New nomenclature of Races of *Plasmopara halstedii* (Sunflower Downy Mildew)

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Summary:

A proposal, formulated by pathologists from six countries, is made for a standardised set of nine, publicly available lines to be used as "differential lines» to identify races of sunflower downy mildew (*Plasmopara halstedii*). In addition, a nomenclature system using a triplet code, which is concise and easy to use, is presented. It should allow pathologists to "name" unambiguously a new race and at the same time convey information on its virulence pattern. This new nomenclature was used to replace those used previously (US system: races 1, 2, 3…., French system: races A, B…). At present, 10 races have been described: 100, 300, 310, 330, 700, 703, 710, 711, 730 and 770.

Résumé :

Les pathologistes de 6 pays, se sont mis d'accord pour proposer une nouvelle nomenclature des races de *Plasmopara halstedii*. Elle est basée sur l'utilisation de lignées hôtes-différentielle disponibles au près des sélectionneurs publics. Cette nomenclature qui utilise un système de codification à trois chiffres va permettre de supprimer toutes ambiguïté dans la caractérisation des races et assurer la transparence dans l'information scientifique. Cette nouvelle nomenclature est a utiliser en remplacement des nomenclatures antérieures (nomenclatures américaine : races 1, 2, …et française : races A, B, …). Actuellement, 10 pathotypes ont été décrites : les races 100, 300, 310, 330, 700, 703, 710, 711, 730 et 770.

Introduction:

Until 1980, the situation with races of *Plasmopara halstedii*, the obligate pathogen causing downy mildew of sunflower, was very simple, because only two races were known. Since that time, however, there have been a multitude of races identified, first in North America (Gulya et al., 1991[b]; Rashid, 1993), and subsequently in several European countries following extensive surveys (Gulya et al., 1991 [b]; Spring et al., 1994; Viranyi and Gulya, 1995; Roeckel-Drevet et al., 1997), and ultimately in all countries where sunflower downy mildew has been recorded. The use of a variable number of lines to characterise races, and the absence of a universally accepted nomenclature system has led inevitably to a situation where information on races was not readily understandable by sunflower researchers in other countries. Sackston et al. (1990) summarised sources of resistance known in 1990 and what was known of the genetics, and suggested a system of race nomenclature in which the virulence pattern (a list of the total number of resistance genes attacked) was used for race identification. Gulya et al. (1991[a]) expanded upon this by proposing a set of 15 lines, and continued the practice of identifying races according to a list of the genes they were virulent upon. When resistance genes in a given line had not been verified, a letter designating the anticipated gene was used. Thus, what was once referred to as race 5, a very virulent race, was designated as race 1,2,3,4,5,a,b,c,d,e. This denoted that race 5 was virulent on *Pl*1, *Pl*2, *Pl*3, *Pl*4, and *Pl*5, and genes a, b, c, d, and e, whose inheritance had not been demonstrated conclusively in printed research.

While this nomenclature system presented exact information on virulence, it obviously led to cumbersome race "labels" which would be difficult to remember and awkward to use in print or speech. In addition, segregation studies have shown that some *Pl* genes are, in fact, clusters of genes, each giving resistance to one or a few races. (Vear et al, 1997). It is therefore not possible, at present, to use gene names to name races. Gulya et al. (1998) suggested adopting a triplet coding system as proposed by Limpert and Muller (1994) for plant pathogens in general. This nomenclature has been applied for European and American races.

Materials and methods

Standardised set of differentials:

Several requirements for ideal differential lines were proposed. The genotypes should be (1) publicly available, (2) fixed inbred lines, rather than hybrids or composites, (3) offer consistent downy mildew reactions, (4) and preferably have had some classical or molecular research into the genetics of downy mildew resistance. In some cases, lines with supposedly identical downy mildew reactions are known (e.g. Rha-265, Rha-266, and AD-66 all resistant only to European race and have been used interchangeably by researchers. From the standpoint of uniformity, however, the group agreed that a single line, with no substitutions, should be used. Additionally, because many of the proposed lines were originally released up to 25 years ago, with the possibility of divergent selections existing, it was agreed that a single seed source be used for distribution. Requests for seed should therefore be made directly to the breeders (J. Miller, USDA; D. Skoric, IFVC; F. Vear, INRA).

The nine differentials will be grouped into three sets of three lines. The first set will consist of three USDA lines. HA-304 is proposed as the "universal susceptible". Rha-265 has resistance to European race. Rha-274 is known to confer resistance to European races and Red River race.

The second and third set of differentials are different from those previously proposed. The second set will consist of PMI3, PM-17 and 803-1. PMI3, an INRA selection of the USDA composite DM-2, was felt to offer more uniformity in downy mildew reaction because it is a fixed line as contrasted with DM-2. PM-17, a line selected from PI 406022 by Rama Urs of Dahlgren, is proposed as a substitute for IS-2000, the latter a proprietary line of Interstate (now Advanta). When Pioneer Hi-Bred Int. acquired Dahlgren research and its seed inventory, Pioneer officials agreed to let this line be distributed publically as a differential line. The line 803-1 was selected by the breeding staff of the Institute of Field & Vegetable Crops, Novi Sad, Yugoslavia.
The third set of differentials consists of HAR-4, QHP1 and HA-335. HAR-4 is a USDA synthetic bred from an Argentine genotype developed by INTA (Saenz Pena 74-1-2). QHP1 was selected from a cross including HAR-5 made by INRA. Lastly, the line HA-335 is proposed as the line, which at the moment is resistant to all known mildew races.
We have decided to give the nine lines new designations to simplify the system. The differentials will be referred to as ***"D-1***" to ***"D-9***". Since the USDA and INRA system of naming germplasm is very different, and there are already two lines with very similar designations (PMI3 and PM-17), we felt that it would be advisable to give the differentials a uniform and simple label. The new designations and pedigrees of the differentials are found in Table 1.

Table 1: Sunflower differential lines for downy mildew race identification.

Designation	Original Name	Pedigree	Source of Resistance	Supplier
D-1	**HA-304**	Commander	None	USDA (J. Miller[1])
D-2	**Rha-265**	Peredovik/953-102	953-102 (Canada)	USDA (J. Miller[1])
D-3	**Rha-274**	HA-119/HA-62	953-88 (Canada)	USDA (J. Miller[1])
D-4	**PMI3**	selection of DM-2	Novinka (Russia)	INRA (F.Vear[2])
D-5	**PM-17**	PI 406022	? (Iran)	USDA (J. Miller[1])
D-6	**803-1**	*H. tuberosus* cross	*H. tuberosus* (Yugo)	IFVC (D. Skoric[3])
D-7	**HAR-4**	Saenz-Pena 74-1-2 selec.	? (Argentina)	USDA (J. Miller[1])
D-8	**QHP1**	HAR-5 x PRS7 Selec.	Guayacan INTA (Argentina)	INRA (F. Vear[2])
D-9	**HA-335**	HA-89 x *H. annuus*	wild *H. annuus*	USDA (J. Miller[1])

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Addition of new differentials in the future:

To insure continuity in race naming, it is suggested that the present nine differentials be used from now on. Once it becomes apparent that new lines are necessary to characterise new mildew virulence patterns, a complete new set of three lines will be added in order to continue using the triplet code. The differential lines can be reviewed and discussed every four years at the general meeting of the International Sunflower Association.

Triplet coding system:

An essential component of this proposal is the triplet coding system, which numerically assigns a value if a differential is susceptible, and thus conveys information on the virulence of an isolate. If the first line of a set of three is susceptible, it imparts a value of "1"; if the second line

is susceptible it imparts a value of "2"; and if the third line is susceptible it imparts a value of "4". The virulence code is additive within each set, so that if the first and second line are susceptible, they impart a value of 1 + 2, or 3. To continue the example, if the first and third lines are susceptible, they would impart a virulence code of 1 + 4, or 5, while if all three lines were susceptible the virulence code would total a "7". This is graphically illustrated in Table 2, where the triplet code for historical races: European race and Red River race is given, along with a fictitious race. In this system the higher the triplet code is, the more virulent the race. The final virulence code or "race name" of an isolate will be a 3-digit code, one digit from each of the three sets of lines.

Table 2: Triplet coding system for defining sunflower downy mildew races adapted from Limpert and Muller (1994).

Differential Line	*D-1*	*D-2*	*D-3*	*D-4*	*D-5*	*D-6*	*D-7*	*D-8*	*D-9*	*D-10…*	Triplet Code
Value if **S**.	1	2	4	1	2	4	1	2	4	…	
European race	**S**	**R**	**R**	**R**	**R**	**R**	**R**	**R**	**R**	…	
	1+0+0=**1**			0+0+0=**0**			0+0+0=**0**			**…**	**100 …**
Red River race	**S**	**S**	**R**	**R**	**R**	**R**	**R**	**R**	**R**	…	
	1+2+0=**3**			0+0+0=**0**			0+0+0=**0**			**…**	**300 …**
Race ?xyz?	**S**	**S**	**S**	**S**	**R**	**S**	**R**	**S**	**R**	…	
	1+2+4=**7**			1+0+4=**5**			0+2+0=**2**			**…**	**752 …**

S = susceptible; **R** = resistant; Race "xyz" = hypothetical race

Testing methods and interpretation of results:

In order for independent laboratories to characterise downy mildew isolates for their race reaction and achieve the same results, there must be some standardisation in methods and interpretation, but this does not imply that everyone must follow exactly the same procedure. There are currently many variations in the whole seedling immersion technique, with differences in inoculum concentration, inoculation period, growing medium, etc. Gulya *et al*. (1991), Mouzeyar *et al*. (1993, 1994), Gulya (1996) and Gulya *et al*. (1998) have reviewed this topic.

In the infection by seedling immersion, some resistant lines show a variable amount of sporulation on cotyledons (Cotyledon Limited Infection). However a non-virulent race never sporulates on the true leaves and this must be the main criterion for race characterisation. In the results, S is susceptible with sporulation on true leaves, R is resistant with no sporulation or sporulation on cotyledons only.

Results:

The new differential lines have been analysed in different countries, the results are presented in table 3.

Table 3: Reaction of sunflower differential lines to downy mildew race and triplet codes.

	Old Race Denomination														
	American name											French name			
	1	2	3	4	5	6	7	8	9	10	11	A	B	C	D
Line															
SET ONE															
D-1	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**
D-2	R	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**	**S**
D-3	R	R	**S**	**S**	**S**	R	R	**S**	R	**S**	**S**	**S**	**S**	**S**	R
SET TWO															
D-4	R	R	R	**S**	**S**	**S**	**S**	**S**	**S**	R	**S**	**S**	R	R	R
D-5	R	R	R	**S**	**S**	R	**S**	R	**S**	R	R	R	R	R	R
D-6	R	R	R	R	**S**	R	R	R	R	R	R	R	R	R	R
SET THREE															
D-7	R	R	R	R	R	R	R	R	R	**S**	**S**	R	**S**	R	R
D-8	R	R	R	R	R	R	R	R	R	**S**	R	R	**S**	R	R
D-9	R	R	R	R	R	R	R	R	R	R	R	R	R	R	R
New race	**1**	**3**	**7**	**7**	**7**	**3**	**3**	**7**	**3**	**7**	**7**	**7**	**7**	**7**	**3**
Denomination	**0**	**0**	**0**	**3**	**7**	**1**	**3**	**1**	**3**	**0**	**1**	**1**	**0**	**0**	**0**
	0	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**3**	**1**	**0**	**3**	**0**	**0**

In tests at Clermont-Ferrand, the resistance reaction of *D-4*, *D-5*, *D-6* and *D-8* included Cotyledon Limited Infection (CLI) when tested with some of the races. This shows the need for careful interpretation of tests before deciding the pathotype of a downy mildew isolate.
Races 7 and 9 cannot be distinguished with the differentials defined in “Materials and Methods”. From the late 1980’s until the early 1990’s, the USDA used a proprietary line from Interstate Seed Company: IS-2000 and race 7 was R and race 9 was S. PM-17 was substituted in the same triplet position because IS-2000 is not available to all research programmes. No publicly available line shows different reaction to the two races

Conclusion

To help learning the new names of the known downy mildew races and the equivalences are given in table 4. We hope that these new names will be adopted in all new publications.

Table 4: The new denomination of race of *Plasmopara halstedii.*

International Name	**American Name**	**French Name**	**Old Name**
Race 100	race 1		European race
Race 300	race 2	race D	Red River race
Race 310	race 6		
Race 330	races 7 & 9		
Race 700	race 3	race C	
Race 703	race 10	race B	
Race 710	race 8	race A	
Race 711	race 11		
Race 730	race 4		
Race 770	race 5		

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