

Long distance spread and evolution of the yellow rust pathogen *P. striiformis* f.sp. *tritici* in NW-Europe and its relevance for plant breeding

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Introduction

Wheat production in Denmark is based on a rapid change in cultivar distribution to gain maximum benefit from the increase in yield potential and enhanced level of resistance to fungal pathogens in new cultivars. From 1985 to 1999 the Danish wheat area (harvested) increased significantly and large changes in distribution of the most commonly grown cultivars occurred (HOVMØLLER, 2001). In the mid 1980s, *Yr1* was deployed over a relatively large area in the cvs Kraka and Longbow. These cultivars were replaced by with Sleipner carrying *Yr9* resistance, which later was replaced by two cvs, Haven (*Yr9* and *Yr6*) and Pepital with highly efficient resistance in adult plants (APR-resistance). In 1993, *Yr17* was introduced in Denmark, first in cvs Hussar and Brigadier and subsequently in Lynx, the former two combining *Yr17* and *Yr9*, and the latter probably combining *Yr17*, *Yr9* and *Yr6*. Recently, *Yr1* was reintroduced on a large scale in Ritmo. The changes in distribution of cultivars from 1985 to 1999 resulted in large changes in the distribution of resistance genes, and thereby the selection forces affecting the pathogen population.

The diversity of 'races' or 'pathotypes' is often low in populations of *Puccinia striiformis* f.sp. *tritici*, the causal agent of yellow rust on wheat, although the diversity may vary considerably between region, year and host cultivar. However, the low genetic diversity does not prevent the fungus from a rapid evolution of new pathotypes, and thereby cause previously resistant wheat cultivars to become susceptible to the yellow rust disease. So despite the mechanisms by which new variation is created is not fully understood, mutation from avirulence to virulence may occur fairly frequent.

Materials and methods

More than 350 isolates collected in the Danish virulence survey between 1993 and 2000 were pathotyped (HOVMØLLER, 2001). They were collected from a wide range of locations and cultivars in field trials and from farmers' fields. If possible, at least two samples at each location and year were taken from the susceptible standard, Anja, in order to obtain isolates that had not been subject to direct selection by their host. A total of 15 different pathotypes were found.

76 of the isolates were selected for an AFLP analysis (JUSTESEN et al., 2002). The isolates represented most Danish locations and cultivars where yellow rust was observed in these years so that all combinations of year of collection, geographical origin and source cultivar were represented. The isolates were collected from leaves with low disease severity, as detached leaf segments each containing a single lesion. The leaf segments were incubated on water agar in petri dishes immediately after sampling, as described by HOVMØLLER, 2001. In order to investigate migration of *P. striiformis* within NW Europe, additional samples from the UK, France and Germany were obtained. The 12 isolates from the UK represented eight pathotypes that were detected in the previous Danish study (JUSTESEN et al., 2001) and one unique pathotype. The six French and six German isolates represented all known *Yr17* virulent pathotypes from these countries in 1997-1998.

Results and discussion

Virulence- and AFLP assays

All isolates possessed virulence for *Yr3* and the resistance in Strubes Dickkopf (SD), and avirulence for *Yr5*, *Yr7*, *Yr8*, *Yr10* and the resistance in Spaldings Pro-

lific (SP). Polymorphic loci were indicated by the following pathotype code: virulence for *Yr1* as '1', virulence for *Yr2* as '2', virulence for *Yr3b+Yr4b* as '4', virulence for *Yr6* as '6', virulence for *Yr9* as '9', virulence for *Yr17* as '[17]' and virulence for Carstens V as '[CV]'. Avirulence for these resistances is shown by '-' (Figure 1). On average, no differences in pathotype frequencies were observed whenever samples were collected from Anja or from other cultivars. However, some of the most rare pathotypes were only observed on cultivars with at least some matching host resistance genes, e.g. pathotype 1,2,4,6,9,17 collected from Ritmo (*Yr1*), Haven (*Yr6*, *Yr9*), Lynx and Madrigal (both *Yr6*, *Yr9* and *Yr17*), and pathotype 1,2,CV that were only observed on Kraka (*Yr1*, Carstens V).

The AFLP-technique gave informative DNA fingerprints. Twenty-one +2 primer combinations were selected, which produced reproducible and polymorphic fragments that were easy to score. The 21 combinations revealed 28 AFLP-markers (polymorphic fragments) when all 76 isolates were examined. Of the 28 AFLP markers, 21 occurred at frequencies between 10 and 90% (JUSTESEN et al., 2002). The 28 markers identified 16 AFLP phenotypes among the isolates, which were located at four main branches in a phenetic tree (Figure 2). Two of these branches contained *Yr17* virulent isolates; one with isolates of pathotype 12--9[17]- (AFLP group K) and the other with isolates of pathotype 124-9[17]- (AFLP group O and P). A third branch had mainly isolates of the -24-9- pathotype (AFLP groups E and G). The fourth branch contained more diverse isolates, some combining virulence for *Yr6* and *Yr9* (groups A, B, D and R), and some with virulence for *YrCV* (groups S and T).

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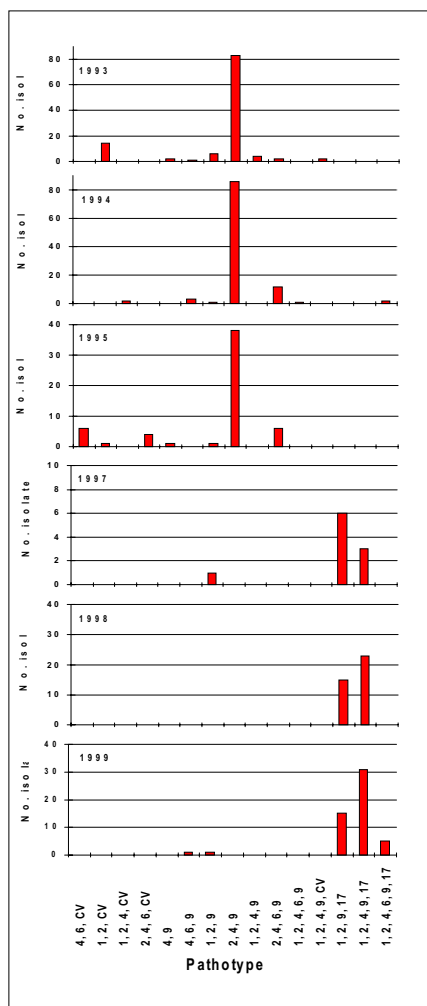


Figure 1: Number of isolates of the 15 *P. striiformis* pathotypes observed in Denmark from 1993 to 1999 (HOVMØLLER, 2001). Pathotype code: virulence for *Yr1* as '1', virulence for *Yr2* as '2', virulence for *Yr3b+Yr4b* as '4', virulence for *Yr6* as '6', virulence for *Yr9* as '9', virulence for *Yr17* as '[17]' and virulence for Carstens V as '[CV]'.

HOVMØLLER et al., 2002 showed that there was effectively a single *P. striiformis* population in the UK, northern France, Germany and Denmark, up to 1700 km apart, consistent with a 'continent-island' model in which Denmark was mainly the recipient of migrants from other countries. In five cases, specific pathogen clones were dispersed between the UK and Denmark, and on at least two recent occasions, clones were also spread

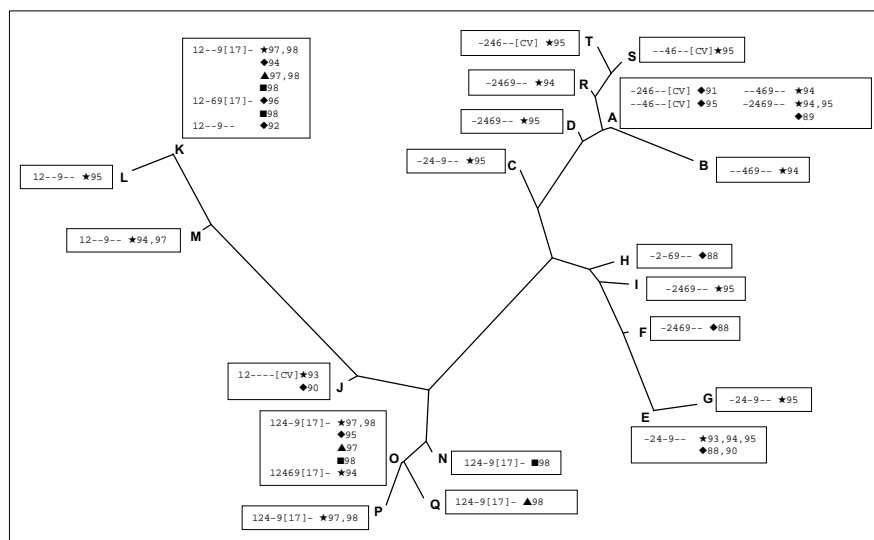


Figure 2: Unrooted tree of *Puccinia striiformis* f.sp. *tritici* representing AFLP variation in NW Europe between 1988 and 1998 (HOVMØLLER et al., 2002). Isolates were grouped in 21 AFLP-phenotypes, positions shown by capital letters. Symbols refer to country of origin of isolates (u UK; n Germany; ▲ France; * Denmark), and number prior to symbol pathotype code. Numbers after the symbols indicate years of sampling. The length of the bar equals one polymorphism.

from the UK to Germany and France, causing outbreaks of yellow rust on wheat cultivars that were previously resistant to the disease in these countries. The agronomic consequences of migration were enhanced because of the limited genetic diversity for yellow rust resistance in wheat cultivars in the area. These results demonstrate that long-distance migration of pathogen clones, coupled with low diversity in the host species, may cause previous useful resistance genes to become ineffective for disease control on a continental scale. This may lead to the following conclusions:

- Low amount of genetic diversity at scales from single field to NW-European countries; in DK particularly in years following re-appearance of the fungus
- Data consistent with hypothesis of new pathogen variation due to single step mutation, i.e. loss/gain of AFLP fragments and virulence/avirulence function

- A specific virulence character may evolve more than once
- In two recent cases, particular pathogen clones spread from UK to Denmark (and France and Germany), causing outbreaks of yellow rust on previously resistant varieties (*Yr17*) in these countries
- Pyramiding R-genes probably not a sustainable solution in the yellow rust/wheat pathosystem
- Long-term yellow rust resistance breeding must rely on germplasm with high levels of partial resistance.

References

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