Prediction of genotypic values for yield in rubber tree-clone test trials using REML/BLUP procedure

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ABSTRACT

The present study was carried out to estimate variance components, prediction of genotypic values and repeatability for yield trait in rubber tree clones [*Hevea brasiliensis* (Willd. Adr. ex Juss.) Muell.-Arg.], through the procedure REML/BLUP. In the study were utilized twenty clones established in the Experimental Station of Votuporanga, São Paulo, Brazil, in the randomized block design with three replications. Each plot was represented by six trees, planted in the spacing of 7.0 m between lines and 3.0 m among plants. Data of yield of dry rubber were obtained when the trees reached a girth of 45.0cm at 1.20m above the bud union. The tapping system used was 1/2S d/4 6d/7. ET 2.5%. The results showed, a high genetic variability among clones, which can inferred on the estimates of the genotypic variation among clones (σ^2_g=92.79) and broad sense heritabilities based on mean annual tapping yield (h^2_{b1}=0.82) and of mean of five annual tapping yield (h^2_{b5}=0.89). The broad sense individual heritability (h^2_{bi} = 0.22), showed the presence of dominance for the trait and heterose should be explored with larger emphasis in the studied population. The clones IAC 301, IAC 56 and IAN 3156 presented superior genotypic values when compared to the average of the population. Based on the values of the repeatability coefficient and the found efficiency results it is supposed that there is no advange in more than three annual tapping, considering the necessary time for the selection and the obtained additional progress.

KEY WORDS: *Hevea brasiliensis,* components of variance, repeatability, variability.

INTRODUCTION

The rubber tree belonging to the genus *Hevea*, of the family Euphorbiaceae, has in the [*Hevea brasiliensis* (Willd. ex Adr. of Juss.) Muell.-Arg.] the most important species of the genus. Although it is an original crop of tropical and humid climate region (Bastos and Diniz, 1975; Varghese and Mydin, 2000), its cultivation is dispersed for several areas of Brazil, including the non traditional, whereas commercial exploration became viable due to the absence of South American Leaf Blight (SALB) incidence caused by *Microcyclus ulei* (P. Henn.) von Arx (Ortolani et al., 1983).

Brazil, although the country of the species of the genus (Gonçalves et al., 2002), contributed, the year of 2000, with only 1% of the world production of natural rubber, with 6,629 thousand tons and consumed 3%, of 7,361 tons of the world demand (International Rubber Study Group, 2001).

With the concern of increasing the rubber production, trials have been established in different environments, being considered the components of variances and other genetic parameters (Meenattor et al., 1991; Tan, 1995, Gonçalves et al., 1998a, 1998b, 1998c; Gonçalves, 1999; Costa et al., 2000) and repeatability (Alika, 1985; Gonçalves et al., 1990; Vasconcellos et al, 1985; Resende et al., 1996a) among others. However, these trials approached the several aspects of the genetic evaluation using traditional methods, based on the analysis of variance (least squares) considering the mean values of the individual progenies. However, it is necessary to evaluate candidates for selection at the individual level by predicted additive genetic values.

Accurate prediction of genetic values of individual candidates for selection is an essential part of forest breeding programs (Resende and Fernandes, 1999). The random prediction procedure named BLUP (best linear unbiased prediction) or methodology of mixed lineal models presented formally by Henderson (1975), it is a great procedure of prediction of genetic values (Henderson, 1984; Gianola and Hammond, 1990; Robinson, 1991; Searle et al., 1992). Also the REML (restricted maximum livelihood) method (Paterson and Thompson, 1971) is a optimum procedure for estimating variance components especially for unbalanced data.

For situations of unbalanced data (due to mortality during time), the methodology of mixed models has just been applied for families' means (White and Hodge, 1989; Resende et al., 1993; Resende et al., 1996, 1996b; Resende, 1997; Bueno Filho, 1997). However it is necessary to assess individual candidates for selection, fact that will lead to maximization of the genetic gain from selection.

In Brazil, in balanced data situations, the selection of individuals has been carried out using the multi-effect index method presented by Resende and Higa (1994a), which has proved superior to other forms of combined selection (Bueno Filho, 1992; Resende and Higa 1994b; Pires et al., 1996; Costa et al., 1999; Costa et al., 2000). The multi-effect index is an optimum selection procedure for balanced data situations, as it considers all the random effects of the statistical model associated to the randomized complete block design with several plants per plot, leading to identical results provided by BLUP in these situations (Resende and Fernandes, 1999). In rubber tree the individual BLUP procedure using variance components estimated by least mean squares was used by Costa et al. (1999) and the REML/BLUP procedure for Costa et al. (2001).

It is important to stand out that the genetic evaluation involving individuals of perennial species should base on models that consider the additional effect denominated effect of permanent environment, as well as, the parameter associated to the phenotypic correlation among repeated measures in a same individual, which is named repeatability. Repeatability coefficient measures the capacity of the individuals in repeating the expression of the character, by several periods of time (or in the space), along of their lives (Resende et al., 1995; Resende, 2002).

The present study was carried to estimate the variance components, and to predict the genotypic and repeatability values for the character rubber yield in *Hevea*, through the procedure REML/BLUP.

MATERIAL AND METHODS

Clonal material

Genetic materials used in the experiment were 20 *Hevea* genotypes (clones) from different origins, which were introduced in a small-scale trial (Table 1) for evaluation. The clones were budded on to established rootstocks (Tjir 1 x Tjir 16) in a nursery. One-and-a-half-year-old rootstock seedlings raised in nurseries were used to bud graft the clonal materials.

Table 1. Rubber tree clones and parental of 20 clones that belongs to the trial established in the Experimental Station of Votuporanga, SP[1/], Brazil.

Clones	Parental
IAN 4493	Fx 4421 (F 4573 x PB 86) x Tjir 1
IAC 316	AVROS 1328 (A VROS 214 x A VROS 317) x RR1M 608 (Tjir 33 x Tjir 1)
Fx 3899	F4542 x A VROS 363
Fx985	F315 x AVROS 183
IAC 300	RRIM 605 (Tjir 1 x PB 49) x AVROS 363
IAC 301	RR1M 501 (Pi149 x Lun N) x AVROS 1518
IAC 302	RRIM 501 (Pil A 49 x Lun N) x AVROS 353
IAC 303	RR1M 511 (pil B 84 x Pi1 A 44) x AVROS l.218 (AVROS 214 x A VROS 216)
IAC 306	AVROS 49 x RRIM 509 (Pil A 44 x Pil 246)
IAC 309	RR1M626 (Tjir 1 x RRIM 600)xFx25 (F 351 x AVROS 49)
IAC 312	RR1M 600 (Tjir 1 x PB 86) x Fx 25 (F 351 x AVROS 49)
IAC 313	RRlM 626 (Tjir 1 x PB 49)x Fx 25 (F 351 x AVROS 49)
IAC 56	RRIM 608 (A VROS 33 x Tjir 1) x Fx 3810 (F 4542 x AVROS 363)
IAN 3156	Fx 5l.6 (F 4542 x AVROS 363) x PB 86
IAN3193	Fx 516 (F 4542 x AVROS 363)x PB 86
IAN 3703	Fx 4371 (F 4542 x PB 86) x PB 86
IAN 6323	Tjir 1 x Fx 3810 (F 4542 x AVROS 363)
IAN 6721	Fx 43-655 [Fx 2.l3 (F 4542 x A VROS 183) x AVROS 183] x PB 86]
RO 45	Primary clone
RRlM 600	Tjir 1 x PB 86

[1/] IAN: Instituto Agronômico do Norte; F: Ford (primary clone); Fx: Cruzamento Ford; AVROS: Algemene Vereniging Rubber Planters Oostkust Sumatra; Tjir: Tjirandji; PB: Prang Besar; RO: Rondônia and RRIM: Rubber Reseach Institute of Malaysia.

The successful bud grafts were uprooted and planted in plastic bags. The experiment was planted in the field after the first flush of leaves.

The site

This work was done at the Votuporanga Experimental Station located in northwestern São Paulo State (Brazil) at 20º25'S latitude, 49º59'W longitude and 450m elevation. Mean monthly temperatures varied from 20 to 25ºC, and annual rainfall totals for the duration of the experiment ranged from 1,087mm to 1,537mm. The winter drought varied from four to six dry months, with an average water deficiency of 180mm.

The experimental design was a randomized complete block with three replications, using six trees per row with 7.0m x 3.0m spacing. Missing plants were replaced with spares during the first two years after planting to maintain plantation density, but were not scored. One row of the clone RRIM 600, acquired from a commercial nursery, was planted around the plot Annual fertilizations consisted of 400g of 10-10-10 formula NPK per plant, according to Bataglia and Gonçalves (1996).

Measurements

Trees that reached a girth of 45.0cm or more were opened for tapping at a height of 1.20m above the bud union. The used tapping system was ½ S d/4 6d/7. ET 2.5% (a half spiral cut tapped in each four days, six day in tapping followed by one day of rest, stimulated with 2.5% ethefon) Yield was recorded on the days when normal tapping, which starts around 6:30 AM, was possible. After tapping, latex was allowed to collect in plastic cups provided for each recording tree. Once the latex was flow stopped, rubber was coagulated in the cup itself adding two percent of acetic acid solution and stirring it well. The coagulated rubber in each cup was them made into a "biscuit" which were dried ranged by a wire tied in each tree for about 30 days, was weighed and the dry rubber content for each tree was recorded.

Statistical analysis

I model lineal mixed (Additive dominant univariate model I – of repeatability)

y = Xb + Za + Wc + Sd +Tp + e, where:

y, b, a, c, d, p e e: are vectors of data, of the effects of blocks (fixed), additive genetic effects (random), plot effects (random), genetic effects of dominance, effects of environment and random errors, respectively.

X, Z, W, S e T: are incidence matrixes for b, a, c, d and p, respectively.

Structures of means and variances

$$E\begin{bmatrix} y \\ a \\ c \\ d \\ p \\ e \end{bmatrix} = \begin{bmatrix} Xb \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \end{bmatrix};\ Var\begin{bmatrix} y \\ a \\ c \\ d \\ p \\ e \end{bmatrix} = \begin{bmatrix} V & ZA\sigma_a^2 & WI\sigma_c^2 & SI\sigma_d^2 & TI\sigma_p^2 & I\sigma_e^2 \\ A\sigma_a^2 Z' & A\sigma_a^2 & 0 & 0 & 0 & 0 \\ I\sigma_c^2 W' & 0 & I\sigma_c^2 & 0 & 0 & 0 \\ I\sigma_d^2 S' & 0 & 0 & I\sigma_d^2 & 0 & 0 \\ I\sigma_p^2 T' & 0 & 0 & 0 & I\sigma_p^2 & 0 \\ I\sigma_e^2 & 0 & 0 & 0 & 0 & I\sigma_e^2 \end{bmatrix}$$

Mixed model equations

$$\begin{bmatrix} X'X & X'Z & X'W & X'S & X'T \\ Z'X & Z'Z + A^{-1}\lambda_1 & Z'W & Z'S & Z'T \\ W'X & W'Z & W'W + I\lambda_2 & W'S & W'T \\ S'X & S'Z & S'W & S'S + I\lambda_3 & S'T \\ T'X & T'Z & T'W & T'S & T'T + I\lambda_4 \end{bmatrix} \begin{bmatrix} \hat{b} \\ \hat{a} \\ \hat{c} \\ \hat{d} \\ \hat{p} \end{bmatrix} = \begin{bmatrix} X'y \\ Z'y \\ W'y \\ S'y \\ T'y \end{bmatrix}$$

where:

$$\lambda_1 = \frac{\sigma_e^2}{\sigma_a^2} = \frac{1-\rho}{h^2}; \quad \lambda_2 = \frac{\sigma_e^2}{\sigma_c^2} = \frac{1-\rho}{c^2}; \quad \lambda_3 = \frac{\sigma_e^2}{\sigma_f^2} = \frac{1-\rho}{(h_a^2 - h^2)}; \quad \lambda_4 = \frac{\sigma_e^2}{\sigma_p^2} = \frac{1-\rho}{p^2}$$

h^2 : individual narrow sense heritability in the block;

h_a^2 : individual broad sense heritability in the block;

A : Matrix of additive genetic correlation among the individuals in evaluation

D : Matrix of genetic correlation of dominance among the individuals in evaluation.

REML estimates of the variance components via EM algorithm

$$\hat{\sigma}_e^2 = [y'y - \hat{b}'\ X'y - \hat{a}'\ Z'y - \hat{c}'\ W'y - \hat{f}'\ S'y - \hat{p}'\ T'y]/[N - r(x)]$$

$$\hat{\sigma}_a^2 = [\hat{a}' A^{-1}\ \hat{a} + \hat{\sigma}_e^2\ tr\ (A^{-1}\ C^{22})]/q$$

$$\hat{\sigma}_c^2 = [\hat{c}' c + \hat{\sigma}_e^2\ tr\ C^{33}]/s$$

$$\hat{\sigma}_d^2 = [\hat{d}' D^{-1} \hat{d} + \hat{\sigma}_e^2\ tr\ (D^{-1} C^{44})]/q$$

$\hat{\sigma}_p^2 = [\hat{p}'\hat{p} + \hat{\sigma}_e^2\ tr\ C^{55}]/q$, where:

C : Matrix of the coefficients of the mixed model equation;

tr : trace of a matrix;

r(x) : rank of the x matrix;

N, q, s : total data number, individual and plot number respectively.

The genotypic predicted values of the clones were obtained by $\hat{g} = \hat{a} + \hat{d}$. The genotypic variance among clones is equal $\hat{\sigma}_g^2 = \hat{\sigma}_a^2 + \hat{\sigma}_d^2$.

The selective efficiency, in terms of genetic gain with selection, of the use of *m* measurements in each plant in relation to the use of just one was estimated by the following expression:

$$Efficiency = \left[\frac{m}{[1+(m-1)\rho]}\right]^{1/2}$$, Where m = number of

measurements and s = coefficient of repeatability. That expression is valid so much for both generative and vegetative strategy propagation strategy of the selected material.

RESULTS AND DISCUSSION

The results regarding the estimates of the genetic parameters for the yield of dry rubber of the clone population are presented in the Table 2.

It was observed a high genetic variability among clones, as it can be inferred of the estimates of the genotypic variation among clones (h_g^2=92.79) and broad sense heritabilities based on mean of annual tapping yield level (h_{b1}^2=0.82) and based on mean of five annual tapping yield level (h_{b5}^2=0.89). These heritabilities values are in agreement, with those found by Paiva et al. (1982), Vasconcellos et al (1985) and Gonçalves et al. (1995).

The broad sense based on individuals (h_{bi}^2=0,22), it is almost the same value of that presented by Resende et al. (1996a), denoting to be a population with narrow genetics base. In spite of this, this result demonstrates presence of dominance for yielding trait. So heterosis should be explored with larger emphasis in the population in study. This procedure can be maximized being adopted outlines of reciprocal recurrent selection.

It is essential the knowledge of the three different of heritabilities values to planning the strategy of improvement of perennial species that be possible allow the vegetative propagation and the obtaining of repeated measurements in each individual, for the trait of interest (Resende et al., 1996a), in the case in study, yield trait.

The coefficient of genetic variation, which express in percentage of the general mean ($\overline{x}$=39,17), the amount of existent genetic variation, showed a considerable value (CVg=24.59) for the trait in study. This value shows coherence with the result obtained by Costa et al. (2000) for half siblings' progenies in the same

Table 2. Estimates of broad sense heritabilities, genotypic and phenotypic components of variances for yield trait of 20 clones of rubber tree grown in Votuporanga, São Paulo State, Brazil.

Parameters	Values
Broad sense heritability individual (h_{bi}^2)	0.2240
Broad sense heritability for annual tapping mean level (h_{b1}^2)	0.8257
Broad sense heritability for five annual mean level (h_{b5}^2)	0.8992
Genotypic variance among clones (σ_g^2)	92.7942
Phenotypic variance (σ_y^2)	414.3508
Variance among plots (σ_c^2)	6.2347
Environmental durable variance (σ_{ep}^2)	108.7526
Environmental variance among plots (σ_e^2)	206.5693
General mean ($\overline{x}$)	39.1738
Experimental coefficient of variation (CVe%)	36.6891
Genotypic coefficient of variation (CVg%)	24.5903

*General mean of genotypic values

location where the present study was carried out.

It is important to stand out that the selection based in clonal test is the method more accurate and it should lead to smallest variances of the predicted genetic gains (Resende et al., 1995), meaning larger probability of accomplishment of estimates gains.

In the Table 3, the phenotypic values and predicted genotypic of the clones are presented in study, genetic gain predicted of the three better clones, individual repeatability and at five annual tapping level for the yield of dry rubber.

It is observed that the clones IAC 301, IAC 56 and IAN 3156 presented genotypic values superior to the other evaluated clones (53.9159), (52.9750) and (51.3457), respectively. These values are expressive compared to the average obtained in the population (39.1738), what maximizes the genetic gains with selection, as presented in the Table 3. The genetic gain (G) of the three better clone are 37.6327, 35.2308, 31.0714, respectively.

Concerned to the repeatability (individual and at the level of five annual tapping), it is observed values considered high, suitable with countless quantitative characters found in the literature for perennial species, with prominence for yield of rubber tree ($\hat{p}_5$=0.84) (Resende et al., 1996a). Considering that the individual heritability in the broad sense it is around ($\hat{p}_1$=0.50) for rubber, it is deduced that there is a strong environmental component permanent for the species.

The maximization of the efficiency of the selection occurring more than an phenotypic observation of each genotype is desirable, considering that the average of more than a annual tapping tends to represent the genotypic value of the tree better than a annual tapping, and that the best individuals in a annual tapping harvest are not necessarily the best ones in other. The repeatability coefficient serves exactly to measure the high or small capacity, which the plants have to repeat the expression of the character (Vencovsky, 1972). Therefore, the use of the repeatability coefficient for the yield trait, associate to the vegetative propagation consists of an breeding strategy for the rubber tree, the one which, when applied to the available germoplasm, it allows fast genetic gain compared to the obtained by strategies based in progeny tests.

In the Table 4, the selective efficiencies are contained with base in five annual tappings for the yield trait.

For the studied trait, with repeatability coefficient of five annual tapping level ($\hat{p}_5$=0.84) it is had efficiencies of the selection from E=1.15 to E=1.31 for *m* varying from two to five annual tapping evaluations. Based in these values of repeatability coefficient and of the referred found efficiencies it is not advantageous to collect data of rubber yield besides three annual tapping considering the necessary time for the selection and the obtained additional progress. Resende et al. (1995), emphasizes that the efficiencies obtained with information of families, progenies or clonal tests they are reduced

Table 3. Phenotypic and genotypic values genetic gain predicted for the three better clones, individual ($\hat{\rho}_i$) and at the level of five annual tapping mean ($\hat{\rho}_5$) repeatability for yield trait of rubber tree in Votuporanga, São Paulo State, Brazil.

Clones	Number of data	Phenotypic values	Genotypic values ($\hat{\mu}+\hat{g}$)	Genetic gain (G) (Three better clones)
IAC 301	84	55.5678	53.9159	37.6327
IAC 56	78	54.4899	52.9750	35.2308
IAN 3156	78	52.1595	51.3457	31.0714
RO 45	74	51.0895	49.6601	-
IAC 300	75	49.9119	49.2880	-
RRIM 600	75	48.1591	48.0752	-
IAC 303	85	46.1438	45.7443	-
FX 3899	69	39.6708	40.1169	-
IAN 4493	82	39.3013	39.4292	-
IAC 302	79	38.5656	38.1891	-
IAC 313	82	36.9153	37.6643	-
FX 985	61	34.5848	34.5561	-
IAN 6721	82	33.2675	33.9734	-
IAC 309	85	32.1255	32.8250	-
IAN 3193	75	28.5412	30.0433	-
IAC 306	75	28.0644	29.7508	-
IAN 6323	79	28.2731	29.4270	-
IAN 3703	79	28.9783	29.3321	-
IAC 316	74	26.8096	28.7214	-
IAC 312	70	27.0445	28.4429	-
$\hat{\rho}_1$			0.5015	
$\hat{\rho}_5$			0.8400	

Table 4. Efficiency of selection (E) based on five annual tapping for yield trait of rubber tree clones in Votuporanga, São Paulo State, Brazil.

Number of harvests (m)	Efficiency of the selection (E)
02	1.1540
03	1.2240
04	1.2640
05	1.2900
06	1.3080

in comparison with the efficiencies made for mass selection calculations based in yield of dry rubber for annual tapping.

RESUMO

Predição de valores genotípicos para produção de borracha em clones de seringueira usando o procedimento REML/BLUP

O presente estudo objetivou estimar os componentes de variância, predição de valores genotípicos e repetibilidade para o caráter produção de borracha em clones de seringueira [*Hevea brasiliensis* (Willd. ex Adr. de Juss.) Muell.-Arg.], através do procedimento REML/BLUP. Vinte clones foram estabelecidos na estação experimental de Votuporanga, SP, no delineamento de blocos ao acaso com três repetições; cada clone foi representado por seis plantas por parcela linear, plantadas no espaçamento de 7,0 m entre linhas e 3,0 m entre plantas. Dados de produção de borracha seca foram obtidos a partir do sétimo ano de idade de todas as plantas da parcela que apresentaram perímetro do caule superior a 45 cm, a 1,20 m acima do calo de enxertia, utilizando-se o sistema 1/2S d/4 6d/7. ET2.5%. Verificou-se, de maneira geral, uma alta variabilidade genética entre os clones avaliados, conforme pode ser depreendido das estimativas da variação genotípica entre clones (92,7942g/árvore/sangria) e das herdabilidades no sentido amplo ao nível de média em um ano de sangria (0,8257) e de média em cinco ano de sangria (0,8992). A herdabilidade individual no sentido amplo (0,2240), demonstra que existe dominância para o caráter produção de borracha seca e que a heterose deve ser explorada com maior ênfase na população em estudo. Os clones IAC 301, IAC 56 e IAN 3156 apresentaram valores genotípicos superiores aos demais avaliados quando comparados à média da população, o que maximiza os ganhos genéticos com seleção. Com base nos valores do coeficiente de repetibilidade e das referidas eficiências encontradas não é vantajoso coletar dados de produção de borracha além de 3 safras, considerando o tempo necessário para a seleção e o progresso adicional obtido.

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