

# Mixed model with spatial variance-covariance structure for accommodating of local stationary trend and its influence on multi-environmental crop variety trial assessment

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## Abstract

The most common procedure for analyzing multi-environmental trials is based on the assumption that the residual error variance is homogenous across all locations considered. However, this may often be unrealistic, and therefore limit the accuracy of variety evaluation or the reliability of variety recommendations. The objectives of this study were to show the advantages of mixed models with spatial variance-covariance structures, and direct implications of model choice on the inference of varietal performance, ranking and testing based on two multi-environmental data sets from realistic national trials. A model comparison with a  $\chi^2$ -test for the trials in the two data sets (wheat data set BW00RVTI and barley data set BW01RVII) suggested that selected spatial variance-covariance structures fitted the data significantly better than the ANOVA model. The forms of optimally-fitted spatial variance-covariance, ranking and consistency ratio test were not the same from one trial (location) to the other. Linear mixed models with single stage analysis including spatial variance-covariance structure with a group factor of location on the random model also improved the real estimation of genotype effect and their ranking. The model also improved varietal performance estimation because of its capacity to handle additional sources of variation, location and genotype by location (environment) interaction variation and accommodating of local stationary trend.

**Additional key words:** multi-environmental trials; chi-squared test; spatial variance-covariance; consistency ratio test; wheat; barley.

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## Introduction

National multi-environmental yield trials (MET), allow assessment of the potential yield performance of different varieties across a range of environments (locations and possibly over years, as well as combination of the two). These trials play an important part in crop variety evaluation in breeding programs and varietal recommendations for plant production. It is therefore vital that the statistical methods used to design the studies and analyse data from national yield trial evaluation programs are as accurate, efficient and informative as possible. Although the development of statistical methods for analysing variety trial data has

a long history, due to the complexity of varietal and environmental interactions there is no specific model that is generally suitable for analysing combined data sets from national trials. Spatial variability often exists in field experiments due to factors such as moisture, fertility, pH and structure of the soil, as well as the pressure of diseases and pests (Davidoff & Selim, 1988; Scharf & Alley, 1993; Wu & Dutilleul, 1999; Stroup, 2002). Multi-environment crop variety trials and field evaluations are a particularly well-known example of this. Failure to effectively control for spatial variability greatly increases the risk of misleading interpretations or erroneous inferences (Mo & Si, 1986; Stroup, 2002; Yang *et al.*, 2004).

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Abbreviations used: AIC (Akaike's information criteria); AR (autoregressive); BIC (Bayesian information criterion); CS (compound symmetry); d.f. (degrees of freedom); LL (log-likelihood); LRT (likelihood-ratio test); MET (multi-environmental trial); MVN (multivariate normal); NNA (nearest neighbour adjustment); RCBD (randomized complete block design); REML (restricted maximum likelihood); SLMM (spatial liner mixed model).

Historically, the analysis of variance (ANOVA), along with randomised block designs (including complete, incomplete blocks), has been used to deal with the spatial variability of these trials. Numerous studies have shown that such design-based control of the spatial variation of field trials are often not optimal and results in poor analysis efficiency (Yang *et al.*, 2004). Statistical procedures that account for spatial variation between plots within trials have been proposed to address the topic of modelling spatial variation in crop evaluation trials using polynomial trend analysis, nearest neighbour analysis and a model with correlated errors.

The problem with the ANOVA method as a means to analyse multi-environmental crop variety trials is that it requires the assumption of homogenous variance-covariance structures across locations or environments. This homogeneity of variance and covariance may be unrealistic in many circumstances (Kempton, 1984; Piepho, 1999a). As a result, a range of more complex and informative models that can account for variance or/and covariance heterogeneity have been proposed for analysing MET data. While other models are available, the problem of how the models should be assessed and which model is more suitable for a given trial's data has not been solved. This restricts the applicability of the models and model choice. Therefore, a linear mixed model approach with flexible spatial variance-covariance structures is proposed. Correspondingly, model-based approaches for analysing field trials that focus on the need to control spatial variation have been put forward. These approaches include nearest neighbour adjustment (NNA) analysis and its modifications (Bartlett, 1978; Cullis & Gleeson, 1991; Clarke & Baker, 1996; Yang *et al.*, 2004). Other options include linear mixed models with spatial covariance structures such as those used in geostatistics (Zimmerman & Harville, 1991; Gilmour *et al.*, 1997; Stroup, 2002). The efficiency of spatial approaches has been compared with the no spatial analyses found in the literature (Brownie & Gumpertz, 1997; Wu & Dutilleul, 1999; Smith *et al.*, 2001; Yang *et al.*, 2004; Hong *et al.*, 2005).

However, most comparisons of efficiency in the literature appear to focus on the nearest neighbour adjustment (including its modification or extensions) and/or the linear mixed model with one special covariance structure (usually the first order autoregressive model, AR(1)) against the analysis of variance of block designs. There have been few

comparisons of mixed models with different spatial covariance structures. Now a migration seems to be taking place from the NNA to a fully-fledged mixed model analysis with different spatial components for spatial variability because of the flexibility, simplicity of use and other advantages of mixed model analysis (Piepho *et al.*, 2008). Recently, linear mixed models have become well developed, and range from simple variance component models that provide information similar to ANOVA, to models with complex variance-covariance structures that aim to explore complex sources of variability and better accommodate interactions. Specifically, different analytical models can be cast in a unified mixed modelling framework (Denis *et al.*, 1997; Piepho, 1998, 1999b). Within such a framework, different models can be handled as mixed models with different variance-covariance structures. Thus candidate models can be assessed and selected for MET data analyses, which can result in high accuracy when estimating and testing varietal effects.

Within advanced experimental designs, many spatial methods were proposed for adjusting the spatial trend (Bartlett, 1978; Wilkinson *et al.*, 1983; Schwarzbach, 1984; Williams, 1986; Gilmour *et al.*, 1997; Gleeson, 1997; Piepho, 1999a). A common feature of these methods is that plots that are closer together are assumed to have a higher correlation than plots farther apart. Via such models the precision of genotypic value estimates can be improved through both blocking and the adjustment of spatial trend in one or two dimensions.

With regard to the practical application of the linear mixed model with a spatial component, various unsolved problems must be dealt with. Among other issues, these are concerned with the selection of a suitable covariance model, *i.e.*, a model with criteria that form the basis for a user's choice of whether or not to use a spatial model at all. Another point in this regard is the fact that the covariance parameters are unknown in practice and the estimated values based on observed data have to be used. In this case the statistical tests about the fixed effects of linear mixed models are generally not exact and their degrees of freedom must be determined by approximation. For some types of mixed models, the available methods for approximating degrees of freedom have been well examined (Schaalje *et al.*, 2002; Spilke *et al.*, 2004, 2005). For mixed models with spatial covariance structures, however, the use of the approximation methods has to be undertaken with care. In addition of the approxi-

mation, further consideration has to be given to the question of what influence the various spatial models have on the statistical tests used for, ranking and selection of lines in cultivar trial evaluations, apart from on efficiency vis a vis standard errors for line effect estimations. In MET, the local spatial tendency within trials and the residual heterogeneity between trials can be jointly modelled in the context of linear mixed models. By using a two-dimensional coordinate system at each trial, it is possible to define the plot location in a field, for example by specifying the latitude and longitude of plot centres (Casanoves *et al.*, 2005, 2013).

The main objectives and contribution of this paper were (1) to highlight the advantages of mixed effect models in the data analysis of a national MET; (2) to show the importance of several main spatial variance-covariance structures, and direct implications of model choice for the inference of varietal performance, ranking and testing based on two data sets from real national trials by comparing blocking without spatial effect (ANOVA) model and a model with a block and spatial effect; the mixed models with spatial variance-covariance structure models were fitted using restricted maximum likelihood (REML) approach; and finally (3) to compare parameter estimates, ranking the varieties and ranking order and tests of varietal effects between the ANOVA model with only block effects and the mixed effects model with a block effect with selected spatial variance-covariance structure.

## Material and methods

Linear mixed models have become well developed, and range from simple variance component models that provide information similar to ANOVA, to models with complex variance-covariance structures that aim to explore or better accommodate interactions. Specifically, different analytical models can be cast in a unified mixed modelling framework (Denis *et al.*, 1997; Piepho, 1998, 1999b). Within such a framework, different models with specific variance-covariance structures can be formulated. Thus candidate models can be assessed and selected for MET data analyses, which result in high accuracy when estimating and testing varietal effects. Although there are already some general reviews of crop breeding analysis and variety evaluation trials (Davidoff & Selim, 1988; Smith *et al.*, 2001, 2005), as well as studies on the

analysis of MET data using the mixed models (Bartlett, 1978; Piepho, 1997; Kelly *et al.*, 2007; Piepho & Möhring, 2010; Stefanova & Buirchell, 2010), most references just contain some examples for demonstration, or contain just one specific type of mixed model in data analysis.

Both traditional block design ANOVA models and spatial effect models can take the general form of the linear mixed model:

$$\mathbf{y} = \mathbf{X}\boldsymbol{\beta} + \mathbf{Z}\mathbf{u} + \mathbf{e} \quad [1]$$

where  $\mathbf{y}$  stands for the vector of observations,  $\mathbf{X}$  is a matrix of constants associated with the fixed effects contained in the vector  $\boldsymbol{\beta}$ ,  $\boldsymbol{\beta}$  is a vector of unknown fixed effects,  $\mathbf{Z}$  is a matrix of constants associated with the random effects,  $\mathbf{u}$  is a vector of random effects, and  $\mathbf{e}$  is a vector of random residual errors. The random effects are assumed to be distributed as multivariate normal (MVN) or more precisely  $\mathbf{u} \sim \text{MVN}(0, \mathbf{G})$  and the residual errors ( $\mathbf{e}$ ) distributed as  $\text{MVN}(0, \mathbf{R})$ . It follows that the vector of observations is distributed as  $\mathbf{y} \sim \text{MVN}(\mathbf{X}\boldsymbol{\beta}, \mathbf{V})$  where  $\mathbf{V} = \mathbf{Z}\mathbf{G}\mathbf{Z}' + \mathbf{R}$ . The matrix  $\mathbf{G}$  is the covariance matrix among random effects,  $\mathbf{R}$  is the covariance matrix among the random residual errors, and  $\mathbf{V}$  is the covariance matrix of  $\mathbf{y}$ . For block designs, block effects may be regarded as fixed or random effects. A random block analysis makes additional use of the so-called inter-block information and is generally the preferred approach (Littell *et al.*, 2006). In this article, block effects will be considered random in a combined analysis of data from different location. In this situation,  $\mathbf{u}$  is the vector of block effects, and  $\mathbf{Z}$  corresponds to the block effect design.

For analysis of variance models for block designs, block effects are assumed to be iid  $\sim N(0, \sigma_b^2)$ , and residual errors are assumed to be iid  $N(0, \sigma_e^2)$ , where iid denotes independent and identically distribution, and  $\sigma_b^2$  and  $\sigma_e^2$  are variance components of blocks and residual errors, respectively. Hence,  $\mathbf{G} = \mathbf{I}_b \sigma_b^2$  and  $\mathbf{R} = \mathbf{I}_n \sigma_e^2$ , where  $\mathbf{I}_b$  is an identity matrix whose dimension equals the number of blocks,  $\mathbf{I}_n$  is an identity matrix whose rank equals the number of observations. The main feature of analysis of variance models for block designs is that random variables located in the same block have the same covariance regardless of the extent of spatial variation; random variables not located in the same block have a covariance of zero.

In spatial effect models,  $\mathbf{R}$  takes the form  $\mathbf{R} = \mathbf{I}_n \sigma_s^2 + \sigma_s^2 \mathbf{F}$ , where  $\sigma_s^2$  is the covariance parameter of spatial structure variation,  $\mathbf{F}$  is a square matrix with

**Table 1.** Spatial covariance structures

Structures	Description	Parameters	(i,j) elements
SP(EXP)( <i>c-list</i> )	Exponential	2	$\sigma^2 \exp\{-d_{ij} / \theta\}$
SP(EXPA)( <i>c-list</i> )	Anisotropic Exponential	2c + 1	$\sigma^2 \prod_{k=1}^c \exp\{-\theta_k d(i, j, k)^{\rho_k}\}$
SP(EXPGA)( <i>c<sub>1</sub>,c<sub>2</sub></i> )	2D Exponential, Geometrically Anisotropic	4	$\sigma^2 \exp\{-d_{ij}(\theta, \lambda) / \rho\}$
SP(GAU)( <i>c-list</i> )	Gaussian	2	$\sigma^2 \exp\{-d_{ij}^2 / \rho^2\}$
SP(GAUGA) ( <i>c<sub>1</sub>,c<sub>2</sub></i> )	2D Gaussian, Geometrically Anisotropic	4	$\sigma^2 \exp\{-d_{ij}(\theta, \lambda)^2 / \rho^2\}$
SP(LIN)( <i>c-list</i> )	Linear	2	$\sigma^2 (1 - \rho d_{ij}) 1(\rho d_{ij} \leq 1)$
SP(LINL)( <i>c-list</i> )	Linear Log	2	$\sigma^2 (1 - \rho \log(d_{ij})) \times 1(\rho \log d_{ij} \leq 1)$
SP(MATERN)( <i>c-list</i> )	Matérn	3	$\sigma^2 \frac{1}{\Gamma(\nu)} \left(\frac{d_{ij}}{2\rho}\right)^\nu 2K_\nu\left(\frac{d_{ij}}{\rho}\right)$
SP(MATHSW)( <i>c-list</i> )	Matérn (Handcock-Stein-Wallis)	3	$\sigma^2 \frac{1}{\Gamma(\nu)} \left(\frac{d_{ij}\sqrt{\nu}}{2\rho}\right)^\nu 2K_\nu\left(\frac{2d_{ij}\sqrt{\nu}}{\rho}\right)$
SP(POW)( <i>c-list</i> )	Power	2	$\sigma^2 \rho^{d_{ij}}$
SP(POWA)( <i>c-list</i> )	Anisotropic Power	c + 1	$\sigma^2 \rho_1^{d(i,j,1)} \rho_2^{d(i,j,2)} \dots \rho_c^{d(i,j,c)}$
SP(SPH)( <i>c-list</i> )	Spherical	2	$\sigma^2 \left[1 - \left(\frac{3d_{ij}}{2\rho}\right) + \left(\frac{d_{ij}^3}{2\rho^3}\right)\right] 1(\rho d_{ij} \leq \rho)$
SP(SPHGA)( <i>c<sub>1</sub>,c<sub>2</sub></i> )	2D Spherical, Geometrically Anisotropic	4	$\sigma^2 \left[1 - \left(\frac{3d_{ij}(\theta, \lambda)}{2\rho}\right) + \left(\frac{d_{ij}(\theta, \lambda)^3}{2\rho^3}\right)\right] \times 1(d_{ij}(\theta, \lambda) \leq \rho)$

a dimension reflecting the number of observations, whose ij<sup>th</sup> element is f(d<sub>ij</sub>), in which d<sub>ij</sub> is the Euclidian distance between spatial observation points *i* and *j*. Suppose (x<sub>i</sub>,y<sub>i</sub>) and (x<sub>j</sub>,y<sub>j</sub>) describe the coordinates of the median points of plots for observations *i* and *j*, respectively, then their distance is:

$$d_{ij} = \sqrt{(x_i - x_j)^2 + (y_i - y_j)^2} \quad [2]$$

where *x* and *y* denote horizontal and vertical directions. The variable f(d<sub>ij</sub>) is generally a function of d<sub>ij</sub> and its form is dependent on the spatial model used, which is dependent on the characteristics of spatial variation. The spatial covariance structures available for analysing field trials are listed in Table 1. In Table 1

*c-list* contains the names of the numeric variables used as coordinates of the location of the observation in space, and is the Euclidean distance between the *i*<sup>th</sup> and *j*<sup>th</sup> vectors of these coordinates, which correspond to the *i*<sup>th</sup> and *j*<sup>th</sup> observations in the input data set. For SP(POWA) and SP(EXPA), *c* is the number of coordinates, and *d*(*i*, *j*, *k*) is the absolute distance between the *k*<sup>th</sup> coordinate, *k* = 1 ..., *c*, of the *i*<sup>th</sup> and *j*<sup>th</sup> observations in the input data set. For the geometrically anisotropic structures SP(EXPGA), SP(GAUGA), and SP(SPHGA), exactly two spatial coordinate variables must be specified as *c<sub>1</sub>* and *c<sub>2</sub>*. Geometric anisotropy is corrected by applying a rotation *1* and *İ* scaling to the coordinate system, *d<sub>ij</sub>*(*θ*, *λ*) which represents the Euclidean distance between two points in the

transformed space. SP(MATERN) and SP(MATHSW) represent covariance structures in a class defined by Matérn (see Matérn, 1986; Handcock & Stein, 1993; Handcock, 1994). The function  $K_\nu$  is the modified Bessel function of the second kind of (real) order  $\nu > 0$ ; the parameter governs the smoothness of the process (for further detail see SAS 9.3 help and documentation). The five spatial-variance covariance structures presented above belong to isotropic models, *i.e.*, the variation properties are the same in both directions  $x$  and  $y$ ; the other models, as their names show, belong to anisotropic covariance structures, *i.e.*, the variation properties can be different in directions  $x$  and  $y$ .

Estimation and statistical test of varietal effects for the classical analysis of block designs uses ANOVA, which is, equating the observed mean squares to the expected mean squares with the assumption of independence, normality and homogeneity of the variances of the residuals. While spatial models analyses use REML for estimating variance components. Estimable functions  $L\beta$  of linear contrast of fixed effects (variety) are estimated based on  $L\hat{\beta} = L(X'V^{-1}X)^{-1}X'V^{-1}y$  with  $V$  being replaced by a REML estimate  $\hat{V}$ . The variance of  $L\hat{\beta}$  is determined based on  $\text{var}(L\hat{\beta}) = L(X'\hat{V}^{-1}X)^{-1}L'$  (Hartley & Rao, 1967; Harville, 1977). Null hypotheses of the form of  $H_0: L\beta = 0$  are tested using the statistic

$$t = \frac{L\hat{\beta}}{\sqrt{\text{var}(L\hat{\beta})}} \sim t(d.f.) \quad [3]$$

In general, the test statistic in [3] is only approximately t-distributed and its degrees of freedom must be estimated. The approximate degrees of freedom in this research were determined using the Kenward-Roger method (Kenward & Roger, 1997). This approximation also uses the basic idea of Satterthwaite (1941). Its extension relative to the Satterthwaite method of Giesbrecht & Burns (1985) and Hrong-Tai Fai & Cornelius (1996) is an asymptotic correction of the estimated standard error of fixed effects due to Kackar & Harville (1984) in small and/or unbalanced data structures.

### Statistical tools for model selection and test of consistency

Two questions in the analysis of practical trials are whether there is significant spatial variability and

whether spatial models should be used (and if so, which models are most appropriate for data analysis). To answer these questions, statistical tools include likelihood-based methods (Oman, 1991; Wolfinger, 1993). The likelihood-ratio test (LRT) allows the comparison of the model's fit, provided that one of the models is hierarchically subordinated to the other or similarly the smaller model is nested with the larger one. This is the case if one model can be seen as a special case of a more general model due to certain model restrictions. The LRT then results from

$$LRT = -2(\ln LL_g - \ln LL_s) \sim \chi^2(d.f.) \quad [4]$$

where  $\ln LL_g$  and  $\ln LL_s$  denote the log likelihood of the general model  $g$  and the special models, respectively. Given certain regularity conditions, the LRT testing statistic asymptotically follows a  $\chi^2$  distribution, with the degrees of freedom ( $d.f.$ ) resulting from the number of restrictions that are necessary to transform the general model  $g$  into the special model (Fahrmeier & Hammerle, 1984; Greene, 2003). The general model fit, when compared to the special model, is considered better if  $LRT > \chi^2(1-\alpha, d.f.)$  with a significant level of  $\alpha$ . If the model comparison focuses on the covariance structure of a constant expectation structure, the likelihoods are employed via the REML method (Wolfinger, 1993). This can be used for the first question. In this case,  $g$  corresponds to the model with spatial correlations among observations, and corresponds to the model without spatial correlation among observations. The LRT based on formula [4] can also be used for testing the difference between the block design ANOVA model (block effects as random) and the model without correlations among observations, because the latter is also a special model variation of the former. Thus, it can be used for testing the difference between the spatial models with and without block effects.

As mentioned above, the LRT is only applicable when comparing two nested models. For model comparisons that do not require hierarchical models, there are a number of analytical criteria. These are so-called «Information Criteria» based on likelihood estimations. In the current work Akaike's Information Criterion (AIC) is used for comparing the covariance structures for an identical expectation structure using the REML estimation methods and is generally given by:

$$AIC = -2 \ln LL + 2q \quad [5]$$

where  $\ln LL$  is the log-likelihood same as in formula [4] and  $q$  is the number of the parameters of the variance-

covariance structure. Thus, the formula of the information criteria is given in such a way that the model with the smaller value for the AIC is preferred (Bozdogan, 1987; Burnham & Anderson, 2002). For un-nested model we prefer to use the AIC but we note that there are other available information criteria, such as the Schwarz Bayesian Information Criterion (BIC) (Schwarz, 1978). Guerin & Stroup (2000) compared the performance of AIC and BIC on covariance model selection for repeated measures and stated that AIC tends to select a more complex model but with a better control of type I error than the BIC. To assess consistency (or inconsistency) in the statistical tests on varietal effects between two models one can use the test consistency ratio, which is computed as follows:

$$\text{test consistency ratio} = \frac{\text{number of significant varietal differences tested simultaneously in two models}}{\max(\text{number of significant varietal differences tested under the two models considered})} \quad [6]$$

## Data set and analysis

The data sets used in this study are taken from the Ethiopian Agricultural Research Institute National Variety Trials for Bread Wheat (BW00RVTI data) and Barley Trial (BW01RVII data) of 2006-2008. Some 20 bread wheat (*Triticum aestivum* L.) varieties were tested in at six locations (environments) on the first year (2006/7) and five locations (environments) among the six of the first used on the second year (2007/8). Similarly 25 barley (*Hordeum vulgare* L.) varieties were tested in five locations (environments) in 2007/8. All the trials in each location were laid out as a randomized complete block (RCB) design with four replicates. There are two approaches to analysing MET data using mixed model, the so-called one- and two-stage approaches (Welham *et al.*, 2010). In a one-stage analysis, individual plot data from all trials are combined in a single analysis (Cullis *et al.*, 1998). In a two-stage analysis, variety means are first obtained from the separate analysis of individual trials (Stage I), and are then combined in an overall mixed model analysis (Stage II). The two-stage analysis can be unweighted (*e.g.*, Patterson & Silvey, 1980) or weighted to reflect the relative precision of variety means from each trial (*e.g.*, Smith *et al.*, 2001). A one-stage approach provides the most accurate predictions of variety performance, but it can be computationally difficult to use when the variance models involved are complex. With the steady improvements in computing power, single-stage analyses are becoming feasible.

Apart from computational speed, the main advantages of the two-stage approach are that one can carefully analyse each trial individually, taking into account any specifics of the design or field trends.

In this study we used two approaches for analysis; the first one was a separate individual analysis of each location of the BW00RVTI data set of wheat and BW01RVII data set for barley. The second one was a one-stage analysis, individual plot where data from all trials (locations) are combined in a single analysis of a two year BW00RVTI data set of wheat and a one year BW01RVII data set of six location. Each data set was separately fitted per location and per year using the mixed model with fourteen variance-covariance structures. The mixed model with compound symmetry (CS) variance-covariance structures was identical to the ANOVA model. The optimally fitted spatial model and the ANOVA model are used for further varietal effect assessment and statistical tests (or inference). The single-stage analysis was applied to each of the data sets by fitting one spatial-variance covariance structure at a time for all location. Putting location as random group factor on SAS (proc mixed) analysis gave a different random parameter estimate for each location. All the analyses were conducted using standard SAS software version 9.3. The results from the two models were compared and used to assess consistency (or inconsistency) in statistical tests on varietal effects between the two models, using consistency ratio defined earlier.

## Results and discussion

Model fit statistics from ANOVA and the mixed model with various spatial variance-covariance structures and results of possible LRT and AIC for all models are summarised in Tables 2, 3 and 4. Note that “—” denotes the failure of a model to converge. This occurred with the sp(lin) and sp(linlog) structures in any of the locations, which shows that these models are not suitable for that trial data (Schabenberger & Pierce, 2002). The smallest AIC value (bold in Tables 2, 3 and 4) indicates that for BW00RVTI trial data set year 1 and 2 support the anisotropic power [spa (powa)] and exponential [spa (exp)] variance-covariance structures as the best compared to the ANOVA model for seven trials (locations) out of eleven. Similarly for the BW00RVTI trial five different spatial variance-covariance structures [sp(pow),

**Table 2.** Related fitting statistics for the ANOVA model and the linear mixed model with spatial variance-covariance structures for the first year BW00RVTI data set

Model	Location-1			Location-2			Location-3			Location-4			Location-5			Location-6		
	LL	AIC	Pr> $\chi^2$	LL	AIC	Pr> $\chi^2$												
RCBD	385.1	387.1	—	344.2	346.2	—	398.8	400.8	—	426.4	<b>428.4</b>	—	421.7	<b>423.7</b>	—	377.3	379.3	—
sp(sph)	408.5	412.5	1	360.8	364.8	1	407.5	411.5	1	442.1	446.1	1	441.4	445.4	1	364.4	368.4	0.0003
sp(exp)	385.1	387.1	1	343.7	347.7	0.4869	394.2	398.2	0.0303	426.4	<b>428.4</b>	1	421.7	<b>423.7</b>	1	361.8	365.8	<.0001
sp(gau)	384.9	388.9	0.6537	343.8	347.8	0.5743	394.7	398.7	0.0425	426.3	430.3	0.746	421.7	<b>423.7</b>	1	365.5	369.5	0.0006
sp(pow)	385.1	389.1	0.865	343.7	347.7	0.4869	394.2	398.2	0.0303	426.2	430.2	0.63	421.7	425.7	0.9213	361.8	365.8	<.0001
sp(mat)	385.1	387.1	1	343.7	349.7	0.7806	394.1	400.1	0.0953	426.1	432.1	0.8603	421.6	427.6	0.9648	359.7	365.7	0.0001
sp(EXPA)	—	—	—	—	—	—	386.4	396.4	0.0144	—	—	—	—	—	—	—	—	—
sp(EXPGA)	385.1	391.1	0.0608	334.5	342.5	0.0179	386.1	394.1	0.0747	426.4	432.4	0.2721	421.7	427.7	0.1855	353	361	0.0034
sp(GAUGA)	379.3	387.3	0.0339	344.2	352.2	1	398.8	404.8	1	426.4	432.4	0.3575	421.7	427.7	0.2295	363.9	371.9	0.1199
sp(MATHSW)	385.1	389.1	1	343.7	349.7	0.7806	394.1	400.1	0.0953	426.4	430.4	1	421.7	425.7	1	359.7	365.7	0.0001
sp(POWA)	372.7	<b>378.7</b>	0.002	332.5	<b>338.5</b>	0.003	386.4	<b>392.4</b>	0.002	424.8	430.8	0.4545	420.1	426.1	0.4665	349	<b>355</b>	<.0001
sp(SPHGA)	393.8	399.8	1	—	—	—	—	—	—	—	—	—	439.2	447.2	1	355.6	363.6	<.0001

—: denotes the failure of a model to converge. Bold values indicate smallest AIC (Akaike’s information criteria). LL: log-likelihood. Locations 1, 2, 3, 4, 5 and 6 are Kulumsa, Adet, Bekoji, Sinana, Holeta, and DeberZeit (Ethiopia), respectively.

**Table 3.** Related fitting statistics for the ANOVA model and the linear mixed model with spatial variance-covariance structures for the second year BW00RVTI data set

Model	Location-1			Location-2			Location-3			Location-4			Location-5		
	LL	AIC	Pr> $\chi^2$												
RCBD	370	372	—	334.2	336.2	—	345.6	<b>347.6</b>	—	395.2	397.2	—	284.1	<b>286.1</b>	—
sp(sph)	382.4	386.4	1	351.9	355.9	1	366.4	370.4	1	394.2	398.2	0.317	303	307	1
sp(exp)	367.1	<b>371.1</b>	0.091	333.5	337.5	0.428	345.6	<b>347.6</b>	1	387.3	<b>391.3</b>	0.005	284.1	<b>286.1</b>	1
sp(gau)	368.3	372.3	0.195	332.4	336.4	0.189	345.6	<b>347.6</b>	1	388.7	392.7	0.011	284.1	<b>286.1</b>	1
sp(pow)	367.1	<b>371.1</b>	0.091	333.5	337.5	0.428	345.5	349.5	0.659	387.3	<b>391.3</b>	0.005	284	288	0.762
sp(mat)	365.8	371.8	0.124	—	—	—	—	—	—	386.9	392.9	0.016	—	—	—
sp(EXPA)	366.2	374.2	0.29	322.9	<b>330.9</b>	0.01	343.9	353.9	0.79	384.9	394.9	0.036	—	—	—
sp(EXPGA)	363.8	371.8	0.339	323.1	331.1	0.003	340.5	348.5	0.108	384.6	392.6	0.232	284.1	290.1	0.234
sp(GAUGA)	370	378	1	325.6	333.6	0.032	344	352	0.976	395.2	403.2	1	284.1	290.1	0.594
sp(MATHSW)	365.8	371.8	0.124	—	—	—	—	—	—	386.9	392.9	0.016	284.1	288.1	1
sp(POWA)	367.9	373.9	0.367	322.7	328.7	0.003	344.1	350.1	0.464	386.3	392.3	0.011	283.6	289.6	0.773
sp(SPHGA)	382.4	390.4	1	—	—	—	356.1	364.1	1	391.3	399.3	0.273	—	—	—

—: denotes the failure of a model to converge. Bold values indicate smallest AIC (Akaike’s information criteria). LL: log-likelihood.

sp(expga), sp( mathsw), sp(expga) and sp(powa)] models were selected as the best compared to the ANOVA model for the five location BW01RVII trial data set.

A model comparison between a block effect without spatial structure (ANOVA) and a model with a block and spatial effect using the LRT  $\chi^2$ -test for the trials for the two (BW00RVTI and BW01RVII) data sets suggested that the selected spatial variance-covariance structure fitted the data significantly better than the ANOVA model. However the optimally-fitted spatial

variance-covariance structures were not the same from one location to the other. The optimally fitting spatial variance-covariance structure was spatial power [sp(powa)] for most of the locations. These results showed that assuming a homogeneous variance-covariance structure in the ANOVA model is generally not realistic, and therefore using a linear mixed model with spatial variance-covariance is necessary to improve the efficiency of the data analysis and accommodation of local stationary trend of MET data.

It appears the year to year effect on variance-

**Table 4.** Related fitting statistics of ANOVA model and linear mixed model with spatial variance-covariance structures for the one year BW01RVII data set

Model	Location-1			Location-2			Location-3			Location-4			Location-5		
	LL	AIC	Pr> $\chi^2$												
RCBD	475.5	477.5	—	454.5	456.5	—	421.6	423.6	—	506.9	508.9	—	524	526	—
sp(sph)	475.5	477.5	1	454.5	456.5	1	427.7	431.7	1	506.9	508.9	1	551.3	555.3	1
sp(exp)	468.5	<b>472.5</b>	0.0083	452.3	456.3	0.142	410.9	414.9	0.001	503.7	507.7	0.0712	524	526	1
sp(gau)	471	475	0.0341	452.4	456.4	0.145	414.1	418.1	0.006	504.8	508.8	0.1409	523.9	527.9	0.689
sp(pow)	468.5	<b>472.5</b>	0.0083	452.3	456.3	0.142	410.9	414.9	0.001	503.7	507.7	0.0712	524	528	0.8947
sp(mat)	—	—	—	452	458	0.283	407.7	<b>413.7</b>	0.001	502.9	508.9	0.1344	524	526	1
sp(EXPA)	—	—	—	452.4	462.4	0.711	—	—	—	—	—	—	—	—	—
sp(EXPGA)	466.7	474.7	0.347	447.7	<b>455.7</b>	0.07	421.6	429.6	1	493.2	<b>501.2</b>	0.0141	524	530	0.0323
sp(GAUGA)	469	477	0.3427	449.6	457.6	0.157	410.1	418.1	0.275	498.6	506.6	0.0619	529.9	530.5	0.1424
sp(MATHSW)	—	—	—	452	458	0.283	407.7	<b>413.7</b>	0.001	502.9	508.9	0.1344	524	528	1
sp(POWA)	467.4	473.4	0.0179	458.8	459.2	0.435	413.7	419.7	0.02	497.1	503.1	0.0071	519.2	<b>525.2</b>	0.0873
sp(SPHGA)	475.5	481.5	1	458.7	466.7	1	420.4	428.4	0.742	495	503	0.0077	—	—	—

—: denotes the failure of a model to converge. Bold values indicate smallest AIC (Akaike’s information criteria). LL: log-likelihood.

**Table 5.** The number of significant and highly significant variety contrasts of t-test for trials of the BW00RVTI and BW01RVII data sets and the consistency ratio test between the ANOVA model and the spatial linear mixed model with optimally fitting spatial variance-covariance structure (SLMM)

	Data set BW00RVTI								Data set BW01RVII				
	Year-1				Year-2				Year-1				
	ANOVA	SLMM	Consistency		ANOVA	SLMM	Consistency		ANOVA	SLMM	Consistency		
			No.	Ratio (%)			No.	Ratio (%)			No.	Ratio (%)	
Location-1	60	78	53	67.94	44	40	37	84.09	Location-1	46	65	43	66.15
Location-2	53	50	37	69.81	33	35	26	74.29	Location-2	67	79	60	75.95
Location-3	10	18	6	33.33	*	*			Location-3	97	157	94	59.87
Location-4	*	*			45	50	41	82	Location-4	47	36	22	46.8
Location-5	*	*			*	*			Location-5	36	45	26	57.78
Location-6	45	20	15	33.33									
Average	42	41.5	22.75	51.11	40.67	41.66	34.67	80.13	Average	58.6	76.4	49	61.31

\*: the optimally fitting model is ANOVA.

covariance of varieties is greatly exhibited in the BW00RVTI data set. This is shown through the variance-covariance structures being mostly consistent for different locations in the same year, but obviously not consistent between years as shown in Table 2 and 3. This result is easily understood by realising that within a year we expect only between location differences, but between years there could be differences in environments (years). The failure of some spatial variance-covariance structures to converge may indicate that they are not suitable or compatible with

the structure of the current MET data but could work with other data sets.

To examine the impact of the spatial variance-covariance structures on estimates on test of varieties, the number of significant (at  $\alpha = 0.05$ ) varietal differences by the t-test are given in Table 5. Using the ANOVA model and mixed model with the optimally-fitted spatial variance-covariance for each location, we assessed the consistency between these two models. The number of significant varietal differences by t-test is not the same between the ANOVA model and the

**Table 6.** The first eight genotype ranking comparison between the ANOVA model and the optimally fitting spatial variance-covariance structure (SLMM) of five trials of data set BW01RVII location by location and a single-stage analysis

Rank	Location-1		Location-2		Location-3		Location-4		Location-5		All
	ANOVA	SLMM	SLMM								
1	G23	G23	G21	G23	G23	G23	G23	G23	G23	G23	G23
2	G13	G13	G23	G21	G4	G4	G11	G2	G2	G2	G21
3	G5	G21	G3	G3	G17	G17	G19	G1	G4	G1	G4
4	G17	G3	G17	G2	G2	G21	G15	G5	G8	G19	G13
5	G21	G17	G7	G17	G21	G10	G2	G19	G13	G10	G2
6	G4	G4	G11	G13	G10	G2	G5	G8	G14	G4	G17
7	G3	G5	G9	G16	G8	G8	G8	G15	G1	G3	G8
8	G19	G15	G13	G9	G6	G15	G13	G11	G19	G13	G3

mixed model with optimally fitted spatial variance-covariance structures. The consistency ratio test between the two models falls in the range of 33-84%. From the average of all trials (locations), the test consistency ratio of two models is approximately 64%, which means that approximately 36% of the varietal differences being tested as significant or very significant in one model cannot be tested as significant or very significant by the other model.

### Varietal ranking

Apart from contrasts between new varieties, the ranking of varietal productivity and a comparison of new varieties with standard variety is also important for variety trials. We consider the trial from the five locations of BW01RVII data to compare variety mean ranking between the ANOVA model and the optimal spatial variance-covariance model. A trial corresponds to a single experiment at a single location. Table 6 shows the ranking for the first eight entries from the optimal spatial variance-covariance mixed model compared to the ranking from the ANOVA model across the locations. The model with spatial structure is relatively more consistent in its top eight ranking than the ANOVA model. The ranking are different for different locations and differ between the spatially structured model and ANOVA. A rank difference of genotype between the locations is showing the presence of genotype by environment interaction. This also indicates the advantage of single stage spatial models on the handling of the spatial trend and variation of the trials.

The simple homogenous variance-covariance structures implied by ANOVA models, which assume

that the interaction effects of varieties are independent, is mostly not appropriate for data analyses of MET. The fact that the goodness of fit of one variance-covariance structure was different for various trial data sets, and that none fitted all trial data sets optimally throughout, indicates that the heterogeneous characteristics of variance-covariance are not identical across the trials. Therefore, the arbitrary use of a homogeneous variance-covariance structure (*e.g.* ANOVA model) to analyse the MET cannot ensure a high degree of accuracy. In this study, the ANOVA model, as a special case form of mixed models, showed obvious inconsistency in estimates and tests of varietal effects compared to the linear mixed model with the optimally-fitted spatial variance-covariance structures.

Both effective experimental designs and spatial analyses can have an important role in improving the reliability and precision of experiment results. The importance of spatial variability to be expected from a logical and subjective-related perspective is confirmed in a variety of experiments. As presented in much of the literature, spatial analysis may lead to higher efficiency with regard to standard error of estimation of fixed effects than a non-spatial analysis, provided that spatial variability is present. Based on this work, the commonly used ANOVA mixed model is not an appropriate model for data analysis of MET trials. The spatial variance-covariance models are more useful in a practical sense, given that they can describe actual existing variance-covariance characteristics more accurately than the ANOVA model. Of course, with one-stage analyses, the proposed spatial variance-covariance models are expected to yield identical mean yields for balanced data, and differences are expected only for unbalanced data. Even so, a selection of

variance-covariance structures based on the mixed model framework is important since the standard error of varietal effect estimates (*i.e.* the accuracy of varietal effect estimates) is different under the various models, and unbalanced data is common in MET (Möhring & Piepho, 2009). The advantage and validity of using spatial variance-covariance structure depends on the present spatial variability. Most of the investigated spatial models showed better data fitting and smaller standard error for variety contrasts than the ANOVA model.

The main purposes of the present paper was to show the importance of variance-covariance structure selection and to illustrate that the classical ANOVA model is inferior to more elaborate mixed models in the analysis of MET data. This does not imply that the models considered in this paper are appropriate for any situation. For example, in some locations (trials) the ANOVA model still optimally fitted the data better than the spatial models.

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