Theor Appl Genet (1989) 78: 567–580

Non-additive gene effects in populations under different methods of selection *

E. A. Carbonell **, A. E. Bell and J. J. Frey** ***

Department of Animal Sciences, Purdue University, West Lafayette, IN 47907, USA

Received September 1, 1988; Accepted March 6, 1989
Communicated by J. S. F. Barker

Summary. The genetic parameters of two quantitative traits, 13-day larval weight and pupal weight, in *Tribolium* populations developed by reciprocal recurrent selection (RRS) and by within-line purebred selection (WLS) were compared each with the other and also with the parameters of the unselected base populations using the genetic model of Carbonell, Nyquist and Bell. The variability for two- and three-way crosses of inbred lines derived from "companion" populations (two strains, breeds, or varieties used for a terminal cross or hybrid) was analyzed into genetic effects: autosomal additivity (*g), autosomal heterosis (*s), sex-linked additivity (L), sex-linked heterosis (LL), general maternal (m), specific maternal or reciprocal (r), additive by additive epistasis (aa), and deviations from the model due, among other causes, to higher order epistasis (dev). One series of crosses involved companion populations with diverse origins. For contrast, a second series of crosses involved companion populations originating from a common heterogenous base population. For the heterotic trait larval weight, *g and *s effects were equally important and accounted for over 50% of the total variation. The aa epistasis contributed another 20% and was followed in importance by higher order epistasis and general maternal effects. For the more highly heritable trait, pupal weight, *g effects were most important with *s, aa, and m effects having smaller but significant influences. Sex-linked and reciprocal effects were statistically significant for many crosses, but they were relatively unimportant overall. In general, the unselected base populations showed higher *g variation than either RRS or WLS populations with the reverse true for *s effects. In agreement with theoretical expectations, RRS was more effective than WLS in exploiting *s effects. The aa epistatic effects for larval weight were of major importance in the unselected populations, but RRS and WLS did not differ significantly for exploiting superior aa gene combinations. Companion populations with diverse origins revealed significantly larger variation due to *g and *s effects in crosses than did populations initiated from a common heterogeneous base.

Key words: Epistasis – *Tribolium* – Recurrent selection – Heterosis – Maternal effects

Introduction

Many investigators have attempted to quantify the relative importance of hereditary and environmental effects and their interactions. Estimation procedures for additive gene effects are well known today, but those for dominance, epistatic, sex-linked, and maternal effects are less well documented. Additional information as to the nature of and how to effectively utilize these non-additive gene effects is important for the design of optimum selection procedures in animal and plant improvement (Bell 1981, 1982b).

Carbonell et al. (1983) provided a model for the estimation of maternal, sex-linked, and additive × additive epistatic effects in addition to the usual additive and dominance effects. Subsequently, Carbonell et al. (1985) demonstrated the application of this model in a genetic study of a heterotic trait (13-day larval weight) and an

* Journal Paper No. 11559 from Purdue University Agricultural Experimental Station
** Present address: Instituto Valenciano de Investigaciones Agrarias, Apartado Oficial, E-46113 Moncada (Valencia), Spain
*** Present address: Pfizer Inc., 235 E. 42nd St., New York, NY 10017, USA

additive trait (pupal weight) in two unrelated base populations of *Tribolium*.

Recently, Melchinger et al. (1986) used three methods – (1) generation means analysis, (2) diallel analysis of generation means, and (3) analysis of single and three-way crosses – to estimate genetic parameters, including epistasis and maternal effects. The maternal effects were attributed to a heterozygous seed parent and, hence, were defined only in F_2 and some backcrosses. Therefore, their maternal effects are analogous to the "heterotic maternal effect" as defined by Carbonell et al. (1983). Dickerson's (1969) concept of "recombination loss" recognizes the importance of epistasis in crossbreeding systems, as does Sheridan's (1980) "parental epistasis," but neither can be equated unequivocally to generalized gene effects. Kinghorn (1982) presented statistical models for the estimation of epistatic effects in animal populations, but he ignored sex-linked effects. Subsequently, Kinghorn (1983) used his models to analyze additive and non-additive effects in three inbred strain of mice and to discuss the relationship between additive × additive interaction and recombination loss. Using data from Sewall Wright's early work, Kinghorn (1987) found that the model equivalent to additive × additive epistasis gave the best general fit over 7 biological models of 2-locus interaction for the 6 out of 11 traits in which epistasis effects were apparent. Diallel cross analyses for estimating reciprocal and specific reciprocal effects have been used by Eisen et al. (1985) to study body weight and litter size of mice, but they assumed sex-linked effects to be absent. Dearborn et al. (1987) have estimated maternal heterosis and grandmaternal effects for preweaning traits in beef cattle.

The main objective of the above studies has been to provide "static" estimates of genetic effects in order to predict cross-bred performance using specific breeds or lines. Once a superior cross or hybrid has been identified, various selection schemes (e.g., Comstock et al. 1949; Bell et al. 1952) have been proposed to improve the performance of the cross-bred. Numerous selection studies empirically comparing such schemes have been reviewed (Bell 1982 b), yet none attempted to compare them as to their effectiveness for exploiting the different types of gene effects.

In the present paper, we will use the Carbonell Model to identify long-term changes in the relative magnitude of both additive and non-additive gene effects in populations under reciprocal recurrent selection and we will contrast them to comparable changes in populations under purebred or within-line selection.

Materials and methods

The genetic material for this study consisted of a large number of inbred lines derived from 26 populations with distinct selection histories. These populations had originated from two large unrelated heterogeneous base populations of the flour beetle, *Tribolium castaneum*, which have been used extensively for model experiments in quantitative genetics (Bell 1982a). The Purdue Black Population is genetically marked with the partially dominant black (*b*) body color mutant and the Purdue Pearl Population is marked with the recessive pearl (*p*) eye color mutant. Two replicate samples were taken from each base population and were designated I and II; thus, BI denotes replicate I of a selection line originating from Purdue Black. In order to simulate or model "improved" purebred lines or varieties, each of these sub-populations was subjected to within-line selection (WLS) for 30 generations using the purebred family mean 13-day larval weight as the selection criterion (see McNew and Bell 1974, for details).

After the 30 generations of WLS, the four selected lines (BI, BII, PI, PII), collectively designated as C_1 populations, were random mated separately for an additional 5 generations (Fig. 1 shows the formation of BI and PI lines). Then BI and PI were reciprocally crossed to each other as were BII and PII. Their cross-bred offspring were randomly reproduced en masse for three generations, at which time each cross-bred population (BI × PI or BII × PII) was subdivided according to body color segregants, black or wild-type red. The new cross-bred populations of black individuals were denoted XBI and XBII according to their origin, and those with the "normal" or wild-type red body color were denoted XNI and XNII. These four new heterogeneous populations were collectively designated as C_2 populations.

In addition to the two original base populations (B and P), each of the above C_1 and C_2 populations was continued with relaxed selection by randomly mating approximately 1500 offspring each generation. Subsequently, two sub-populations were taken at random from each of the C_1 and C_2 populations and selected for an additional 23 generations, with one of the sub-populations subjected to continued within line selection (WLS) and the other to reciprocal recurrent selection (RRS). For both WLS and RRS, the term "companion populations" is used to identify two specific populations with contrasting body colors (black or red) paired within selection method and replication, and repeatedly crossed over generations for the determination of crossbred performance. This crossbred performance was the selection criterion for RRS with purebred performance within each of the companion populations observed as a correlated response. For WLS, the reverse was true. The details of this earlier selection study are reported by McNew and Bell (1976). The population codes are summarized in Fig. 1.

According to their origins and selection histories, the populations described above are grouped as follows:

1 *Unselected Base Populations* – B and P

(a) Unselected, randomly mated diverse populations, Black and Pearl, referred to as "UNSR" companion populations when crossed B × P.

2 *C_1 Populations* – BI, PI, BII, and PII (30 generations of WLS)

(a) Relaxed Selection – Four C_1 populations reproduced without further selection, referred to as "WLSR1" and "WLSR2" companion populations when paired as BI × PI and BII × PII, respectively.
(b) WLS – Continued WLS in four C_1 populations for an additional 23 generations, referred to as "WLSC1" and "WLSC2" companion populations when crossed as BI × PI and BII × PII, respectively.

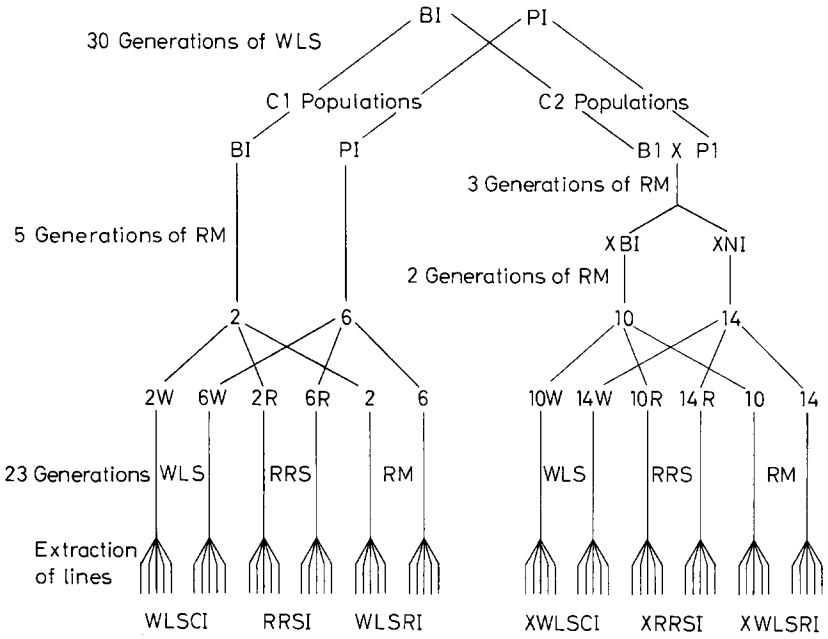

Fig. 1. Schematic history of Replication I inbred lines with companion populations connected by dashed lines. Replication II inbred lines were extracted similarly from Populations 4, 8, 12, and 16 (McNew and Bell 1976)

(c) RRS – 23 generations of RRS in four C_1 populations referred to as "RRS1" and "RRS2" companion populations when paired as BI × PI and BII × PII, respectively.

3 *C_2 Populations* – XBI and XNI (from BI × PI); XBII and XNII (from BII × PII)

(a) Relaxed Selection – Four C_2 populations reproduced without further selection, referred to as "XWLSR1" and "XWLSR2" companion populations when paired as XBI × XNI and XBII × XNII, respectively.
(b) WLS – 23 generations of WLS in four C_2 populations referred to as "XWLSC1" and "XWLSC2" companion populations when crossed as XBI × XNI and XBII × XNII, respectively.
(c) RRS – 23 generations of RRS in four C_2 populations, referred to as "XRRS1" and "XRRS2" companion populations when paired as XBI × XNI and XBII × XNII, respectively.

The 26 populations outlined above were expanded and random mated for 5 generations to dissipate the linkage disequilibrium arising from selection. Then 50 inbred lines were initiated from each population by single pair full-sib matings for 6 generations. Crosses among the surviving inbred lines were then made by random sets of four lines taken from the two "companion" populations unique to each selection scheme and replication.

The mating design for any two companion populations called for 12 independent sets of 4 inbred lines, with 2 random black and 2 random pearl (or wild-type red body color) lines constituting a set. The four lines in a set were arbitrarily coded as 1, 2 and 3, 4, respectively. A balanced set consisting of 12 two-way and 24 three-way crosses, avoiding backcrosses but including reciprocal crosses, was made among the four inbred lines within each set (Table 1). In addition, four of the crosses, chosen randomly, were repeated within each set in order to have an estimate of experimental error. Also, a second repetition of each set was carried out.

A 24-hour egg collection in a ¾-ounce creamer containing 4 g of standard medium (whole wheat flour enriched with 5% dried brewers' yeast) was taken from a mass mating of four males and four females for each cross. This egg collection was cultured for 13 days in an environmental chamber controlled at approximately 33 °C and 70% relative humidity. At that time, the larvae were screened and a group weight of ten randomly chosen larvae per mating was recorded in decamicrograms (dμg). All larvae from each mating were returned to the environmental chamber until most had pupated, at which time a group weight of ten randomly chosen unsexed pupae per mating was recorded. If a mating produced fewer than ten offspring, all were weighed and the observed value was converted to a basis of ten progeny.

Table 1. The two-way and three-way crosses made among the 4 inbred lines of each set. Inbred lines 1 and 2 from the Black Population and lines 3 and 4 from the Pearl (or Red) Population were randomly selected for each set of crosses. The reciprocal crosses $C_{i'i}$ and $C_{i'i'',i}$ were made but are not listed here

Two-way crosses ($C_{ii'}$)	Three-way crosses ($C_{i,i'i''}$)	
1 × 2	1 × (3 × 2)	3 × (1 × 2)
1 × 3	1 × (2 × 4)	3 × (1 × 4)
1 × 4	1 × (3 × 4)	3 × (2 × 4)
2 × 3	2 × (3 × 4)	4 × (1 × 2)
2 × 4	2 × (1 × 3)	4 × (3 × 1)
3 × 4	2 × (1 × 4)	4 × (3 × 2)

Statistical model

Data from each set were analyzed according to the following statistical model:

$$Y_{pqr} = \mu + R_p + w_{(p)} + C_q + RC_{pq} + e_{r(pq)}$$

where Y_{pqr} is the observed value of the analyzed trait (larval or pupal weight); μ is the overall mean; R_p is the effect (random) of

the p^{th} repetition ($p=1, 2$); $w_{(p)}$ is the restriction error in the randomization (Anderson 1970); C_q is the effect (fixed) of the q^{th} cross ($q=1, \ldots 36$); and $e_{r(pq)}$ is the within repetition and cross error. The sum of squares due to crosses was divided into the following genetic components using the models developed by Carbonell et al. (1983):

$$C_{ii'} = {}^*\mu + {}^*g_i + {}^*g_{i'} - m_i + m_{i'} + {}^*s_{ii'} + r_{ii'} + aa_{ii'}$$

$$C_{i,i'i''} = {}^*\mu + {}^*g_i + 1/2({}^*g_{i'} + {}^*g_{i''}) - m_i + 1/2(m_{i'} + m_{i''}) + 1/2({}^*s_{ii'} + {}^*s_{ii''}) + 1/2(r_{ii'} + r_{ii''}) + 1/4\, aa_{i'i''} + 1/2(aa_{ii'} + aa_{ii''})$$

$$C_{i'i'',i} = {}^*\mu + {}^*g_i + 1/2({}^*g_{i'} + {}^*g_{i''}) + m_i - 1/2(m_{i'} + m_{i''}) - 1/4\, L_{i'} + 1/4\, L_{i''} + 1/2({}^*s_{ii'} + {}^*s_{ii''}) - 1/2(r_{ii'} + r_{ii''}) - 1/4\, LL_{ii'} + 1/4 LL_{ii''} + 1/4\, aa_{i'i''} + 1/2(aa_{ii'} + aa_{ii''})$$

where $C_{ii'}$ is the single cross between lines i (male) and i' (female), $C_{i,i'i''}$ is the three-way cross between male line i and female single cross $i'i''$, $C_{i'i'',i}$ is its reciprocal, and

$${}^*g_i = g_i - aa_i^h$$
$$L_i = 1/2(a_i^s + d_i^s) + h_i^s$$
$${}^*s_{ii'} = s_{ii'} - aa_{ii'}^h$$
$$LL_{ii'} = hh_{ii'}^s \text{ and}$$
$$aa_{ii'} = aa^h + aa_i^h + aa_{i'}^h + aa_{ii'}^h$$

Furthermore, g_i and $s_{ii'}$ are the well-known general and specific combining abilities for autosomal genes, and m_i and $r_{ii'}$ are the general and specific maternal effects; L_i and $LL_{ii'}$ have the same meaning as ${}^*g_i$ and ${}^*s_{ii'}$ but applied to sex-linked genes. The $aa_{ii'}$ is the additive × additive epistatic effect present when two lines are crossed as defined by Eberhart and Gardner (1966). As they indicate, $aa_{ii'}$ can be subdivided into its components, the average effect aa^h, the two general effects for both lines aa_i^h and $aa_{i'}^h$ and the specific effect $aa_{ii'}^h$. The additive and intraline dominance effects of sex-linked genes as defined by Carbonell et al. (1983) are denoted a_i^s and d_i^s for these respective effects when expressed in females. The sex-linked interline dominance effect $h_{ii'}^s$ arises when the two populations i and i' are crossed and the sex-linked dominance contribution of the cross deviates from the mean of the sex-linked dominance contribution of both parental populations, i.e., it represents the heterosis due to sex-linked genes. This effect can be subdivided into its components in similar fashion to that of $aa_{ii'}$. As noted in the "Materials and methods" section, the offspring from the various crosses were unsexed; hence, one should assume a 1:1 sex ratio in order to be able to estimate the sex-linked effects (see comments in Carbonell et al. 1983, p. 611). The general maternal effect of any crossed dam is assumed to be the average of that of the grandmaternal lines. The difference between the sums of squares due to the models and that for crosses will provide an indirect estimation of higher order epistasis (and possibly linkage and any other effects not considered in the statistical model or assumed to be absent).

According to the definitions of the parameters, $*g$ and L effects are primarily additive and intraline dominance effects; $*s$ and LL effects are the interline dominance effects resulting from the cross of two lines; m effects are due to differences between a line entering as a paternal or maternal parent in a cross and are a function of the sex-linked additive female effects and the non-frequency dependent maternal effects (cytoplasmic, maternal ability, etc.); r effects are the specific maternal reciprocal effects and do not contain sex-linked effects; and aa are the additive × additive epistatic effects. $*s$, LL and aa provide the basis for the expression of heterosis.

Even though heterosis is an observed phenomenon and not a genetic mechanism, we will use in short the word "autosomal heterosis" for the $*s$ effects, and "sex-linked heterosis" for the LL effects to stress the fact they are expressed when two lines are crossed, as compared with the intraline dominance present in $*g$ and expressed in the lines themselves. "Additive × additive epistasis" will refer to the aa effects and "higher order epistasis" to deviations from the model. The $*g$ and L effects will be referred as to "autosomal additive" and "sex-linked additive" effects, respectively. The precise meaning of each of these effects and words should be clear from the above statistical definitions and from Carbonell et al. (1983, 1985).

Results

The results from the statistical analyses of larval and pupal weights for two- and three-way crosses among inbred lines from the two unselected Black and Pearl base populations were reported by Carbonell et al. (1985). They will not be included here except when the relative magnitude of various gene effects in these two unselected base populations (UNSR) is contrasted in a later section with those from the selected C_1 and C_2 populations.

Larval weight

C_1 populations. Mean larval weights for C_1 two-way and three-way crosses averaged over sets are presented in Table 2. Mean values of the two-way crosses are broken down into those that represent crosses among inbred lines "within" each companion population (1×2 and 3×4) and those among inbred lines "between" companion populations (1×3, 1×4, 2×3, and 2×4). Likewise, three-way crosses are split into those where the female parents were either crossbred or inbred.

Two-way crosses for all populations showed greater heterosis for crosses involving "between" companion populations than those using "within" companion populations with relative differences ranging from 47% for RRS1 to 24.8% for WLSC2. On the average, populations under RRS had the largest relative difference, whereas populations under continued WLS had the smallest difference. Similarly, three-way crosses with F_1

Table 2. Mean larval weights (dμg) for C_1 two- and three-way crosses with a comparison of within- and between-companion population two-way crosses and inbred versus F_1 dams for three-way crosses

Population	Two-way crosses			Three-way crosses		
	Within	Between	Mean	Inbred dam	F_1 dam	Mean
WLSC1	312±4	401±3	371±3	390±1	413±1	401±1
WLSC2	278±3	347±2	324±2	337±1	346±1	341±1
RRS1	259±4	381±3	340±3	365±2	390±2	377±2
RRS2	272±4	348±2	323±3	334±1	350±1	342±1
WLSR1	237±3	331±2	300±2	317±1	318±1	317±1
WLSR2	235±3	312±2	286±2	302±1	317±1	309±1

Table 3. Analyses of variance for C_1 larval weight, pooled over sets and over populations with similar selection histories

Source	*df*	MS	*df*	MS	Pooled+ MS
	WLSC1		WLSC2		WLSC
Reps	12	3,578.0	12	2,137.1	2,857.6
Cross	420	5,655.3**	420	2,939.8**	4,297.5**
*g	36	15,121.9**	36	8,857.6**	11,989.7**
L	36	3,121.3**	36	1,487.3*	2,304.3**
*s	24	21,082.6**	24	14,049.3**	17,566.0**
LL	24	1,123.2	24	1,036.3	1,079.7
m	36	9,942.2*	36	3,196.1**	6,569.2**
r	36	1,992.5*	36	1,249.1	1,620.8
aa	72	6,759.0**	72	2,466.9**	4,613.0**
dev	156	1,753.0*	156	1,037.8	1,395.4**
R × C	420	1,285.2	420	903.0	1,094.1
	RRS1		RRS2		RRS
Reps	12	10,969.4	12	6,400.8	8,685.1
Cross	420	9,092.1**	420	5,470.2**	7,281.2**
*g	36	22,447.4**	36	17,664.2**	20,055.8**
L	36	5,822.4**	36	3,977.2**	4,899.8**
*s	24	37,384.8**	24	26,941.0**	32,162.9**
LL	24	3,485.5**	24	1,765.6	2,625.5**
m	36	12,361.8**	36	5,093.7**	8,727.7**
r	36	3,277.5**	36	2,316.6**	2,797.0**
aa	72	10,400.9**	72	3,755.2**	7,078.0**
dev	156	3,257.7**	156	1,873.7**	2,565.7**
R × C	420	1,745.6	420	1,267.0	1,506.3
	WLSR1		WLSR2		WLSR
Reps	12	7,282.7	12	4,341.2	5,812.0
Cross	420	4,044.5**	420	3,462.3**	3,753.4**
*g	36	10,247.9**	36	11,100.9**	10,674.4**
L	36	2,979.5**	36	1,965.3**	2,472.4**
*s	24	28,238.9**	24	15,973.3**	22,106.1**
LL	24	913.3	24	587.1	750.2
m	36	4,679.3**	36	6,302.0**	5,490.7**
r	36	1,023.1	36	501.1	762.1
aa	72	2,137.3**	72	2,782.9**	2,460.1**
dev	156	1,048.2**	156	904.2**	976.2**
R × C	420	722.6	420	636.3	679.5

+ *df* of the "Pooled MS" are the sum of *df* of the populations to be pooled

* and ** in MS denote significance at 0.05 and 0.01 level of probability, respectively (as in all other tables)

Table 4. Number of significant sets at 5% level (*N**) and 1% level (*N***) out of 12 sets and percentage of total variation for C_1 larval weight among crosses atributable to genetic effects

Effect	*N**	*N***	(%)	*N**	*N***	(%)
	WLSC1			WLSC2		
*g (autosomal additive)	12	10	22.9	11	11	25.9
L (sex-linked additive)	6	2	4.7	2	0	4.3
*s (autosomal heterosis)	12	11	21.3	12	12	27.3
LL (sex-linked heterosis)	1	0	1.1	0	0	2.0
m (general maternal)	12	9	15.0	6	5	9.3
r (specific maternal)	2	0	3.0	3	1	3.6
aa (A × A epistasis)	11	8	20.5	3	3	14.4
dev (higher order epistasis)	0	0	11.5	2	0	13.1
	RRS1			RRS2		
*g (autosomol additive)	12	11	21.2	12	12	27.7
L (sex-linked additive)	7	3	5.5	7	3	6.2
*s (autosomal heterosis)	12	12	23.5	12	12	28.1
LL (sex-linked heterosis)	2	1	2.2	0	0	1.8
m (general maternal)	10	9	11.7	8	4	8.0
r (specific maternal)	2	2	3.1	3	2	3.6
aa (A × A epistasis)	10	9	19.6	6	4	11.8
dev (higher order epistasis)	5	2	13.3	3	2	12.7
	WLSR1			WLSR2		
*g (autosomal additive)	11	11	21.7	12	12	27.5
L (sex-linked additive)	8	5	6.3	6	3	4.9
*s (autosomal heterosis)	12	12	39.9	12	12	26.4
LL (sex-linked heterosis)	1	0	1.3	0	0	1.0
m (general maternal)	12	9	9.9	11	11	15.6
r (specific maternal)	2	0	2.2	0	0	1.2
aa (A × A epistasis)	8	4	9.1	11	7	13.8
dev (higher order epistasis)	5	2	9.6	3	0	9.7

dams were heavier than those where the female parents were inbred. The relative difference ranged from 7% for RRS1 to 1% for WLSR1. Overall, three-way crosses had larger values than those for two-way crosses.

The analyses of variance for C_1 larval weights pooled over sets and over populations with similar selection histories are presented in Table 3. Since these analyses were based on many observations, a substantial number of degrees of freedom are associated with the various effects. Consequently, rather small *F* ratios are sufficient to declare statistical significance. Hence, the number of sets with significant effects at 5% level (*N**) and 1% level (*N***) out of the 12 sets is more indicative of the relative importance of effects (Table 4). In addition, a relative measurement of variation based on the percentage of the sum of squares attributable to each of the different effects is included in the table as a second criterion to evaluate the relative importance of the effects over populations.

The pooled analyses showed all genetic effects, except *LL* and *r*, to be highly significant, with autosomal additivity (*g) and autosomal heterosis (*s) accounting for over 50% of the total variation. Autosomal heterosis (*s) was the most important effect for both RRS populations, whereas for WLSC and WLSR populations, the relative importance of *s effects was not consistent over replications. All populations showed significant additive sex-linked (*L*) effects, but only RRS1 showed significant sex-linked heterosis (*LL*). Specific cytoplasmic (*r*) effects were generally unimportant, whereas maternal effects

(*m*) were an important source of variation in all populations. Additive × additive epistasis (*aa*) was highly significant in all populations with slightly higher values in those that had undergone longer selection histories. In general, higher order epistasis (*dev*) was of minor importance.

C_2 populations. Mean larval weights for C_2 two- and three-way crosses averaged over sets are presented in Table 5 and are summarized similarly to those for C_1 populations.

All populations again showed greater heterosis for "between" companion population crosses than that observed for "within" crosses. Likewise, three-way crosses involving cross-bred dams had larger body weights than those with inbred dams. Except for XWLSC2, all populations showed three-way crosses with higher larval weights than the corresponding two-way crosses. As expected from their selection histories, both XWLSC and XRRS had much higher mean larval weights than the relaxed selection populations (XWLSR). Replication I consistently had higher values than replication II; however, the differences between replications were confounded with environmental time trends.

The analyses of variance pooled over sets and populations with similar selection histories are shown in Table 6. Again, rather small *F* ratios are found to be statistically significant and all genetic effects, other than *LL* and *r* effects, were highly significant. The number of significant sets and percentage of variation (Table 7) is more indicative of the relative importance of different effects. In general, the autosomal additive (**g*) and autosomal heterotic (**s*) effects accounted for a smaller portion of the total variation than was observed for the C_1 populations. However, the *aa* and higher order epistatic effects have increased to about 40% of the variation. The other genetic effects were relatively small and similar to the C_1 values. The exceptionally large amount of *aa* epistasis shown in Table 7 for the XWLSC2 population will be discussed in a later section.

Pupal weight

C_1 populations. Mean pupal weights for C_1 two-way and three-way crosses averaged over sets are presented in Table 8 with a similar breakdown as for larval weight.

Although the values for three-way crosses were consistently higher than those for two-way crosses, their relative differences were much smaller than for larval weight. Similarly, "between" companion population two-way crosses were heavier than "within" populations crosses but their relative difference was always less than 11%. In addition, except for WLSR1, all three-way crosses with F_1 dams were heavier than those from inbred dams.

Table 5. Mean larval weights (dμg) for C_2 two- and three-way crosses with a comparison of within- and between-companion population two-way crosses and inbred versus F_1 dams for three-way crosses

Population	Two-way crosses			Three-way crosses		
	Within	Between	Mean	Inbred dam	F_1 dam	Mean
XWLSC1	352±4	418±3	396±4	406±2	414±2	410±2
XWLSC2	285±4	331±3	316±4	284±2	304±2	294±2
XRRS1	348±3	403±2	384±3	395±1	417±1	406±1
XRRS2	299±3	350±2	332±4	346±1	366±1	356±1
XWLSR1	261±3	302±2	288±3	296±1	317±1	305±2
XWLSR2	231±3	272±2	258±3	268±1	285±1	276±1

Table 6. Analyses of variance for C_2 larval weight, pooled over sets and over populations with similar selection histories

Source	*df*	MS	*df*	MS	Pooled MS
	XWLSC1		XWLSC2		XWLSC
Reps	12	8,815.9	12	4,757.4	6,786.7
Cross	420	4,712.3	420	6,177.2**	5,444.7**
**g*	36	13,587.3**	36	7,012.1**	10,299.7**
L	36	3,823.6**	36	15,109.6**	9,466.6**
**s*	24	14,312.7**	24	3,017.0**	8,664.8**
LL	24	3,006.5*	24	2,381.5*	2,694.0*
m	36	6,177.2**	36	3,822.7**	4,999.9**
r	36	1,899.2	36	1,887.7	1,893.5
aa	72	4 085.1**	72	15,282.8**	9,683.9**
dev	156	2,255.4**	156	2,323.9**	2,289.7**
R × C	420	1,686.0	420	1,570.8	1,628.4
	XRRS1		XRRS2		XRRS
Reps	11	1,839.8	10	725.3	1,282.6
Cross	385	3,426.6**	350	3,759.8**	3,593.2**
**g*	33	7,560.6**	30	10,749.0**	9,154.8**
L	33	2,347.8**	30	3,156.1**	2,751.9**
**s*	22	10,985.0**	20	10,238.3**	10,611.6**
LL	22	1,885.4**	20	1,566.2**	1,725.8**
m	33	2,907.1**	30	5,100.5**	4,003.8**
r	33	1,204.7	30	2,729.0**	1,966.8**
aa	66	4,550.2**	60	3,490.0**	4,020.1**
dev	143	1,909.8**	130	1,680.1**	1,795.0**
R × C	385	954.1	350	859.5	906.8
	XWLSR1		XWLSR2		XWLSR
Reps	13	4,703.4	12	2,015.5	3,359.4
Cross	455	2,770.0**	420	2,349.5**	2,559.7**
**g*	39	6,414.0**	36	3,395.0**	4,904.5**
L	39	2,141.7**	36	1,718.8**	1,930.8**
**s*	26	6,115.8**	24	8,323.9**	7,219.8**
LL	26	1,377.1	24	910.7	1,143.9
m	39	4,089.2**	36	4,626.4**	4,357.8**
r	39	725.9	36	1,178.8	952.4
aa	78	3,304.4**	72	2,330.6**	2,817.5**
dev	169	1,693.9**	156	1,309.3**	1,501.6**
R × C	455	96.7	420	879.3	938.0

Table 7. Number of significant sets at 5% level (N^*) and 1% level (N^{**}) out of 12 sets and percentage of total variation for C_2 larval weight among crosses attributable to genetic effects

Effect	N^*	N^{**}	(%)	N^*	N^{**}	(%)
	XWLSC1			XWLSC2		
*g (autosomal additive)	10	8	24.7	6	5	9.7
L (sex-linked additive)	3	1	7.0	12	11	21.0
*s (autosomal heterosis)	12	10	17.4	1	0	2.8
LL (sex-linked heterosis)	2	0	3.7	0	0	2.2
m (general maternal)	8	5	11.2	3	2	5.3
r (specific maternal)	0	0	3.5	0	0	2.6
aa (A × A epistasis)	5	3	14.9	12	12	42.4
dev (higher order epistasis)	2	0	17.2	2	2	14.0
	XRRS1[a]			XRRS2[b]		
*g (autosomal additive)	9	9	18.9	9	8	24.5
L (sex-linked additive)	3	1	5.9	5	4	7.2
*s (autosomal heterosis)	10	9	18.3	10	9	15.6
LL (sex-linked heterosis)	2	1	3.1	2	0	2.4
m (general maternal)	6	5	7.3	9	5	11.6
r (specific maternal)	2	0	3.0	3	3	6.2
aa (A × A epistasis)	11	9	22.8	6	5	15.9
dev (higher order epistass)	4	3	20.7	2	2	16.6
	XWLSR1[c]			XWLSR2		
*g (autosomal additive)	8	8	19.9	6	4	12.4
L (sex-linked additive)	3	1	6.6	3	1	6.3
*s (autosomal heterosis)	10	8	12.6	9	7	20.2
LL (sex-linked heterosis)	0	0	2.8	1	0	2.2
m (gemeral maternal)	7	5	12.7	10	6	16.9
r (specific maternal)	0	0	2.3	1	0	4.3
aa (A × A epistasis)	9	7	20.5	6	5	17.0
dev (higher order epistasis)	6	2	22.7	1	0	20.7

[a] based in 11 sets
[b] based in 10 sets
[c] based in 13 sets

The analyses of variance for C_1 pupal weights pooled over sets and over populations with similar selection histories are presented in Table 9. The number of significant sets and percentage of total variation attributable to specific gene effects are shown in Table 10.

When pooled over populations, all effects were significant in WLSC, whereas for RRS the specific maternal *r* effects were not important; for WLSR, no significance was found for sex-linked heterosis, specific maternal effects, or higher order epistasis. Except for RRS1, autosomal additivity (*g) was the major source of variation in all populations as expected for an additive trait. Autosomal heterosis (*s) and general maternal effects (*m*) were moderately important sources of variation. Apparently, the maternal effects present in early stages of development are still important for pupal or adult weight in *Tribolium*. In general, epistasis (*aa* and *dev*) was less important for pupal weight than for larval weight.

Table 8. Mean pupal weights (dμg) for C_1 two- and three-way crosses with a comparison of within- and between-companion population two-way crosses and inbred versus F_1 dams for three-way crosses

Population	Two-way crosses			Three-way crosses		
	Within	Between	Mean	Inbred dam	F_1 dam	Mean
WLSC1	357±2	388±1	378±1	382±1	387±1	384±1
WLSC2	296±2	327±1	317±1	325±1	331±1	328±1
RRS1	349±2	383±1	372±1	376±1	377±1	377±1
RRS2	317±2	340±1	332±1	337±1	340±1	338±1
WLSR1	301±2	324±1	316±1	321±1	319±1	320±1
WLSR2	284±2	309±1	301±1	305±1	310±1	308±1

Table 9. Analyses of variance for C_1 pupal weight, pooled over sets and over populations with similar selection histories

Source	*df*	MS	*df*	MS	Pooled MS
	WLSC1		WLSC2		WLSC
Reps	12	353.3	12	747.9	550.6
Cross	420	875.7**	420	1,338.4**	1,107.0**
*g	36	2,246.1**	36	7,119.6**	4,682.9**
L	36	427.0**	36	430.4**	428.7**
*s	24	1,954.4**	24	3,255.8**	2,605**
LL	24	412.7*	24	521.3**	467.0**
m	36	2,556.6**	36	1,987.7**	2,272.2**
r	36	417.5**	36	469.6**	443.4**
aa	72	671.4**	72	793.1**	732.2**
dev	156	378.5**	156	346.8*	362.7**
R × C	420	235.9	420	254.1	245.0
	RRS1		RRS2		RRS
Reps	12	3,257.1	12	2,063.8	2,660.4
Cross	420	897.8**	420	1,233.7**	1,065.8**
*g	36	1,774.6**	36	6,503.9**	4,139.3**
L	36	627.7**	36	808.9**	718.3**
*s	24	3,668.3**	24	1,987.2**	2,827.8**
LL	24	581.3**	24	550.2	565.8**
m	36	1,964.0**	36	1,793.8**	1,878.9**
r	36	356.7	36	213.0	284.9
aa	72	499.4**	72	653.4**	576.4**
dev	156	443.1**	156	478.8**	460.9**
R × C	420	257.8	420	411.6	334.7
	WLSR1		WLSR2		WLSR
Reps	12	1,127.9	12	335.3	731.6
Cross	420	561.6**	420	1,239.5**	900.6**
*g	36	1,564.8**	36	5,562.8**	3,563.8**
L	36	509.1**	36	440.5**	474.8**
*s	24	2,090.8**	24	2,039.4**	2,065.1**
LL	24	155.8	24	171.0	163.4
m	36	1,224.0**	36	2,846.9**	2,035.4**
r	36	215.9	36	306.1	261.0
aa	72	237.2	72	445.1**	341.2**
dev	156	253.7*	156	211.7	232.7
R × C	420	198.0	420	247.3	222.6

Table 10. Number of significant sets at 5% level (N^*) and 1% level (N^{**}) out of 12 sets and percentage of total variation for C_1 pupal weight among crosses attributable to genetic effects

Effect	N^*	N^{**}	(%)	N^*	N^{**}	(%)
	WLSC1			WLSC2		
*g (autosomal additive)	10	9	22.0	12	12	45.6
L (sex-linked additive)	2	1	4.2	6	3	2.8
*s (autosomal heterosis)	8	7	12.8	12	10	13.9
LL (sex-linked heterosis)	1	0	2.7	5	3	2.2
m (general maternal)	12	11	25.0	11	7	12.7
r (specific maternal)	3	2	4.1	3	1	3.0
aa (A × A epistasis)	5	3	13.2	10	5	10.2
dev (higher order epistasis)	3	1	16.1	3	1	9.6
	RRS1			RRS2		
*g (autosomol additive)	9	6	16.9	11	10	45.2
L (sex-linked additive)	5	3	6.0	4	2	5.6
*s (autosomal heterosis)	12	10	23.3	9	8	9.2
LL (sex-linked heterosis)	2	1	3.7	0	0	2.5
m (general maternal)	11	11	18.8	9	7	12.5
r (specific maternal)	2	0	3.4	0	0	1.5
aa (A × A epistasis)	5	3	9.5	3	1	9.4
dev (higher order epistasis)	3	1	18.3	1	0	14.4
	WLSR1			WLSR2		
*g (autosomal additive)	12	10	23.8	12	12	44.7
L (sex-linked additive)	5	3	7.7	4	2	3.5
*s (autosomal heterosis)	11	11	21.2	12	10	10.9
LL (sex-linked heterosis)	0	0	1.6	0	0	0.9
m (general maternal)	12	9	18.6	12	11	22.9
r (specific maternal)	2	0	3.3	1	1	2.5
aa (A × A epistasis)	0	0	7.2	6	4	7.2
dev (higher order epistasis)	3	0	16.7	1	0	7.4

C_2 populations. Mean pupal weights for C_2 two-way and three-way crosses are found in Table 11. Weights for the continued selection populations, XWLSC and XRRS, were consistently higher than those for the unselected base, XWLSR, indicating that a correlated response had resulted from the larval weight selection. In general, three-way crosses have slightly higher values than two-way crosses. In all populations, the "between" companion population two-way crosses were higher than the "within" crosses, and the greatest relative difference was observed for the XRRS populations which had been selected for high "between" crossbred performance. Similarly, three-way crosses with F_1 dams were heavier than those with inbred dams, with XRRS again showing the largest difference and XWLSC the smallest one. These differences were much smaller than those observed for larval weight.

The pooled analyses of variance for C_2 pupal weights are shown in Table 12 and the number of significant sets and percentage of variation in Table 13. Autosomal addi-

Table 11. Mean pupal weights (dμg) for C_2 two- and three-way crosses with a comparison of within- and between-companion polation two-way crosses and inbred versus F_1 dams for three-way crosses

Population	Two-way crosses			Three-way crosses		
	Within	Between	Mean	Inbred dam	F_1 dam	Mean
XWLSC1	399±2	423±1	415±2	416±1	418±1	417±1
XWLSC2	361±2	373±1	369±1	358±1	364±1	360±1
XRRS1	366±1	393±1	384±2	388±1	396±1	392±1
XRRS2	340±1	361±1	354±2	359±1	367±1	363±1
XWLSR1	326±1	338±1	334±1	334±0	341±0	337±1
XWLSR2	293±1	307±1	302±1	305±0	309±0	307±1

Table 12. Analyses of variance for C_2 pupal weight, pooled over sets and over populations with similar selection histories

Source	*df*	MS	*df*	MS	Pooled MS
	XWLSC1		XWLSC2		XWLSC
Reps	12	3,542.4	12	967.9	2,255.1
Cross	420	1,041.0**	420	881.2**	961.1**
*g	36	4,981.2**	36	1,610.2**	3,295.7**
L	36	845.6**	36	1,605.3**	1,225.5**
*s	24	2,030.2**	24	580.4**	1,305.3**
LL	24	619.1**	24	319.2	469.2**
m	36	1,324.7**	36	435.0*	879.9**
r	36	244.9	36	299.0	271.9
aa	72	723.6**	72	1,992.4**	1,358.0**
dev	156	354.2	156	403.0**	378.6**
R × C	420	291.9	420	285.5	288.7
	XRRS1		XRRS2		XRRS
Reps	11	156.3	10	918.9	537.6
Cross	385	886.2**	350	778.7**	832.4**
*g	33	3,669.9**	30	3,198.3**	3,434.1**
L	33	483.3**	30	730.3**	606.8**
*s	22	2,694.5**	20	1,830.8**	2,262.4**
LL	22	531.9**	20	324.8**	428.4**
m	33	754.4**	30	1,246.7**	1,000.6**
r	33	181.3	30	203.0	192.2
aa	66	786.2**	60	607.9**	697.1**
dev	143	352.3**	130	243.3*	297.8**
R × C	385	156.8	350	180.3	168.5
	XWLSR1		XWLSR2		XWLSR
Reps	13	750.5	12	765.2	757.8
Cross	455	467.6**	420	482.0**	474.8**
*g	39	2,316.6**	36	2,019.7**	2,168.1**
L	39	302.2**	36	205.8**	254.0**
*s	26	562.5**	24	1,308.8**	935.7**
LL	26	148.3	24	118.8	133.5
m	39	749.8**	36	956.6**	853.2**
r	39	120.4	36	183.2	151.8
aa	78	326.2**	72	266.9**	296.6**
dev	169	193.9**	156	178.3*	186.1**
R × C	455	141.8	420	138.3	140.1

Table 13. Number of significant sets at 5% level (N^*) and 1% level (N^{**}) out of 12 sets and percentage of total variation for C_2 pupal weight among crosses attributable to genetic effects.

Effect	N^*	N^{**}	(%)	N^*	N^{**}	(%)
	XWLSC1			XWLSC2		
*g (autosomal additive)	11	11	41.0	8	8	15.7
L (sex-linked additive)	6	4	7.0	11	10	15.6
*s (autosomal heterosis)	10	6	11.1	5	1	3.8
LL (sex-linked heterosis)	5	1	3.4	2	1	2.1
m (general maternal)	9	5	10.9	5	2	4.2
r (specific maternal)	1	0	2.0	1	0	2.9
aa (A × A epistasis)	7	5	11.9	12	10	38.8
dev (higher order epistasis)	1	0	12.6	5	0	17.0
	XRRS1[a]			XRRS2[b]		
*g (autosomol additive)	11	10	35.5	10	8	35.2
L (sex-linked additive)	4	4	4.7	8	6	8.0
*s (autosomal heterosis)	10	10	17.4	10	8	13.4
LL (sex-linked heterosis)	5	1	3.4	2	1	2.4
m (general maternal)	7	3	7.3	8	5	13.7
r (specific maternal)	1	0	1.8	0	0	2.2
aa (A × A epistasis)	11	8	15.2	6	5	13.4
dev (higher order epistasis)	6	4	14.8	3	2	11.6
	XWLSR1			XWLSR2		
*g (autosomal additive)	13	11	42.5	10	9	35.9
L (sex-linked additive)	5	3	5.5	0	0	3.7
*s (autosomal heterosis)	6	4	6.9	7	6	15.5
LL (sex-linked heterosis)	0	0	1.8	1	1	1.4
m (general maternal)	9	8	13.7	10	8	17.0
r (specific maternal)	1	0	2.2	2	0	3.3
aa (A × A epistasis)	5	2	12.0	4	3	9.5
dev (higher order epistasis)	2	1	15.4	4	1	13.7

[a] based in 11 sets; [b] based in 10 sets; [c] based in 13 sets

tivity (*g) again was the most important effect for all populations except for XWLSC2. The latter exhibited unusually large additive × additive and sex-linked effects similar to what was observed for larval weight (Table 7). The XRRS populations consistently had large autosomal heterosis (*s) values for pupal weight, while the XSLSR populations had the largest values for maternal effects. On the other hand, the selected WLS and RRS populations showed larger aa epistatic effects than was observed for the unselected XWLSR base.

Discussion

Background

Since our primary goal here is the comparison of different selection schemes as to their effectiveness for exploiting the different types of quantitative gene effects which determine cross-bred performance, it is worthwhile to consider the appropriateness of our genetic model and the populations examined.

The general genetic model proposed by Carbonell et al. (1983) assumed disomic species, a fixed set of random mating populations, Wright's genotypic frequencies at every locus both autosomal and sex-linked, linkage equilibrium, and additive × additive epistasis among autosomal loci.

The genetic mechanisms in *Tribolium castaneum* are known to include diploid inheritance, nine pairs of autosomes, plus a single X or sex chromosome in males, with gene recombination occurring in both sexes. Since the number of genes influencing *Tribolium* larval and pupal weights has been estimated to be as large as 600 (Bell 1969), many will assuredly be linked.

When this study was initiated, the unselected B and P base populations (UNSR crosses) had been random mated for more than 100 generations, and the relaxed selection C_1 and C_2 populations (WLSR and XWLSR crosses, respectively) had been random mated for 28 generations. Therefore, these populations qualify well for the assumptions of linkage equilibrium and Wright's genotypic frequencies. The continued selection of C_1 and C_2 populations, whereby RRS and WLS were compared over 23 generations and their respective crosses (WLSC, RRS, XWLSC and XRRS), satisfied less well the above two assumptions. Nevertheless, the random mating of all selected populations for 5 generations prior to the extraction of inbred lines would have brought these populations close to equilibrium, except for tightly linked loci, and made the theoretical assumptions more tenable.

In regard to statistical analyses, the crosses between four inbred lines which constituted a "set" (Table 1) can be considered as fixed, given the way the inbred lines were chosen, i.e., two lines from each of two "companion" populations. The large number of sets per replication provides a rather powerful test when pooled over sets. And when populations with similar selection histories are pooled, inferences can be made as to the genetic architecture of the trait and comparisons between selection methods are valid. Furthermore, given that no significant "Replication × cross" interactions were found in our preliminary analyses, tests for statistical significance are the same, regardless of the random or fixed nature of the "Cross" effects.

The genetic parameters found in this study for the various populations with different selection histories provide important evidence as to the nature of quantitative gene action and how different gene effects responded to different selection methods. Yet the results from specific sets of crosses involving only four inbred lines (Table 1) varied as one should expect. This fact points to the importance of evaluating many sets. The analysis pooled over sets will be more indicative of the general genetic situation for each population or selection method. Since small F values for these analyses were enough to declare statistical significance, alternative

Table 14. Ratios of F values testing the significance of genetic effects in populations with different selection histories. Larval weight

Effect	RRS/WLSC	RRS/WLSR	RRS/UNSR	WLSC/WLSR	WLSC/UNSR	WLSR/UNSR
*g	1.21	0.85	0.69*	0.70*	0.57**	0.81
L	1.54*	0.89	1.16	0.58**	0.75	1.30
*s	1.53*	0.66*	4.89**	0.49**	3.67**	7.44**
LL	1.76*	1.58*	0.68	0.90	0.39**	0.43**
m	0.97	0.72*	0.98	0.74*	1.02	1.37*
r	1.26	1.66*	1.26	1.32	1.00	0.76
aa	1.11	1.30	0.67*	1.17	0.60**	0.52**
dev	1.33**	1.18	0.85	0.89	0.64**	0.72
	XRRS/XWLSC	XRRS/XWLSR	XRRS/UNSR	XWLSC/XWLSR	XWLSC/UNSR	XWLSR/UNSR
*g	1.60**	1.93**	0.52**	1.21	0.33**	0.27**
L	0.52**	1.47*	1.09	2.82**	2.88**	0.74*
*s	2.20**	1.52*	2.68**	0.69*	1.22	1.76**
LL	1.15	1.57*	0.74	1.36	0.64*	0.48**
m	1.44*	0.94	0.75	0.66*	0.52**	0.79
r	1.86**	2.14**	1.47	1.15	0.78	0.69
aa	0.75	1.48*	0.63**	1.98**	0.85	0.43**
dev	1.40*	1.24	0.99	0.88	0.71*	0.80
	RRS/XRRS	WLSC/XWLSC	WLSC/XWLSC1	WLSR/XWLSR		
*g	1.32	1.73**	1.36*	3.00**		
L	1.07	0.36**	0.93	1.77**		
*s	1.82**	3.02**	1.89**	4.22**		
LL	0.92	0.60	0.56*	0.90		
m	1.31	1.95**	1.64**	1.74**		
r	0.86	1.28	1.31	1.10		
aa	1.06	0.71*	1.74**	1.21		
dev	0.86	0.91	0.96	0.90		

measures are needed to be more indicative of the relative importance of various gene effects. Comparisons based on the number of significant sets and the percentage of total variation were presented in Tables 4, 7, 10 and 13. In addition, a comparison between the magnitude of the corresponding F values for each genetic effect can be made between populations and selection methods by using the method presented by Bradley and Schumann (1956) and Schumann and Bradley (1959). This method is preferred to that by Stuber and Moll (1971) using ratios of corresponding genetic mean squares. The latter method does not account for the relative importance of the pooled error mean squares in the two experiments; hence, the comparisons might be misleading when the error mean squares are substantially different. Furthermore, it provides no significance level to be stated. The Bradley-Schumann Method is used when, as in our case, two parallel experiments are distinct and independent, and have separate samples from the same treatments. The method is based on the comparison of two independent non-central variance ratios from the two parallel experiments and is simply calculated by the ratio of the two corresponding F values from both ANOVA tables. Ratios of F values for genetic effects between populations for larval weight are presented in Table 14 and for pupal weight in Table 15.

Larval weight comparisons

In the upper third of Table 14 the comparisons for the C_1 populations reveal that the unselected UNSR populations had significantly higher variation due to *g effects than WLSC or RRS but similar variation to the relaxed selected population WLSR. The selected RRS and WLSC populations had similar values. These results can be explained by the simple model proposed by McNew and Bell (1976) for these populations. They suggested two kinds of gene loci – one consisting of additive loci with partial or complete dominance and a second with overdominant or heterotic loci. Both RRS and WLS are predicted to move the gene frequencies of both additive and dominant alleles towards homozygosity and WLS was more effective than RRS. On the other hand, frequencies in WLSR have regressed towards those in UNSR due to the relaxation of selection.

The fact that selected populations had lower variation due to *g effects than unselected ones agrees with findings by Stuber and Moll (1971) for grain yield in

Table 15. Relations of *F* values testing the significance of genetic effects in populations with different selection histories. Pupal weight

Effect	RRS/WLSC	RRS/WLSR	RRS/UNSR	WLSC/WLSR	WLSC/UNSR	WLSR/UNSR
*g	0.65*	0.77	0.82	1.19	1.26	1.06
L	1.23	1.01	1.52*	0.82	1.24	1.51*
*s	0.79	0.91	4.45**	1.15	5.59**	4.88**
LL	0.88	2.32**	2.45**	2.62**	2.77**	1.06
m	0.61**	0.61**	1.06	1.01	1.75**	1.73**
r	0.47**	0.73	0.56*	1.55*	1.19	0.77
aa	0.58**	1.12	1.01	1.95**	1.76**	0.90
dev	0.93	1.31	1.05	1.41*	1.13	0.80
	XRRS/XWLSC	XRRS/XWLSR	XRRS/UNSR	XWLSC/XWLSR	XWLSC/UNSR	XWLSR/UNSR
*g	1.78**	1.32	1.34	0.74	0.75	1.02
L	0.85	1.99**	2.55**	2.34**	3.01**	1.28
*s	2.97**	2.01**	7.06**	0.68*	2.38**	3.52**
LL	1.57*	2.67**	3.68**	1.71*	2.34**	1.38
m	1.95**	0.98	1.12	0.50**	0.58**	1.15
r	1.21	1.06	0.75	0.87	0.62*	0.71
aa	0.88	1.95**	2.44**	2.22**	2.76**	1.25
dev	1.35*	1.33	1.35*	0.98	1.00	1.02
	RRS/XRRS	WLSC/XWLSC	WLSC/XWLSC1	WLSR/XWLSR		
*g	0.61**	1.67**	1.20	1.03		
L	0.60**	0.41**	0.60**	1.18		
*s	0.63**	2.35**	1.52*	1.39		
LL	0.67	1.18	0.90	0.77		
m	0.94	3.04**	2.19**	1.50*		
r	0.74	1.93**	2.15**	1.08		
aa	0.42**	0.64**	1.20	0.72*		
dev	0.78	1.13	1.22	0.79		

maize. Although their pooled *F* values for three generations of RRS were slightly higher than those for the unselected population, they were really due to a few very significant sets in RRS. Many more of their unselected sets had significant **g* effects than were observed among the RRS sets.

All C_1 populations revealed important **s* effects for larval weight (Table 3 and 4), with the Table 14 contrasts showing UNSR to be the smallest and, surprisingly, the relaxed WLSR populations to be the largest. Apparently, selection on purebred performance had acted favorably on heterotic as well as additive effects, yet the heterotic loci had not reached fixation. RRS had significantly higher **s* effects than WLS, as expected from the earlier study by Rich and Bell (1980) with these same C_1 populations. They observed a significantly greater heterosis for larval weight of RRS crossbred cultured in both good and poor nutritional environments.

Maternal effects and *aa* epistatic effects were highly significant in all populations (Table 3 and 4), but they seem to be more important in the unselected than in the selected populations, as contrasted in Table 14. No significant ratios were found among the RRS/WLCS selected populations. Apparently, selection was not successful in identifying good *aa* epistatic combinations for better crossbred performance. It is interesting to note that among the selected populations, RRS accumulated more higher order epistatic combinations (*dev*) than was observed for the rest of the selected populations (assuming the rest of causes mentioned at the end of the "Statistical model" section to be of similar magnitude through populations).

For the C_2 populations, the ratios between *F* values for larval weight as listed in the mid-section of Table 14 indicate that selected C_2 populations had lower variation due to **g* effects than unselected UNSR, and XWLSR showed the smallest value. When the C_1 populations were crossed to initiate these heterogeneous C_2 populations, gene frequencies in the newly formed crossbred populations were expected to be the average of the parental frequencies. Loci that had the same allele fixed in the companion C_1 populations would remain fixed in the new population; however, previous selection could have fixed different desirable alleles in the two populations. The latter would result in gene frequencies close to 0.5. for desirable alleles at unfixed loci in the new C_2 populations. Renewed selection would again increase gene frequencies of desirable alleles with corresponding

changes in genetic variances depending on degree of dominance. Theoretically, WLS would be more effective than RRS for exploiting additive gene effects. As found from the subsequent selection experiment (McNew and Bell 1976), considerable response resulted from WLS. After 23 generations of selection response plateaus were not apparent, suggesting that favorable alleles were still away from fixation. Theoretically, RRS should be operative for heterotic *s effects as well as for additive ones. Yet, the overall response of XRRS was less than that of XWLSC, even though selection intensities were the same. Apparently, gene frequencies at additive loci were changed less in the XRRS populations, which in turn might result in the higher additive variances, provided one assumes the initial gene frequencies were near 0.5. Also, as RRS moved frequencies at heterotic loci from their equilibrium values, additive variation would theoretically increase. Evidence supporting this point comes from the significantly larger variation due to *g effects found in the mid-third of Table 14 for XRRS in comparison with XWLSC and WLSR.

The variation due to *s effects for XRRS in Table 14 was significantly higher than that for XWLSC or XWLSR and all of them were higher than those for unselected UNSR, indicating that the relative importance of autosomal heterotic effects for crossbred performance was enhanced as a consequence of RRS selection. It seems clear that in the new C_2 populations, RRS was more efficient than WLS in moving apart gene frequencies of dominant or overdominant loci, so that XRRS had larger *s values than XWLSC.

The interpretation of the *aa* epistatic effects is difficult because of the unique behavior of the XWLSC2 population for both larval and pupal weights (Tables 7 and 13). If one excludes the results from XWLSC2, then similar patterns as those for C_1 populations were found for *aa* effects, with selected RRS and WLS populations showing similar values and all lower than those of the unselected populations. The peculiar response for XWLSC2 merits further discussion.

Given that epistatic combinations are characteristic of a population, then an extremely good or bad epistatic combination might be brought about by chance when two populations are crossed. One important point indicating that some unusual gene combinations were present in the XWLSC2 populations is the fact that both larval and pupal weights for three-way crosses were always smaller for XWLSC2 than those for two-way crosses, while the reverse was true for all other populations (Tables 5 and 11). It can be shown that the difference between three- and two-way crosses is $(aa_{12}+aa_{13}+aa_{14}+aa_{23}+aa_{24}+aa_{34})/24-(L_1+L_3)/24$. Upon calculation of the values of the *aa* and L effects for these populations (not shown here), it was found that all *aa* values were positive for all populations except for XWLSC2. The XWLSC2 population had positive values for aa_{13} and aa_{24} and negative ones for the remaining *aa* terms. Hence, with such values, a high additive × additive epistatic variance will be obtained. Furthermore, both L_1 and L_3 were also positive, and so the above expression will have a negative sign. Unfortunately, the XWLSC2 inbred lines were discarded before these extreme epistatic combinations were identified. Otherwise a search would have been made for major gene involvement. In an earlier study of *Tribolium* growth on high and low nutrition, we were able to identify a single autosomal gene (*cos*) as responsible for an unexpectedly large G × E interaction response (Costantino et al. 1967).

When C_1 and C_2 populations are compared in the lower part of Table 14, it is found that the diverse and long-time selected C_1 populations had slightly higher variation due to *g effects in all paired comparisons than in the newly formed heterogeneous C_2 populations. This may have resulted from the more diverse origins for the C_1 populations, or it could have arisen from higher frequencies for favorable alleles at more loci in the C_2 selected poulations. When the C_1 populations were crossed to form the initial C_2 populations, new genetic variation resulted for each of the new heterogeneous populations. Furthermore, the highly selected C_1 populations most likely had fixation for some undesirable alleles which were linked to favorable ones. Such linkage combinations may have been broken by recombination in the C_2 populations. Thereby new selection limits would be possible at higher levels of gene frequencies. Results from McNew and Bell (1976) indicate that a larger response to selection was actually found for C_2 as compared with C_1 populations.

Regarding heterotic variation due to *s effects, C_1 populations again had larger values across all comparisons. Since the paired "companion" C_2 populations had been isolated as black versus wild-type body color segregants from the same advanced-generation C_1 crossbreds, they should have been genetically similar except for genes closely linked to the *b* locus. Hence, heterosis should be less important for C_2 crosses than for C_1 crosses that had genetically divergent origins. This fact is clear from the experimental results reported by McNew and Bell (1976). Crossbred responses for C_1 populations were consistently superior to purebred responses, whereas purebred and crossbred C_2 responses were more similar. This suggests that C_2 selections were mostly based on additive effects, whereas C_1 crossbred responses were based to a larger degree on heterotic loci. Gene frequencies at the heterotic loci were moved towards fixation of different alleles in the companion populations much more efficiently in the RRS than in XRRS populations, so that the cross would show larger heterosis. This is confirmed when comparing the mean larval weight differences for "within" and "between" companion population two-

way crosses for both populations (Tables 2 and 5). The relative increment for C_1 RRS is 37.3%, whereas for C_2 XRRS it is only 17.6%.

No consistent differences for variation due to *aa* epistatic effects are found in Table 14 between C_1 and C_2 populations under RRS. If XWLSC2 is excluded, then C_1 WLS populations had significantly larger *aa* variation but the opposite is true for pooled populations. Therefore, no general conclusion can be found for this type of gene action.

Pupal weight comparisons

Regarding pupal weight ratios (Table 15), the differences between base populations (UNSR) and selected populations (RRS and WLS) were not as large as those observed for larval weight. This arises in part from the fact that pupal weight values are correlated responses. In addition, the trait has its own unique genetic architecture.

For the C_1 ratios (upper part, Table 15), the only significant difference observed for $*g$ effects was between RRS and WLSC populations. Presumably, additive gene frequencies had been less than 0.5 in the base populations, and the greater response from WLS had increased the frequencies of favorable alleles to a point where the additive genetic variance was higher than that for the other populations. Both WLS and RRS had operated upon heterotic loci in such a way that $*s$ effects had been enhanced over the unselected populations. Yet only the WLSC populations showed a significant increase for *aa* epistasis.

For the C_2 comparisons (mid-section, Table 15), the general trends for pupal weight were also similar to those observed for larval weight, with minor exceptions. The $*g$ and $*s$ variations were again more important for populations under RRS; however, $*s$ variation was never larger than the $*g$ one, while the reverse was true for larval weight. Also, epistatic action was relatively less important for pupal weight than for larval weight.

When comparing C_1 and C_2 populations, it is clear that because of the newly created variability in the C_2 populations, gene frequencies were moved more efficiently in C_2 than in C_1 populations. The smallest pupal weight $*g$ variation was observed for XWLSC, which implies that additive gene frequencies had moved toward fixation as a correlated response to selection.

While larval weight is considerably influenced by non-additive as well as additive gene effects, pupal weight is primarily influenced by additive effects, with dominance and epistatic effects playing minor roles. This point was demonstrated conclusively by the long-term pupal weight selection studies of Bell and Moore (1972). They found WLS much superior to RRS for improving this highly heritable trait. Improved RRS crossbred performance was highly correlated with purebred performance as expected with additive gene effect. Theoretically, this correlation can be negative when overdominance or higher order epistasis prevails (McNew and Bell 1971). Such was the case reported by Brown and Bell (1980) where a negative genetic correlation between purebred and crossbred performance for *Drosophila* egg numbers was confirmed experimentally, in that purebred performance actually declined under RRS selection while crossbred egg numbers increased. Crossing highly selected populations increases the opportunity for WLS to yield still higher responses by exploiting the increased additive genetic variance present in the new crossbred population. For RRS to excel over WLS, the "companion" populations should have diverse origins and non-additive gene effects ($*s$) must be more important than additive effects ($*g$). This latter point was demonstrated clearly by the selection experiments described by Orozco and Bell (1974).

References

Anderson VL (1970) Restriction errors for linear models. (An aid to develop models for designed experiments.) Biometrics 26:255–268

Bell AE (1969) The nature of selection responses in *Tribolium*. Jpn J Genet 44:299–309

Bell AE (1981) On the utilization of total genetic variation, an overview. Génétique quantitative et appliquée des populations croissées, Toulouse. Colloq INRA 10:107–123

Bell AE (1982a) The *Tribolium* model in animal breeding research. Proc 2nd World Congr Genet Appl Livestock Prod 5:26–42

Bell AE (1982b) Selection for heterosis, results with laboratory and domestic animals. Proc 2nd World Congr Genet Appl Livestock Prod 6:206–227

Bell AE, Moore CH (1972) Reciprocal recurrent selection for pupal weight in *Tribolium* in comparison with conventional methods. Egypt J Genet Cytol 1:92–119

Bell AE, Moore CH, Bohren BB, Warren DC (1952) Systems of breeding designed to utilize heterosis in the domestic fowl. Poult Sci 31:11–22

Bradley RA, Schumann DEW (1956) The comparison of the sensitivities of similar experiments: Applications. Biometrics 13:496–510

Brown WP, Bell AE (1980) An experimental comparison of selection alternatives to plateaued responses. Genetics 94:477–496

Carbonell EA, Nyquist WE, Bell AE (1983) Sex-linked and maternal effects in the Eberhart-Gardner general genetics model. Biometrics 39:607–619

Carbonell EA, Frey JJ, Bell AE (1985) Estimation of maternal, sex-linked and additive × additive epistatic gene effects for body size of *Tribolium*. Theor Appl Genet 70:133–137

Comstock RE, Robinson HF, Harvey PH (1949) A breeding procedure designed to make maximum use of both general and specific combining ability. Agron J 41:360–367

Costantino RF, Bell AE, Rogler JC (1967) Genetic analysis of a population of *Tribolium*. 1. Corn oil sensitivity and selection response. Heredity 22:529–539

Dearborn DD, Gregory KE, Cundiff LV, Koch RM (1987) Maternal heterosis and grandmaternal effects in beef cattle: Preweaning traits. J Anim Sci 65:33–41

Dickerson GE (1969) Experimental approaches in utilizing breed resources. Anim Breed Abstr 37:191–202

Eberhart SA, Gardner CO (1966) A general model for genetic effects. Biometrics 22:864–881

Eisen EJ, Hortsgen-Schwark G, Bandy TR, Saxton AM (1985) Diallel cross among lines of mice selected for litter size and body weights: Body composition traits. Z Tierz Züchtungsbiol 102:10–22

Kinghorn B (1982) Genetic effects in crossbreeding. 1. Models of merit. Z Tierz Züchtungsbiol 99:59–68

Kinghorn B (1983) Genetic effects in crossbreeding. 3. Epistatic loss in crossbred mice. Z Tierz Züchtungsbiol 100:209–222

Kinghorn B (1987) The nature of 2-locus epistatic interactions in animals: evidence from Sewal Wright's guinea pig data. Theor Appl Genet 73:595–604

McNew RW, Bell AE (1971) The nature of the purebred-crossbred genetic covariance. Genet Res 18:1–7

McNew RW, Bell AE (1974) Crossbred response from purebred selection, and experimental check on selection theory with *Tribolium*. Theor Appl Genet 44:100–105

McNew RW, Bell AE (1976) Comparison of cross bred and purebred selection for a heterotic trait in highly selected populations of *Tribolium*. J Hered 67:275–283

Melchinger AE, Geiger HH, Schnell FW (1986) Epistasis in maize (*Zea mays* L.). 2. Genetic effects in crosses among early flint and dent inbred lines determined by three methods. Theor Appl Genet 72:231–239

Orozco F, Bell AE (1974) Reciprocal recurrent selection compared to within-strain selection for increasing rate of egg lay of *Tribolium* under optimal and stress conditions. Genetics 77:143–161

Rich SS, Bell AE (1980) Genotypic–environment interaction effects in long-term selected populations of *Tribolium*. J Hered 71:319–322

Schumann DEW, Bradley RA (1959) The comparison of the sensitivities of similar experiments: Model II of the analysis of variance. Biometrics 15:405–416

Sheridan AK (1980) A new explanation for egg production in crosses between White Leghorns and Australorps. Br Poult Sci 21:85–88

Stuber CW, Moll RH (1971) Epistasis in maize (*Zea mays* L.). 2. Comparison of selected with unselected populations. Genetics 67:137–149