	-	26	-	37	-
167 316		1	-	-	-	3	3bg	3ka	10	13	14a	15	-	17	-	-	-	17b	37	-	-
167 317		1	-	-	-	3	3bg	3ka	10	13	14a	15	-	17	-	-	26	17b	37	-	-
167 335		1	-	-	-	3	3bg	3ka	10	13	14a	15	-	17	20	-	26	-	37	-	-
167 337		1	-	-	-	3	3bg	3ka	10	13	14a	15	-	17	20	-	26	17b	37	-	-
167 357		1	-	-	-	3	3bg	3ka	10	13	14a	15	-	17	-	23	26	17b	37	-	-
Fam 167 vir24/28	167 336 3	1	-	-	-	3	3bg	3ka	10	13	14a	15	-	17	20	-	-	17b	37	24	28
	167 337 3	1	-	-	-	3	3bg	3ka	10	13	14a	15	-	17	20	-	26	17b	37	24	28
	167 377 3	1	-	-	-	3	3bg	3ka	10	13	14a	15	-	17	20	23	26	17b	37	24	28
Fam 016 022 3	016 022 3	-	-	-	2c	-	-	-	10	13	-	-	-	-	20	-	-	17b	-	24	28
	016 122 3	-	-	-	2c	-	-	-	10	13	14a	-	-	-	20	-	-	17b	-	24	28
	016 126 3	-	-	-	2c	-	-	-	10	13	14a	-	-	-	20	-	-	17b	37	24	28
Triticale	012 002	-	-	-	2c	-	-	-	10	-	-	-	-	-	-	-	-	17b	-	-	-
	012 042	-	-	-	2c	-	-	-	10	-	-	-	-	-	-	23	-	17b	-	-	-
	012 102	-	-	-	2c	-	-	-	10	-	14a	-	-	-	-	-	-	17b	-	-	-
	016 106	-	-	-	2c	-	-	-	10	13	14a	-	-	-	-	-	-	17b	37	-	-
Durum	Different differential set																				

Alignment efforts

Project partners met for a week at the end of February 2019 to exchange knowledge and results for the national virulence surveys. A total of 25 yellow rust samples from five European rust diagnostic laboratories, and 15 stem rust samples from three labs were submitted to GRRRC prior to the workshop: United Kingdom (yellow rust), France (yellow rust), Germany (yellow and stem rust), Poland (yellow and stem rust), Czech Republic (stem rust) and Slovakia (stem rust). Results are reported in milestone report M1.6 (October 2019). 16 leaf rust samples from 3 laboratories were received at INRAE and alignment is ongoing.

In general, SSR genotyping results were highly useful for resolving cases with admixture of isolates of different genotypes/races, which is a continuous challenge when working with incoming samples of rust infected plant material, in particular under high disease load where individual lesions/pustules may be difficult to distinguish from one another. The genotyping results proved consistent across labs and replications. At present time, additional alignment between SSR genotyping and amplicon sequencing in the UK is in progress.

The virulence phenotyping experiments confirmed the influence of environment and assessment practice on the final result, in particular for incompatible 'resistance'/'avirulence' combinations conferring intermediate infection types (e.g., IT5-6 on a 0-9 scale), i.e., relatively minor quantitative differences between

‘virulent/avirulent’ isolates, syn. ‘resistant/susceptible’ host reactions. It is therefore recommended to include at regular basis standard isolates (if available) for which ‘virulence’/‘avirulence’ to critical resistance genes has been confirmed in repeated tests.

The set-up and procedures in RustWatch addresses these challenges and allows us to put together pathogen genotyping/phenotyping results from different labs without creating bias in results arising from ‘technical/experimental’ differences among labs and environments.

The alignment experiments in spring 2020 is in progress.

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	Karin Andersson
	Kristian Barck

Kristian Jochnick
Lars Johansson
Lina Norrlund
Louise Aldén
Lovisa Eriksson
Lukas Hallberg
Robert Dinwiddie

Acknowledgements

Submission of samples of rust infected plant material in 2019 was facilitated by a huge number of collaborators from multiple organisations and countries. Sampling from EC-VCU network trials was coordinated by Valérie Cadot (Geves), France (WP3). Additional sampling from several areas was based on bilateral agreements, e.g., Jordbruksverket, Sweden (Gunilla Berg), Latvia (Jānis Jaško), IRTA, Spain (Dolors Villegas). Les Szabo, CDL, Minnesota, provided reference DNA samples of previously defined stem rust genetic groups based on SNP analyses.