

2015

# Thin Plate Spline Regression Model Used at Early Stages of Soybean Breeding to Control Field Spatial Variation

M. Sun  
*Monsanto Inc.*

A. S. Goggi  
*Iowa State University, susana@iastate.edu*

K. Matson  
*Monsanto Inc.*

R. G. Palmer  
*Iowa State University*

K. Moore  
Follow this and additional works at: [https://lib.dr.iastate.edu/agron\\_pubs](https://lib.dr.iastate.edu/agron_pubs)  
*Iowa State University, kjmoore@iastate.edu*

 Part of the [Agriculture Commons](#), [Agronomy and Crop Sciences Commons](#), and the [Plant Breeding and Genetics Commons](#)  
*See next page for additional authors.*

The complete bibliographic information for this item can be found at [https://lib.dr.iastate.edu/agron\\_pubs/512](https://lib.dr.iastate.edu/agron_pubs/512). For information on how to cite this item, please visit <http://lib.dr.iastate.edu/howtocite.html>.

---

This Article is brought to you for free and open access by the Agronomy at Iowa State University Digital Repository. It has been accepted for inclusion in Agronomy Publications by an authorized administrator of Iowa State University Digital Repository. For more information, please contact [digirep@iastate.edu](mailto:digirep@iastate.edu).

---

# Thin Plate Spline Regression Model Used at Early Stages of Soybean Breeding to Control Field Spatial Variation

## Abstract

Yield variation observed in Soybean (*Glycine max*) progeny-row yield trial (PRYT) is the final result of line genotypic merit, field spatial pattern, and experimental error. The spatial variation in field tests could confound the estimates of genetic merits. The objectives of this research were to: i) quantify non-genetic yield variation in a soybean breeding PRYT; and ii) determine efficiency of the Thin Plate Spline Regression (TPSR) model in adjusting yield because of variation caused by field spatial pattern. The 3rd objective was to evaluate if the use of the TPSR model could improve the selection accuracy of PRYT unreplicated yield tests. Uniformity Study, Early Generation Test, and Confirmation Study were conducted. Our results indicated that large spatial variations in soybean PRYT field could be present as evaluated by the Uniformity Study conducted with two commercial lines. In this experiment, the use of the TPSR proved to be effective in reducing the error variance and the coefficient of variability, with an improvement in relative efficiency (IRE) of 37.9%. In Early Generation Tests, 2565 lines were evaluated within test-sets along with three checks. The TPSR model also was effective in the Early Generation Tests, the IRE was 40.4%. The correlation coefficients calculated between yield estimates obtained in two-year Early Generation Tests and Confirmation Study improved by 0.21 points compared with results from the non-TPSR experiments. The results indicated that the use of TPSR model was effective in accounting for some of the spatial variation in field tests.

## Keywords

Progeny-row yield trial, genetic gain, two-dimensional thin plate spline regression, best linear unbiased prediction, soybean breeding

## Disciplines

Agriculture | Agronomy and Crop Sciences | Plant Breeding and Genetics

## Comments

This is a manuscript of an article published as Sun, M., A. S. Goggi, K. Matson, R. G. Palmer, K. Moore, S. R. Cianzio. 2015. Thin plate spline regression model used at early stages of soybean breeding to control field spatial variation. *J. of Crop Improvement* 29 (3): 333-352. doi: [10.1080/15427528.2015.1026623](https://doi.org/10.1080/15427528.2015.1026623). Posted with permission.

## Authors

M. Sun, A. S. Goggi, K. Matson, R. G. Palmer, K. Moore, and S. R. Cianzio

# **Thin Plate Spline Regression Model Used at Early Stages of Soybean Breeding to Control Field Spatial Variation**

M. Sun<sup>1</sup>, S. A. Goggi<sup>2</sup>, K. Matson<sup>1</sup>, R. G. Palmer<sup>2</sup>, K. Moore<sup>2</sup>, S. R. Cianzio<sup>2\*</sup>

<sup>1</sup> *Monsanto Inc., St. Louis, Missouri 63017, USA*

<sup>2</sup> *Department of Agronomy, Iowa State University, Ames, Iowa 50011-1010, USA*

*Address correspondence to Dr. Cianzio at 2107 AGRON Hall, Ames, IA 50011-1010, USA. E-mail: [scianzio@iastate.edu](mailto:scianzio@iastate.edu)*

## ABSTRACT

Yield variation observed in Soybean (*Glycine max*) progeny-row yield trial (PRYT) is the final result of line genotypic merit, field spatial pattern, and experimental error. The spatial variation in field tests could confound the estimates of genetic merits. The objectives of this research were to: i) quantify non-genetic yield variation in a soybean breeding PRYT; and ii) determine efficiency of the **Thin Plate Spline Regression (TPSR)** model in adjusting yield because of variation caused by field spatial pattern. The 3<sup>rd</sup> objective was to evaluate if the use of the **TPSR** model could improve the selection accuracy of PRYT un-replicated yield tests. Uniformity Study, Early Generation Test, and Confirmation Study were conducted. Our results indicated that large spatial variations in soybean PRYT field could be present as evaluated by the Uniformity Study conducted with two commercial lines. In this experiment, the use of the **TPSR** proved to be effective in reducing the error variance and the coefficient of variability, with an improvement in relative efficiency (IRE) of 37.9%. In Early Generation Tests, 2565 lines were evaluated within test-sets along with three checks. The **TPSR** model also was effective in the Early Generation Tests, the IRE was 40.4%. The correlation coefficients calculated between yield estimates obtained in two-

year Early Generation Tests and Confirmation Study improved by 0.21 points compared with results from the non-**TPSR** experiments. The results indicated that the use of **TPSR** model was effective in accounting for some of the spatial variation in field tests.

**KEYWORDS** Progeny-row yield trial, genetic gain, two-dimensional thin plate spline regression, best linear unbiased prediction, soybean breeding

**ABBREVIATIONS AND NOMENCLATURE** (PRYT) progeny-row yield trial; (CV) coefficient of variation; (SRMSD) square root of mean square deviation; (SRMSE) square root of mean sum of squares for error; (**TPSR**) two-dimensional thin plate spline regression; (IRE) improvement in relative efficiency; (IQR) interval quartile range; (yld\_adj) yield with the **TPSR** spatial effect adjustment; (BLUP) Best linear unbiased prediction; (RM) relative maturity.

In this publication, data from Monsanto Inc. have been used with permission.

## INTRODUCTION

The major goal in soybean breeding programs is to select cultivars with superior yield. Development of new cultivars in a breeding cycle involve parental selection, crossing between the desired parents to generate genetic variation in the segregating population, two to three generations of inbreeding, a planting for maturity classification, and multi-stage yield tests conducted across locations and years (Fehr, 1993; Orf et al., 2004). Public release and/or commercialization begin after the superior genotypes have been identified,

and their performance has been evaluated across a range of environments. One of the most important steps in the breeding cycle is the yield test conducted at early stages of the program.

Soybean yield tests at early stages generally consist of progeny-row yield tests (PRYTs) (Fehr, 1993). Plots in the tests are usually single-rows planted with seed from an individual plant. The limited seed numbers produced by an individual plant imposes limitations to plot sizes and also to number of replicates. The PRYTs are the initial indicator of yield potential for  $F_3$ - or  $F_4$ -derived lines, because each line represents a unique genetic entity from a population developed by crossing the two desired parents. A widely used layout of PRYT fields in North American soybean breeding programs consists of unreplicated one-row plots, usually 1.5 m in length with a 0.8 m space between rows planted at one location (Cianzio, unpublished information, Iowa State University, 2008).

The use of PRYTs to predict yield potential at early generations has shown to be effective in predicting the yield potential for later tests conducted at multi-locations (St. Martin et al., 1990; Hegstad et al., 1999). Bernardo (2003) conducted a simulated study for self-pollinated crops and concluded that early generation tests and PRYT-based selection of lines were expected to be effective in predicting performance of genotypes, unless non-genetic effects were large relative to the true genetic merits of the tested lines. Bernardo's observations suggest that test size may be an important factor that could affect the prediction value of the early generation tests. According to Bernardo (2003), the observed yield variation in PRYT results from the genotype of the soybean lines, and the addition of variable non-genetic effects, such as field spatial patterns, and experimental error. The

variation due to non-genetic effects could be a confounding factor in determining the genetic variation in yield of the test lines (Becker, 1995).

The heterogeneity and spatial trend within the test field, caused by factors such as soil heterogeneity, agricultural practices, pest pressure variation, among others, may be contributors to the non-genetic effects (Gilmour et al., 1997). The field spatial trend is usually measured as a yield correlation among plots (Brownie et al., 1993; Federer, 1956), and this can have large effects on line rank and hence bias the accuracy of selection (Brownie et al., 1993; Stroup et al., 1994). Lussenden (personal communication, Redwood Falls, MN, Monsanto Inc., 2011) observed that yield variation caused by non-genetic effects can be as high as 383.3 kg ha<sup>-1</sup> in soybean PRYT fields near Redwood Falls, MN, with a mean performance of lines equal to 1748.5 kg ha<sup>-1</sup>.

The experimental error is a random variable contributing to biasing the estimates of genetically superior lines. The spatial pattern in yield is, however, often predictable, and may be removed from the observed yield by proper experimental design used in conjunction with spatial models (Stroup et al., 1994; Becher, 1995; Stringer and Cullis, 2002; Robbins et al, 2012; Edmé et al., 2007).

For non-replicated experiments with a large number of lines under evaluation as is often the case in PRYT, the augmented design (Federer, 1956) along with proper modeling (Scott and Milliken, 1993) has been effective in removing the spatial trend in one dimension (Federer, 1961). Spatial trends in an experimental field, however, are not limited to a single dimension, and the augmented design may not be sufficient to capture spatial trends in two dimensions (Federer, 1961). The **Thin Plate Spline Regression (TPSR)** model can be used to address this problem where splines are piecewise functions

fitted through control points, referred to as knots, and thereby allow complex functions to be approximated by relatively simple functions connected by knots (Bookstein, 1989; Bazen and Gerez, 2002; Robbins et al, 2012). The **TPSR** model has been effective in engineering for modeling deformation in two-dimensional planes (Bazen and Gerez, 2002). Robbins et al. (2012) introduced the **TPSR** to maize non-replicated yield trials and validated the use of the **TPSR** using simulated data. The authors indicated that the **TPSR** effectively dissected the spatial trend from the genetic effects, and reduced the error variance by modeling the spatial variation.

There are no published studies in which the effect of the **TPSR** model in capturing and removing field spatial trend in PRYT for soybean yield has been investigated. The objectives of this research were to quantify non-genetic yield variation in a soybean breeding PRYT, and determine the efficiency of the **TPSR** model in adjusting yield due to variations caused by field spatial pattern using a standard analysis of variance. A 3<sup>rd</sup> objective was to evaluate if the application of the **TPSR** model could increase the selection accuracy in unreplicated PRYTs.

## MATERIALS AND METHODS

The research consisted of three separate experiments: 1) the Uniformity Study; 2) the Early Generation Tests; and 3) the Confirmation Study. The Uniformity Study was performed to quantify the possible yield variation within a PRYT test-set, and to determine the efficiency of the **TPSR** model by the improvement in relative efficiency (IRE) between the actual yield test and the adjusted yield test. The Early Generation Tests were designed to assess the efficiency of the **TPSR** model by obtaining a correction factor for the spatial

effects. The Confirmation Study was designed to assess the efficiency of the **TPSR** model by comparing the performance of the bi-parental populations and the individual lines evaluated in the Early Generation Tests and their later evaluation in multiple location tests. The line and population performance estimates obtained from the Confirmation Study were assumed to provide closer estimates to the true genetic performance of the lines, since the estimates were derived from replicated multi-location field tests.

In the conduct of the three experiments, field plots were planted and harvested mechanically, utilizing computer technology to automatically record grain harvest and grain moisture. The agronomic data recorded in each individual plot were relative maturity (RM) at reproductive stage R7 (Fehr et al., 1971), and grain yield measured in kg ha<sup>-1</sup> at reproductive stage R8 (Fehr et al., 1971) with a moisture correction factor to express yield at 13.5% moisture content. The individual plots of each experiment were each harvested in bulk.

#### Uniformity Study

Two lines, MON1230 and MON1431, were selected for this study. The lines were commercial pure lines with stable yield performance from 2010 to 2012 (Lussenden, personal communication, Redwood Falls, Monsanto Inc. 2012). MON1431 yielded on average 190.1 kg ha<sup>-1</sup> more than MON1230 based on 737 within test-set comparisons at 199 locations evaluated from 2010 to 2012 in Monsanto Inc. yield tests.

In 2012, the Uniformity Study was conducted at the Monsanto Inc. soybean research station located near Redwood Falls, MN. The soil at the research location is a silt clay loam, and the sowing date was June 11. Each line was planted in 48 row-plots, arranged in a grid

that was four ranges deep and 12 columns wide. The 48 row-plots of each line were considered a separate test-set. Each test-set was planted in a block, according to the field layout of a randomized complete-block design (RCBD) with four blocks and two replicates within each block. The row-plots within test-sets were 1.5 m in length, spaced 0.8 m between rows, planted with 40 seeds, resulting in a sowing population of 27 seeds m<sup>-1</sup>.

### Early Generation Test

The Early Generation Tests were conducted in 2010 and in 2012 at the Monsanto Inc. soybean research station located near Huxley, IA. The soil at the research location is a sandy clay loam. The sowing date was May 5 in 2010, and May 10 in 2012. The weather observed between the two years was quite different, particularly in reference to rain fall received during the growing seasons (Table 1).

Nineteen bi-parental populations, each represented by 135 F<sub>3</sub>-derived lines in F<sub>3:5</sub> generation, were evaluated every year for a total of 2565 F<sub>3:5</sub> lines. The 135 lines per population were randomly divided into three groups of 45 lines each, to which three checks were added, bringing the total to 48 entries per test-set. Each of the 19 bi-parental populations was represented by three test-sets as three replicates. Each of the three checks formed a check-set with three replicates. Each check-set contained one check replicated 46 times, and two other checks replicated only once. Total number of checks used in the experiments was three.

According to the field layout of the RCBD, the 19 test-sets and the three check-sets were assigned to three blocks. Within a block, the test-sets and the check-sets were arranged randomly. Similar to the Uniformity Study, the 48 entries within a test-set also

were arranged in a grid that was four ranges deep and 12 columns wide. To accommodate the auto selection controlled by the computer based on yield advantage to average yield of the checks during mechanical harvest, the three checks were placed always at ranges 1 and 2 of column 1, and the third check at range 1 of column 2. The same check arrangement was also used within check-sets. Row-plots were planted at 40 seeds per plot in 1.5 m row length, spaced 0.8 m between rows, resulting in 27 seeds m<sup>-1</sup>.

### Confirmation Study

The seeds of each of the 135 F<sub>3:5</sub> lines per population (19 populations) harvested in bulk from the 2010 Early Generation Test, were used to plant the Confirmation Study in 2011. The study was conducted at four locations (Huxley, Dayton, Williamsburg, and Winterset, Iowa) at the Monsanto Inc. research sites. The soil at four locations is a sandy clay loam, a clay loam, a silt clay loam, and a clay loam, respectively. The sowing date was May 10, May 9, May 7, and May 10, respectively. The 2565 lines were classified into three maturity groups, defined on the basis of the RM recorded in the 2010 PRYT. The group with RM from 2.0 to 2.4 was composed of 405 lines and identified as F24S. The second RM group was from RM 2.5 to 2.8, composed of 1215 lines, and identified as F26S. The third group was identified as F29S, had RM from 2.9 to 3.1, and was composed of 945 lines. Within each RM group, 45 F<sub>3</sub>-derived lines in F<sub>3:6</sub> were assigned randomly to one test-set, and five checks were added, bringing the total to 50 entries per test-set. Within a test-set, the 50 entries were randomly planted in the field in a grid that was five ranges deep and 10 columns wide. Plots were two-row plots, 3.7 m in length, and 0.8 m between rows, resulting in a sowing population of 30 seeds m<sup>-1</sup>.

## Statistical Analysis

The assumptions of normality and homogeneity of variance in the error term of yield data sets from each of the three experiments were evaluated before any parametric statistical analysis was conducted. In all experiments, the yield data were continuous with near normal distribution based on the QQPLOT in the PROC UNIVARIATE procedure of SAS version 9.3.2 (SAS Institute, Cary, NC).

Box-plots based on the interval quartile range (IQR) method (Tukey, 1977) were implemented for outlier identification. The IQR was calculated on a whole-experiment basis for the Uniformity Study and the Early Generation Test. The IQR for the Confirmation Study was calculated for each individual location. On the basis of the research conducted by Tukey (1977) and Eo et al. (2012), if an observed yield value was less than  $(q1 - 1.5*iqr)$  or larger than  $(q3 + 1.5*iqr)$ , it was treated as an outlier. An outlier was, therefore, treated as a missing value too, otherwise, avoid distorting the statistical inference. For a given data set,  $q1$  is the 25% sample quartile,  $q3$  is the 75% sample quartile, and IQR is the difference between  $q3$  and  $q1$ . The yield data from each of the three experiments were analyzed using the SAS 9.2.3 statistical package (SAS Institute, Cary, NC), and R FIELDS package (Nychka, 2013).

In the Uniformity Study, the yield variation observed within a test-set was assumed to consist of the field spatial patterns and the experimental error. The variation observed across the whole experimental field was assumed to consist of the field spatial pattern, the experimental error, and the genetic variation between the two test lines. The yield data were first fitted to the general linear model with line as fixed effects:

$$y_{ijk} = \mu + v_k + e_{ij} \quad [1]$$

where  $y_{ijk}$  is the observed yield of the line  $k$  at range  $i$  and column  $j$ ;  $\mu$  is the overall mean;  $v_k$  is the line  $k$  effect; and  $e_{ij}$  is a random error having a normal distribution  $e_{ij} \sim$  independent  $N(0, \sigma_e^2)$  with mean = 0 and variance =  $\sigma_e^2$ . Assuming the estimated residual of each plot, from fitted model, as a combination of spatial patterns and experimental error, the **TPSR** was implemented with the residuals to separate the spatial patterns from the experimental error.

The **TPSR** model is a semi-parametric spatial model (Bookstein, 1989). Under the **TPSR** model, the spatial trend effect at each progeny-row plot can be estimated as a function of its neighboring check plots, referred to as knots, by using a localized interpolation function (Robbins et al., 2005). The resulting **TPSR** model corresponded to a mixed linear model for yield of line  $i$  in range  $j$  and column  $k$  given as:

$$y_{kijl} = \mu + \sum_{l=1}^n W_{ijl} \beta_l + v_k + r_i + c_j + e_{kijl} \quad [2]$$

where  $\mu$  is the overall mean;  $\beta_l$  is a fixed effect for the  $l$ -th knot;  $W_{ijl}$  is the weight for the  $l$ -th knot at range  $i$  and column  $j$ ;  $v_k$  is the genetic effect for the  $k$ -th line;  $r_i$  is the random effect for the  $i$ -th range;  $c_j$  is the random effect for the  $j$ -th column; and  $e_{kijl}$  is a random error for the plot at range  $i$  and column  $j$  with  $e_{kijl} \sim$  independent  $N(0, \sigma_e^2)$  with mean = 0 and variance =  $\sigma_e^2$  (Robbins et al., 2012).

The weight  $W_{ijl}$  is defined as:

$$W_{ijl} = \frac{1}{\|R_l - R_i, C_l - C_j\|}, \quad [3]$$

where  $R_l$  and  $C_l$  are the range and column for the  $l$ -th knot, respectively; and  $R_i$  and  $C_j$  are the range and column for the plot on which the spatial effect  $\beta_l$  will be estimated, respectively; the expression  $\|a, b\| = \sqrt{a^2 + b^2}$  is Euclidian distance. The spatial effects were estimated using the R package FIELDS of version 6.9.1 (Nychka, 2013).

The spatial patterns obtained from the Uniformity Study were used to adjust the observed yield on the basis of progeny-row in the Uniformity Study. The observed yield after adjustment of the spatial effects predicted from the **TPSR** was denoted as `yld_adj`. The same notations were applied in the Early Generation Tests as well.

In the Uniformity Study, the analysis of variance for a RCBD was conducted with the following model:

$$y_{ij} = \mu + v_i + b_j + vb_{ij} + e_{ij} \quad [4]$$

where  $y_{ij}$  is either the yield or the `yld_adj` of the line  $i$  in the block  $j$ ;  $\mu$  is the overall mean;  $v_i$  is the line  $i$  effect;  $b_j$  is the block  $j$  effect;  $vb_{ij}$  is the line  $i$  and block  $j$  interaction effect; and  $e_{ij}$  is a random error having a normal distribution  $e_{ij} \sim$  independent  $N(0, \sigma_e^2)$  with mean = 0 and variance =  $\sigma_e^2$ . Data were analyzed using the PROC GLM statement of SAS version 9.3.2 (SAS Institute, Cary, NC). The mean difference between the lines was tested for significance by Fisher's least significant difference (LSD) test (Fisher, 1935).

In the Early Generation Tests, the yield data set, containing only the checks, was first fitted to the same general linear model with checks as fixed effects as described in equation[1], where  $y_{ijk}$  is the yield of the check  $k$  at range  $i$  and column  $j$ ;  $\mu$  is the overall mean;  $v_k$  is the check  $k$  effect; and  $e_{ij}$  is random error with  $e_{ij} \sim$  independent  $N(0, \sigma_e^2)$  with

mean = 0 and variance =  $\sigma_e^2$ . The assumption was that the estimated residual of each check plot consisted of the spatial pattern and the experimental error. The **TPSR** was implemented with the check residuals to predict spatial patterns as a correction factor across test fields on the progeny-row plot base. The correction factor was then used to adjust yield observations.

For the combined analysis of variance across two years in the Early Generation Tests, the following model with RM as a covariate was used:

$$y_{ijk} = \mu + RM + v_i + y_j + vy_{ij} + e_{ijk} \quad [5]$$

where  $y_{ijk}$  is the yield or the `yld_adj` of the  $k$ -th replicate of line  $i$  in year  $j$ ;  $\mu$  is the overall mean;  $v_i$  is the line  $i$  effect;  $y_j$  is the year  $j$  effect;  $vy_{ij}$  is the line  $i$  and year  $j$  interaction effect; and  $e_{ijk}$  is a random error having a normal distribution  $e_{ijk} \sim \text{independent } N(0, \sigma_e^2)$  with mean = 0 and variance =  $\sigma_e^2$ . Data were analyzed using PROC GLM statement of SAS version 9.3.2 (SAS Institute, Cary, NC). `yield` or `yld_adj` were analyzed using the same model as described in equation [5] with the PROC MIXED statement of SAS version 9.3.2 (SAS Institute, Cary, NC), where  $v$  and  $y$  were the random effects, and RM was used as a covariate. The best linear unbiased prediction (BLUP) of each line in each of the 19 bi-parental populations was predicted based on the two-year data. The performance of each population was the mean performance of the lines within the population.

For individual year analysis of the Early Generation Tests, the following model was assumed with RM as a covariate and the effects of line and block as random effects:  $y_{ijk} =$

$$\mu + RM + v_i + b_j + e_{ijk} \quad [6]$$

where  $y_{ijk}$  is the yield or  $yld\_adj$  of the  $k$ -th replicate of line  $i$  in block  $j$ ;  $\mu$  is the overall mean;  $v_i$  is the line  $i$  effect;  $b_j$  is the block  $j$  effect; and  $e_{ijk}$  is random error having a normal distribution  $e_{ijk} \sim$  independent  $N(0, \sigma_e^2)$  with mean = 0 and variance =  $\sigma_e^2$ . The BLUP of each line was estimated by year, and the performance of the population was estimated by year as well using the mean performance of the lines within the population.

In the Confirmation Study, yield data were analyzed across locations using the linear mixed model with RM as a covariate and the effects of lines and locations as random effects:

$$y_{ij} = \mu + RM + v_i + l_j + e_{ij} \quad [7]$$

where  $y_{ij}$  is the yield of line  $i$  in location  $j$ ;  $\mu$  is the overall mean;  $v_i$  is the line  $i$  effect;  $l_j$  is the location  $j$  effect; and  $e_{ij}$  is random error having a normal distribution

$e_{ij} \sim$  independent  $N(0, \sigma_e^2)$  with mean = 0 and variance =  $\sigma_e^2$ . Yield was analyzed using the PROC MIXED statement of SAS version 9.3.2 (SAS Institute, Cary, NC). The BLUP was estimated for each line. As in the combined two-year analysis, the performance of the population was the mean performance of all the lines tested within the bi-parental population.

To assess the efficiency of using the **TPSR** model in correcting yield in the Uniformity Study, the coefficient of variation (CV) and the square root of mean squared deviation (SRMSDs) on the test-set base were compared between the **TPSR** adjustment and non-**TPSR** adjustment.

$$SRMSD = \sqrt{\frac{1}{n-1} \sum_{i=1}^n (y_i - \bar{y})^2} \quad [8]$$

Where  $n$  is the total number of observations for yield in a test-set from the Uniformity Study;  $y_i$  is the yield or the  $yld\_adj$  of the  $i$ -th observation and  $\bar{y}$  is the mean of the observations in the test-set.

To identify the magnitude in the error reduction, the IRE was calculated as:

$$IRE_{TPSR} = \frac{(SRMSE_{non-TPSR} - SRMSE_{TPSR})}{SRMSE_{non-TPSR}} \times 100\%, \quad [9]$$

where  $SRMSE_{non-TPSR}$  is the square root of mean sum of squares for error (SRMSE) from the model without the **TPSR** adjustment, and  $SRMSE_{TPSR}$  is SRMSE from the model with the **TPSR** adjustment, multiplied by 100%, to express the value as percentage.

In the Early Generation Tests,  $IRE_{TPSR}$  was also calculated from  $SRMSE_{non-TPSR}$  and  $SRMSE_{TPSR}$  based on the combined two-year data analysis using the model as described in equation [5] for each yield and  $yld\_adj$ . The Pearson correlation coefficients, between the BLUPs estimated from the Early Generation Tests and those estimated from the Confirmation Study on the basis of the bi-parental population performance, were calculated to measure the efficiency of the **TPSR** model. The rank correlation coefficients, between the BLUPs estimated from the Early Generation Tests and those estimated from the Confirmation Study on the basis of the individual line performance, were calculated to assess effectiveness of selection of the superior lines in the Early Generation Tests after adjustment by the **TPSR**. The Pearson and the rank correlation coefficients were calculated using the PROC CORR statement of SAS version 9.3.2 (SAS Institute, Cary, NC).

## RESULTS AND DISCUSSION

## Uniformity Study

Quantification of Non-genetic variation in the Test-set Groups. The results from the Uniformity Study indicated that the spatial variation in the field could be important and might bias the yield estimates when not taken into consideration (Table 2). The two lines used in the Uniformity Study were pure stable commercial lines with proven performance (Lussenden, personal communication, Redwood Falls, Monsanto Inc. 2012). Yield variations, however, were observed within test-sets suggesting the variation might be non-genetic variation. The CV for yield ranged from 8.6% to 18.4% for Mon1230, and from 10.5% to 21.6% for Mon1431 (Table 2). The average of the CV in the test-sets was 11.6% for Mon1230 and 14.9% for Mon1431.

For individual test-sets, the SRMSDs for yield were as large as 664.0 kg ha<sup>-1</sup>, with a mean of 482.2 kg ha<sup>-1</sup>, and ranged from 339.2 kg ha<sup>-1</sup> to 664.0 kg ha<sup>-1</sup> for MON1230. For MON1431, the SRMSDs were also large, 944.3 kg ha<sup>-1</sup>, with a mean of 661.0 kg ha<sup>-1</sup> and range from 446.9 kg ha<sup>-1</sup> to 944.3 kg ha<sup>-1</sup> (Table 2). The CV and the SRMSD values indicated that the Uniformity Study had a large component of non-genetic variation within the field on a test-set base. The variations observed among CVs and among SRMSDs in the test-sets were also large, suggesting that the magnitudes of the spatial patterns across the whole test field could be also substantial. These observations agreed with previous results (Duncan, 1969; Tovey et al, 1973). Tovey et al. (1973) reported similar results in sugar yields conducted with sugarcane [*Saccharum spp.*], in which plot variations were measured in small test plots. The authors indicated different reasons to explain the presence of non-genetic variation. According to Duncan (1969) and Tovey et al. (1973), potential bias and variation in yield might be caused from competition effects among neighboring test lines.

Variation from non-controlled environmental factors, agricultural practices, and human errors also may contribute to the unexplained variation among plots as mentioned by Wishart and Sanders (1955).

**TPSR Efficiency in Determining Spatial Effects.** The **TPSR** model for field spatial effect adjustment was applied to the yield observations of the Uniformity Study to calculate the CVs and SRMSDs for the yld\_adj within test-sets (Table 2). There were reductions observed for both the CVs and SRMSDs. For individual test-sets, the CV for yld\_adj ranged from 4.1% to 10.0%, with a mean value of 6.7% for Mon1230. For Mon 1431 the range was from 6.8% to 14.6%, with a mean of 9.9% for Mon1431. The mean reduction in CVs from the **TPSR** adjustment was 4.9% (P = 0.002) for MON1230, and 5.0% (P = 0.008) for MON1431; both highly significant. The SRMSDs for yld\_adj ranged from 172.1 kg ha<sup>-1</sup> to 414.8 kg ha<sup>-1</sup> with a mean of 281.1 kg ha<sup>-1</sup> for MON1230. For MON1431, the SRMSDs for yld\_adj ranged from 297.1 kg ha<sup>-1</sup> to 641.7 kg ha<sup>-1</sup> with a mean of 436.5 kg ha<sup>-1</sup>. The mean reduction of SRMSD from the **TPSR** adjustment was 201.1 kg ha<sup>-1</sup> (P = 0.001) for MON1230, and 224.5 kg ha<sup>-1</sup> (P = 0.011) for MON1431, also both significant. The reduction rates in mean CV and mean SRMSD attributable to the **TPSR** adjustment were 37.1% and 37.2%, respectively. The variation still observed in the yld\_adj was assumed to be attributable to the experimental error.

The results of the analysis of variance for the yield and yld\_adj indicated that yld\_adj had a significantly smaller mean square error than yield (Table 3). The IRE<sub>TPSR</sub> was 37.9%, suggesting that the use of the **TPSR** model could improve line comparisons, and

also improve the probability of detecting smaller yield differences between lines because of the increase in estimate precision.

Prior to data adjustment by the **TPSR** model, the yield differences between the lines MON1230 and MON1431 could not be detected, although on average MON1431 yielded 242.0 kg ha<sup>-1</sup> more than MON1230. The yield of each of the two lines was not significantly different, because of a large block mean square and large line x block interaction mean square (Table 3), which indicated spatial heterogeneity across blocks and within-blocks. After the **TPSR** adjustment, the mean squares from block and line x block interaction were minimal (P = 0.977, for block effect of zero, and P = 0.987 for line x block interaction effect of zero). The **TPSR** model adjustment effect was reflected on the comparison of the line means. The F-value for line effect was much higher under the yld\_adj and highly significant (P = 8.9701E-18). After the **TPSR** adjustment, the analysis was performed with yld\_adj, and it could be concluded that MON1431 yielded 242.0 kg ha<sup>-1</sup> more than MON1230. The superiority of the **TPSR** analysis was evident from the greater genotypic discrimination ability in the test. The results from the Uniformity Study suggested that spatial variation might also be present in the PRYT field tests and that using the **TPSR** model could improve the efficiency of the test. Stinger and Cullis (2000), suggested that since genotypic adjustment for the effects of the relative line position in the field was environmental in nature, the use of the **TPSR** model could lead to improved progeny selection and greater breeding efficiency.

Early Generation Test

Quantification of Spatial Patterns Using Checks in Large Scale Early Generation Tests.

In the 2010 and 2012 Early Generation Tests, a trend in yield variation was observed; i.e., the yield of lines increased from the upper to the lower area of the test field (Figure 1). Observed yields could result from the combination of line genetic merit, field spatial pattern, and experimental error (Bernardo, 2003).

The field spatial patterns in the Early Generation Tests were evaluated using a subset of data only containing the three common checks. The analysis of residuals from the linear model with check as a fixed effect revealed that the yield distribution of the checks was affected by their spatial location within the field. In the 2010 and the 2012 field tests, the residuals for the checks placed in the upper area of the field were smaller compared with the residuals of the checks in the lower area.

To quantify the field spatial patterns in yield separately for each year (2010 and 2012), the spatial effects across the test fields were predicted based on the **TPSR** model used with check residuals (Figure. 2). In both years, the spatial effects on yield were larger in the lower half area and smaller in the upper half area. In 2010, the spatial effects ranged from  $-1396.1 \text{ kg ha}^{-1}$  to  $1820.7 \text{ kg ha}^{-1}$ , with a mean of  $620.9 \text{ kg ha}^{-1}$  and a SRMSD of  $599.9 \text{ kg ha}^{-1}$  in the left side of the lower area. In the upper area the range was from  $-2473.8 \text{ kg ha}^{-1}$  to  $804.0 \text{ kg ha}^{-1}$ , with a mean of  $-502.3 \text{ kg ha}^{-1}$  and a SRMSD of  $671.2 \text{ kg ha}^{-1}$ . In 2012, the spatial effects ranged from  $-509.4 \text{ kg ha}^{-1}$  to  $1306.8 \text{ kg ha}^{-1}$ , with a mean of  $407.8 \text{ kg ha}^{-1}$  and a SRMSD of  $465.7 \text{ kg ha}^{-1}$  in the lower area. In the upper area the range was from  $-1339.0 \text{ kg ha}^{-1}$  to  $307.8 \text{ kg ha}^{-1}$ , with a mean of  $-467.7 \text{ kg ha}^{-1}$  and a SRMSD of  $337.0 \text{ kg ha}^{-1}$ . The patterns in estimates of spatial effects of the **TPSR** model (Figure 2) were similar to the field spatial trends in yield (Figure 1), and in check residuals (Figure 2). The observations

indicated that field spatial trends attributable to heterogeneous environments could be identified and removed by the use of the **TPSR** model. In 2012, the severe drought in the Midwest of North America during the pod-filling season (Table 1) might have also affected the field spatial trend that was observed for the year.

**TPSR Efficiency in Determining Spatial Effects.** Data with and without spatial adjustment (yld\_adj and yield) in 2010 and in 2012 Early Generation Tests were fitted to the general linear model for the analysis of variance using the model in equation [5]. The mean square for error from the analysis of yld\_adj was smaller than that of the yield (Table 4), and the  $IRE_{\text{TPSR}}$  was 40.4%. The value was similar to the IRE observed in the Uniformity Study (Table 3). The results of the Early Generation Tests indicated that the **TPSR** model was effective in capturing the spatial patterns in a large field, similarly as it was effective at a smaller scale in the Uniformity Study. The reduction in the error variance also indicated that the **TPSR** model could improve line comparisons, allowing detection of small differences between lines by reducing the LSD. Robbins et al. (2012) describing work conducted with un-replicated maize trials also concluded that the **TPSR** model could effectively reduce the error variance by accounting for spatial variations.

#### Confirmation Study

**Selection Efficiency Improvement Using the **TPSR** Model.** In a soybean breeding program, each row of the un-replicated PYRT test would be bulk-harvested and the superior high-yielding genotypes would be selected for planting larger plots at multiple locations during

the following year. In the study reported here, all genotypes tested in the Early Generation Test were planted in 2011 at each of four locations in Iowa, in replicated field tests with two-row plots for every genotype. All lines from the Early Generation Tests were planted in the Confirmation study to be able to compare the individual line yield performance in each of the tests planted.

The first yield performance comparison was made by calculating the Pearson correlation coefficients between the mean performance of the individual bi-parental populations in the individual years of the Early Generation Tests, and combined across years for the observed yield and the yld\_adj of the same lines evaluated in the Confirmation Study (Table 5). In the data-set from the year 2010, the correlation coefficient of the estimated yield increased from 0.43 (P = 0.065) to 0.54 (P = 0.018) after the use of the **TPSR** adjustment. This represented a positive change of 0.11 points in the correlation because of the use of the **TPSR** model. In the year 2012, similar results were observed. The correlation increased from 0.24 (P = 0.324) to 0.57 (P = 0.011), an improvement of 0.33 points in the correlation value because of the use of the **TPSR** adjustment.

Additional improvement in the size of the correlation value was also observed when the combined years of the data-set of the Early Generation Tests were used (Table 5). The correlation coefficient with the **TPSR** adjustment was 0.61 (P = 0.006) and without the **TPSR** adjustment was 0.40 (P = 0.094). The improvement in the correlation value attributable to the **TPSR** adjustment was of 0.21 points. Similar results were also reported by Edmé et al. (2007) in the sugarcane work.

To assess if the **TPSR** model adjustment used in the Early Generation Tests would have an effect on identifying individual line performance of yield, 1% selection intensity was applied to the lines tested in the Early Generation Tests, both on the basis of actual yield and the yld\_adj. The lines were ranked according to yield as evaluated in the Early Generation Test from 1 to 26 (data not shown). The ranking of the selected lines was then compared with the rank of the same lines evaluated in the Confirmation Study. The selection was practiced across populations by individual years, 2010 and 2012, and also for the yield averaged across the two years. The rank correlations were calculated between the actual yield and adjusted yield for each of the selection environments and the yield selection criteria. None of the rank correlations among the studies were significant indicating that the line ranking at each test was different. When the selections were made using the 2010 yield data of the Early Generation Test, only four of the selected lines were among the superior 26 genotypes in the Confirmation Study. None of the lines selected in the Early Generation test in 2012 could be found among the 26 best lines of the Confirmation Study. When selections were based on the combined yield data of 2010 and 2012, none of the selected lines was among the best 1% of the lines identified in the Confirmation Study. These observations suggested that although the **TPSR** adjustment was effective in controlling spatial variation, the adjustment might still not be enough to reflect an improvement in the prediction of line performance of the Early Generation tests on the basis of the progeny-row.

## CONCLUSIONS

This research was conducted to determine if the **TPSR** model could effectively remove variation attributable to non-genetic spatial trends from the early generation tests. The underlying assumption was that the error term would be reduced by the adjustment, thus allowing a more precise comparison among yields of lines that were genetically diverse. The results of two of the studies, namely the Uniformity Study and the Early Generation Test, indicated that the **TPSR** model was an effective tool for removing non-genetic spatial variation in soybean PRYT field tests. The use of the **TPSR** adjustment in both studies effectively reduced the coefficient of variation and the square root of the mean square deviation in each test. Additionally, the efficiency in hypothesis testing was increased as determined by the IRE values.

The third study, the Confirmation Study, was designed to test if the removal of the non-genetic variation in field tests done by adjusting yield using the **TPSR** model would be reflected in an increase in the efficiency of line selection. If the lines selected after use of the **TPSR** adjustment for yield, would have also been among the superior lines in the Confirmation Study, this would have been an indication of the effectiveness of the **TPSR** adjustment to increase the precision of the yield estimation at the early stages of breeding. This was not observed. The results suggested that use of the **TPSR** model as performed in this research would not increase selection precision of lines at the early generations. The **TPSR** model adjustment did increase the Pearson correlation coefficients among the yield results from the Early Generation yield tests and the yield results from the Confirmation Study on the basis of bi-parental population performance. The positive change in the correlation values, however, was not reflected in the individual line performance *per se*. When a selection intensity of 1% was used to identify the highest yielding lines in the Early

Generation Tests, and their performance in the Confirmation Study was determined, the number of superior lines in both studies was equal to six or less regardless of the **TPSR** adjustment. These results indicated that under the conditions of this study, there was no improvement in the selection efficiency after using the **TPSR** adjustment.

A factor to be considered in interpreting the results, however, is the number of checks used to model spatial variation. In this experiment, three checks replicated throughout the field were used. The total number of lines evaluated in a test-set of the Early Generation Test was 45, and the total number of checks was three. The ratio of number of lines to checks was 15 to 1. It might be that the number of checks might have been too small compared with the number of lines evaluated. This ratio, in turn, might have caused an underestimation of the field spatial variation, which could have contributed to the lack of prediction of the Early Generation Tests compared with line performance evaluated in the Confirmation Study. Research is in progress to determine if an increase in the number of checks in the Early Generation Tests could increase the selection efficiency at early stages of soybean breeding.

#### ACKNOWLEDGEMENTS

This research was supported by Monsanto Inc.. The authors thank all the people who participated in the execution of the field and lab studies. Special appreciations are due to Dr. Daniel Nordman for the statistical review of the paper, and special thanks are due to Dr. Christiana Wiebbecke, Dr. Roger Lussenden, and Dr. Andrew Nickell of Monsanto Inc. who greatly contributed to this research by suggesting lines, conducting the field tests, and offering suggestions on the manuscript.

## REFERENCES

Bazen, A. M., and S. H. Gerez. 2002. Thin-plate spline model of elastic deformations in fingerprints. In: *Proceedings of the 3rd IEEE Benelux Signal Processing Symposium (SPS-2002)*, edited by M. Moonen, 1-4. Leuven, Belgium. 21-22 Mar. 2002.

Becker, H. H. 1995. On the importance of soil homogeneity when evaluating filed trials. *J. Agron. Crop Sci.* 174:33-40.

Bernardo, R. 2003. On the effectiveness of early generation selection in self-pollinated crops. *Crop Sci.* 43:1558-1560.

Bookstein, F. L. 1989. Principle warps: Thin-plate splines and the decomposition of deformations. *IEEE Trans. Pattern Anal. Mach. Intell.* 11:567-586. doi:10.1109/34.24792.

Brownie, C., D. T. Bowman, and J. W. Burton. 1993. Estimating spatial variation in analysis of data from field trials: a comparison of methods. *Agron. J.* 85:1244-1253.

Duncan, W. G. 1969. Cultural manipulation for higher yields. In: *Physiological Aspects of Crop Yield: Proceedings of a symposium*, edited by J. D. Eastin, F. A. Haskins, C. Y. Sullivan, C. H. M. Van Bavel, and R. C. Dinauer, 327-342. ASA, Madison, WI.

Edmé, S. J., P. Y. P. Tai, and J. D. Miller. 2007. Relative efficiency of spatial analyses for non-replicated early-stage sugarcane field experiments. *J. Amer. Soc. Sugar Cane Tech.* 27:89-104.

Eo, S. H., D. Pak, J. Choi, and H. J. Cho. 2012. Outlier detection using projection quartile regression for mass spectrometry data with low replication. *BMC Res. Notes* 5:236. DOI:10.1186/1756-0500.

Federer, W. T. 1956. Augmented (or hoonuiaku) designs. *Hawaii Plant. Records* 55:191-208.

Federer, W. T. 1961. Augmented designs with one-way alimentation of heterogeneity. *Biometrics* 17: 447-473.

Fehr, W. R. 1993. Development of self-pollinated cultivars. In: *Principles of cultivar development*, edited by W. R. Fehr, 388-400. Macmillan Publishing Company. New York

Fehr, W. R. 1993. Early-generation testing. In: *Principles of cultivar development*, edited by W. R. Fehr, 339-346. Macmillan Publishing Company. New York.

Fehr, W. R., C. E. Caviness, D. T. Burmood, and J. S. Pennington. 1971. Stage of development descriptions for soybeans, *Glycine max* (L.) Merrill. *Crop Sci.* 11:929-931.

Fisher, R. A. 1935. *The Design of Experiments* (9th ed.). London: MacMillan & Co. Ltd.. ISBN 0-02-844690-9.

Gilmour, A. R., B. R. Cullis, and A. P. Verbyla. 1997. Accounting for natural and extraneous variation in the analysis of field experiments. *J. Agric. Biol. Environ. Stat.* 2:269-293.

Hegstad, J. M., G. Bollero, and C. D. Nickell. 1999. Potential of using plant row yield trials to predict soybean yield. *Crop Sci.* 39:1671-1675.

Lin, C. S., and M. R. Binns, H. D. Voldeng, and R. Guillemette. 1993. Performance of randomized block designs in field experiments. *Agron. J.* 85:168-171.

Magnussen, S. 1993. Bias in genetic variation estimates due to spatial autocorrelation. *Theor. Appl. Genet.* 86:349-355.

Nychka, D. 2013. Package 'fields' of R, version 6.9.1. Tools for spatial data. URL <http://www.image.ucar.edu/Software/Fields>.

Orf, J. H., B. W. Diers, and H. R. Boerma. 2004. Genetic improvement: conventional and molecular-based strategies. In: *Soybeans: Improvement, production, and uses*, 3rd ed. Agron. Monogr. 16, edited by H. R. Boerma and J. E. Specht, 417-450. Madison, WI.

Robbins, K. R., J. E. Backlund, and K. D. Schnelle. 2012. Spatial corrections of unreplicated trials using a two-dimensional spline. *Crop Sci.* 52:1138-1144. doi: 10.2135/cropsci2011.08.0417.

Robbins, K. R., I. Misztal, and J. K. Bertrand. 2005. Joint longitudinal modeling of age of dam and age of animal for growth traits in beef cattle. *J. Anim. Sci.* 83:2736-2742.

Rosielle, A. 1980. Comparison of lattice designs, check plots, and moving means in wheat breeding trials. *Euphytica* 29:129-133.

SAS Institute. 2012. SAS 9.3.2 user's guide. Carry, NC: SAS Institute, Inc. <http://www.sas.com/publishing>.

Scott, R. A. and G. A. Milliken. 1993. A SAS program for analyzing augmented randomized complete-block designs. *Crop Sci.* 33:865-867.

Stringer, J. K., and B. R. Cullis. 2002. Application of spatial analysis techniques to adjust for fertility trends and identify interplot competition in early stage sugarcane selection trials. *Aust. J. Agric. Res.* 53:911-918.

Stroup, W. W., P. S. Baezinger, and D. K. Mullitze. 1994. Removing spatial variation from wheat yield trials: a comparison of methods. *Crop Sci.* 34:62-66.

Tovey, D. A., K. T. Glasziou, R. H. Farquhar, and T. A. Bull. 1973. Variability in radiation received by small plots of sugarcane due to differences in canopy heights. *Crop Sci.* 13:240-242.

Tukey, J. W. 1977. Easy summaries - - numerical and graphical. In: *Exploratory data analysis*, edited by J. W. Tukey, 27-58. Addison-Wesely. Menlo Park, CA.

Wishart, J., and H. G. Sanders. 1955. Principles and practice of field experimentation. Tech. Comm. No. 18, Commonwealth Bureau of Plant Breeding and Genetics, Cambridge, U.K.



## Tables

**Table 1** Weather information for the research location of Monsanto Inc. near Huxley, IA that was used in the conduct of the 2010 and 2012 Early Generation Tests.

Weather	Year	April	May	June	July	August	Sept.
Rainfall (mm)	2010	107.4	96.1	304.4	134.9	313.4	211.1
	2012	107.2	57.5	71.9	56.6	67.3	54.6
	1997 to 2011†	97.3	141.1	137.3	110.9	118.7	80.7
Temperature‡ (°C)	2010	12.9	15.7	22.0	24.0	24.0	17.7
	2012	11.5	19.2	22.4	26.7	21.8	17.1
	1997 to 2011†	9.9	15.8	20.9	23.6	22.2	17.7

† Averages of rainfalls and daily temperatures from 1997 to 2011

‡ Daily temperature was the mean of daily maximum and minimum temperatures

Note: The soil at the research location near Huxley, IA is sandy clay loam. Sowing date was May 5 in 2010; sowing date was May 10 in 2012.

**Table 2** Variation within test-set in the yield observations recorded in the Uniformity Study, without the **TPSR** adjustment (yield), and with the **TPSR** adjustment (yld\_adj).

Test-set Code	yield			yld_adj		
	Mean (kg ha <sup>-1</sup> )	SRMSD <sup>†</sup> (kg ha <sup>-1</sup> )	CV <sup>†</sup> (%)	Mean (kg ha <sup>-1</sup> )	SRMSD <sup>†</sup> (kg ha <sup>-1</sup> )	CV <sup>†</sup> (%)
<u>MON1230‡</u>						
1	3601.8	664.0	18.4	4169.6	414.8	9.9
2	4377.8	380.5	8.7	4184.4	254.8	6.1
3	4352.4	444.9	10.2	4161.6	236.5	5.7
4	4442.4	584.6	13.2	4168.6	354.0	8.5
5	3646.1	352.3	9.7	4177.8	187.6	4.5
6	3943.2	339.2	8.6	4184.4	172.1	4.1
7	4319.1	466.3	10.8	4162.0	305.6	7.3
8	4732.2	625.7	13.2	4190.1	323.5	7.7
Grand mean	4176.9	482.2	11.6	4174.8	281.1	6.7
<u>MON1431‡</u>						
1	3997.2	466.7	11.7	4428.6	323.1	7.3
2	4254.9	589.5	13.9	4419.2	390.2	8.8
3	4599.4	660.1	14.4	4426.2	474.6	10.7
4	4731.9	811.6	17.2	4417.2	473.1	10.7
5	4182.9	904.3	21.6	4426.7	570.5	12.9
6	4393.8	464.4	10.6	4404.3	321.6	7.3
7	4256.6	446.9	10.5	4388.8	297.1	6.8
8	4888.9	944.3	19.3	4411.0	641.7	14.5
Grand mean	4413.2	661.0	14.9	4415.3	436.5	9.9

† CV = coefficient of variation; SRMSD = square root of the mean square deviation.

‡ MON1230 and MON1431: soybean commercial cultivars released by Monsanto Inc. in 2010 and 2012, respectively.

**Table 3** Analysis of variance of the Uniformity Study based on yield observations without the **TPSR** adjustment (yield) and with the **TPSR** adjustment (yld\_adj).

Source	DF	Mean square (kg ha <sup>-1</sup> )	F-value	P-value†
<u>yield</u>				
Line (v)	1	10724151.0***	28.91	1.01E-07
Block (b)	3	23375835.7***	63.01	2.18E-36
v x b	3	1263073.0*	3.40	0.017
Error	760	370992.2		
<u>yld_adj</u>				
Line (v)	1	11100877.7***	77.47	8.97E-18
Block (b)	3	9673.7	0.07	0.977
v x b	3	6631.0	0.05	0.987
Error	760	143283.9		
IRE <sub>TPSR</sub> ‡ = 37.9%				

\*, \*\*, \*\*\* Significant at P = 0.05, P = 0.01, and P = 0.001, respectively.

† 1.01E-07 = 1.01 x 10<sup>-7</sup>; 2.18E-36 = 2.18 x 10<sup>-36</sup>; 8.97E-18 = 8.97 x 10<sup>-18</sup>.

‡ IRE<sub>TPSR</sub> = ((SRMSE<sub>yield</sub> - SRMSE<sub>yld\_adj</sub>)/SRMSE<sub>yield</sub>) x 100%.

**Table 4** Analysis of variance of the Early Generation Test combined over years with relative maturity (RM) as a covariate based on yield observations without the **TPSR** adjustment (yield) and with the **TPSR** adjustment (yld\_adj).

Source	DF	Mean square (kg ha <sup>-1</sup> )	F-value	P-value
<u>yield</u>				
RM	1	485050.0	0.87	0.350
Year (y)	1	582775396.0***	2745.43	<.0001
Line (v)	2567	1266547.0***	7.42	<.0001
y x v	2567	544476.0***	3.85	<.0001
Error	1199	566068.0		
<u>yld_adj</u>				
RM	1	176049.0	0.86	0.355
Year (y)	1	552382770.0***	1029.52	<.0001
Line (v)	2567	1492321.0***	2.24	<.0001
y x v	2567	775079.0	0.96	0.786
Error	1199	201201.0		
IRE <sub>TPSR</sub> <sup>†</sup> = 40.4%				

\*, \*\*, \*\*\* Significant at P = 0.05, P = 0.01, and P = 0.001, respectively.

† IRE<sub>TPSR</sub> = ((SRMSE<sub>yield</sub> - SRMSE<sub>yld\_adj</sub>)/SRMSE<sub>yield</sub>) × 100%.

**Table 5** Analysis on the basis of bi-parental population performance for Pearson correlation coefficients (top) and P-values (bottom) for significance between the Confirmation Study, and the Early Generation Tests for two years and by year with yield observations without the **TPSR** adjustment (yield) and with the **TPSR** adjustment (yld\_adj), respectively.

Population performance	BLUP1012 _TPSR <sup>†</sup>	BLUP1012 _un <sup>‡</sup>	BLUP10 _TPSR <sup>§</sup>	BLUP10 _un <sup>¶</sup>	BLUP12 _TPSR <sup>#</sup>	BLUP12 _un <sup>††</sup>
BLUP_true <sup>‡</sup>	0.61 0.006**	0.40 0.094 <sup>ns</sup>	0.54 0.018*	0.43 0.065 <sup>ns</sup>	0.57 0.011*	0.24 0.324 <sup>ns</sup>

Note: BLUP stands for the best linear unbiased prediction for individual populations calculated by the mean of the individual line BLUPs within population.

\*, \*\* Significant at P = 0.05 and P = 0.01, respectively; ns F test non significant at P = 0.05.

† Population BLUPs were estimated based on yld\_adj from two-year Early Generation Tests.

‡ Population BLUPs were estimated based on yield from two-year Early Generation Tests.

§ Population BLUPs were estimated based on yld\_adj from the Early Generation Test in 2010.

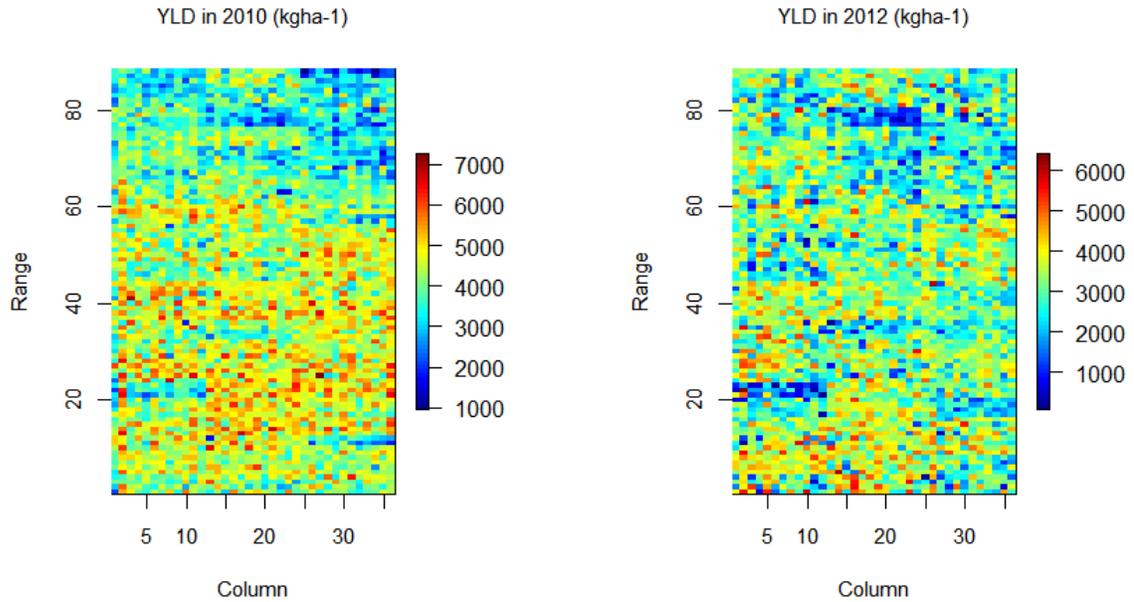
¶ Population BLUPs were estimated based on yield from the Early Generation Test in 2010.

# Population BLUPs were estimated based on yld\_adj from the Early Generation Test in 2012.

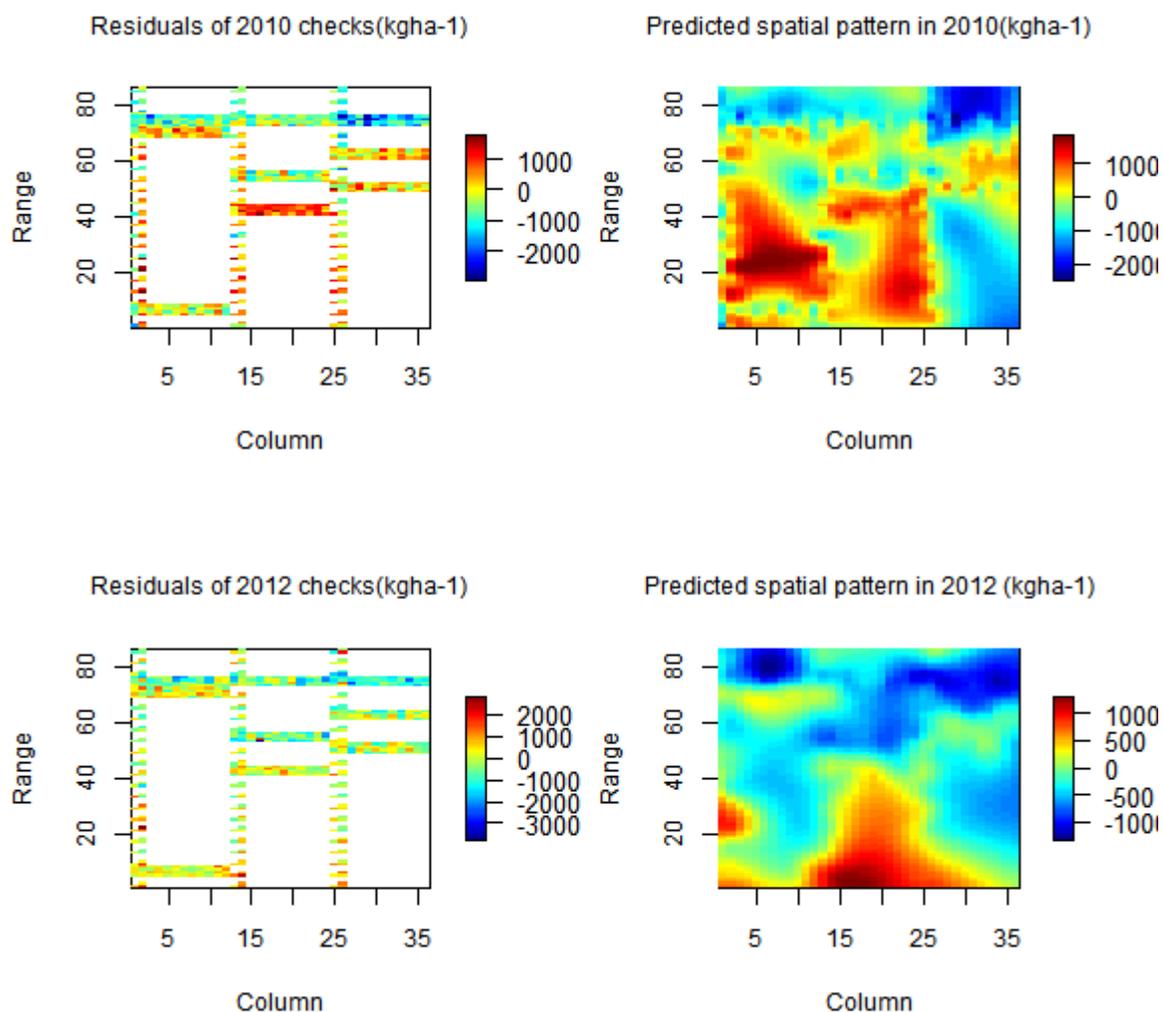
†† Population BLUPs were estimated based on yield from the Early Generation Test in 2012.

‡ Population BLUPs were estimated based yield from the Confirmation Study.

## Figures



**Fig. 1** Heat map for yield observations without the **TPSR** model adjustment (yield) of the Early Generation Tests in 2010 (left) and 2012 (right), respectively.



**Fig. 2** Heat map of check residuals estimated from the linear model with check as fixed effect based on the check plot yield observations from the Early Generation Test in 2010 (top/left), and the heat map of the predicted spatial patterns using the **TPSR** model based on the check's residuals from 2010 Early Generation Test (top/right); the heat map of check residuals estimated from the linear model with check as fixed effect based on the check plot yield observations from the Early Generation Test in 2012 (bottom/left), and

the heat map of the predicted spatial patterns using the **TPSR** model based on the check's residuals from 2012 Early Generation Test (bottom/right).