Neth. J. Pl. Path. 72 (1966): 12-19

ON THE DANGERS OF ARTIFICIAL INFECTION WITH YELLOW RUST TO THE BARLEY CROP OF THE NETHERLANDS; A QUANTITATIVE APPROACH[1]

Het gevaar van kunstmatige infecties met gele roest voor het gerstgewas in Nederland; een kwantitatieve benadering

J. C. ZADOKS

Department of Phytopathology of the State Agricultural University, Wageningen, The Netherlands

In the Netherlands, some barley breeders perform artificial infections with barley yellow rust in their fields to test their selections for resistance. The question has been raised wether this artificial infection of breeders fields can be justified in view of the danger of causing an epidemic. In 1962, conditions were favourable to arrive at an answer to this question. A rust survey provided the data from which the amount of rust in artificially and in spontaneously infected fields could be computed using a new, quantitative method. Artificial and spontaneous infection yielded approximately equal amounts of rust on the reference date, July 1st, 1962. In years with a severe epidemic, the total amount of spontaneously occurring rust is at least a thousandfold of the amount present in 1962, a year with exceptionally little rust. This leads us to the conclusion, that artificial infection of breeders fields does not materially contribute to spontaneous epidemics of yellow rust of barley in the Netherlands.

INTRODUCTION

In 1961, there was a severe epidemic of yellow rust on barley (YRB) (*Puccinia striiformis* Westend. f.sp. *hordei* Eriks.) in the Netherlands. The common view that YRB was only of minor importance turned into an anxious attitude.

In the spring of 1962, the Board of the Netherlands Grain Centre raised the following points:

1. To screen for resistance, Netherlands barley breeders must test their material in artificially infected fields.
2. The winter 1961–1962 has killed all or a large portion of the YRB inoculum in the Netherlands.
3. The artificial infection increases the amount of YRB inoculum.
4. Is it justified to increase the amount of inoculum by artificial infection when nature has practically eradicated the inoculum caused by spontaneous infection?

The question of point 4 must be answered on the base of objective and preferably quantitative data. These were provided by means of a survey.

EPIDEMIOLOGY OF YRB

During the years 1956–1961, the epidemiology of YRB in the Netherlands was studied simultaneously with that of yellow rust on wheat (ZADOKS, 1961). The life cycles of the two *formae speciales* are identical.

[1] Accepted for publication 25 October, 1965.

YRB passes from the main crop to pre-harvest late tillers and pre-harvest volunteers, hence to post-harvest late tillers and post-harvest volunteers. There is a slight difference of emphasis between YRB and yellow rust of wheat, since the former relies mainly on pre-harvest late tillers and the latter mainly on pre-harvest volunteers. Summer and autumn carry-over take place on late tillers and volunteer plants, not only in the fields, but also on farm yards, around barns and along the roads. Overwintering occurs on autumn-sown winter barley and on volunteers. In the Netherlands, there is no evidence indicating that other cereals and grasses act as accessory hosts, nor is there any evidence for an intermediate host.

In Europe, two races of YRB are known. In the register of yellow rust races, where the races are numbered irrespective of the *formae speciales* to which they may belong, these YRB races bear the numbers 23 and 24 (FUCHS, 1956). During the 1961 epidemic, race 24 was predominant (DANTUMA, 1964). The artificial infections have been done with both races. There is no evidence, that the two YRB races differ much in their epidemiological behaviour. For this reason and because race identification during the survey was impracticable the question of races is not considered in the following account. The conclusions are thought to be valid for the f.sp. *hordei*, as it exists at present in the Netherlands.

International exchange of inoculum is probable, because the YRB races 23 and 24 are common to most West European countries (STUBBS & FUCHS, 1965). Nevertheless, influx of inoculum into the Netherlands must be of an incidental nature, because there is no evidence of air-borne epidemics. Netherlands YRB epidemics are of the endemic type.

Spring barley volunteers die from frosts below –5° C lasting one or a few days and the YRB dies with them. During the cold winter 1961–1962 practically all spring barley inoculum was killed. Crops and volunteer plants of winter barley survive severe frosts. In 1961, the spring barley acreage was 92,000 ha against a winter barley acreage of 7,400 ha. This implies that most of the YRB inoculum of the Netherlands must have died before the spring of 1962.

YRB SURVEY

The question remains to be answered: how much inoculum was left after the winter? A survey was organised about July 1st. In each of the agricultural areas where barley is an important crop, a sampling area was designated (Fig. 1, Table 1). Per sampling area about 20 fields were to be inspected. Six trained observers received a special instruction for this survey.

In each field, a narrow strip was chosen at random and scrutinized for YRB; the inspection was to last ten minutes. The following notes were taken: 1. date of inspection, 2. community and/or locality, 3. variety or habit (winter/spring), 4. growth stage GS according to FEEKES (1941), 5. YRB severity S (= percentage of attack) or at low infection levels the number N of YRB lesions, 6. length and width of inspected strip or, in case of artificially infected fields, total area of field.

July 1st was chosen as a reference date, because it could be expected that on this date spontaneous and artificial infection were well enough established without spreading to such a degree that they interfered with each other and,

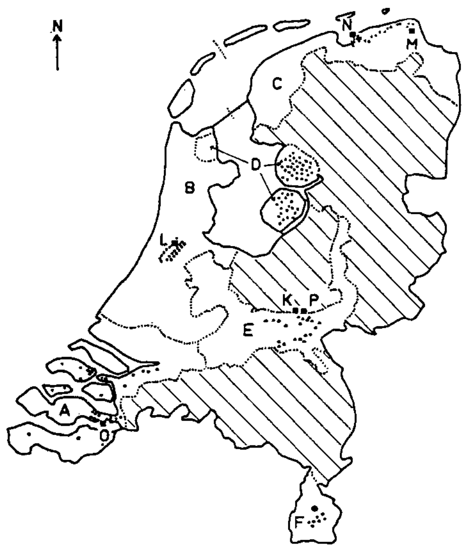


FIG. 1. Map of the Netherlands with agricultural areas, sample areas, sampled fields and artificially infected fields.
Kaart van Nederland met landbouwgebieden, monstergebieden, bemonsterde percelen en kunstmatig geïnfecteerde proefvelden.
A–F Agricultural areas, see Table 1/ *Landbouwgebieden volgens tabel 1*
■, K–P Artificially infected breeders fields/ *Kunstmatig geïnfecteerde kwekersproefvelden*
● Sampled field/*Bemonsterd perceel*
○ Sampled trial field/*Bemonsterd proefveld*
\\\\ Not-inspected agricultural areas/ *Niet-bemonsterde landbouwgebieden*
..... Borderline of inspected agricultural area/*Grens van bemonsterd landbouwgebied*

TABLE 1. Agricultural and sampling areas of YRB survey, 1962.
Landbouwgebieden en monstergebieden van de YRB-enquête, 1962.

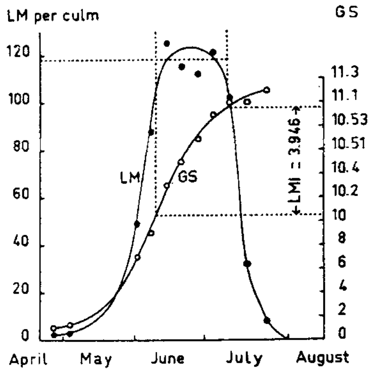
Agricultural area *Landbouwgebied*	Sampling area *Monstergebied*	Number of fields *Aantal percelen*	Date *Datum*
A. South-western sea clay *Zuidwestelijke zeeklei*	(dispersed) *(verspreid)*	30	4—7
B. Mid-western sea clay *Centrale zeeklei*	Haarlemmermeerpolder	13	30—6
		7	7—7
C. Northern sea clay *Noordelijke zeeklei*	Noord-Groningen	19	3—7
D. IJsselmeerpolders	Noordoostpolder	33	3/4—7
	Oostelijk Flevoland	20	3—7
E. River clay *Rivierklei*	Betuwe and adjacent districts/*Betuwe en aangrenzende gebieden*	20	5—7
F. Löss	Zuid-Limburg	23	1—7
	total	165	

consequently, with the interpretation of the observations. The results justified the choice of this reference date. Only one of the inspected fields showed an infection which was attributed to wind-borne inoculum from an artificially infected field at 500 meters distance. This field was discarded from the survey.

TRANSFORMATION OF GROWTH STAGE GS INTO LEAF MASS LM

The LM of a plant, plot or field is the projected living leaf area of the plant(s) in square centimeters. The leaf mass index LMI of a crop is the LM of the crop expressed in square centimeters per hectare (ZADOKS, 1961).

FIG. 2. Reference plot at Wageningen, spring barley cultivar 'Topper', artificially infected with yellow rust. The development of "leaf mass" LM per stem, "leaf mass index" LMI and growth stage GS are plotted against time.
Referentie-proefveld te Wageningen met 'Topper'-zomergerst, kunstmatig geïnfecteerd met gele roest. De ontwikkeling van de bladmassa LM per halm, de bladmassaindex LMI en het groeistadium GS zijn uitgezet tegen de tijd.



In Wageningen, a reference plot of spring barley cv. 'Topper' was grown. At weekly intervals LM per stem and the number of stems per square meter were determined. From these data, LMI's for successive growth stages were determined (see Fig. 2). The calculated LMI's were taken to be representative for barley crops in the inspected areas.

For each agricultural area, the median growth stage on the observation date was determined. Using Fig. 2, the probable growth stage on July 1st was estimated. This item served to determine the LMI for the agricultural area on July 1st, which was entered in Table 2. For each agricultural area, the barley acreage BA, derived from the List of Varieties of Field Crops, 1963, is entered in Table 2. The reduced leaf mass per agricultural area is computed as: $LM_{red} = LMI \times BA$.

TABLE 2. Rust mass RM of naturally infected fields on July 1st, 1962.
Roestmassa RM van de spontaan geïnfecteerde percelen op 1 juli, 1962.

Agricultural area/*Landbouwgebied*	GS_{red}	LMI $\times 10^{-8}$	BA in ha	x_{red} $\times 10^{8}$	RM_{red} $\times 10^{4}$
A	10.53	3.946	38,500	0.61	9.27
B	10.4	3.946	7,000	0.0	0.00
C	10.4	3.946	10,100	16	63.80
D	10.4	3.946	3,600	3,700	5,254.00
[1]	10.5	3.946	3,600	19	24.98
E	10.4	3.946	7,300	1.4	4.03
F	10.1	3.946	3,000	11	13.02
Netherlands, rest/*Nederland, rest*			27,100	0.0	0.00
Netherlands, total/*Nederland, totaal*			100,200	-	5,369.10

BA = Barley Area/*Gerstareaal*
GS = Growth Stage/*Groeistadium*
LMI = Leaf Mass Index/*Bladmassaindex*
red = Data reduced to July 1st/*Gegevens herleid tot 1 juli*
RM = Rust Mass/*Roestmassa*
x = Rusted fraction of leaf area/*Met roest bezet deel van het bladoppervlak*

[1] Fields of Zuyderzee Development Authority in Oostelijk Flevoland not included in the statistics of the List of Varieties of Field Crops./*De percelen van de Rijksdienst voor de IJsselmeerpolders in Oostelijk Flevoland zijn niet begrepen in de rassenstatistiek van de Rassenlijst.*

In the case of artificially infected fields, the same procedure was followed for each field individually. LM is entered in Table 3. Only those fields where artificial infection with YRB was successful were included in the table.

TRANSFORMATION OF RUST RECORDS INTO RUST MASS RM

The RM of a plant, plot or field is the projected living and visibly rusted leaf area of the plant(s) in square centimeters. This value is found by multiplying LM with the rust fraction x, the rust fraction x being that fraction of the living leaf area which is visibly rusted. The point is to find the fractions x.

For each field, GS of the crop and length and width of the inspected crop are known. Using Fig. 2 for the GS to LMI conversion, the inspected LM was calculated. When the rust severity S was specified, the inspected rust mass was computed as RM $= S \times LM / 100$. When the number of rust lesions N was specified, the inspected rust mass was found as RM = N, taking the average area of a YRB lesion to be 1 cm². For each agricultural area RM and LM of individual fields were summarised into ΣRM and ΣLM. The fraction x per agricultural area at the observation date is: $x_{obs} = \Sigma RM / \Sigma LM$.

The 'Topper' reference plot in Wageningen was artificially infected with YRB. The rust fraction x was determined at weekly intervals. In Fig. 3, logit $x = \ln \frac{x}{1-x}$ was plotted against time. The course of the epidemic is adequately represented by the logit line, the slope of which is a measure for the speed of the epidemic (VAN DER PLANK, 1963; ZADOKS, 1961).

It is assumed, that the epidemic of the reference plot is representative for the epidemics in the inspected areas. This assumption permits us to reduce the fractions x_{obs} per agricultural area at their various observation dates to fractions x_{red} per agricultural area on July 1st, using the formula of the logit line from the reference plot: a = 0.268b – 10.172 (see Fig. 3). RM per agricultural area reduced to July 1st is calculated as: $RM_{red} = x_{red} \times LM_{red}$. Results are entered in Table 2.

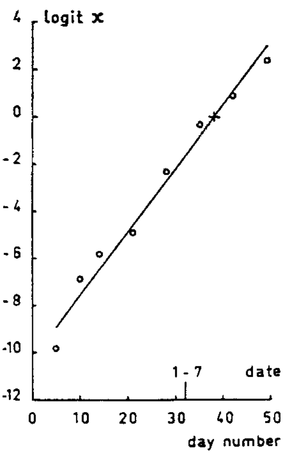


FIG. 3. Reference plot at Wageningen, spring barley cultivar 'Topper', artificially infected with yellow rust. The development of the rust epidemic is well represented by the logit line.
Referentie-proefveld te Wageningen met 'Topper'-zomergerst, kunstmatig geïnfecteerd met gele roest. De ontwikkeling van de roestepidemie wordt goed weergegeven door de logit-lijn.

logit x = a
day number = b/*dag nummer* = b
formula of the logit line: a = 0.268b – 10.172
formule van de logit-lijn: a = 0,268b – 10,172

TABLE 3. Rust mass RM of artificially infected fields on July 1st, 1962.
Roestmassa RM van de kunstmatig geïnfecteerde proefvelden op 1 juli, 1962.

Field *Proefveld*	Observation date/*Waarnemingsdag*	GS_{red}	$LMI_{red} \times 10^{-8}$	Area ha	x_{red}	$RM_{red} \times 10^{4}$
K	28–6	10.5	3.946	0.0862	0.358	1,215
L	4–7	10.4	3.946	0.4	0.101	1,592
M	3–7	10.3	3.946	0.15	0.467	2,760
N	3–7	10.3	3.946	0.1	0.0055	22
O	4–7	10.52	3.946	1.0	0.0004	4
P	16–7	10.53	3.946	0.3	0.0045	53
Totals/*Totaal*				2.0362		5,646

GS = Growth Stage/*Groeistadium*
LMI = Leaf Mass Index/*Bladmassaindex*
red = Data reduced to July 1st/*Gegevens herleid tot 1 juli*
RM = Rust Mass/*Roestmassa*
x = Rusted fraction of leaf area/*Met roest bezet deel van het bladoppervlak*

RM of artificially infected fields was calculated in the same way as for naturally infected fields. The infection of breeders fields is usually quite irregular. The rust severity data were the estimates of the average rust severity over a whole field. Reduction from observation date to July 1st was done as described above. Results are given in Table 3.

RESULTS

The results published here differ somewhat from those given in a preliminary report (ZADOKS, 1963) mainly because of the reduction of data from artificially infected fields to July 1st. The changes in the figures do not influence the reasoning or the conclusion. In Table 2, it is assumed that the non-inspected areas, hatched in Fig. 1, have little or no YRB even during severe epidemics in the rest of the country. Moreover, incidental inspections in the hatched areas did not yield YRB in 1962. According to Table 2, the total rust mass in the Netherlands due to spontaneous infection on July 1st was approximately:

$$RM = 5.4 \times 10^{7}.$$

Table 3 shows, that the total rust mass of the Netherlands due to artificial infection on July 1st was approximately:

$$RM = 5.6 \times 10^{7}.$$

In 1962, a year with an exceptionally low level of spontaneous infection, the proportion of artificial to spontaneous inoculum was approximately 1:1. Between July 1st and harvest time, YRB did not increase much. In 1961, a year with an exceptionally high level of spontaneous infection leading to a severe epidemic, the above proportion was at least 1:10,000. Since the amount of artificially introduced inoculum is nearly constant through the years, this amount is quite small when compared with to the inoculum necessary for a large scale epidemic. Consequently, the amount of artificially introduced inoculum does not contribute materially to the development of a general and severe epidemic.

DISCUSSION

The problem has two aspects: first, the accuracy of the data and second, the subjective decision of what has to be done next year.

The greatest source of variability is the sampling method. The number of samples (165) and the number of sampled areas (7) were small. Another source of variability is the choice of the inspected strip, especially when the fraction x is below 10^{-5}. One observer may find a small rust focus in his inspection strip whereas the other observer, choosing his inspection strip a few meters further away, just misses the focus. This, indeed, happened during an inspection trip made by two observers for control purposes. Underestimation is a systematic error at low infection levels. A third source of variability is the difference between observers. Control experiments showed that agreement between observers was adequate for the present purpose.

Errors of another nature are inherent in the assumptions underlying the reduction of data at observation date to reduced data at July 1st. Fortunately, the reduction usually covers only a few days, a time span too short to introduce great errors.

The overall accuracy of the method is such that the order of magnitude of the measured phenomenon is correct and that the end figure itself may be correct within limits of approximately $\pm$ 100%. In the case of the spontaneous infection there remains an error of underestimation which cannot be expressed numerically.

The subjective decision can now be discussed more clearly. By comparing the figure for artificial infection, which is fairly constant over the years, with the figure for spontaneous infection, which varies at least a 10,000-fold over the years, the conclusion is reached that the artificial infection does not materially influence the course of natural epidemics.

This is true when the Netherlands, covered by a large scale epidemic, are regarded as a whole. On a smaller scale, however, artificially infected fields may well endanger their environment. Nearly every year rust has spread from the artificial foci to neighbouring fields, but no claims for damage were ever made or complaints received.

The possibility that artificial field infection re-introduces races which have been eliminated by natural processes must not be overlooked. Experience teaches that races exist in nature as long as their compatible cultivars are grown. When these "carrier" cultivars are no longer grown, the race disappears. Re-introduction of the race is of little consequence when a compatible cultivar is no longer present. Therefore, artificial field infection with YRB is not dangerous to barley crops with regard to an eventual re-introduction of old rust races.

After considering the risks of artificial infection in the field the advantages must be set forth. It is imperative that breeders test their material against YRB to select resistant lines. Up to the present, greenhouse testing of seedlings is not yet possible on a large scale, nor is it fully adequate. There is no other alternative than field testing. Since heavy spontaneous infections are rare in the Netherlands, waiting for them is a waste of time. Artificial infection must be accepted as a reliable substitute.

In the writers opinion, the computed results in combination with considerations of a more general nature warrant the subjective conclusion that artificial infection of breeders fields with yellow rust of barley should be continued.

SAMENVATTING

In 1961 bleek, dat gele roest van gerst (YRB) gevaarlijk kon zijn voor de praktijk. Na de koude winter 1961–1962 moest men aannemen, dat het merendeel van het YRB-inoculum was doodgevroren. Het Nederlands Graan-Centrum stelde toen de vraag, of het kunstmatig infecteren van kwekersproefvelden met YRB onder deze omstandigheden nog wel verantwoord kon worden. Deze vraag werd beantwoord met behulp van een enquête, die op een nieuwe manier bewerkt werd, zodat kwantitatieve resultaten beschikbaar kwamen (Tabel 1, Fig. 1). Berekend werd de roestmassa (RM), dit is het aantal cm^2 levend roestig blad per object (Fig. 2, Fig. 3). De resultaten, samengevat voor heel Nederland, waren per 1 juli 1962: spontane infectie in praktijk (Tabel 2) $RM = 5{,}4 \times 10^7$; kunstmatige infectie in kwekersproefvelden (Tabel 3) $RM = 5{,}6 \times 10^7$.

In het jaar 1962 met een zeer lichte epidemie was dus de verhouding kunstmatig: spontaan ongeveer 1:1. In het jaar 1961 met een zeer ernstige epidemie was deze verhouding naar schatting ongeveer 1:10.000. De hoeveelheid kunstmatige infectie in deze beide jaren was ten naaste bij gelijk. De kunstmatige infectie is dus vrijwel niet van belang bij het ontstaan van epidemieën.

Op grond van de berekende gegevens en van een aantal nadere overwegingen (zie ook ZADOKS, 1961) kan naar de mening van de schrijver de – uiteraard subjectieve – beslissing genomen worden dat de kunstmatige infecties van kwekersproefvelden met gele roest van gerst ook in de toekomst verantwoord zijn.

ACKNOWLEDGEMENTS

Thanks are due to the Netherlands Grain Centre, which raised the problem and supported the survey and to the surveyors Dr. S. BROEKHUIZEN, Mr. A. VAN ESSEN, Mr. W. HOOGKAMER, Mr. H. VECHT and Ir. W. WILTEN. Mr. T. LIMONARD provided the data from the reference plot.

REFERENCES

DANTUMA, G., – 1964. Sources of resistance to mildew and stripe rust in breeding spring barley. Euphytica 13: 245–249.

FEEKES, W., – 1941. De tarwe en haar milieu. Versl. tech. Tarwe Comm. 12:523–888.

FUCHS, E., – 1956. Der Stand der Rassenspezialisierung beim Gelbrost Puccinia glumarum (Schm.) Erikss. et Henn. in Europa. NachrBl. dt. PflSchutzdienst, Stuttg. 8:87–93.

PLANK, J. E., VAN DER, – 1963. Plant diseases: Epidemics and control. Academic Press, New York.

STUBBS, R. W. & E. FUCHS, – 1965. Report on the "Yellow Rust Trials Project" in 1963. Tech. Ber. Sticht. Ned. Graan-Cent. 14.

ZADOKS, J. C., – 1961. Yellow rust on wheat. Studies in epidemiology and physiologic specialization. Tijdschr. PlZiekt. 67:69–256.

ZADOKS, J. C., – 1963. Enquête naar het voorkomen van gele roest op gerst in 1962. Ned. Graan-Cent., stencilled report, unpublished.