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Effect of Dwarf Apple Rootstocks on Average 'Gala' Fruit Weight at Six Locations over Three Seasons

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Abstract

The influence of rootstock on average fruit weight of 'Gala' apple [*Malus* x *sylvestris* (L) var. *domestica* (Borkh.) Mansf] was evaluated for a subset of data from the 1994 multi-location NC-140 apple rootstock trial. Data for 10 dwarfing rootstocks (M.9 EMLA, M.26 EMLA, M.9 RN29, M.9 Pajam1, M.9 Pajam2, B.9, O.3, V.1, Mark, and M.9 NAKBT337) at six locations for three years were used. Analysis of covariance was used to evaluate the effect of rootstock on average fruit weight when crop density was included in the linear model as a covariate. For five of the 18 data sets, rootstock variances were not equal, so a heterogeneous variance model was used. Rootstock significantly affected average fruit weight in 16 of the 18 data sets, but the rootstock by crop density interaction was significant for only one data set. Trees on M.9RN29 (also known as M.9Nic29), B.9, and M.9T337 (usually referred to as M.9NAKBT337) tended to produce the largest fruit and trees on Mark tended to produce the smallest fruit. Although rootstock significantly influenced average fruit weight at 16 of the 18 location-year combinations, results were not very consistent from one location to another or from year to year within a location. Possible explanations for these unexpected results are discussed.

As the apple [*Malus* x *sylvestris* (L.) var. *domestica* (Borkh.) Mansf.] industry continues to move towards high density orchards there is need for rootstocks that control tree size, provide excellent tree survival, and produce consistent crops of high quality fruit. Wholesale produce buyers pay premiums for large fruit, so apple producers are interested in rootstock effects on fruit size. In previous rootstock trials, fruit size was inconsistently influenced by rootstock. In some trials rootstock did not influence fruit size (3, 4, 5, 18), but other reports indicate that average fruit size was affected by rootstock (2, 11). In one trial, trees on P.22 and B.9 produced the smallest fruit and trees on C.6 produced the largest fruit (18). Since crop load can influence fruit size, and rootstock can influence crop load, average fruit weight values must be adjusted for crop load to interpret the data properly. In an attempt to account for varying crop loads, least squares means (LSmeans), adjusted for crop density (CD), were reported for several rootstock experiments (2, 6, 7, 8, 16). These results may not be valid for several reasons: 1) these reports provided no indication that the assumptions required for the analysis of covariance (normally distributed residuals, homogeneous variances, over-lapping ranges for CD, and homogeneous slopes) were tested; 2) data were usually pooled over several seasons without regard for possible year x rootstock x CD interactions; 3) a general linear models (GLM) procedure, based on least squares, was used to analyze the data. In most rootstock experiments replication is unequal because trees die over the course of a 10-year experiment. When the rootstock x CD interaction is significant in a mixed effects model, the analysis of covariance provided by PROC GLM is not adequate because it does not utilize the between-block information about the slopes (13). Most rootstock experiments used randomized complete block designs with block a random effect, resulting in a mixed-effects model. Marini et al. (14) used SAS's MIXED procedure to test for a year x CD x rootstock

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interaction and for equality of slopes. Since there was a strong 3-way interaction, analyses of covariance were performed for each of two years. For two of the eight data sets, slopes were not homogeneous, so LSmeans were compared at three levels of CD.

The purpose of this paper is to present results of the effect of 10 dwarfing apple rootstocks on average fruit weight of 'Gala' at six locations for three seasons. Also presented in this paper are slopes for the relationship between fruit weight (FW) and CD estimated from the solution vector requested with the solution option in the model statement using the MIXED procedure.

Materials and Methods

Data were from six locations in the NC-140 1994 dwarf rootstock trial (15). Locations were selected to provide a wide range of growing conditions. All selected locations had good tree survival and within every combination of year by location the ranges for CD overlapped. The experimental design was a randomized complete block at each location, where one tree per rootstock was randomly assigned to each of ten blocks per location on the basis of initial trunk cross-sectional area (TCA). Fourteen rootstocks were evaluated at all locations, but for this study only the ten rootstocks with commercial potential were included in the analyses (M.9 EMLA, M.26 EMLA, M.9 RN29, M.9 Pajam1, M.9 Pajam2, B.9, O.3, V.1, Mark, and M.9 NAKBT337). All trees were propagated by TRECO, Inc., Woodburn, OR and the scion was 'Treco Red Gala #42'. Each year cooperators submitted data for crop density (CD; number of fruit·cm^{-2} TCA) and average fruit weight (FW). To account for potential variations in seasonal crop loads and weather conditions, data were used for the final three years of the study. Some locations did not report yield data every year, so the final three years of cropping data were reported in this study. Because the interaction of location x year x rootstock was highly significant, each combination of location and year was analyzed separately.

For each combination of location and year, an analysis of covariance (ANCOVA) was performed with the MIXED procedure of SAS (13), where FW was the response variable, rootstock was the class variable or the indicator variable, CD was the covariate, and block was designated as a random effect. The approach outlined by Littell et al. (13) and used by Marini et al. (14) was used to test the hypotheses that all slopes were equal to zero and that all slopes were homogenous. When the hypothesis that all slopes were equal to zero was not rejected ($P > 0.05$), an analysis of variance was performed and rootstock LSmeans were compared with PDIFF. When the hypothesis that all slopes were equal to zero was rejected, an ANCOVA was performed and the model included a term to test the rootstock x CD interaction. If the interaction was not significant, a normal ANCOVA was performed and rootstock LSmeans were compared with PDIFF. When the interaction was significant, indicating that the effect of rootstock on FW depended on the level of CD, rootstock LSmeans were compared at three levels of CD. The CD levels selected for comparison included the overall mean CD for the data set plus values near the minimum and maximum for that data set.

The Shapiro-Wilk W-statistic, generated with SAS's Univariate Procedure (19), was used to test normality and all data sets adequately satisfied the assumption of normality. Levene's Test (17) was used to evaluate heterogeneity of variances and about one-third of the data sets had unequal variances. In those cases, the Akaike Information Criterion (AIC) was used to decide whether or not a model that accounted for heterogeneous variances for rootstock was desirable. ANCOVAs were performed with and without the statement "repeated/group = rootstock". The repeated statement specifies that the experimental units for each treatment are a repeated measurement and a separate residual variance is estimated for each group. When the repeated statement is not included, the

MIXED procedure uses the homogeneous residual variance. The AIC values produced by the two analyses were compared to determine if modeling heterogeneous variances was appropriate and the analysis with the smallest AIC value was used. Although slopes were usually equal, slopes for each rootstock were estimated from the solution vector requested with the solution option in the model statement. The model statement used was: MODEL FW = STOCK CD STOCK*CD / SOLUTION HTYPE=1. Unlike the GLM procedure, HTYPE=1 requests Type I tests in the MIXED procedure and this provides valid SE for treatment effects.

Results and Discussion

For 17 of the 18 location*year combinations, the rootstock*CD interaction was not significant, so a normal ANCOVA was performed for each combination and LSmeans were compared (Table 1) with PDIFF. Rootstock significantly influenced FW at 16 of the 18 locations-year combinations, but results were not very consistent from one location to another or from year to year within a location. For example, in Illinois in 2000 the largest fruit were produced by M.9 Pajam 2, M.9RN29, B.9 and M.9T337. However in 2001 B.9 produced medium size fruit and in 2002 rootstocks did not significantly affect FW. The two rootstocks that were most often (15 of 18 location-year combinations) in the group with the largest fruit were M.9RN29 and M.9T337. In contrast, Mark was in the group with the largest fruit only once, but had significantly smaller fruit than all other rootstocks in 12 of the 18 situations.

A significant rootstock x CD interaction would indicate that the relationship between CD and FW is not equal for all rootstocks. In this study the interaction was significant only for British Columbia in 2003; where at the low level of CD Mark produced the smallest fruit, but at the high level of CD Mark and M.26 produced the smallest fruit (Table 2). In a previous trial where 8 dwarf rootstocks budded to 'Gala' were compared, Marini et al. (14) found that there was a rootstock x CD interaction at only one of the four locations; and at one location rootstock did not influence FW. It is difficult to compare results with the previous trial because 3 of the 8 rootstocks were not common to both trials, but in the previous trial trees on P.1 consistently produced small fruit, trees on Mark produced intermediate sized fruit, and trees on B.9, M.9 EMLA, and Mac.39 produced the largest fruit. Results from both trials indicated that trees on B.9 produce large fruit.

If the relationship between FW and CD differed for rootstocks, this would indicate that certain rootstocks should be thinned differentially to ensure adequate fruit size. Slopes for FW as a function of CD, estimated from the solution vector, are presented in Table 3. Although it is well-established that the relationship between FW and CD is negative, some of the slopes were positive. Scatter plots of FW against CD verified that large fruit were sometimes associated with high CD and trees with low CDs sometimes produced small fruit (Fig. 1). The nature of the relationship between FW and CD was further investigated with simple linear regression and in most cases the coefficients of determination were less than 0.1 and were not significant at the 5% level, indicating that the relationship between FW and CD is often very poor in rootstock experiments where there is an attempt to thin trees to ideal crop loads. Had the range of CD been greater, the relationship between CD and FW would likely have been stronger and negative. In an attempt to summarize the slope data, the slopes were analyzed with a Friedman's rank sum test (10), where each location-year combination was considered to be a block. Slopes were not significantly affected by rootstock ($P > 0.15$), providing further evidence that the relationship between FW and CD was not consistently influenced by rootstock although for specific locations and years there was a strong effect of CD on FW.

It is difficult to compare results from different rootstock trials because experimental

Table 1. Average fruit weight of 'Gala' apple on 10 dwarfing rootstocks at five locations for three seasons. Values are least-squares means, adjusted for missing observations and crop density (CD).[z]

	Illinois			Maine			Michigan			New York - Geneva			Virginia		
Stock	2000	2001	2002	2001	2002	2003	2000	2001	2002	2001	2002	2003	2000	2001	2002
M.9 EMLA	185 b	178 ab	145	142 a	121 ab	144 a	178 ab	162 b	149 b	142 a	121 ab	144 a	178 ab	162 b	149 b
M.26 EMLA	187 b	175 b	147	136 ab	121 ab	136 ab	176 bc	159 b	152 ab	136 ab	121 ab	136 ab	176 bc	159 b	152 ab
M.9RN29	192 ab	177 ab	147	135 ab	118 abc	144 a	181 ab	159 b	149 b	135 ab	118 abc	144 a	181 ab	159 b	149 b
M.9 Pajam1	188 b	180 ab	143	138 ab	118 abc	141 a	182 a	164 b	148 b	138 ab	118 abc	141 a	182 a	164 b	148 b
M.9 Pajam2	198 a	186 ab	145	128 bc	116 bc	134 ab	181 ab	163 b	149 b	128 bc	116 bc	134 ab	181 ab	163 b	149 b
B.9	198 a	172 b	164	142 a	124 ab	144 a	171 cd	162 b	150 b	142 a	124 ab	144 a	171 cd	162 b	150 b
O.3	187 b	171 bc	141	142 a	126 a	125 b	181 ab	158 b	156 a	142 a	126 a	125 b	181 ab	158 b	156 a
V.1	183 b	173 b	157	135 a	117 bc	136 ab	174 bc	153 b	154 ab	135 a	117 bc	136 ab	174 bc	153 b	154 ab
Mark	185 b	156 c	137	125 c	108 c	125 b	161 d	148 c	136 c	125 c	108 c	125 b	161 d	148 c	136 c
M.9T337	193 ab	187 a	146	140 a	122 ab	135 ab	179ab	173 a	161 a	140 a	122 ab	135 ab	179ab	173 a	161 a
P-value from ANCOVA															
Stock	0.001	0.001	0.491	0.001	0.001	0.055	0.004	0.001	0.011	0.007	0.024	0.047	0.001	0.001	0.018
CD	0.007	0.003	0.001	0.003	0.019	0.021	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
CD*Stock	0.544	0.281	0.347	0.136	0.261	0.313	0.519	0.431	0.304	0.329	0.225	0.873	0.938	0.223	0.169

[z] LSmeans within location and year were compared with PDIFF, P=0.05.

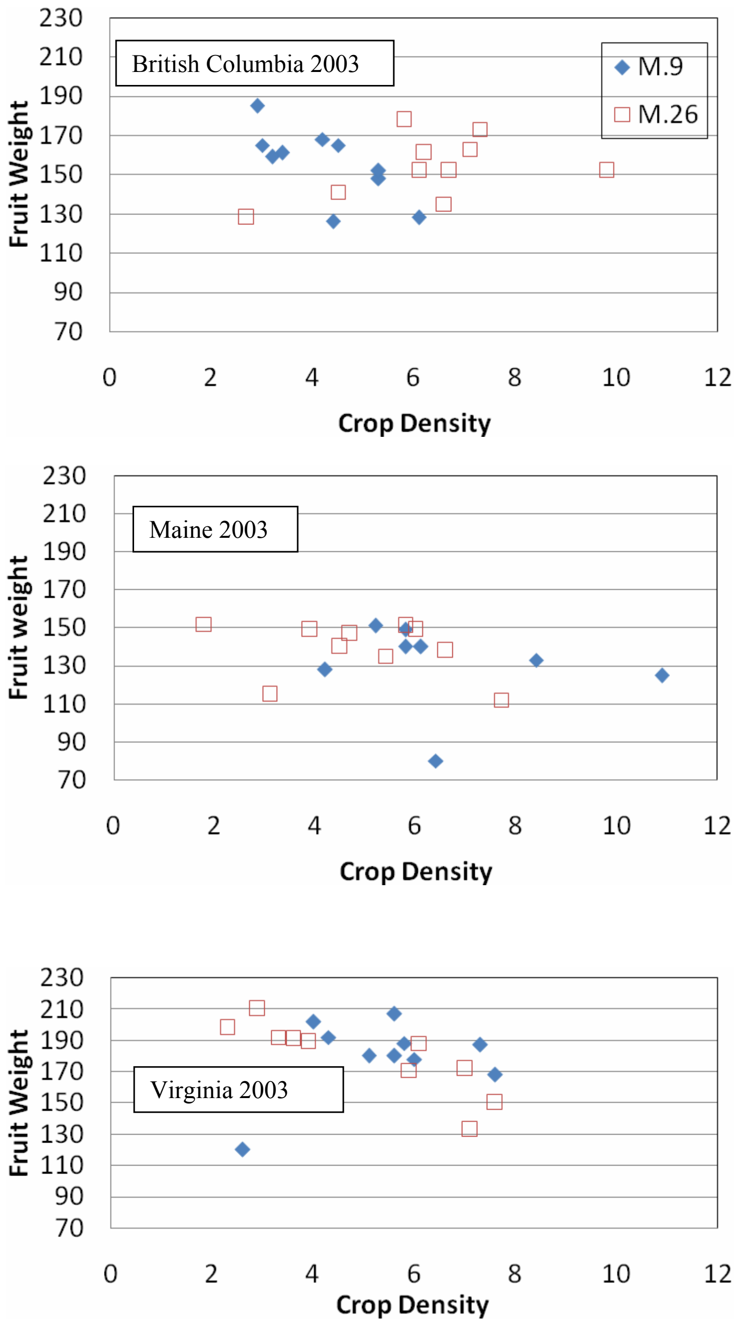

Figure 1. Scatter plots for average fruit weight (g) on crop density (no. of fruit per cm^2 of trunk cross-sectionional area) for two rootstocks at three locations. Scatter plots show the relationship is sometimes poor.

Table 2. Average fruit weight of 'Gala' apple on 10 dwarfing rootstocks at British Columbia for three seasons. Values are least-squares means, adjusted for missing observations and crop density (CD). In 2003 there was a significant rootstock by CD interaction, so least squares means were compared at three CDs (3.0, 5.6, and 7.0 fruit•cm^{-2} TCA).[z]

Stock	2001	2002	2003		
			CD = 3.0	CD = 5.6	CD = 7.0
M.9 EMLA	190 a	177 b	142 b	151 ab	156 a
M.26 EMLA	194 a	180 ab	169 ab	143 b	129 b
M.9RN29	202 a	197 a	156 ab	159 a	161 a
M.9 Pajam1	197 a	187 ab	180 a	161 a	151 a
M.9 Pajam2	195 a	189 ab	166 ab	157 a	153 a
B.9	194 a	185 ab	153 b	154 ab	155 a
O.3	196 a	181 ab	169 ab	162 a	158 a
V.1	196 a	187 ab	165 ab	152 a	144 ab
Mark	161 b	137 c	122 c	129 b	134 b
M.9T337	203 a	197 a	165 ab	161 a	159 a

[z] LSmeans within columns were compared with PDIFF, P=0.05.

designs, scion cultivars, rootstocks and statistical methods may vary. This is the second trial with 'Gala' where trees on B.9 produced relatively large fruit. However, these results also contradict those of the previous trial (14), where trees on Mark produced intermediate sized fruit and trees on M.26 EMLA consistently produced small fruit. The positive relationships between FW and CD, as indicated by the positive slopes, were unexpected because there are many reports of a negative relationship between these two variables (1, 9, 12, 20). There are several possible explanations for these unexpected results. 1) Some cooperators may have thinned trees too late in the season to substantially improve fruit size. 2) The number of replications may have been too low to obtain the true relationship because unusual observations can be highly influential when there are few replicates. 3) The unexpected results most likely resulted from the narrow range of crop loads. In most thinning experiments, treatments are selected that will produce a wide range of crop loads. However in rootstock studies, cooperators use various fruit thinning techniques to obtain crop loads that would encourage good fruit size and adequate return bloom. For these reasons, typical rootstock trials and orchard observations may not be appropriate for evaluating the influence of rootstocks on fruit size. Perhaps the influence of crop load on FW is relatively minor and inconsistent when trees are thinned adequately. Ideally, experiments should be designed specifically to evaluate the influence of rootstock and cultural practices on fruit size. Such experiments would involve wide ranges in CD and overlapping CDs for all rootstocks or treatments. NC-140 cooperators are currently conducting a study to evaluate the effects of rootstock on FW over a wide range of CDs and results from that study may help explain previous inconsistent results.

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Table 3. Slopes of average fruit weight on crop density (CD) for 'Gala' apple on 10 rootstocks at six locations for three seasons. Slopes indicate the change in average fruit weight (g) for an increase of one fruit•cm^{-2} in CD. Slopes were estimated from the solution vector provided by SAS's MIXED procedure.

	IL			ME			MI			NY-G			VA			BC		
Stock	2000	2001	2002	2000	2002	2003	2001	2002	2003	2001	2002	2003	2000	2001	2002	2001	2002	2003
M.9 EMLA	-3.6	-3.5	-3.8	1.4	-4.6	-1.3	-4.4	-4.5	-4.5	-4.0	-3.6	-3.3	-2.9	-2.0	-2.9	1.9	0.1	3.5
M.26 EMLA	-5.4	-9.0	-3.4	2.7	-6.0	0.5	-4.1	-2.1	-3.5	-2.0	-3.2	-6.6	-3.9	-2.9	-13.9	-4.6	-4.9	-10.6
M.9RN29	-6.1	3.2	-9.7	9.1	-6.2	5.3	-2.2	-4.7	-6.6	-3.5	-3.4	-3.1	-2.4	-4.2	-1.9	-4.6	-9.3	1.6
M.9 Pajam1	-4.0	-3.4	-8.3	-0.2	-1.2	-4.2	-1.4	-2.4	-0.5	-6.5	-2.4	-6.2	-3.8	3.4	3.8	-0.4	-3.8	-6.8
M.9 Pajam2	-7.1	-2.5	-7.6	7.6	1.8	-3.7	-2.4	-3.8	-8.4	-4.9	-7.6	-5.3	-2.8	-3.8	-2.6	-1.9	-3.6	-3.4
B.9	-11.2	-2.9	-10.6	8.6	-2.7	-6.6	-1.6	-1.2	-8.4	-0.9	-2.2	-6.2	-7.7	-4.1	-8.1	-2.7	-5.5	0.7
O.3	-6.8	-6.4	-4.5	1.3	-6.3	-5.7	-2.6	-1.4	-3.4	-1.5	-1.8	-5.4	-4.4	-5.2	-0.6	-3.9	-7.4	-3.2
V.1	-3.6	-1.9	-2.9	0.1	-8.4	-0.3	-4.3	-2.7	-10.6	-6.1	-8.3	-8.5	-1.2	-4.0	-3.6	2.6	-5.9	-5.3
Mark	-7.6	-5.6	-6.3	-4.4	-8.6	-1.6	-2.2	-0.9	-5.1	-1.9	-5.0	-8.5	-9.7	-4.0	-21.7	-0.3	12.1	2.7
M.9T337	-1.8	-3.6	-8.6	6.4	6.1	-12.3	0.1	-1.4	-7.7	-2.3	-3.4	-3.6	-4.1	-2.6	-3.9	-6.3	-0.8	-0.8

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