

**BUREAU OF SUGAR EXPERIMENT STATIONS
QUEENSLAND, AUSTRALIA**

**FINAL REPORT
SRDC PROJECT BS45S
OPTIMUM FAMILY SELECTION
FOR NET MERIT GRADE
IN STAGE 2 TRIALS**

**by
M C Cox
SD96007**

Principal Investigator: Dr M C Cox
Senior Research Officer
BSES
PO Box 651
BUNDABERG QLD 4670

This project was funded by the Sugar Research Council/Sugar Research and Development Corporation during the 1990/91, 1991/92, 1992/93 and 1993/94 financial years.

**BSES Publication
SRDC Final Report SD96007**

August 1996

CONTENTS

	Page No
1. SUMMARY	1
2. BACKGROUND	1
3. PROJECT OBJECTIVES	2
4. INTRODUCTION	2
5. METHODOLOGY	2
5.1 Origin of genetic material	2
5.2 Stage 3 trials	3
5.3 Statistical analyses	4
6. RESULTS AND DISCUSSION	4
6.1 Stage 2 analyses	4
6.2 Stage 3 analyses of variance	5
6.3 Correlation between stage 2 and stage 3 family means	6
6.4 Family means and clonal distributions	6
6.5 Simulated selection	7
7. DIFFICULTIES	9
8. RECOMMENDATIONS FOR FUTURE RESEARCH	9
9. PUBLICATIONS	10
10. REFERENCES	10
11. ACKNOWLEDGMENTS	11
APPENDIX A - Families, parents and NMG categories in Experiments 1 and 2	12
APPENDIX B - Mean squares and significance levels for individual analyses of variance	13
APPENDIX C - Mean squares and significance levels for combined analyses of variance	14
APPENDIX D - Mean NMG of random and selected clones in stage 3	16
APPENDIX E - Mean NMG of random and selected clones in stage 3	17
APPENDIX F - Family selection in stage 2 - combined results	18
APPENDIX G - Frequency of stage 3 clones with acceptable (>9) NMG	19

1. SUMMARY

Mass selection of individuals in seedling or early clonal stage trials is routinely used in most sugarcane improvement programs throughout the world. It is, however, inefficient as the heritability of cane yield on a single plant basis is low. In Australia, the introduction of mobile truck-mounted weighing equipment offered the opportunity to implement family selection utilising weighed family data. Family selection has been used in some Bureau of Sugar Experiment Stations' (BSES) selection programs since 1986 and is now routinely used in all regional selection programs. This research has demonstrated that a combination of family and mass selection in stage 2 of selection will result in larger genetic gains and a higher frequency of superior clones in later stages than mass or family selection alone. This combination allows improved efficiency since fewer resources are required to select only within superior families in the first ratoon crop than to mass select individuals across the entire population. A liberal family selection rate (about 40%) balances genetic gain and the need to maintain a broad genetic base. While mass selection rates did not vary in this work, it is believed that differential selection rates within families should be used so that more clones are selected out of the best families. The availability of objective family data also allows more accurate estimation of the breeding value of parents utilising best linear unbiased predictors (BLUP). This results in better genetic combinations through crossing and provides more objective information on new parents.

2. BACKGROUND

Selection in early stages of a sugarcane breeding program has been described as being very inefficient (Skinner, 1971). Selection on an individual plant basis (mass selection) is likely to be confounded by environmental effects. Thus genotypic values are difficult to assess and heritability is low. Consequently selection rates in seedling stages of Australian cane breeding programs are high (10 to 30%) to reduce the possibility of discarding superior varieties.

In Stage 2 4-sett plots harvested at Bundaberg Sugar Experiment Station in 1989, mean Net Merit Grade (NMG) for almost 200 families ranged from 1.5 to slightly greater than 10. The mean of the standard clones, also planted as for family plots, is given a NMG of 10.

A considerable number of experimental crosses are planted each year, and must be evaluated for suitability. If family selection on the basis of mean NMG is to be efficient, information is required on the level of NMG below which families should be rejected. If the cut off point is too high, families capable of producing superior clones will be lost. If it is too low, the efficiency of the breeding program is reduced because of wasted resources on families unlikely to produce superior clones. It is believed that fairly liberal selection of families may be necessary, but this needs to be defined more precisely.

Since visual selection will be practised within families as a normal part of the program, quantification of the response to this selection is needed.

3. PROJECT OBJECTIVES

- Determine the optimum family mean NMG in Stage 2 (4-sett plots) to select with efficiency the families most likely to produce superior sugarcane clones.
- Determine the response to visual selection within families.

4. INTRODUCTION

Breeding of more productive sugarcane clones is widely acknowledged as one of the most important methods for improving the profitability of sugarcane industries. The investment in breeding is large and optimising the efficiency of programs is essential to maximise the return on that investment. The success of a clonal improvement program is highly dependent on an efficient selection system in the early stages of the program.

5. METHODOLOGY

5.1 Origin of genetic material

The families and clones for the two experiments were derived from bunch seedlings (stage 1) planted in 1987 (Experiment 1) and 1988 (Experiment 2). Clones selected from these seedlings in 1988 and 1989 were planted in replicated family plots, where each family plot consisted of 10 clones (stage 2). Each clone within a family plot was planted as a 2 m (4-sett) plot with a small gap (0.2 m) between clones. Thus each family plot consisted of a single row 22 m in length (2.2 x 10). The number of replications of each family varied, depending on the number of seedlings available for selection. Standard clones, planted as for the family plots, were included in the trials.

The plant crops of these stage 2 families were harvested in 1989 and 1990. Family plots were sampled for CCS just prior to harvest. Five stalks, one from each of five randomly chosen clones per family plot were crushed to give family based juice measurements. Whole family plots were harvested and weighed. From these data, cane yield (TCH), CCS, sugar yield (TSH), and NMG were calculated. These data were used to choose families from a range of NMG categories to examine the effectiveness of family selection.

Six families were randomly chosen from each of six NMG categories in each year. These NMG categories for Experiment 1 (derived from 1987 seedlings) and Experiment 2 (derived from 1988 seedlings) are shown in Table 1. In 1990, there were insufficient families with NMG <4 and the lowest category had to be raised to <5.

Table 1
Stage 2 Net Merit Grade (NMG) categories of families in Experiments 1 and 2

NMG Category	
Experiment 1	Experiment 2
<4	<5
4-5	5-6
5-6	6-7
6-7	7-8
7-8	8-9
>8	>9

The families, their parents, and the NMG Category are given in Appendix A.

In the first ratoon crop of the 36 families in each experiment, 12 clones were randomly chosen while the best 12 clones based on visual grade were also selected. Thus a total of 864 clones were selected for planting in each experiment as stage 3 (20 sett) plots.

5.2 Stage 3 trials

In the stage 3 trials, a split plot design was used. The whole plot was allocated to families, represented by eight clones, four of which were randomly selected and four of which were visually selected. Thus there were three replications of each family to make up the 24 clones per family. Clones were not replicated. In Experiment 1, a randomised complete block design was used while in Experiment 2, a completely randomised design was inadvertently used. Each clone was planted in a row 10 m in length with 1.5 m between rows, giving a family plot consisting of 8 rows x 10 m. In both experiments, three standard varieties (CP51-21, Q137, and Q141) were included. These were planted as for a family plot with 8 rows x 10 m plots of each in each replicate.

Both experiments were planted on the Bundaberg Sugar Experiment Station which has a Krasnozem (red volcanic) soil type. Experiment 1 was planted in spring (August 8, 1990) while Experiment 2 was planted in autumn (April 17, 1991). Both experiments were planted with 300 kg/ha of CropKing 33 and received a further 200 kg/ha of urea on December 5, 1990 (Exp 1) and October 14, 1991 (Exp 2). The plant crops were harvested on August 26, 1991 and October 15, 1992, respectively. In the first ratoon crops, 300 kg/ha of CropKing was applied about one month after harvest and a further 300 kg/ha of urea was applied on October 30, 1991 (Exp 1) and December 2, 1992 (Exp 2). The first ratoon crops were harvested on November 3, 1992 and August 4, 1993.

Both experiments were grown under irrigated conditions. Experiment 1 received eight irrigations in the plant crop totalling 1 250 mm while the first ratoon crop received five irrigations totalling 518 mm. Experiment 2 received nine irrigations in the plant crop totalling 956 mm while the first ratoon crop received nine irrigations totalling 554 mm. Both spray (early) and flood irrigation were used.

Each clone was tested for CCS by taking a two-stalk sample just prior to harvest. The stalks were crushed in a small mill and normal juice measurements taken (BSES, 1984). The trials were harvested with a mechanical harvester and plot weights were recorded. From this cane yield, sugar yield (cane yield * CCS/100) and Net Merit Grade (NMG) were calculated.

5.3 Statistical analyses

Analyses of variance for each crop-year and over crop-years were conducted and family main effects and interactions were further partitioned into NMG groups and families within NMG groups. Correlations between stage 2 and stage 3 family data were estimated. Simulated selection of families with and without mass selection was evaluated on a mean NMG basis (relative to random selection) and on the basis of frequency of elite clones.

6. RESULTS AND DISCUSSION

6.1 Stage 2 analyses

Stage 2 family plots harvested in 1989 and 1990 normally had variable replication. The NMG of 203 families in 1989 ranged from 1.5 to 10.2, while in 1990 the NMG of 193 families ranged from 2.5 to 11.8. In 1989, 31 of the 36 families were selected from block K3 where 59 families were grown in a randomised complete block trial with four replications. One standard variety, repeated five times per replication was also included. The analysis of variance of NMG for this trial is shown in Table 2.

Table 2
Analysis of variance of NMG in a stage 2 family trial (K3, harvested 1989)

Source	df	Mean square
Blocks	3	8.03**
Families	59	9.10**
Error	193	1.30

** $P \leq .01$

Coefficient of variation = 16.8%

The other five families in Exp. 1 were in two other blocks (B4 and K2). One way analyses indicated families differences were highly significant. Coefficients of variation were 20.9% and 24.3%. Similarly in 1990, families with variable replication were grown in several blocks, with the 36 families in Exp. 2 coming from three blocks (K1, G, K9). In one-way analyses of variance of NMG, family differences were highly significant ($P \leq 0.01$) and coefficients of variation ranged from 16.4% to 20.7%.

6.2 Stage 3 analyses of variance

Analyses of variance of the plant and first ratoon crops of each experiment were conducted. Differences among families were highly significant ($P < 0.01$) for cane yield (TCH), CCS, sugar yield (TSH) and net merit grade (NMG) in each analysis. Family effects were partitioned into those due to NMG groups and those due to families within NMG groups. Both sources of variation were again highly significant ($P < 0.01$) for all traits in each analysis, indicating that there were differences in the stage 3 performance of groups of families with similar NMG in stage 2, but that there were also significant differences among families within the same NMG group.

The effect of selection type (random or visually selected) was significant for TCH, TSH and NMG ($P < 0.05$) in all cases except for TCH in the plant crop of Exp 1. As expected, there were no significant selection type effects for CCS. Family x type interactions were generally not significant, except for TSH (Exp 1, P crop), and CCS and NMG (Exp 2, 1R crop). Coefficients of variation ranged from 25 to 31% for TCH, 6.4 to 7.7% for CCS, 26.2 to 32.1% for TSH, and 29.0 to 36.3 % for NMG. Mean squares and significance levels are shown in Appendix B.

Concern about the lack of blocking in Exp 2 was investigated through the analysis of the standard varieties (which were blocked correctly) and through the analysis of eight families which were blocked correctly by chance. The probability of a significant block effect together with the coefficients of variation for these analyses are shown in Table 3. These indicated that, in general, significant block main effects were not common except in the first ratoon crop. However, no significant block effects were found for NMG, the main selection criterion.

Table 3
Probability of F for blocks, and coefficients of variation from analyses of variance

Trait	Standard Varieties		8 Families	
	P	1R	P	1R
TCH	0.309 (13.0) ¹	0.383 (13.4)	- (27.1)	0.009 (31.1)
CCS	0.216 (2.6)	0.028 (3.5)	0.049 (6.5)	0.020 (7.6)
TSH	0.376 (13.5)	- (13.8)	0.252 (27.9)	0.024 (32.8)
NMG	0.415 (13.9)	- (14.3)	0.200 (30.1)	0.057 (37.0)

¹ Probability (Coefficient of variation)

- F value < 1.0

Combined analyses of variance over years were conducted for each experiment and mean squares and significance levels are shown in Appendix C. Highly significant main effects of years and families occurred for all traits. The family x year interaction was not significant for TCH and was significant for CCS and TSH in Exp 1 only. However family x year was significant for NMG in both experiments. Selection type (selected based on visual appraisal of yield, or random) was significant for TCH but not for CCS, as would be expected. There were significant family x type interactions for TCH, TSH, and NMG in both experiments. These interactions were associated with families within groups in Exp 1, but in Exp 2 were associated with NMG groups. This indicated that, in Exp 2, there was a differential response to visual selection depending on NMG category of the families. As will be shown later, the greatest response to visual selection appeared to be in families in the lowest (<5) and highest (8-9, >9) NMG categories and least in intermediate groups. There were no significant type x year or family x type x year interactions for any trait.

6.3 Correlation between stage 2 and stage 3 family means

Correlation analyses were conducted between stage 2 family means and the mean stage 3 performance of each family (based on 24 clones) for each trait (Table 4).

Table 4
Correlation coefficients between stage 2 and stage 3 family means

Trait	Experiment 1			Experiment 2		
	P	1R	Mean	P	1R	Mean
TCH	0.67**	0.54**	0.67**	0.61**	0.42*	0.57**
CCS	0.70**	0.54**	0.69**	0.65**	0.73**	0.73**
NMG	0.62**	0.44**	0.60**	0.42*	0.38*	0.45**

These data showed that family performance data based on 4-sett plots (stage 2) predicted the average performance of clones in stage 3 quite well, particularly given that each stage was grown in a different year, using very different family plot structure.

6.4 Family means and clonal distributions

The mean NMG of random and selected clones in each Stage 2 NMG group is shown in Appendix D, with the mean over plant and first ratoon crops plotted in Appendix E. The results clearly show an improved average performance of clones in families with higher stage 2 NMG. Random and selected clones improved by about 20% from lowest to highest NMG group in Exp 1, and by about 14% in Exp 2. There were some anomalous results in the random clones in Exp 2, with the 8-9 NMG group having a low mean NMG in stage 3 in both plant and first ratoon crops. This anomaly could be due to population sampling limitations. Twelve random clones represented each family, and it is possible that, by chance, this NMG group (6 families, 12 clones/family) was a biased sample, lower than the true population mean. This was supported to some extent by the larger increase in NMG of

selected clones over the random clones in this NMG group (13%) compared with other NMG groups (average of 4%). As mentioned previously (6.2), response to mass selection appeared to greater in poor (<5) and good (>8) stage 2 families than intermediate families in Exp 2 (Appendix E). Combining the results of the two experiments yields five stage 2 NMG groups (<5, 5-6, 6-7, 7-8, >8). The mean NMG for random and selected stage 3 clones is shown in Appendix F. This again illustrates the superiority of families with higher stage 2 family mean NMG and also that visual selection is effective.

The frequency of clones that would be considered for selection from stage 3 (first clonal stage) to stage 4, arbitrarily taken as those with a NMG greater than 9, was plotted against the stage 2 NMG (Appendix G). Data from both experiments was pooled. For random clones (family selection only), the frequency of these clones rose from 25% in the lowest stage 2 NMG group to 38% in the highest group. For selected clones (family and mass selection), the frequency rose from 26% to 52%.

6.5 Simulated selection

The percent gain in NMG over the random population for mass selection, family selection, and combined family and mass selection is shown in Table 5. These results show that gains in NMG from family selection alone were of the order of 2-6% except where selection intensity was high (0.17, 6-9%), while gains of 9-16% resulted from a combination of family and mass selection. These gains occurred when family selection rates of 17-50% were used. Mass selection rates were confined to 50% in these experiments.

Table 5
Percentage gain in net merit grade (NMG) over the NMG of randomly selected stage 3 clones for different selection strategies [proportion selected in parentheses]

Stage 2 selection		Gain (%) over random stage 3 clones	
Family	Mass	Exp 1	Exp 2
No	Yes	3.4	5.3
Yes (0.50)	No	4.0	2.6
Yes (0.33)	No	5.6	1.5
Yes (0.17)	No	8.5	5.7
Yes (0.50)	Yes	8.9	10.0
Yes (0.33)	Yes	9.7	12.9
Yes (0.17)	Yes	10.6	15.6

The percentage of elite clones in stage 3 (defined as having a mean NMG over plant and first ratoon crops of ≥ 11) resulting from different selection strategies is shown in Table 6.

Table 6
Percentage of elite stage 3 clones (NMG \geq 11) from random, mass, family, and combined family and mass selection in stage 2

Stage 2 selection		% of elite (NMG \geq 11) stage 3 clones	
Family	Mass	Exp 1	Exp 2
No	No	11.3	13.2
No	Yes	9.0	13.4
Yes (0.50)	No	11.1	16.7
Yes (0.33)	No	11.8	13.2
Yes (0.17)	No	15.3	13.9
Yes (0.50)	Yes	10.6	16.7
Yes (0.33)	Yes	12.5	19.4
Yes (0.17)	Yes	15.3	22.2

This showed that a substantially higher proportion of elite clones were found when family selection (0.33 or lower) was combined with mass selection (13-22%). The response was considerably higher in Experiment 2. Combining the families and clones from the two experiments and selecting families with stage 2 NMG \geq 8 (family selection rate of 25% overall) combined with mass selection resulted in almost 50% more elite clones in stage 3 (Table 7).

Table 7
Percentage of elite stage 3 clones (NMG \geq 11) from random, mass, family (NMG \geq 8), and combined family and mass selection (combined results from two experiments)

Stage 2 selection		% of elite stage 3 clones
Family	Mass	
No	No	12.2
No	Yes	11.9
Yes	No	13.9
Yes	Yes	18.1

The implications of this for a selection program where 2 000 clones progress into stage 3 are shown in Figure 1. This shows that an additional 118 elite clones can be expected through combined family and mass selection compared with random (or indeed) mass selection.

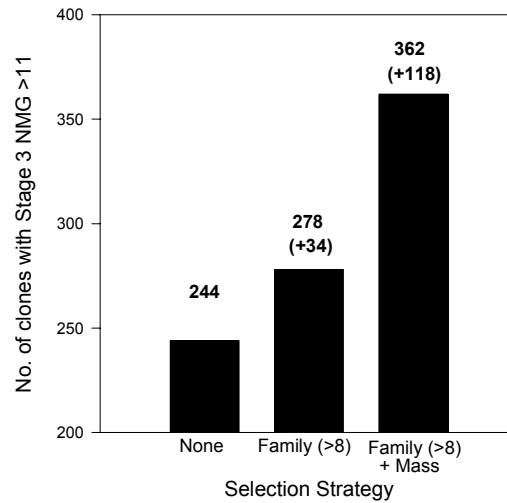


Figure 1 - Number of elite clones (NMG \geq 11) expected from a population of 2 000 clones

7. DIFFICULTIES

No major difficulties were encountered in this project. Experiment 2 was inadvertently planted to a completely randomised design instead of a randomised complete block design. Standard varieties were blocked and analysis indicated that lack of blocking was not a major problem. Some ratoon stunting disease was found in the first ratoon crop of Experiment 2, but we consider that, given the lack of water stress with nine irrigations, this would have had a minor effect.

8. RECOMMENDATIONS FOR FUTURE RESEARCH

Research on family selection to date has shown that combined family and mass selection is effective and efficient. Further work is currently in progress (BSES and SRDC) which will look at the effect of differential mass selection rates on genetic gain. BSES currently uses this selection method, taking more clones from better families and fewer from intermediate families. Thus adequate research into family selection methods has been or is being done.

9. PUBLICATIONS

- Cox, M C and Hogarth, D M** (1993). The effectiveness of family selection in early stages of a sugarcane improvement program. In 'Focussed Plant Improvement'. *Proceedings of The Tenth Australian Plant Breeding Conference Vol 2*, 53-54.
- Cox, M C, McRae, T A, Bull, J K and Hogarth, D M** (1996). Family selection improves the efficiency and effectiveness of a sugarcane improvement program. In *Towards a Sweeter Future: Sugar 2000 Symposium*, In Press.

10. REFERENCES

- BSES** (1984). Laboratory Manual for Australian Sugar Mills. Vol. 1 (BSES:Brisbane).
- Bull, J K, Hogarth, D M and Basford, K E** (1992). Impact of genotype x environment interaction on response to selection in sugarcane. *Australian Journal of Experimental Agriculture* **32**, 731-737.
- Cox, M C and Hogarth, D M** (1993). The effectiveness of family selection in early stages of a sugarcane improvement program. In 'Focussed Plant Improvement'. *Proceedings of The Tenth Australian Plant Breeding Conference Vol 2*, 53-54.
- Hogarth, D M** (1971). Quantitative inheritance studies in sugarcane. II. Correlation and predicted response to selection. *Australian Journal of Agriculture Research* **22**, 103-109.
- Hogarth, D M, Braithwaite, M J and Skinner, J C** (1990). Selection of sugarcane families in the Burdekin district. *Proceedings of The Australian Society of Sugar Cane Technologists* **12**, 99-104.
- Hogarth, D M and Mullins, R T** (1989). Changes in the BSES plant improvement program. *Proceedings of The International Society of Sugar Cane Technologists* **20**, 956-961.
- Jackson, P A, McRae, T A, and Bull, J K** (1995). The role of family selection in sugarcane breeding programs and the effect of genotype x environment interactions *Proceedings of The International Society of Sugar Cane Technologists* **22**, (in press).
- McRae, T A, Hogarth, D M, Foreman, J W and Braithwaite, M J** (1993). Selection of sugarcane seedling families in the Burdekin district. In 'Focussed Plant Improvement'. *Proceedings of The Tenth Australian Plant Breeding Conference Vol 1*, pp77-82.

Pollock, J S (1982). Variety selection in the Burdekin. *Proceedings of The Australian Society of Sugar Cane Technologists* **4**, 121-129.

Skinner, J (1971). Selection in sugarcane: a review. *Proceedings of The International Society of Sugar Cane Technologists* **14**, 149-162.

Stringer, J K, McRae, T A and Cox, M C (1996). Best linear unbiased prediction as a method of estimating breeding value in sugarcane. In *Towards a Sweeter Future: Sugar 2000 Symposium*, In Press.

11. ACKNOWLEDGMENTS

The following staff contributed to this project:

Dr D M Hogarth who recognised the possibilities of family selection for sugarcane improvement.

Mr P B Hansen who organised most of the field and laboratory work and data entry.

Mr J F Reimers who ensured that crop management (irrigation, fertilisation, pest control) was optimal.

Field staff at Bundaberg Sugar Experiment Station.

APPENDIX A

Families, parents and NMG categories in Experiments 1 and 2

Experiment 1			Experiment 2		
Code	Female x Male	NMG	Code	Female x Male	NMG
C114	65C286 x 59S55	<4	D210	66N2008 x H69-9103	<5
C123	66C807 x Co331	<4	D227	78N430 x 66N2008	<5
C130	Co740 x Co6602	<4	D291	Q138 x Q121	<5
C151	CP70-1527 x CP50-11	<4	D296	49R3863 x 59S55	<5
C159	F161 x 66N2008	<4	D300	59S55 x 62C476	<5
C199	61N567 x Q153	<4	D301	59S55 x Co331	<5
C133	Co740 x 59S55	4-5	D154	CP51-21 x NA56-79	5-6
C202	62N1659 x L62-86	4-5	D181	H56-752 x 63S782	5-6
C217	68N1797 x CP53-19	4-5	D200	60N1853 x R65-142	5-6
C231	NA56-79 x N14	4-5	D242	Q96 x 63B48	5-6
C235	Q79 x 66N2008	4-5	D311	60S7540 x H49-3666	5-6
C241	Q96 x 66C807	4-5	D313	61S145 x 61N567	5-6
C109	64C386 x 66N2008	5-6	D104	BN64-4051 x Q121	6-7
C145	CP51-21 x 66N2008	5-6	D126	66C789 x CP50-11	6-7
C146	CP51-21 x Q121	5-6	D147	Co740 x 59S55	6-7
C245	Q96 x Q142	5-6	D295	Q141 x 66N2008	6-7
C268	Q117 x 65S1102	5-6	D302	59S55 x Co440	6-7
C278	Q125 x CP71-1240	5-6	D334	TUC65-29 x CP51-21	6-7
C108	64C386 x CP53-19	6-7	D101	64A490 x 66N2008	7-8
C126	68C771 x CP53-19	6-7	D118	Q153 x 72C494	7-8
C153	CP72-1312 x Q96	6-7	D156	CP51-21 x Q121	7-8
C187	60N1853 x CP72-1370	6-7	D172	F161 x Q96	7-8
C322	67S9144 x Q117	6-7	D257	Q117 x CP56-59	7-8
C323	67S9144 x Q121	6-7	D320	70S448 x Q117	7-8
C165	H56-752 x Q96	7-8	D205	62N1659 x CP50-11	8-9
C218	68N1797 x 66N2008	7-8	D214	68N1797 x CP51-21	8-9
C224	71N437 x 66N2008	7-8	D232	PR975 x Q162	8-9
C259	Q117 x CP53-19	7-8	D241	Q95 x Q142	8-9
C265	Q117 x 70N959	7-8	D294	Q141 x Q153	8-9
C277	Q124 x Q142	7-8	D309	60S7493 x 66C807	8-9
C117	Q153 x H56-752	>8	D119	Q153 x 75C139	>9
C119	Q153 x Q121	>8	D166	F151 x Q142	>9
C174	58N829 x 66N2008	>8	D195	58N829 x 66N2008	>9
C212	67N2254 x 59S55	>8	D223	73N1254 x Q142	>9
C255	Q113 x Q142	>8	D260	Q117 x H58-8070	>9
C296	59S55 x Q142	>8	D282	Q128 x 72S1058	>9

APPENDIX B

Mean squares and significance levels for individual analyses of variance

Trait	Source of variation	Experiment 1		Experiment 2	
		P	1R	P	1R
TCH	Families	1963.0**	1954.0**	2225.0**	2134.0**
	:Groups	4725.0**	2625.0**	3929.0**	2162.0 ¹
	:Families/Groups	1502.0**	1842.0**	1941.0**	2130.0**
	Types	813.0 ¹	4256.0**	6309.0**	4150.0**
	Families x Types	428.0 ¹	652.3	673.0	397.7
	:Groups x Types	256.0	459.1	965.4	1290.0**
	:Fam/Gr x Types	457.0*	684.5	624.3	248.9
CCS	Families	13.93**	7.960**	8.427**	15.43**
	:Groups	21.96**	11.04**	27.59**	32.07**
	:Families/Groups	12.59**	7.447**	5.232**	12.65**
	Types	1.470	0.026	1.672	1.257
	Families x Types	1.218	1.167	1.285	2.352*
	:Groups x Types	1.673	1.145	0.693	2.447
	:Fam/Gr x Types	1.142	1.171	1.384	2.336*
TSH	Families	36.83**	60.99**	43.93**	42.95**
	:Groups	107.0**	101.7**	72.96**	32.78*
	:Families/Groups	25.13**	54.19**	39.09**	44.65**
	Types	27.09*	79.27**	105.6**	98.00*
	Families x Types	9.850*	14.78	15.76	9.189
	:Groups x Types	3.556	20.30	17.62	31.48**
	:Fam/Gr x Types	10.90*	13.86	15.44	5.475
NMG	Families	19.42**	17.82**	14.13**	26.97**
	:Groups	54.90**	31.11**	26.44**	23.89**
	:Families/Groups	13.51**	15.60**	12.08**	27.49**
	Types	15.02*	17.17*	25.74*	54.56*
	Families x Types	4.754 ¹	3.563	4.893	5.659
	:Groups x Types	1.476	2.145 ¹	4.611	18.29**
	:Fam/Gr x Types	5.300*	1.161	4.940	3.554

¹ P = 0.05 - 0.10

* P ≤ .05

** P ≤ .01

APPENDIX C

Mean squares and significance levels for combined analyses of variance

Trait	Source of variation	Exp 1	Exp 2	
TCH	Year	309400.**	418582.**	
	Family	3178.**	3511.**	
	:Group	5855.**	5591.**	
	:Family/Group	2723.**	3164.**	
	Family x Year	763.	848.	
	:Group x Year	1495.*	499.	
	:Family/Group x Year	622.	907.	
	Type	4395.**	10345.**	
	Family x Type	884.**	847.**	
	:Group x Type	308.	2092.**	
	:Family/Group x Type	959.**	639.*	
	Type x Year	792.	113.	
	Family x Type x Year	241.	224.	
	CCS	Year	680.20**	603.14**
		Family	17.70**	20.88**
:Group		29.63**	56.02**	
:Family/Group		16.09**	15.02**	
Family x Year		4.42**	2.97	
:Group x Year		3.37	3.64	
:Family/Group x Year		3.95*	2.86	
Type		0.64	0.02	
Family x Type		1.83	2.63**	
:Group x Type		1.75	2.49	
:Family/Group x Type		1.60*	2.66	
Type x Year		0.84	2.92	
Family x Type x Year		0.90	1.00	

APPENDIX C (Cont)

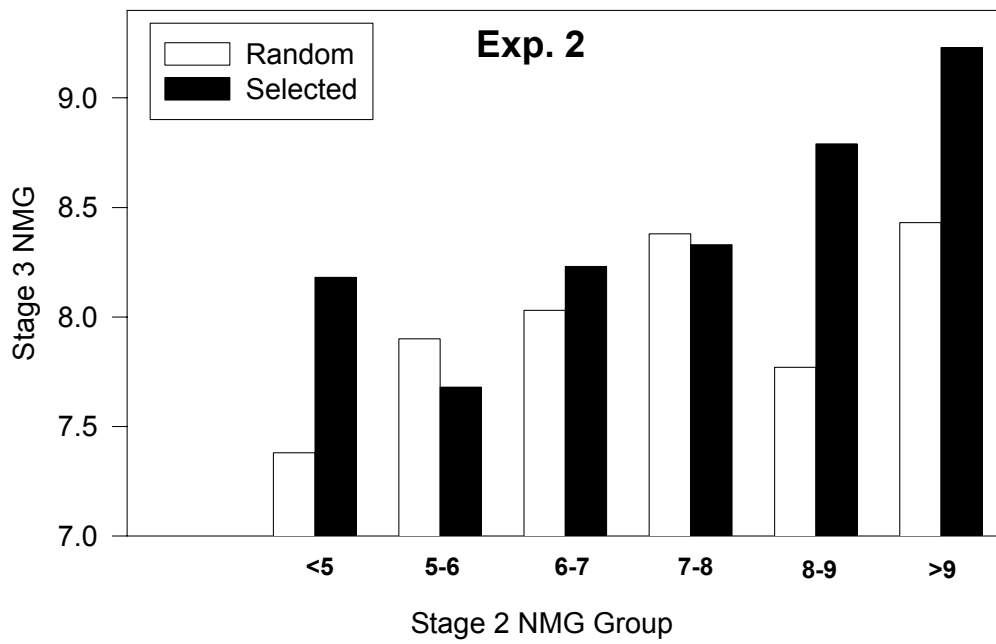
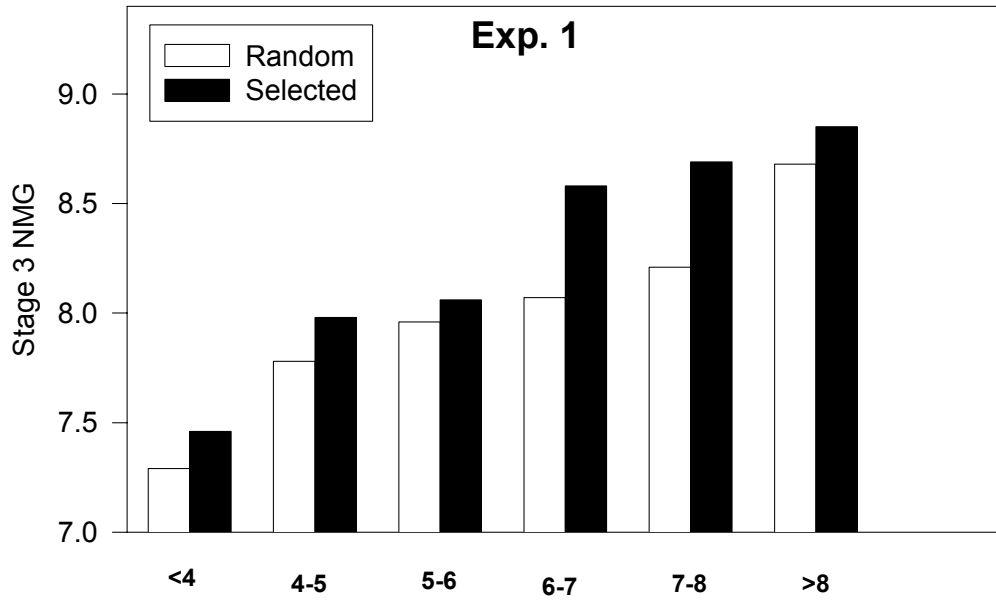
Mean squares and significance levels for combined analyses of variance

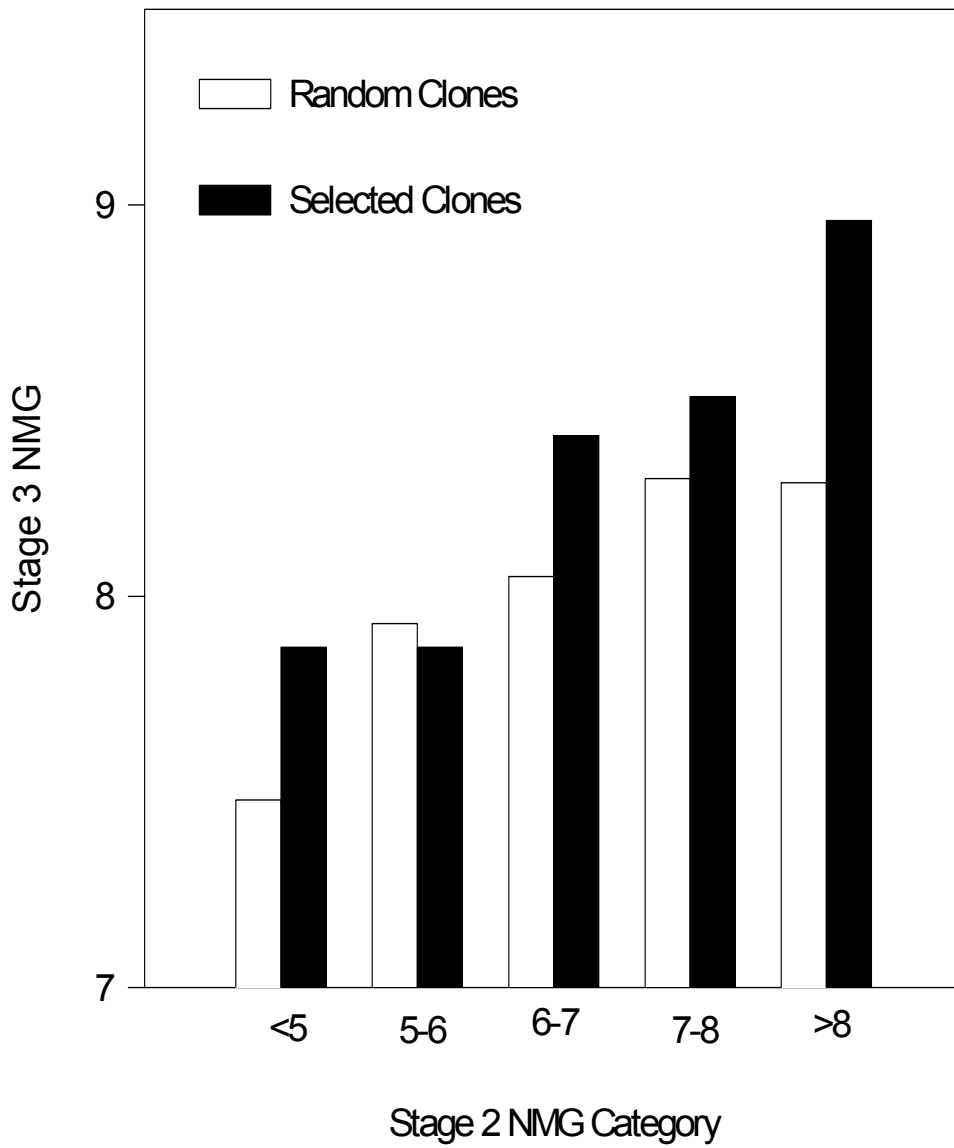
Trait	Source of variation	Exp 1	Exp 2	
TSH	Year	11970.0**	13735.3**	
	Family	77.3**	66.4**	
	:Group	165.3**	91.8**	
	:Family/Group	59.4**	62.2**	
	Family x Year	25.1**	20.5	
	:Group x Year	43.5**	13.9	
	:Family/Group x Year	19.9	21.6	
	Type	106.5**	203.6**	
	Family x Type	19.3**	18.8*	
	:Group x Type	8.0	44.2**	
	:Family/Group x Type	20.0**	14.5	
	Type x Year	8.7	0.1	
	Family x Type x Year	7.7	6.2	
	NMG	Year	52.07**	548.46**
		Family	29.23**	31.04**
:Group		68.27**	44.84**	
:Family/Group		22.07**	28.75**	
Family x Year		8.96**	10.06**	
:Group x Year		17.74**	5.52	
:Family/Group x Year		7.04	10.82**	
Type		34.03**	77.63**	
Family x Type		6.20**	7.88*	
:Group x Type		2.28	19.11**	
:Family/Group x Type		6.61**	6.01	
Type x Year		0.12	2.67	
Family x Type x Year		2.59	2.67	

APPENDIX D

Mean NMG of random and selected clones in stage 3

Stage 2 NMG Group	Mean Stage 3 NMG					
	Random			Selected		
	P	1R	Mean	P	1R	Mean
Exp 1						
<4	6.89	7.70	7.29	7.22	7.71	7.46
4-5	7.75	7.82	7.78	8.26	7.71	7.98
5-6	7.43	8.49	7.96	7.73	8.39	8.06
6-7	7.82	8.32	8.07	8.20	8.97	8.58
7-8	8.08	8.34	8.21	8.23	9.16	8.69
>8	8.94	8.42	8.68	8.87	8.84	8.85
Exp 2						
<5	8.03	6.74	7.38	8.60	7.75	8.18
5-6	8.44	7.37	7.90	8.49	6.86	7.68
6-7	8.28	7.77	8.03	8.59	7.87	8.23
7-8	9.12	7.63	8.38	8.97	7.69	8.33
8-9	8.44	7.10	7.77	9.27	8.30	8.79
>9	9.20	7.67	8.43	9.65	8.81	9.23

APPENDIX E**Mean NMG of random and selected clones in stage 3**

APPENDIX F**Family Selection in Stage 2 - Combined Results**

APPENDIX G**Frequency of stage 3 clones with acceptable (>9) NMG**