**Crop-Weed Relationship Studies in Additive Design: Selecting the Top Model**

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**ABSTRACT**: Crop-weed competition is extensively studied in weed research. The additive design, where weed varied, and the crop is kept constant, is the most used design for competition studies. However, most of the competition research are conducted by non-statistician, which could make the wrong conclusion based on lack of data analysis knowledge. The objective of this study is to show the top model for describing the crop-weed relationship in additive design. We evaluated three models routinely used to interpret competition studies, including polynomial quadratic, logistic, and rectangular hyperbola. Based on statistical criteria and in meaningful parameters, we demonstrated the rectangular hyperbola is the top model to describe crop-weed competition studies in additive design. In addition, we showed that at low densities *C. benghalensis* is more competitive than *Richardia brasiliensis* in corn, but both compete similarly at higher densities. Therefore, weed management will increase in areas of *C. benghalensis*. We proposed the use of the rectangular hyperbola as a standardized model for crop-weed competition in additive design.

**Keywords**: AIC, competition, corn, rectangular hyperbola, *C. benghalensis*, *R. brasiliensis*

**Introduction**

The commonly used criteria for model selection of linear and nonlinear regression models is the equation with higher R-squared (R2). However, R2 tests the goodness of fit and is statistically inadequate for nonlinear model selection (Archontoulis and Miguez 2015; Zuur et al. 2007). There are several appropriate statistical criteria for selecting the best nonlinear model for datasets: Alkaike’s information criterion (AIC), Bayesian information criterion (BIC), F-test, and likelihood ratio (Anderson 2008; Lewis et al. 2011; Zucchini 2000). For non-nested (different structure) models, AIC and BIC are indicated for model selection. However, for the nested model (models is a special case of another), F-test or any of the model selection techniques described above are applicable. From the practical standpoint, the model should be selected in a balance between statistics and biological relevance, which will scientists answer their research questions (Archontoulis and Miguez 2015). Therefore, in crop-weed relationship studies in additive design, the model that provides a good fit and meaningful biological parameters are considered a strong candidate model.

Studies have described the relationship function of crop yield loss in response to weed density using additive design. In the weed science literature, despite the revisions and recommendation in using the rectangular hyperbola (Ritz et al. 2015; Swanton et al. 2015), there is no common sense of which model is the most appropriate (Silva et al. 2015; Strieder et al. 2007; Trezzi et al. 2015; Voll et al. 2002). Four major regressions curves are commonly used: linear (Figure 1A), polynomial quadratic (Figure 1B), sigmoid (Figure 1C), and rectangular hyperbola (Figure 1D).

The advances in statistical software will result in the use of more standardized and complex methods nonlinear regression analysis that could be carried out by non-statistician (Knezevic et al. 2007). Here, we compare three non-nested candidate models (polynomial quadratic, logistic, and a rectangular hyperbola) for describing the crop-weed relationship. We used data from an experiment of corn (*Zea mays* L.) in competition with two weed species, *Richardia brasiliensis,* and *Commelina benghalensis*. First, we want to show the top model to describe the crop-weed relationship for each weed species. Second, we test the hypothesis if *C. benghalensis* and *R. brasiliensis* compete similarly with corn.

**Material and Methods**

**Plant Material**. On March 2011, inflorescences of *R. brasiliensis* were harvest on roadsides near Diamantina, Minas Gerais, Brazil. Inflorescences of *R. brasiliensis* were dried at room temperature (25 °C), cleaned, and stored at 5 °C until the onset of the experiment. Ten days before the experiment began (September 2011), stolon (vegetative propagules) of *C. benghalensis* were collected in wetlands, near Diamantina, MG. Seeds of *R. brasiliensis* and stolon of *C. benghalensis* were seeded and transplanted to separate trays (1210 cm3) filled with red latosol (pH 6.1 and 1% organic matter). A single seed of glyphosate-resistant (GR) corn (AG8088) were sown in 8 dm3 plastic pots filled with the identical soil described above. The soil was fertilized following the local recommendations, and N was applied at 15 and 30 DAE (days after corn emergence) at a rate of 55 mg dm-3 of ammonium sulfate. Greenhouse conditions were 26/19 °C day/night, and pots were watered daily.

**Experimental procedures**. The experiment was conducted under greenhouse conditions at the Federal University of Jequitinhonha and Mucuri, MG. In this study, the additive design for competition studies was used, where weed densities vary, and corn density is kept constant (Swanton et al. 2015). The treatment design was a factorial with two weed species, *R. brasiliensis* and *C. benghalensis*, and five weed densities (0, 1, 2, 3, and 4 plants pot-1), in a completely randomized design with four replications.

Corn biomass was harvested at 60 DAE from each experimental unit. Corn roots were cleaned by emerging into the water and gently with a water pressure hose to remove the soil. Shoot and root biomass were oven-dried at 65 °C until reaching constant weight, and dry weight recorded. The corn biomass (g) data (shoot + root) were converted into yield loss (%) compared with the corn control treatment (no weeds):

eq. (1)

where *Μ* is the mean biomass (g) of the untreated replicates, and is the biomass (g) of an individual treated experimental unit.

**Statistical Analysis**

Three models were fitted to yield loss (%) in response to weed density (plants pot-1):

*Rectangular hyperbola model* proposed by (Cousens 1985):

eq. (2)

where *I* represent YL (yield loss) per unit weed density as *D* (density) approaches 0, and *A* represents YL as D approaches ∞ (or maximum expected yield loss). The rectangular hyperbola model was fitted using the *nls* function of R version 3.3.1 (R Foundation for Statistical Computing, Vienna, Austria).

*Logistic model* (four parameter log-logistic curve):

eq. (3)

where *c* is the (lower limit or YL at low weed density), *d* is the asymptote (upper limit or YL at high weed density), and *e* represents the weed density (weeds pot-2) that cause 50% yield loss (inflection point). The parameter *b* is the relative slope around the parameter *e*, and *D* is the number of weeds pot-1. Parameters for the sigmoidal model (four-parameter logistic) were estimated using the *drm* function of drc package (Ritz and Streibig 2005) in R software.

*Polynomial quadratic model* (second order):

eq. (4)

where *α* is the intercept in the y-axis (maximum YL in the absence of weed), *a* represents the slope of the model. The parameter *b* is the quadratic term of the model, and *D* is the number of weeds plot-1. The parameters for the polynomial quadratic equation were estimated using the *lm* function of R software.

**Best model (non-nested) selection.** AIC was used for model selection, and it is indicated for non-nested model selection. The AIC was calculated according to (Anderson 2008).

eq. (5)

where is the likelihood function and is the number of estimated parameters in the model. The top model results in the lowest AIC value. AIC values for each model were estimated using the *AIC* command in R software.

**Weed competition with corn model (nested) selection.** Assuming that rectangular hyperbola is the top model. The competitiveness of *R. brasiliensis* and *C. benghalensis* on corn yield loss (%) is accessed through the variance-ratio or F-test performed using equation [2] (Lindquist et al. 1996). This statistical procedure evaluates the difference of residual sum squares (RSS) of two considered nested models (Werle et al. 2014).

According to this procedure, the following equation represents the F-test:

eq. (6)

Where RSSFULL RSSRED represents the minimized residual sum squares of the parameters estimated by the full model (step 1) and reduced model (step 2, or 3, or 4), respectively; dfFULL and dfRED represents the degrees of freedom of full model and reduced models, respectively. Large F-value shows the two models are likley different, but small F-value shows the two models are likely similar. In addition, if P-value >0.05, there is no difference between models (the null hypothesis is accepted); therefore a reduced should be used (no difference of parameters *I* and *A* among weed species). However, if P-value <0.05, there is the difference between models (full vs. reduced), the null hypothesis is rejected and that a full model should be used (parameter *I* and *A* for each weed species). The F-test principle for nonlinear regression analysis was calculated for each model using *nls* *ANOVA* command in R software (Ritz and Streibig 2008).

Four major steps need to be completed to compare the parameters using this method (see Supplemental file):

First, fit equation [2] to each dataset individually (*R. brasiliensis* and *C. benghalensis*), this represents the full model, where four parameters (*I* and *A* for each weed species) will be estimated.

Second, pool the entire dataset (*R. brasiliensis* and *C. benghalensis*) and fit equation [2] to the pooled data. This represents the reduced model, where two parameters (*I* and *A* for both weed species combined) are being estimated. This step tests the hypothesis that *I* and *A* do not vary between species, which means that both species compete similarly in corn. If the hypothesis is accepted, stop here. Otherwise, there is two more hypothesis to be tested.

Third, fit equation [2] for each species separately, setting the parameter *I* similar to both data set, but different A. This is a reduced model and thee parameters will be estimated. This step tests the second hypothesis, that weed species compete similarly at low densities (*I*), but different at higher densities (*A*).

Fourth, fit equation [2] for each data set separately, setting the parameter *A* similar to both data set, but different *I*. This is a reduced model, and three parameters will be estimated. This step tests the third hypothesis, that weed species compete similarly at higher densities (*A*), but different at low densities (*I*). Additional AIC was also performed for the nested model selection.

**Goodness-of-fit of the models.** Root mean squared error (RMSE), model efficiency (ME), and R2 were calculated and used to test the goodness-of-fit of non-nested and nested models and (Archontoulis and Miguez 2015; Mayer and Butler 1993; Roman et al. 2000).

eq. [7]

eq. [8]

*R2* eq. [9]

where RSS and RSTare the sums of squares for the residual and total, respectively; is the number of data points, is the number of model parameters. is the observed, is the predicted, and is the mean observed value. In this present study, RSME was an average of *R. brasiliensis,* and *C. benghalensis* fitted curves. The ME values range from -∞ and 1, with values closer to 1 indicating better predictions (Werle et al. 2014). R2 values range from 0 to 1, and it was used only for the polynomial quadratic model, which is a form of linear regression.

**Results**

**Selecting the top (non-nested) model.** The retangular hyperbola model resulted in the lowest AIC (268.3), followed by a sigmoid model (271.3) and a polynomial quadratic model (281.8) (Table 1). The model selected by AIC minimizes the information lost when approximating full reality, which means that it selects the approximating model closest to the unknown reality (Anderson, 2010). Therefore, the Cousens model was the top model to describe the corn yield loss (%) in competition with both *R. brasiliensis* and *C. benghalensis* (Figure 2). In addition to AIC, the Cousens model goodness of fit was RSME of was low (6.1), smaller RMSE indicates the observed values are closer to predicted values. Moreover, the ME is 0.96 and 0.95 for *R. brasiliensis* and *C. benghalensis*, respectively. Four parameters were estimated using the rectangular hyperbola model, *I* and *A* for *R. brasiliensis* and *C. benghalensis*, respectively (Table 2). However, there are two issues with the parameter estimates, *A* of *R. brasiliensis* and parameter *I* and *A* of *C. benghalensis* were estimated over 100% (Table 2). The parameter *I* of *C. benghalensis* curve had a steep inclination, which is likely that at this pot size used, *C. benghalensis* is very competitive in corn. Therefore, bigger pots and lower *C. benghalensis* densities are necessary. Moreover, parameter *A* for both species was over 100%, which is an indication that higher densities (> 4 plant pot-1) should have been included in the study to reach a constant final yield (Weiner and Freckleton 2010). The asymptote (upper limit) was not completely reached using four plants pot-1 in our experiment; therefore, parameter *A* was overestimated (Table 2). Despite this common issue, there is no lack of fit of parameters *I* and *A* estimated for *R. brasiliensis* and *C. benghalensis* (P<0.05).

The logistic model (a sigmoid curve) provided the second best fit for the data according to AIC (Table 1). Moreover, this model provided the lowest RSME (5.7), but the lowest ME for *R. brasiliensis* (0.83) and *C. benghalensis* (0.89). This model contains three parameters that have a biological interpretation (Table 3). The maximum (*d*) corn yield loss caused by the competition of *R. brasiliensis* and *C. benghalensis* was 88.1% and 91%, respectively. The parameter *e* is the weed density (plants pot-1) that caused 50% in yield loss (%). The parameter *e* was 1.3 and 0.76 plants pot-1 of *R. brasiliensis* and *C. benghalensis*, respectively. The common issue of fitting a logistic equation for crop-weed competition study is the systematic lower limit (*c*) lack of fit (Table 3). The P-value for parameter *c* is not significant (P>0.05); therefore this parameter is similar to zero. Also, the standard error is bigger than *c* estimated values, which means that the sample mean is not close to the actual population mean.

The polynomial quadratic model provided the highest AIC (281.8), which means that this model was the least appropriate for fitting our data set (Table 1). Nonetheless, by ME and R2, this model resulted in a good fit for the data. However, ME and R2 are not adequate for model selection. Even so, a quadratic model has a biologically implausible turning point for crop-weed studies (Figure 1B). The parameters estimated for the polynomial quadratic model have no biological meaning in agronomic terms, and there is a lack of fit for the intercept of *R. brasiliensis* and *C. benghalensis* (P>0.05) (Table 4).

**Weed competition with corn model (nested) selection.** The rectangular hyperbola proposed by Cousens (1985) was the top model for fit our data set (Table 1). Thus, we conducted the F-test to evaluate if *R. brasiliensis* and *C. benghalensis* competed similarly with corn. The F-test of the rectangular hyperbola model for corn yield loss (%) indicates that a reduced model (P=0.4864) with different parameter *I* (competition at low weed densities) and similar parameter *A* (competition at higher densities) was the best model to describe corn competition to *R. brasiliensis* and *C. benghalensis* (Table 5). According to the parameter estimates in the rectangular hyperbola, at low densities (*I*), corn yield loss was 56.4 and 159.8% in competition to *R. brasiliensis* and *C. benghalensis*, respectively. However, at higher densities, *R. brasiliensis* and *C. benghalensis* compete similarly, and corn yield loss was 109.5% (Figure 5 and Table 6).

AIC corroborates to F-test (Table 5). The model selected by the F-test (different I, but similar A) resulted in the lowest AIC of 266.8. The best fit was also demonstrated as this model provides the lowest RSME and the highest ME for *R. brasiliensis* and *C. benghalensis*.

**Discussion**

**Selecting the top (non-nested) model.** According to AIC criteria (Table 1), when compared to polynomial quadratic and logistic models, the rectangular hyperbola model was the best model to describe corn yield loss (%) in response to *R. brasiliensis* and *C. benghalensis* (Figure 2) competition. This model was also the best for fitting corn leaf area, height, and stem diameter reduction (data not shown). The rectangular hyperbola model provides two biological parameters that are meaningful in agronomic terms to answer research questions in competition studies, the parameter *I* and *A* (Table 2).

To understand the biology of crop-weed relationship modeling, one needs to comprehend the concept of constant final yield (CFY). The CFY, well revised by Weiner and Freckleton (2010), CFY is described from low to hight densities, whereas the relationship between total biomass per unit area and density is initially linear, but eventually, reaches a constant biomass that does not increase further. Consequently, in crop-weed relationship studies, the concept of CYF is appropriate for the rectangular hyperbola curve (Figure 1D). However, using Cousens (1985) model, the parameter *I* and *A* are only meaningful if CFY is reached. For example, in our study, the CFY was not completely reached, and the parameter *A* for *R. brasiliensis* and *C. benghalensis* was over 100% (Table 2). Similarly, a competition study that shows a linear relationship trend has not reached CFY yet (Figure 1A); therefore, the appropriated weed density for the study was not selected, and it is likely the reason that parameter A for both species, R. brasiliensis and C. benghalensis, was not different in competition with corn.

Regardless of the inadequacy of the data set to provide desired biological process, the logistic model is not appropriate for crop-weed relationship studies (Figure 3). The main reason is that this sigmoid curve of the logistic model is symmetric (S-shape) around the parameter *e* (Figure 1C), which the yield loss (%) or any response (*Y*-axis value) is forced to zero at lower densities. The logistic function assumes that yield loss (%) is near to zero at lower weed densities. Though the logistic model is not recommended for competition studies, it is one of the most commonly used and appropriate models in other weed research topics. Logistic curves are extensively used for herbicide dose-response and critical time for weed removal (Knezevic and Datta 2015; Ritz et al. 2015). For example, in dose-response studies, the parameter *e* is meaningful and important for comparison of herbicide doses that control 50% of weed population (Knezevic et al. 2007; Oliveira et al. 2017).

The polynomial quadratic model had the highest AIC; therefore it was the least relevant model (Table 1). This model does not provide any meaningful biological parameters that would improve the discussion, test hypothesis, and help researchers understand the results from crop-weed competition studies (Figure 4). The polynomial quadratic curve is symmetric around its maximum response value, which makes such response biologically unlikely in a crop-weed relationship (Figure 1B). For example, the maximum corn yield loss (%) is lower at four plants pot-1 than in three plants pot-1 (Figure 2). Therefore, a polynomial quadratic curve should not be encouraged to fit regression in competition studies.

In crop-weed relationship studies, because of wrongly R2 model selection, it is common to find multiple equations fitting response variables (Ferreira et al. 2015; Silva et al. 2015). For example, in a study of two weed species in competition with neotropical trees. Two different equations (one for each weed species) was fitted to describe the biomass (g) of *Senegalia polyphylla* (Monquero et al. 2015). In the same manuscript, other equations were used to describe different response variables. Equations have different parameters, and it would make difficult to compare the competitive effect of weeds.

**Weed competition with corn model (nested) selection.** We rejected the null hypothesis that *R. brasiliensis* and *C. benghalensis* compete similarly in corn (Table 5). The F-test indicated that competition of *R. brasiliensis* was lower than *C. benghalensis* at low densities, but similar at higher densities (Figure 5).

A complete review of model parameter *I* and *A* of the rectangular hyperbola is provided by a (Cousens 1985). Also, this model is recommended for crop-weed studies in weed research (Ritz et al. 2015; Swanton et al. 2015). Many authors have used this model to answer their research questions and improve weed control decision-making (Fischer et al. 2004; Lindquist et al. 1996; Lindquist et al. 1999; Werle et al. 2014). For example, using the rectangular hyperbola model, it was concluded the higher competitiveness of *Amaranthus palmeri* in corn and *Kochia scoparia* in sunflower (Lewis and Gulden 2014; Massinga et al. 2001). Parameters *I* and *A* are also useful for estimating weed competition across different locations and for calculating weed thresholds (Fisher et al. 2004). Additionally, using parameters *I* and *A,* it was demonstrated that organic cropping system has a potential to tolerate great abundance of weeds compared to conventional system (Ryan et al. 2009). Thus, the rectangular hyperbola proposed by Cousens (1985) model and F-test nested model selection are important and useful in crop-weed competition research.

**Conclusions**

Here we demonstrated that the rectangular hyperbola proposed by Cousens (1985) was the best model to fit for crop-weed studies in additive design. We also showed potential Cousens (1985) parameter issues when fitting the data. Nonetheless, the Cousens (1985) model meets the statistical criteria and parameters *I* and *A* are easily interpreted and biologically meaningful in agronomic terms. We propose Cousens (1985) as a standardized model for any data set of crop-weed relationship studies in additive design. If the data trend seems to be linear, the experimental design needs to be adjusted by increasing the weed density to achieve the CFY. Sigmoid curves are recommended to another set of studies in weed research, and polynomial quadratic curves should never be used. We also confirmed that *C. benghalensis* and *R. brasiliensis* have a high potential to cause corn yield loss (%). In areas of *C. benghalensis* infestations, weed management will increase, as *C. benghalensis* showed are competitive even at lower densities.

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