

# RUSTWATCH



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#### **KEY HIGHLIGTS IN 2020**

#### **YELLOW RUST**

- PstS10 is the most prevalent genetic group in Europe. Four races were detected in PstS10, each adapted to local, widely grown wheat varieties.
- A new group provisionally termed ME2018, detected in Egypt in 2018 and Turkey in 2019, was observed in Latvia in 2020.
- Genetic group PstS13 carrying Yr10-virulence detected in Germany, first detected in Poland (2018). PstS13 has severely affected triticale and durum wheat (Europe) and bread wheat (South America and Australia).
- Genetic group PstS7 (Warrior race) and PstS8 (Kranich race) were less prevalent than in previous years. Up to now, PstS7 detected in Europe, northern Africa and South America, and PstS8 in Europe.
- Additional genetic groups were detected in low frequencies, including PstS15 (single genotype and race), present in France since 2017 and Scandinavia since 2019.

#### STFM RUST

- Additional spread of stem rust in Europe, including France, Austria, Ireland and Norway.
- Clade III-B (race TTRTF), widespread in Sicily since 2016, has now been detected in seven countries in Europe and four countries in Africa/Asia. In 2020, first detection in Austria, Czech Republic, Slovakia, and Slovenia.
- Clade IV-F (race TKKTF) has been detected in 10 European countries (2019-2020), and previously in Africa and West/Central Asia.
- Clade IV-B, (race TKTTF and TTTTF) has been detected in five countries in Europe and six countries in Africa. In 2020, TKTTF was detected in France, Ireland and Spain. TTTTF has not been detected in Europe.
- An unknown genetic group (race RFCNC) was observed in Czech Republic. This has previously been detected in Germany, Czech Republic, Slovakia and Hungary.
- Clade I (Ug99) was not detected in Europe.

#### **LEAF RUST**

- Fam 166, a well-known leaf rust family which has become widely spread, was found in every participating country in 2020, following a tendency already observed in 2018 and 2019.
- Fam 106 314 2, the second known largest leaf rust family, was less represented in 2020 than in the last two years, which is possibly a result of the reduced number of samples that were received, along with their origin (Fam 106 314 2 being a family mostly found in western Europe).
- Less prevalent families (Fam 167 and Fam 165 vir24) were found, without any indicator of an increasing frequency.



Summary of SSR genotyping and race phenotyping results from GRRC (2008-2020) is available online (<a href="http://www.wheatrust.org/">http://www.wheatrust.org/</a>), including an updated table showing the relationship between races and genetic groups.

This report comprises results for the wheat yellow/stripe rust (*Puccinia striiformis*), wheat stem rust (*P. graminis* f.sp. *tritici*) and wheat leaf rust (*P. triticina*). The testing of additional samples from 2020 and beginning of 2021 is ongoing with emphasis on alignment of results from different rust diagnostic laboratories and genotyping approaches with the aim of presenting the global landscape for yellow, leaf and stem rust infecting wheat.

In 2020, a total of 615 samples and derived isolates from 19 countries were handled. Each sample generally consisted of multiple rust infected leaves or stems. Representative leaves/stems were selected for alive spore recovery in parallel to genotyping of incoming samples (Table 1, Table 2, and Table 3). More than 300 alive isolates of yellow ,stem rust, and leaf rust were recovered based on submissions in 2020.

The genotyping of yellow rust based on Simple Sequence Repeat (SSR) markers was applied on incoming samples of rust infected plant material without prior recovery, whereas the race phenotyping was based on bioassays using differential wheat lines inoculated with spore samples of alive, pure isolates grown under strict experimental conditions (Hovmøller et al. 2017). Incoming leaf rust samples were recovered and purified, then phenotyped from bioassays on a set of differential wheat lines (Goyeau et al., 2006), and genotyped based on a set of 19 SSR markers (Goyeau et al., 2012). **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. For details, see the rationale for the naming of significant *P. striiformis* races and genetic groups (Ali et al. 2017) and an updated summary of genetic groups and races on the GRRC website. The genetic grouping of stem rust is based on Olivera Firpo et al. (2017), Olivera et al. (2019), and detailed alignment of the SSR and SNP genotyping results between the Cereal Disease Lab (USA) and GRRC; the race grouping is based on 20 internationally agreed wheat lines differentiating virulence in stem rust (Jin et al., 2008).

In yellow rust, we have generally observed a strong correlation between genetic groups and race phenotypes in clonal populations, similarly, most races of wheat stem rust were associated with specific genetic groups ('clades'). 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. However, in sexual populations we observed unique and diverse genotypes and races. These and other infrequent groups are termed "other" in tables and online maps (<a href="www.wheatrust.org">www.wheatrust.org</a>).

For leaf rust, genetic groups are named "families", noted "Fam", followed by a 3 to 7 digits number coding the virulence phenotype, based on an octal code adapted from Gilmour (1973). Thus, a family gathers individuals sharing the same genotype according to the 19 SSR markers used at INRAE, and a similar combination of virulence.

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 (<a href="http://www.wheatrust.org/">http://www.wheatrust.org/</a>), including an updated table showing the relationship between races and genetic groups.

In 2020, 256 (stem rust), 261 (yellow rust), and 98 (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 64 (stem rust), 196 (yellow rust) and 74 (leaf rust) based on submissions from European countries in 2020

For yellow and stem rusts, 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). Diversity being higher in leaf rust than for the two other rusts, purification of isolates before genotyping was necessary.



**Table 1.** Number of stem rust isolates (*P. graminis* f.sp. *tritici*) handled by GRRC in 2019-2020.

		2019			2020							
Country	Dead	Recovered	Total	Dead	Recovered	Total	Grand total					
Austria					3	3	3					
Czech Republic					4	4	4					
Denmark		10	10	1	3	4	14					
France		1	1		4	4	5					
Germany		4	4				4					
Hungary		1	1	1	1	2	3					
Ireland				1	1	2	2					
Italy	12	55	67	8	18	26	93					
Norway					2	2	2					
Poland				1		1	1					
Slovakia				4	5	9	9					
Slovenia				4	9	13	13					
Spain	2	78	80	1	14	15	95					
Sweden	2	6	8				8					
	16	155	171	21	64	85	256					

**Table 2.** Number of yellow rust samples and derived isolates (*P. striiformis*), GRRC 2020.

		20:	20		
Geographic group	Country	Dead	Recovered	<b>Grand Total</b>	
Australasia	Australia	30		30	
Europe	Belgium		13	13	
	Czech Republic	1	2	3	
	Denmark	12	81	93	
	France	4	13	17	
	Germany		3	3	
	Great Britain		7	7	
	Hungary	2	4	6	
	Italy	4	8	12	
	Latvia		2	2	
	Netherlands	1	7	8	
	Norway	8	8	16	
	Poland	2	5	7	
	Slovakia		3	3	
	Spain	10	19	29	
	Sweden	15	20	35	
	Switzerland	6	1	7	
Europe Total		65	196	261	



Number of samples recovered	Samples state				
Country	Dead	None	Recovered	<b>Grand total</b>	
Belgium	1		1		2
Switzerland	2		2		4
Czech					
Republic			11		11
Denmark	8				8
Greece		2	2		4
Italy	5	1	36		42
Norway	2				2
Slovakia	1		5		6
Slovenia	1		8		9
Spain	1		9		10
Grand total	21	3	74		98

**Table 3.** Number of leaf rust samples and derived isolates (*P. triticina*) handled by INRAE in 2020.

## WHEAT STEM RUST RESULTS 2019-2020

A total of 173 samples from 13 countries 2019-2020 were successfully genotyped (Table 4); 205 of these were also race typed using a standard set of 20 wheat lines used to differentiate races of stem rust (Table 5).

Country	Genetic group	2019	2020	Total		
Austria	Clade III-B		2	2		
	Clade IV-F		1	1		
Czech Republic	Clade III-B		1	1		
	Other		2	2		
Denmark	Clade IV-F	6		6		
	Other		2	2		
France	Clade IV-B		4	4		
	Clade IV-F	1		1		
Germany	Clade IV-F	2		2		
Hungary	Other	1		1		
Ireland	Clade IV-B		2	2		
Italy	Clade III-B	22	12	34		
	Clade IV-F	22	11	33		
Norway	Clade IV-F		1	1		
Slovakia	Clade III-B		6	6		
	Clade IV-F		1	1		
Slovenia	Clade III-B		14	14		
	Clade IV-F		2	2		
Spain	Clade III-B	3		3		
	Clade IV-B	1	14	15		
	Clade IV-F	5		5		
	Other	33		33		
Sweden	Clade IV-F	1		1		
	Other	1		1		
		98	75	173		

**Table 4**. SSR genotyping of samples of *P. graminis* f.sp. *tritici* collected in 2019 and 2020. Results are shown as number of isolates within genetic group (clade). Significant races within clades are shown in Table 5 and virulences in Table 6. Graphical presentation of results available on <a href="https://www.wheatrust.org">www.wheatrust.org</a>.



Genetic group	Race_name	2019	2020	<b>Grand Total</b>
Clade III-B	TTRTF	10	10	20
	- a)	15	26	41
Clade III-B Total		25	36	61
Clade IV-B	TKTTF	1	17	18
	- a)		3	3
Clade IV-B Total		1	20	21
Clade IV-F	TKKTF	15	7	22
	- a)	22	9	31
Clade IV-F Total		37	16	53
Other	> 30 races	33	2	35
	- a)	2	5	7
Other Total		35	7	42
<b>Grand Total</b>		98	79	177
a) not tested				

**Table 5.** Relationship between genetic groups and prevalent races detected within these in Europe, 2019-2020

Table 6. Correspondence between race name and virulence on common stem rust differential lines

				\	/irule	nce c	orres	pond	ing to	NA d	ifferer	ntials	1-20 (ı	main F	R-gen	e indica	ated)			
Race-Name	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	<i>36</i>	-	-	17	9a	-	10	-	-	-	-	McN
QCHNC	5	21	-	-	-	-	-	9g	-	9b	-	17	9a	-	10	-	-	-	-	McN
RFCNC	5	21	-	7b	-	-	8a	9g	-	-	-	17	9a	-	10	-	-	-	-	McN
TKKTF	5	21	9е	7b	-	6	8a	9g	-	9b	<i>30</i>	17	9a	9d	10	Tmp	-	-	<i>38</i>	McN
TKTTF	5	21	9e	7b	-	6	8a	9g	-	9b	<i>30</i>	17	9a	9d	10	Tmp	-	-	<i>38</i>	McN
TTKSK	5	21	9е	7b	11	6	8a	9g	-	9b	<i>30</i>	17	9a	9d	10	-	-	<i>3</i> 1	<i>38</i>	McN
TTKST	5	21	9e	7b	11	6	8a	9g	-	9b	<i>30</i>	17	9a	9d	10	-	24	<i>3</i> 1	<i>38</i>	McN
TTKTK	5	21	9е	7b	11	6	8a	9g	-	9b	<i>30</i>	17	9a	9d	10	Tmp	-	<i>3</i> 1	38	McN
TTKTT	5	21	9е	7b	11	6	8a	9g	-	9b	<i>30</i>	17	9a	9d	10	Tmp	24	<i>3</i> 1	<i>38</i>	McN
TTRTF	5	21	9е	7b	11	6	8a	9g	<i>36</i>	9b	-	17	9a	9d	10	Tmp	-	-	<i>38</i>	McN
TTTTF	5	21	9е	7b	11	6	8a	9g	<i>36</i>	9b	<i>30</i>	17	9a	9d	10	Tmp	-	-	<i>38</i>	McN

Clade III-B (TTRTF), present in the epidemics in Sicily in 2016, has been detected in seven countries in Europe and four countries in Africa/Asia. In 2019, first detected in Spain, Tunisia and Iran, and in 2020 first detected in Austria, Czech Republic, Slovakia, and Slovenia.

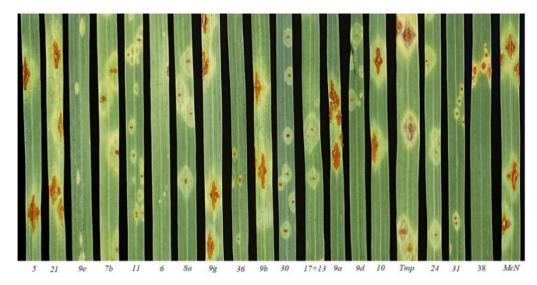
Clade IV-F, containing race TKKTF, was detected in 11 European countries in 2019-2020, and previously also in Egypt, Iran and Tunisia.

Clade IV-B contained two closely related races, TKTTF and TTTTF. In this study, TKTTF was detected in France, Ireland and Spain in 2020, whereas TTTTF was not detected in Europe.

Clade I (Ug99) and associated races have not been detected in Europe. However, multiple Sr31-virulent races, which were not related to Clade I, were detected in a recombining population in Spain in 2019.

Additional unusual races were observed in local areas in Czech Republic (RFCNC) and Denmark (QCHNC), the latter previously detected in Sweden in 2019.





**Figure 1**. Infection type responses on wheat differential lines of a stem rust race (QCHNC) with narrow virulence spectrum.

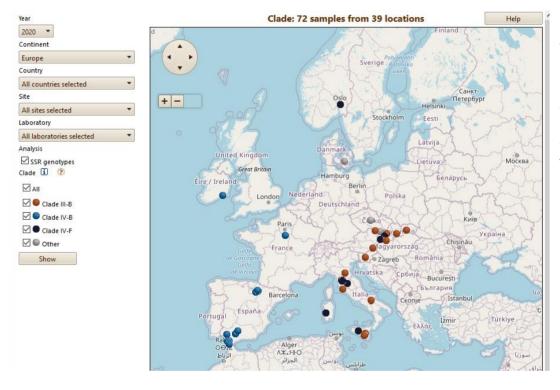


Figure 2. Distribution of genetic groups of stem rust based on samples submitted to Aarhus University, 2020 (samples with GPS information only). Detected races within groups shown in Table 5

## **WHEAT YELLOW RUST RESULTS 2020**

A total of 408 samples collected in 16 European countries in 2019 and 2020 were successfully genotyped (Fig. 3; Table 7). Race typing was done for 55 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, 2020 (samples with GPS information only). Detected races within groups shown in Table 8.

A new genotype in the Middle East (provisionally termed ME2018, 'Other' on the map), first detected in dead samples from Egypt (2018), and at several locations in Turkey (2019), was detected for the first time in Europe, Latvia (2020). This observation was confirmed by independent assays of incoming infected leaves, race typing of recovered isolates, and re-submission of additional samples from the sampling area. Two races differentiated by Yr10-virulence were detected within ME2018.

PstS13 has been detected in multiple countries and continents in recent years. In 2019, a new Yr10-virulent variant was detected in Poland, and in 2020 also in Germany. 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). Alignment experiments based on DNA samples supplied by the University of Sydney, Australia, confirmed PstS13 in Australia, where it was first detected in 2018 (Park et al., 2020). At present, PstS13 is widespread across three continents, Europe, South America and Australia. 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 8). 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.

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 5). However, in recent years, new races adapted to local 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'. So far, it has not been possible to diagnose the new races within PstS10 by molecular techniques nor by standard wheat differential lines. By 2020, four races have been detected in PstS10, typically named after the wheat varieties where they first caused significant epidemics. Alignment experiments with University of Sydney confirmed the presence of PstS10 in Australia since 2017 (Park et al., 2020).

In terms of virulence, the races within PstS10 are similar to the Warrior race (PstS7), except for the resistance



specificity in the varieties Warrior (UK) and Ambition (DK). The "Warrior" race is still present in Europe, but often in low frequency and mainly in Eastern Europe. Likewise, the PstS8 group (containing the "Kranich" race) has become less frequent in Europe. A new race in a new unique genetic group, termed PstS15, was detected in France in 2017-2019, and in Denmark and Sweden in 2019-2020. The significance of this race in terms of potential shifts in rust susceptibility is subject to investigation in the EU-RustWatch project.

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Country	Genetic group	2019	2020	Grand Total		
Belgium	PstS10	15	10	25		
	PstS13	1	1	2		
	PstS7		1	1		
Czech Republic	PstS0		1	1		
	PstS10	3	1	4		
Denmark	PstS10	32	59	91		
	PstS13	12	9	21		
	PstS15	5	4	9		
France	PstS10	12	10	22		
	PstS15	1	1	2		
	PstS7	3	1	4		
Germany	PstS10	6		6		
	PstS13	4		4		
	PstS7	1		1		
	PstS8	1		1		
Great Britain	PstS10		4	4		
Hungary	PstS13		1	1		
	PstS7	4	3	7		
Italy	PstS10	3	8	11		
	PstS13	24	2	26		
	PstS13	1		1		
	PstS14	1		1		
	PstS2	1		1		
	PstS7	2		2		
Latvia	Other		1	1		
	PstS10	1		1		
	PstS14	18		18		
	PstS7	1		1		
Netherlands	PstS10	3	7	10		
	PstS7	1		1		
Norway	PstS10	5	7	12		
	PstS13		2	2		
	PstS7		1	1		
Poland	PstS13	3		3		
	PstS7	2		2		
Slovakia	PstS10	2		2		
	PstS14	1	1	2		
	PstS7	1	2	3		
Spain	PstS10	9	17	26		
	PstS13	11	6	17		
	PstS14	7		7		
	PstS7		3	3		
Sweden	PstS10	16	15	31		
	PstS13	5	9	14		
	PstS7		1	1		
	PstS8		1	1		
Switzerland	PstS14		1	1		
Total		218	190	408		

**Table 7**. SSR genotyping of samples of *P. striiformis* 2019-2020. Results shown as number of isolates within genetic groups in each country, significant races and virulences within each group shown in Table 8.



**Table 8**. Correspondence between genetic groups and prevalent races of *P. striiformis* sampled from epidemic sites since 2000, Global Rust Reference Center, January 2020.

Genetic group	Race	Virulence phenotype*	Prevalence in geographical regio
PstS0	Brigadier	1,2,3,-,-,-,9,-,-,17,-,25,-,-,Av8,-	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
stS1	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
stS2	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	-,2,-,-,6,7,8,9,10,-,-,24,25,-,-,-,AvS,-	East Africa, West Asia
	PstS2,v10,v27	-,2,-,-,6,7,8,9,10,-,-,24,25,27,-,-,AvS,-	West Asia
	PstS2,v3,v10,v27	-,2,3,-,-,6,7,8,9,10,-,-,24,25,27,-,-,AvS,-	East Africa
stS3	PstS3	-,(2),-,-,-,6,7,8,-,-,-,-,-,-,AvS,-	Europe, North Africa, West Asia
	PstS3,v25	-,(2),-,-,-,6,7,8,-,-,-,-,25,-,-,-,AvS,-	Europe, East Africa
stS4	Triticale2006	-,2,-,-,6,7,8,-,10,-,-,24,-,-,-,-	Europe
stS5	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
stS6	PstS6	1,2,-,-,-,6,7,-,9,-,-,17,-,-,27,-,-,AvS,-	East Africa, Central Asia, South Asia
stS7	Warrior	1,2,3,4,-,6,7,-,9,-,-,17,-,25,-,32,Sp,AvS,Amb	Europe, North Africa
stS8	Kranich	1,2,3,-,-,6,7,8,9,-,-,17,-,25,-,32,-,AvS,Amb	Europe
stS9	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
stS10	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
	Amboise	1,2,3,4,-,6,7,-,9,-,-,17,-,25,-,32,Sp,AvS,-	Europe
stS11	PstS11	-,2,-,(4),-,6,7,8,-,-,-,17,-,-,27,32,-,AvS,-	Central and West Asia, East Africa
stS12	Hereford	-,2,3,-,-,6,7,8,-,-,-,17,-,25,-,32,-,AvS,-	Europe
stS13	Triticale2015	-,2,-,-,6,7,8,9,-,-,-,AvS,-	Europe, South America, West Asia
	Triticale2015,v10	-,2,-,-,6,7,8,9,10,-,-,24,-,-,-,AvS,-	Europe
stS14	PstS14	-,2,3,-,-,6,7,8,9,-,-,17,-,25,-,32,(Sp),AvS,-	Europe, North Africa

<sup>\*</sup> 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, Benchmark and Amboise are new races in PstS10, which can only be distinguished by these varieties.

# **WHEAT LEAF RUST RESULTS 2020**

In 2020, a total of 98 samples were received. From the gathered information, they were collected on at least 59 different cultivars (15 of which were durum wheat cultivars). Three of the recorded samples were not considered, with no display of any rust symptoms. 21 samples were not recovered and were probably dead upon arrival.

The 20 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.

A single-pustule isolation was made for the 74 recovered samples. Purified leaf rust samples collected on bread wheat were phenotyped, race typing being adapted from Goyeau *et al.* (2006). All of the samples, including those that could not be recovered, were sent for SSR genotyping. Genotyping results are expected for the end of April.

**Table 9**. Number of leaf rust samples and derived single pustule isolates processed

	Dead (or no LR)	Single-pustule isolated	Total	Sent for SSR genotyping (awaiting results)	Phenotyped
Durum wheat	3 (2)	20	23	23	0
Bread wheat	18 (1)	54	72	72	53
Total	21 (3)	74	95	95	53

Race typing allowed us to postulate five families according to their profile of virulence in 2020, which will be confirmed once the genotyping results are available. Two well-known families were observed this season: Fam 106 314 2 and Fam 166, already identified in France and across Europe (figure 4) as being the two main leaf rust families. Surprisingly, although Fam 166 is still the main family in Europe in 2020 (81% of the samples), Fam 106 314 2 is way less represented than in the previous years (6% of the samples). This could be a result of the reduced number of samples or reduced number of participating countries this year.

Other less prevalent families were found, such as Fam 167 or Fam 165 vir24, with no significant change in frequency in 2020.

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 (see table 10) included in the differential set.

**Table 10**. Pathotypes found in the genetic group Fam166.

Genetic group	Pathotype Code		Virulence phenotype																		
Fam	166 316	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	-	-	-	17b	37	-	-
166	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	-	-



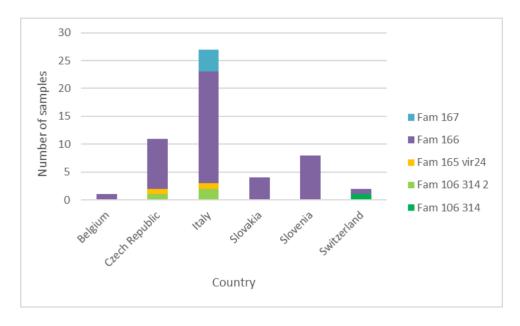


Figure 4. Chart of leaf rust families' distribution across Europe in 2020, based on race typing results

Table 12. Leaf rust virulence phenotypes of 2020 samples

Postulated genetic	Pathotype									V	irulenc	e phe	noty	pe							
group	Code																				
Fam 106 314 2	106 314 2	1	-	-	-	-	-	-	10	13	14a	15	-	17	-	-	-	-	37	-	28
Fam 106 314	106 314	1	-	-	-	-	-	-	10	13	14a	15	-	17	-	-	-	-	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	-	-
	166 347	1	-	-	-	3	3bg	-	10	13	14a	15	-	-	-	23	26	17b	37	-	-
	166 357	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	-	23	26	17b	37	-	-
	166 375	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	20	23	26	-	37	-	-
	166 376	1	-	-	-	3	3bg	-	10	13	14a	15	-	17	20	23	-	17b	37	-	-
Fam 167	167 314	1	-	-		3	3bg	3ka	10	13	14a	15	-	17	-	-	-	-	37	-	-
	167 316	1	-	-	-	3	3bg	3ka	10	13	14a	15	-	17	-	-	-	17b	37	-	-
	167 334	1	-	-	-	3	3bg	3ka	10	13	14a	15	-	17	20	-	-	-	37	-	-
Fam 165 vir24	165 316 1	1	-	-	-	3	3bg	3ka	-	13	14a	15	-	17	-	-	-	17b	37	24	-

# **ALIGNMENT**

Alignment experiments for yellow and stem rust were carried out in Denmark January-March 2020 based on isolates from UK, Germany, Poland and Switzerland. Additional experiments for yellow rust were carried out at INRAE and AU in August-September based on samples collected in France and Denmark, and seed exchange of specific wheat varieties. )

	No. Isolates						
Country	Recovered	Genotyped	Race typed				
France	4		2				
Germany	10	3	6				
Great Britain	17	6	6				
Poland	16	5	10				
Switzerland	1	1	1				
Total	48	15	25				

**Table 13**. Number of yellow rust isolates investigated in alignment experiments in 2020



Overall, the experiments were successful, by far most rust samples appeared pure, two samples of yellow rust showed too low level of infection for race scoring, and two samples were contaminated by leaf/stem rust. The follow-up genotyping results, generally based on 2 independent leaf segments from two contrasting differential lines, were conclusive in all cases, supporting the race typing results, including cases with indication of a low level of mixture (multiple races). The results confirm that the most widely prevalent genetic group in yellow rust, PstS10, contain multiple races, e.g., Warrior(-), Kalmar, Benchmark, and Amboise, and that these are present in multiple countries. Additional races may be identified in case additional wheat varieties were included. The infection type (IT) for such additional cultivars may only show quantitative differences and therefor difficult to conclude unless virulent/avirulent controls are included as references in the experiment. In one case, three genotypes were detected within a single sample, confirming that multiple races may co-exist on individual plants and leaves under field conditions.

Five stem rust samples from Germany were included, most from the 2013 epidemic. The results confirmed the presence of races in the German 2013-epidemic, which have also been observed in Europe in recent years. In conclusion, SSR genotyping was 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. In winter/spring 2021, 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. It is therefore recommended to include 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 present pathogen genotyping/phenotyping results from national rust diagnostic on maps and charts in a common, interactive information system hosted by the Wheat Rust Toolbox, without creating bias in results arising from 'technical/experimental' differences among labs and environments.

The alignment experiments in spring 2021 is at present evaluated by project partners. The 2021 experiments confirmed that the current procedures are appropriate for aligning results from the European rust diagnostic labs. Steps to secure that continued efforts for trans-national alignment of results of national rust surveillance efforts is recommended.

Alignment experiments for leaf rust are being carried out in France since February 2021 based on isolates from UK, Poland, Czech Republic and Slovakia.

	Number of isolates				
Country	Recovered	Ongoing genotyping			
UK	5	5			
Poland	10	10			
Czech Republic	6	6			
Slovakia	4	4			

**Table 14**. Number of leaf rust isolates under investigation in alignment experiments in 2020

All of the samples were successfully recovered; however, phenotyping could not take place yet for work organization and sharing of greenhouse spaces matters. Comparison of virulence profiles with the participating countries is thus still awaiting. DNA extraction was carried out on all of the samples, and DNA samples were sent for genotyping, for which results are expected end of April 2021.

Genotyping in 2020 is made on the usual 19 SSR set used at INRAE, although efforts are being made to build a common European SSR set with the help of Pawel Czembor from IHAR based on his recent publication (Czajowski et al., 2021). Among the 34 SSR markers proposed for leaf rust, 23 have been chosen for their polymorphism and compatibility with



the current set and method. The resulting set of 42 markers has yet to be tested with samples of interest to identify the best combination to represent the European leaf rust diversity.

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# Table 15. Sample collectors

Country	Collectors 2018-2019
Austria	Michael Oberforster
	Thomas Massinger
Belgium	F.De Brouwer; J. Pannecoucque
	G. Jacquemin & R. Meza
Czech Republic	Alena Hanzalova
	Pavel Kraus
Denmark	Ghita Cordsen Nielsen
	Jeppe R. Andersen
	Lise Nistrup Jørgensen
	Susanne Sindberg
	Thomas Olsen
France	Clément Debiton
	Emmanuel Heumez
	Gorichon
	J.P. Maigniel
	Jérome Auzanneau
	Laurent Pageaud
	Marc Leconte
	Mathieu Grare
	Pascal Giraudeau
	S. Barrais; Vallerie Cadot
Germany	Anja Giebel
•	Anne-Kristin Schmitt
	Catherine Cuendet, Getreidezuchtung Peter Kunz
	Cornelia Miersch, SMUL
	E. Kazman
	Friederike Schiermann
	Henning Hartwig, PZO
	Josef Holzapfel
	Kerstin Flath
	Matthias Haas
	Paul Gruner
	Sommerfeldt, Hoffmann
	Thomas Grafe
	Thomas Zschaeckel
	Uta Liesenberg
	Uwe Preiss
Italy	Angela Iori, Fabrizio Quaranta, Andreina Belocchi, Mauro Fornara
•	Anna Maria Mastrangelo
	Antonia Salerno
	Biagio Randazzo
	Daniela Marone
	Emilia Romagna
	Francesca Nocente
	Giovanni Laidò, Rossano Contato
	Giuseppina Goddi
	Luca Fiorilla



Lucio Salafia

Oriana Porfiri

Stefania Stefanelli, Marco Maccaferri

Virgilio Balmas

Latvia Līga Feodorova-Fedotova Netherlands Lubbert van den brink

**Norway** Andrea Ficke

Chloe Grieu

Morten Lillemo

**Poland** Ewelina Piwowarczyk

**Pawel Czembor** 

Slovakia Svetlana Slikova

Alena Hanzalova

**Spain** Carlos Cantero

Dolors Villegas Enrique Can Ibal Elorza Jesús Goñi Joan Serra Luis Urquijo Manuel Calvo

Neus Pulg; Nieves Apa

**Sweden** Alexia von Ehrenheim

Alf Djurberg

Anders Arvidsson
Anders Lindgren
Anna Berlin
Anna Gerdtsson
Anna-Karin Krijger
Charlotte Norén
Elisabeth Bölenius
Elisabeth Lövstad
Erling Christensson

Eva Mellqvist Frans Johnson Gunilla Berg

Gunnel Andersson, Linda af Geijersstam

Johanna Holmblad Jonas Törngren

Julia Dahlqvist, Anna von Heideken

Karin Andersson Kristian Barck Kristian Jochnick Lars Johansson Lina Norrlund Louise Aldén Lovisa Eriksson Lukas Hallberg

Robert Dinwiddie



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