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SPATIAL DEPENDENCY AND BLOCK DESIGNS

*Document prepared by experts from the United Kingdom*

## SPATIAL DEPENDENCE AND BLOCK DESIGNS IN DUS HERBAGE TRIALS

### SUMMARY

Data from nine herbage DUS trials were investigated for evidence of spatial dependence. It was most evident in variates measuring the overall dimensions of the plants, especially late season variates. On average, 45% of the residual variation in the plot means of the spatially dependent variates was estimated to be of a spatial nature. Consequently, it is likely that the efficiency of analysis of these variates would be improved by using spatial analysis instead of non-spatial analysis or by using alpha (incomplete block) designs instead of complete block designs. The spatial information was used to determine the optimal incomplete block sizes and the likely increase in efficiency due to using alpha designs instead of complete block designs.

### INTRODUCTION

Observations made on field trials are prone to spatial dependence between plots caused by spatial variation in fertility, moisture, intercepted light, and other environmental factors (Grondona *et al.* 1996). Conditions in plots spatially close together are more likely to be similar than in plots further apart, so, providing the plants are not in competition, observations made on plants grown on spatially close plots are more likely to be similar in those characteristics that are affected by the environmental conditions than are observations made on plants grown on more distant plots (van Es & van Es 1993).

The main objective of DUS trials is to obtain precise estimates of the differences in character means of pairs of varieties. To estimate the means efficiently, i.e. with small variances, it is necessary to minimise the variation that is residual, i.e. not due to variety effects. When the residual variation is caused by spatial dependence, the experimenter can reduce it 'a posteriori' by using a spatial analysis (see Watson 2000 for further detail). He can also attempt to reduce it 'a priori' by the choice of design, which includes the block design and the block and plot sizes, configurations and orientations (Cressie 1991; Grondona *et al.* 1996; Kristensen & Ersbøll 1992; van Es & van Es 1993). When the data are spatially dependent, the choice of design also affects the efficiency of estimation of variety effects due to the positions of the varieties within the trial (Martin 1986; van Es & van Es 1993).

The conventional analysis of variance of a classical randomized design is non-spatial and is based on ordinary least squares (OLS). In the presence of spatial dependence, OLS estimators of variety effects are unbiased (Bailey 1981). However, they are not efficient in that they do not have minimal error variances. Further, when the data are spatially dependent, the OLS estimates of the variances of the variety estimates are inappropriate because they use an average estimate of error based on a single estimate of the residual variation. This ignores the fact that some variety comparisons involve plots that are a short distance apart and so have a smaller variance than those with plots that are a greater distance apart (van Es & van Es 1993). In consequence, if variety differences are small and OLS is used, strong spatial dependence may reduce the power of an analysis to correctly estimate and identify variety differences.

Clearly, if spatial dependence is likely, the ideal would be to use a spatial design and a spatial method of analysis. However this has cost implications in terms of more complex analysis and interpretation, and the benefits depend on the strength and range of the spatial dependence. The purpose of this paper is to study the prevalence and extent of spatial dependency in data arising from spaced plant herbage DUS trials, and to assess its implication on the design and analysis of DUS trials, which currently ignore any spatial dependence between observations.

## METHODS

### The Field Trial Data

The data came from DUS spaced plant herbage variety trials conducted at Crossnacreevy, Co. Down. There were nine trials comprising three series of ryegrass trials: tetraploid perennial (PRT), tetraploid italian (IRT) and diploid italian (IRD), in each of 1989, 1990 and 1991. The trials are described in Watson (2000). The plants in a trial formed an incomplete grid owing to the presence of guard plants and walkways and the withdrawal of varieties during the trial. Eleven characters or variates were observed on each plant (Table 1).

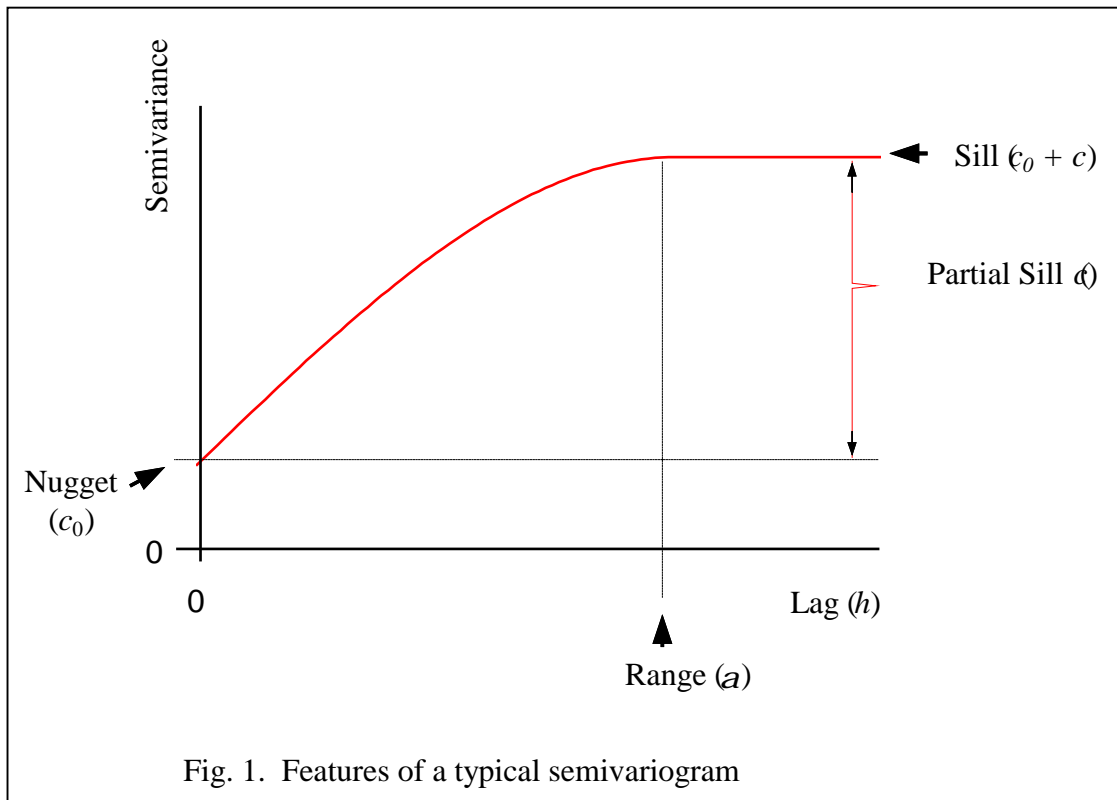
### The Random Field Model

The spatial dependence of the data was investigated by fitting a random field model:

$$Y_i = \mu_i + \varepsilon_i \quad i=1, \dots, n$$

where  $Y_i$  is the random variable observed on plant  $i$ , and  $\mu_i$ , the mean structure for plant  $i$ , is a composite term comprising an overall mean, variety effects and random effects due to location or block effects. The terms  $\varepsilon_i$  represent the spatially dependent random error variables or error structure. They are assumed to have zero expectation and be such that the variance of the differences between pairs of error variables, say  $\varepsilon_i$  and  $\varepsilon_j$ , where plants  $i$  and  $j$  are located distance  $h$  apart, has intrinsic stationarity so that  $V(\varepsilon_i - \varepsilon_j) = 2\gamma(h)$ . This implies that the variance depends only on the distance or lag,  $h$ , between the locations and not on the locations themselves. The function  $\gamma(h)$  relating the variance to  $h$  is known as the semivariogram and the estimate of it for a given value of  $h$  as a semivariance.

The variability of comparisons between observations tends to increase as the separation between plants increases up to, typically, a finite lag  $a$  known as the range of influence of the semivariogram. This marks the limit beyond which observations are spatially independent (Fig. 1). For lags greater than the range, the variance between observations remains constant and is called the sill ( $c_0 + c$ ). As the lag decreases towards 0, the semivariance approaches the nugget effect ( $c_0$ ), which is a measure of the inherent or non-spatial variation (including measurement errors). The partial sill ( $c$ ) (Cressie 1991) is a measure of the strength of the spatial dependence.



### Estimating Semivariances

The data were adjusted for the mean structure using analysis of variance, leaving the residuals. These were placed in an array representing the layout of the plants in the field, from which a set of semivariance estimates at different lags were obtained both from the columns and from the rows. For example, the column semivariance at each lag  $h$  was calculated as the mean over columns of half the squared difference (i.e. the semivariance) of all pairs of non-missing residuals within the same column located  $h$  rows apart. This was done for each of the 11 variates in each of the nine trials.

The resulting semivariance estimates were biased because they were calculated using treatment adjusted residuals (Cressie 1991). The bias could not be adjusted for because it depends on the lag and is different for the row and column semivariances (Watson 1995).

### Fitting Semivariograms

The spherical function model was chosen to model the underlying semivariograms and was fitted to the semivariances by weighted least squares. Details are given in the appendix. The curve was fitted separately to the row and column semivariances, because of their different biases, and only to semivariances at lags of up to half the maximum lag.

### Assessing the spatial dependence

Graphical plots of the row and column semivariances against lag showed that some variates had little or no spatial dependence, whereas others had semivariances that first increased and then levelled off in a way characteristic of spatially dependent variates.

A variate susceptible to spatial dependency would be expected to show evidence of spatial dependency in all years of a particular trial type, although not necessarily in all trial types as different genotypes may not be equally susceptible to spatial dependence. Consequently, only those variate-by-trial type combinations that had at least one of their row or column sets of semivariances satisfactorily fitted by a spherical function curve in all three years were considered as potentially spatially dependent (Table 2). The goodness of fit of the fitted curves was measured by the weighted coefficient of determination,  $R_w^2$  (McBratney & Webster 1986). Fig. 2 shows the 1989 row and column semivariances of the variates considered spatially dependent for the PRT trial type.

## THE SPATIAL ANALYSIS

Six of the 11 variates were considered spatially dependent in one or more trial types. All measure the early or late season overall dimensions of the plants. That the curves in the late season  $PLSE_{e+30}$ ,  $PLS_{e+30}$ ,  $PLSN_{e+30}$  variates tended to fit better might indicate that any spatial dependence is shown more clearly in late season variates than in earlier ones. This is possibly due to the mature plants reflecting more clearly the effects of the spatial dependency factors. The 'date of ear emergence' variate, which is especially important in making DUS decisions about varieties, was not considered spatially dependent. Just one variate was spatially dependent in the IRD trials compared to all six in the PRT trials and five in the IRT trials. This might be due to ploidy (see Watson 2000 for details). The layout of a trial's pairs of blocks (parallel or in series) did not seem to affect the chance of a curve being satisfactorily fitted.

The results for the  $PH_{sp}$  and  $W_{sp}$  variates require caution, as the curves in the former were poorly fitted in the IRT trials and the ranges in the latter are very large suggesting the possible presence of an underlying trend in the data. Excluding these variates, on average the nugget was 85% of the sill and the range was 12 lags (9m).

## DESIGN CONSIDERATIONS

The fitted semivariograms were then used to predict the efficiency that would have been achieved had various alpha (incomplete block) designs (Patterson & Williams, 1976) been used instead of a complete block design. Here efficiency is taken to be the ratio of the average variance of variety differences from the complete block analysis to the average variance from the appropriate incomplete block analysis with recovery of inter-block information. The semivariogram derived from the individual plant observations,  $\gamma(h)$ , was used to calculate  $\gamma_p(h)$ , the semivariance of the plot means of observations on two plots of  $p$  plants arranged in parallel and located  $h$  lags apart (see appendix). As the plots of the spaced plant herbage trials were laid out in a one-dimensional arrangement within the blocks, the  $\gamma_p(h)$  could be used to postblock the trials (Patterson & Hunter 1983). This allowed prediction of the efficiency that would have been likely had each of the blocks in the original complete block design been divided into incomplete blocks and an alpha design used. Details are given in the appendix.

The efficiencies of alpha designs with blocks of 2, ..., 12, 14, 16 and 20 plots in a trial with 96 varieties, 6 replicates and 10 plants per plot were predicted from the average fitted semivariogram of each variate of each trial type. The efficiencies of designs with just one plant per plot were also predicted. The optimum block sizes and corresponding maximum efficiencies are given in Table 2. The percentage of the variation that is likely to be spatial

was calculated for the plot means of 10 plants as the percentage of partial sill to sill using the semivariances  $\gamma_{10}(h)$  and using the semivariances  $\gamma(h)$  for the single plant means. These are shown in Table 2.

The more plants plot means are based on, the less the variation among the plot means, however, the greater the proportion of the total variation among the plot means that is spatial. This causes the optimal block sizes of the alpha design trials with 10 plants per plot to contain fewer plots than the optimal block sizes of the trials with single plant plots (Table 2). See Watson (2000) for details.

## DISCUSSION

On average only 15% of the error variation of the spatially dependent variates was spatial on a single plant basis with a range of 9m or 2.2 times the average of the plot dimensions. However, this study shows that the plot means of 10 plants are likely to be more strongly spatially dependent with on average 46% of the predicted variation of a spatial nature and the same range. Grondona & Cressie (1991) and Gilmour *et al.* (1997) found spatial analysis increased efficiency by 32% and 600% (in a sophisticated analysis) respectively when 40% (inferred from graphs) and 65% of the unexplained variation was spatial with a range 2.4 times and 1.5 times the average of the plot dimensions. Although the extent to which a spatial analysis of the spatially dependent variates would identify distinctnesses more efficiently than the current non-spatial analysis cannot be directly determined from this study, it is unlikely to be negligible.

Needing a reliable semivariogram estimate for a full spatial analysis of variety effects is a potential drawback as, even ignoring the  $W_{sp}$  and  $PH_{sp}$  variates, there was considerable variation in the shape of the fitted semivariogram between trials for a variate. Part of the variation was due to differences in the magnitude of the observations (Watson 2000). Whether the remainder was sampling variation or due to different underlying semivariograms at the different but not distant trial sites can't be told from this study.

A second major drawback is the relative complexity of spatial analysis and the interpretation of the results as the standard errors differ for every pair of varieties compared. However, if a non-spatial analysis is used in the presence of spatial dependence, the different standard errors will still exist even though they are ignored. The greater the spatial dependency, the more potentially misleading the standard errors for decision making concerning variety distinctnesses.

One option is to improve the trial design. It is not practicable to space the plots to ensure spatial independence and the large numbers of varieties involved require prohibitive replication with either the nearest neighbour balanced designs suggested by Martin (1986) or the balanced incomplete block designs suggested by van Es & van Es (1993). However, van Es & van Es (1993) also suggest the use of incomplete block designs such as alpha designs chosen to equalize, as far as possible, the average distance of each variety comparison. This could both improve variety effect estimation and reduce the residual variation.

Given a reliable semivariogram, it may, as seen in this study, be used to compare the efficiencies of different incomplete block designs. Of all the possible alpha designs for a trial with 96 varieties in 6 replicates and 10 plants per plot, those with incomplete blocks of between 4 and 9 plots gave the maximum predicted efficiencies for the spatially dependent variates of the spaced plant herbage trials. These block sizes are similar to the optimal block

size of 8 plots for the same size trial that Ainsley *et al.* (1987) report based on the results of 29 ryegrass spaced plant variety trials grown between 1974 and 1983 at Crossnacreevy. The predicted increases in efficiency of between 9 and 46% in this study are comparable with the 16% reported by Ainsley *et al.* (1987) for the variate  $H_e$ , although  $H_e$  was not classed as spatially dependent in this study.

A reliable semivariogram may also be used to compare the effect of the variety positions on the efficiency of variety effect estimation of a planned design (Cressie 1991) and to assess the effects of altering the plot dimensions and block configurations on the residual variances (Kristensen & Ersbøll 1992). These latter changes would only be worthwhile if they improved the efficiency of estimation of the variety effects of the spatially dependent variates sufficiently to maintain the effectiveness of the DUS decision making process while allowing a reduction in trial costs. By contrast, the use of alpha designs would entail no extra costs and would allow the spatially dependent variates to be analysed more efficiently, possibly by spatial methods. However, it remains to be seen whether the increase in efficiency would be sufficient to make the spatially dependent variates as useful for DUS decision making as the non-spatially dependent ones.

## APPENDIX

### i) The spherical function

The spherical function is given by

$$\gamma(h; \theta) = \begin{cases} 0 & h = 0 \\ c_0 + c \left| \frac{3h}{2a} - \frac{h^3}{2a^3} \right| & 0 < h \leq a \\ c_0 + c & a < h \end{cases}$$

where  $\theta$  is the vector of parameters  $(a, c_0, c)$ . It was fitted to the semivariances by weighted least squares using  $N(h_j) / [\gamma(h_j; \theta)]^2$  as the weight for semivariance  $S(h_j)$ , where  $N(h_j)$  is the number of pairs of residuals from which  $S(h_j)$  is calculated and  $\gamma(h_j; \theta)$  is the value of the spherical function at lag  $h_j, j=1, \dots, k$ .

### ii) Obtaining $\gamma_p(h)$ from $\gamma(h)$

Let  $\gamma_p(h)$  denote the semivariance of the plot-means of observations on two plots, say plot 1 and plot 2, arranged in parallel  $h$  lags apart and each containing a row of  $p$  plants spaced 1 lag apart. Let  $\gamma(h)$  denote the semivariance of observations made on two plants lag  $h$  apart. Then

$$\gamma_p(h) = \frac{1}{p^2} \left[ p\gamma(h) + 2 \sum_{r=1}^{p-1} (p-r)\gamma(\sqrt{r^2 + h^2}) - 2 \sum_{r=1}^{p-1} (p-r)\gamma(r) \right].$$

PROOF:  $\gamma_p(h)$  may be written as  $\frac{1}{2} V(\bar{x}_1 - \bar{x}_2)$  where  $\bar{x}_1 = \frac{1}{p} \sum_{i=1}^p x_{1i}$  and  $x_{1i}, i=1 \dots p$  are the observations on the  $p$  plants in plot 1. Likewise,  $\bar{x}_2$  is the mean of the observations on the  $p$  plants in plot 2.

$$\begin{aligned}
 \gamma_p(h) &= \frac{1}{2} \mathbf{V}(\bar{x}_1 - \bar{x}_2) = \frac{1}{2} \mathbf{V} \left( \frac{1}{p} \sum_{i=1}^p x_{1i} - \frac{1}{p} \sum_{j=1}^p x_{2j} \right) = \frac{1}{2p^2} \mathbf{V} \left( \sum_{i=1}^p (x_{1i} - x_{2i}) \right) \\
 &= \frac{1}{2p^2} \left[ \sum_{i=1}^p \mathbf{V}(x_{1i} - x_{2i}) + \sum_{i=1}^{p-1} \sum_{j=i+1}^p 2\text{Cov}((x_{1i} - x_{2i}), (x_{1j} - x_{2j})) \right] \\
 &= \frac{1}{2p^2} \left[ \sum_{i=1}^p \mathbf{V}(x_{1i} - x_{2i}) + \sum_{i=1}^{p-1} \sum_{j=i+1}^p \left( 2\text{Cov}(x_{1i}, x_{1j}) + 2\text{Cov}(x_{2i}, x_{2j}) \right. \right. \\
 &\quad \left. \left. - 2\text{Cov}(x_{1i}, x_{2j}) - 2\text{Cov}(x_{2i}, x_{1j}) \right) \right] \\
 &= \frac{1}{2p^2} \left[ \sum_{i=1}^p \mathbf{V}(x_{1i} - x_{2i}) + \sum_{i=1}^{p-1} \sum_{j=i+1}^p \left( \mathbf{V}(x_{1i}) + \mathbf{V}(x_{2j}) - 2\text{Cov}(x_{1i}, x_{2j}) \right. \right. \\
 &\quad \left. \left. + \mathbf{V}(x_{2i}) + \mathbf{V}(x_{1j}) - 2\text{Cov}(x_{2i}, x_{1j}) \right. \right. \\
 &\quad \left. \left. + 2\text{Cov}(x_{1i}, x_{1j}) - \mathbf{V}(x_{1i}) - \mathbf{V}(x_{1j}) \right. \right. \\
 &\quad \left. \left. + 2\text{Cov}(x_{2i}, x_{2j}) - \mathbf{V}(x_{2i}) - \mathbf{V}(x_{2j}) \right) \right] \\
 &= \frac{1}{2p^2} \left[ \sum_{i=1}^p \mathbf{V}(x_{1i} - x_{2i}) + \sum_{i=1}^{p-1} \sum_{j=i+1}^p \left( \mathbf{V}(x_{1i} - x_{2j}) \right. \right. \\
 &\quad \left. \left. + \mathbf{V}(x_{2i} - x_{1j}) \right. \right. \\
 &\quad \left. \left. - \mathbf{V}(x_{1i} - x_{1j}) \right. \right. \\
 &\quad \left. \left. - \mathbf{V}(x_{2i} - x_{2j}) \right) \right] \\
 &= \frac{1}{p^2} \left[ \sum_{i=1}^p \gamma(h) + \sum_{i=1}^{p-1} \sum_{j=i+1}^p \left( 2\gamma(\sqrt{(j-i)^2 + h^2}) - 2\gamma(\sqrt{(j-i)^2}) \right) \right] \\
 &= \frac{1}{p^2} \left[ p\gamma(h) + 2 \sum_{r=1}^{p-1} (p-r)\gamma(\sqrt{r^2 + h^2}) - 2 \sum_{r=1}^{p-1} (p-r)\gamma(r) \right]
 \end{aligned}$$

since the observations  $x_{1i}$  and  $x_{2i}$  are made on plants  $h$  lags apart, observations  $x_{1i}$  and  $x_{1j}$  are made on plants  $\sqrt{(j-i)^2}$  lags apart, and observations  $x_{1i}$  and  $x_{2j}$  are made on plants  $\sqrt{(j-i)^2 + h^2}$  lags apart.

### iii) Postblocking and the efficiency of an incomplete block design

The average within-block error mean square,  $MS(k)$ , that would have been obtained had each of the blocks of  $v$  plots of  $p$  plants in the original design been divided into  $s$  incomplete blocks of  $k$  plots of  $p$  plants was obtained by postblocking using:

$$MS(k) = \frac{2 \sum_{h=1}^{k-1} (k-h)\gamma_p(h)}{k(k-1)}$$

The efficiency of using a particular incomplete block design is the ratio of the average variance of variety differences from the complete block analysis to the average variance from the appropriate incomplete block analysis with recovery of inter-block information. Patterson & Hunter (1983) give the efficiency as approximately



$$\lambda \left[ E + \frac{(1-E)(s-1)}{\lambda(v-1) - (v-s)} \right]$$

where  $E$  is the efficiency factor of the design (Patterson *et al.* 1978) which adjusts for the increased variances of the variety differences due to the incomplete blocks, and  $\lambda$  is the ratio  $MS(v)/MS(k)$ .

In predicting the efficiency of an incomplete block design with unequal block sizes, an average value of  $\lambda$  was used.

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Table 1. *The variates observed on the spaced plant variety trials*

	Description and name of variate	When measured
H <sub>sp</sub>	Natural average spring height of plant (cm)*	March
W <sub>sp</sub>	Natural average spring width of plant, calculated as the mean of two widths taken at right angles from above (cm)	March
PH <sub>sp</sub>	Pulled average spring height of plant (cm) <sup>+</sup>	March
H <sub>e</sub>	Natural height of plant at ear emergence (cm)	May/June
W <sub>e</sub>	Width of plant at ear emergence, calculated in same way as W <sub>sp</sub> variate (cm)	May/June
D <sub>e</sub>	Date of ear emergence (Days after 1st March)	May/June
L <sub>f</sub>	Length of flag leaf of longest stem (cm)	June/July
W <sub>f</sub>	Width of flag leaf of longest stem (mm)	June/July
PLSE <sub>e+30</sub>	Pulled length of the longest stem (including the ear) 30 days after the mean date of ear emergence of the within block row (or plot) (cm)	June/July
PLS <sub>e+30</sub>	Same as PLSE <sub>e+30</sub> but excluding the ear (cm)	June/July
PLSN <sub>e+30</sub>	Same as PLSE <sub>e+30</sub> but up to the first node (cm)	June/July

\* Where a variate is described as natural, measurements are taken from the plant in its undisturbed state.

+ Where a variate is described as pulled, the plant's leaves or stems are pulled straight before measurement.

Table 2. Mean parameter estimates,  $R_w^2$  values and the number out of six (2 directions  $\times$  3 years) curves fitted satisfactorily. Predicted optimal block sizes and efficiencies for 96 varieties in six replicates, calculated using the spatial information (see text), are shown for  $p = 10$  and  $p = 1$  plant plots together with estimates of the spatial variation of the means for the two plot sizes

Variate / Trial Type	No / satisf' fitted	mean nugget $c_0$	mean partial sill $c$	mean range $a$	mean sill $c_0 + c$	mean $R_w^2$	Pred' block size p=10	Pred' efficiency p=10	Pred' block size p=1	Pred' efficiency p=1	% * spatial variation p=10	% * spatial variation p=1
PLSE <sub>e+30</sub>												
PRT	6	70.3	14.3	11.5	84.5	51.4	5	146.1	8	103.7	55.1	16.9
IRT	5	120.2	18.5	5.5	138.7	46.7	5	109.3	6	100.7	35.5	13.3
PLS <sub>e+30</sub>												
PRT	6	44.9	8.6	11.4	53.5	46.9	5	142.9	8	103.4	53.4	16.1
IRT	4	89.2	16.3	6.5	105.5	52.3	5	117.0	6	101.5	42.7	15.5
PLSN <sub>e+30</sub>												
PRT	5	33.6	4.2	26.1	37.8	53.7	9	145.8	14	102.7	50.4	11.1
IRT	5	70.2	19.9	5.3	90.1	45.6	4	117.9	5	102.3	49.6	22.1
H <sub>sp</sub>												
PRT	5	36.0	3.2	20.9	39.2	42.5	9	129.1	12	101.4	40.6	8.2
W <sub>sp</sub>												
PRT	4	80.4	34.5	277.9	114.9	42.0	14	146.3	>20	102.8	80.8	30.0
IRD	4	66.1	21.8	118.6	87.9	39.4	10	179.5	>20	105.8	76.0	24.8
IRT	5	85.4	6.6	9.9	92.0	38.5	7	112.0	8	100.5	29.8	7.2
PH <sub>sp</sub>												
PRT	5	41.8	6.7	17.0	48.4	45.7	7	149.2	10	103.3	53.5	13.8
IRT	4	117.0	2.7	18.9	119.6	14.0	11	104.7	16	100.0	14.7	2.3

\* Percentage of mean partial sill to sill

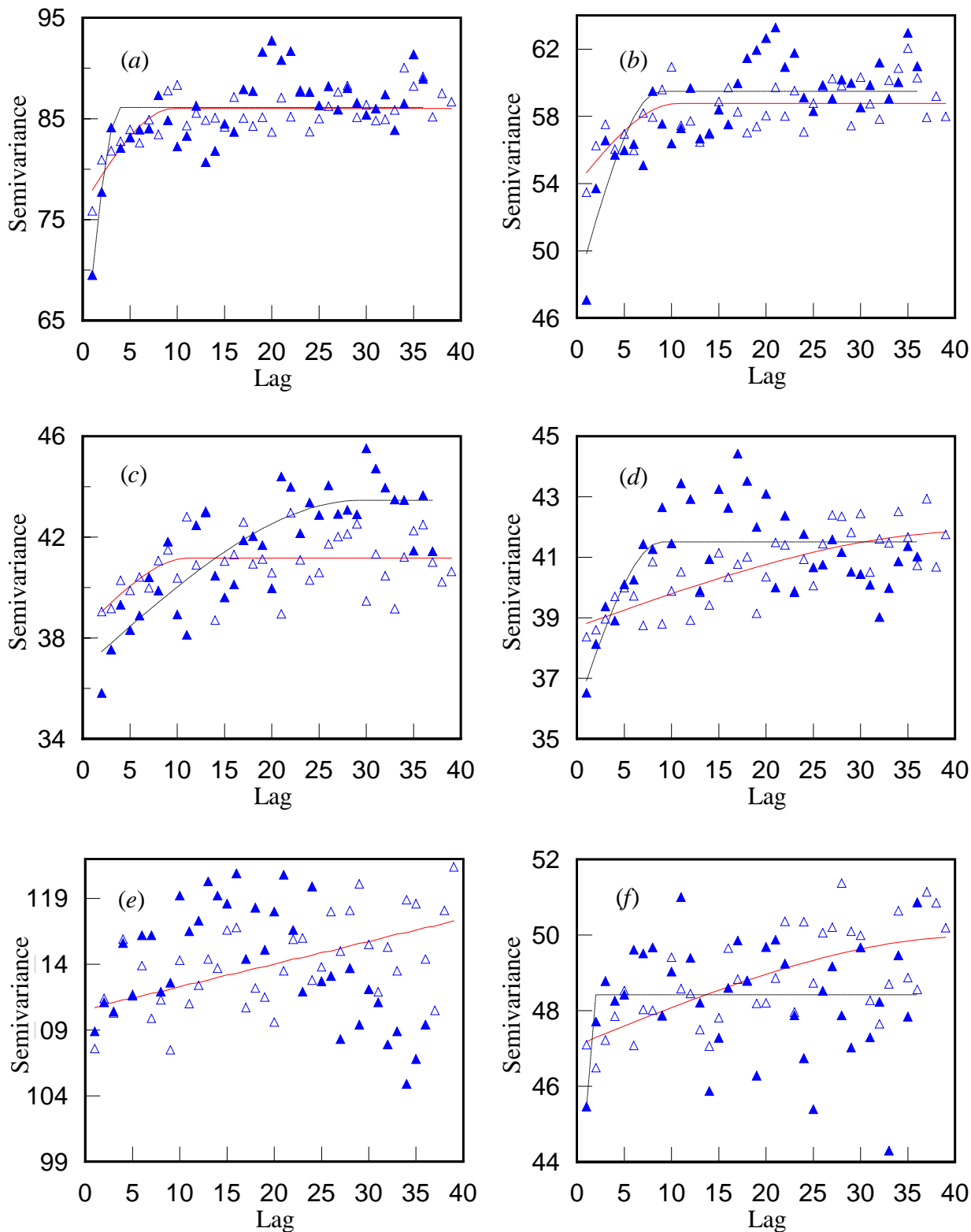


Fig. 2. Row and column semivariances against lag and fitted curves for the tetraploid perennial ryegrass 1989 variates: a) PLSE<sub>e+30</sub>, b) PLS<sub>e+30</sub>, c) PLSN<sub>e+30</sub>, d) H<sub>sp</sub>, e) W<sub>sp</sub>, f) PH<sub>sp</sub>

▲ Row Semivariances      — Fitted Curve (Rows)  
 △ Column Semivariances      — Fitted Curve (Columns)