Allocation of Experimental Resources Used in Potato Breeding to Minimize the Variance of Genotype Mean Chip Color and Tuber Composition

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ABSTRACT

Breeders strive to select superior genotypes despite the challenges that environmental variation and environment × genotype interactions bring in increasing phenotypic variance. Effective selection requires experimental design that minimizes variance of genotype means and therefore allows for the identification of superior genotypes. The variance components calculated from three extensive datasets were used in modeling experiments to determine how experimental replication and sampling affected the variance of genotype mean for composition or fried chip color of tubers from *Solanum* species including the cultivated potato *Solanum tuberosum* L. Maximizing experimental replication over years and locations with limited sampling minimized variance of genotype means for chip color, tuber sugar concentrations, and tuber dry matter. It is suggested that in the early stages of a potato breeding program, more precise evaluations of the genetic potential of individual clones would be achieved through the use of small plots evaluated over several locations and/or years rather than increased replication at one location.

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Abbreviations: EC_{ALL}, environment and clonal propagule for all years and genotypes; EC_{CHIP}, environment, clonal propagule, and chip score; QTL, quantitative trait loci; WC, within clonal propagule.

Potato (*Solanum tuberosum* L.) breeding programs rely heavily on multiple years of phenotypic selection to identify superior genotypes. Biometrical tools used in breeding other crops are only beginning to be used in potato breeding. Most biometrical tools were developed for normally distributed, diploid populations and are unsuitable for commercially bred potatoes, which are highly heterozygous tetraploids. Computational models for use with tetrasomic inheritance have been developed, but to obtain estimates with these requires evaluation of multiple structured families that rapidly become unmanageable when dealing with an asexually propagated crop such as potato. In addition, potato breeders simultaneously select for multiple traits relating to agronomic performance, disease resistance, and tuber quality that are difficult to combine in a single quantitative selection index. Consequently, a typical potato breeding program initiates selection from crosses generating approximately 50,000 to 100,000 seedlings (genotypes) annually. These genotypes are then planted in the field as single-plant (hill) experimental units for visual

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evaluation in comparison to reference clones. Genotypes advanced to subsequent rounds of visual selection are planted in increasingly larger experimental units (4-, 8-, and 16-hill plots) in following years. Timing and intensity of selection varies for each trait. Selections are made in the field without extensive note taking or experimental replication. The goal at early stages of cultivar development is to accurately and rapidly screen a large number of genotypes and to identify progeny with rare combinations of advantageous phenotypes. Replicated field trials of approximately 300 clones selected from the starting population may begin 5 yr after the initial cross, and these trials increase in size and replication as selection reduces the number of clones found to be consistently acceptable after this extensive evaluation. Ultimately, one or two clones of the original 100,000 may be released as varieties after 15 yr of evaluation (Bradshaw and Mackay, 1994).

No or low replication allows the rapid screening of many genotypes in early phases of selection but increases the likelihood that genotypic effects can be confounded with environmental and genotype × environmental effects. This provides an explanation for why visual scores of genotypes grown in single-hill plots had low correlations with scores for the same genotypes in multi-hill plots (Tai, 1975; Neele et al., 1988). Similarly, a lack of consistency has been observed in selecting the best-performing genotypes from replicated single-hill trials (Blomquist and Lauer, 1962). Environmental effects caused extensive variation of diverse genotypes exhibiting a high degree of phenotypic plasticity even when such materials were clonally propagated (McCann et al., 2010). Low replication can also contribute to low heritability estimates, decrease response to selection, and hinder breeding efficiency.

The heritability of tuber chip color and tuber composition in cultivated potato has been examined previously (Chen et al., 2001, Diniz et al., 2006; Li et al., 2008; Menendez et al., 2002; Tai, 1975). Heritability estimates as well as the utility of molecular markers linked to specific traits are dependent on parental clones (Li et al., 2008). Numerous quantitative trait loci (QTL) for tuber sugar composition have been identified (Menendez et al., 2002), but no single locus explained more than 15.5% of phenotypic variance for sucrose, glucose, or fructose. In another report, 17 QTL contributing to tuber starch content were identified (Chen et al., 2001). Given the large effect that environment may have on tuber composition and quality, an examination of how allocation of experimental resources can be used to minimize the variance of genotype means was undertaken. Optimal experimental design will utilize finite resources to accurately measure genotype means while minimizing variance, that is, with precision. Increasing sample size, replication within a location, replication across locations, and replication across years is expected to decrease the variance of genotypic means at the expense of cost and effort. As the variance of genotype means decreases, the ability to detect differences between genotypes using statistical methods increases as does the ability to identify superior genotypes.

MATERIALS AND METHODS

Twelve genotypes displaying a range of light to extremely dark chip color were clonally propagated in 2005, 2006, and 2007 at Madison and Rhinelander, WI. These genotypes included *S. tuberosum* cv. Snowden, four *S. tuberosum* haploid × wild species hybrid clones (AH57.8, GH28.9, H25.51, and H25P3) from the USDA Potato Enhancement Laboratory, Madison, WI, and diverse genotypes that tuberized readily from *S. berthaultii* Hawkes (U.S. Potato Genebank, Sturgeon Bay, WI; accession PI 473239), *S. chacoense* Bitter (PI 175443 and PI 472826), *S. kurtzianum* Bitter & Wittm. (PI 473420), and *S. pinnatisectum* Dunal (PI 347766). Two clones each from *S. berthaultii* PI 473239 and *S. pinnatisectum* PI 347766 were used. Two to twelve clonal propagules, hereafter referred to as replicates (plants), of a genotype grown in a flat at a given environment were maintained and analyzed individually.

Plant production and laboratory methods were as described in McCann et al. (2010). Tubers were removed from cold storage 2 to 3 wk before planting. If no sprouts were visible after a week at room temperature, Rindite (7 parts 2-chloroethanol, 3 parts 1,2-dichlorothanol, and 1 part carbon tetrachloride) was used to break dormancy. All plant materials were started in greenhouses at Madison or Rhinelander, WI, and transplanted into 10 cm pots with Pro-Mix BX (Premier Horticulture, Inc., Quakertown, PA) potting medium and 5 mL of 15–8–11 Osmocote (The Scotts Company, Marysville, OH) extended release fertilizer. The 10 cm pots were used as stressors to initiate tuberization in wild potato species, many of which are not adapted to northern latitudes. Fifteen plants per accession were placed in a flat in a hoophouse at Rhinelander, WI, and in a coldframe in Madison, WI, in a completely randomized design in May when all danger of freezing was past. Growing plants this way tends to minimize environmental effects resulting from extreme temperatures but tends to increase those effects associated with daily fluctuations in soil moisture. Mean daily temperatures from May to September at Rhinelander, WI, were on average 1.6°C lower than at Madison, WI. Temperatures in 2005, 2006, and 2007 were similar. Plants were watered and fertilized as necessary. Water was withheld from visibly senescing plants, and their tubers were harvested when leaves senesced. Plants that did not senesce were allowed to grow until late September or early October at which time water was withheld and tubers were harvested in mid October. Tubers were placed in paper bags and moved to a storage locker at 2°C and greater than 90% relative humidity within a week of harvest.

Harvested tubers were stored at 2°C for 3 mo before chip processing and tuber composition analysis. This storage temperature was chosen to select for extreme resistance to cold-induced sweetening. Two tubers from each clonal propagule were assayed. Each tuber was cut in half longitudinally from stem to bud and a 2 to 3 mm thick slice (chip) removed from each half was fried. Thus, four tuber chips were fried for each clonal propagule. The propagule mean chip color score was recorded in 2005 while each of the four chips was scored individually in 2006 and 2007. Approximately 1 g fresh weight samples from tuber pieces remaining

after chip slices were removed and/or other tubers from the same propagule were used for tuber composition analyses. These samples were used for sugar analysis of each genotype grown in 2005, 2006, and 2007 to measure plant-to-plant variation for each clone. The sugar concentrations of four halves from two tubers for each propagule of *S. tuberosum* cv. Snowden and breeding clones AH57.8, GH28.9, H25.51, and H25P3 were analyzed in 2006, resulting in four sugar analyses for each propagule to measure within-propagule variation.

Statistical Methods

Statistical analysis was conducted using SAS Statistical Software Version 9.1.3 (SAS Institute, Inc., 2007). The experiment was conducted in a randomized complete block design with two locations × 3 yr as blocks. An experimental unit was a flat of 2 to 17 clonal propagules. A random effects model with the REML option and Satterthwaite approximation was used to estimate variance components, and a 95% confidence interval eliminated those variance components that were not statistically significant. These variance components were then divided by a hypothetical number of observations contributing to the genotype mean to determine the variance of a genotypic mean ($s^2_{\bar{\chi}}$).

Three datasets were used to estimate several variance components including location (s^2_L), genotype (s^2_G), genotype × location (s^2_{GL}), and clonal propagules within flats (s^2_C) (Eq. [1], [2], and [3]). Dataset WC (within clonal propagule) consisted of the five genotypes grown in 2006 where four tuber halves were individually analyzed for chip color and tuber sugar concentrations. Sources of variation unique to dataset WC were variance components attributed to clonal propagules within a genotype × location interaction ($s^2_{C(GL)}$), tubers within the three-way interaction of clonal propagules with genotype and location ($s^2_{T(CGL)}$), and number of tuber halves (H) in the residual (s^2_e) (Eq. [1]). Dataset EC_{ALL} (environment and clonal propagule for all years and genotypes) included the propagule mean chip color and tuber composition of the 12 genotypes grown in 2005, 2006, and 2007. Dataset EC_{CHIP} (environment, clonal propagule, and chip score) was a subset of EC_{ALL} and included four chip color scores for each propagule grown in 2006 and 2007. Sources of variation unique to datasets EC_{ALL} and EC_{CHIP} were variance components attributed to year (s^2_Y), location × year (s^2_{LY}), genotype × year (s^2_{GY}), and genotype × location × year (s^2_{GLY}) (Eq. [2] and [3]). The estimated variance of clonal propagules within genotype × location × year ($s^2_{C(GLY)}$) was included in the residual (s^2_e) for the EC_{ALL} dataset. The variance of the four chips scored for a clonal propagule constituted the residual in dataset EC_{CHIP}, which permitted estimation of the variance component attributed to propagules within genotype × location × year ($s^2_{C(GLY)}$) (Eq. [3]). The variance of a genotype mean ($s^2_{\bar{\chi}}$) for chip color and tuber composition was calculated using Eq. [4], [5], and [6] for each respective dataset.

$$\text{Dataset WC: } s^2 = s^2_L + s^2_G + s^2_{GL} + s^2_{C(GL)} + s^2_{T(CGL)} + s^2_e, \quad [1]$$

$$\text{Dataset } EC_{ALL}\text{: } s^2 = s^2_L + s^2_Y + s^2_{LY} + s^2_G + s^2_{GL} + s^2_{GY} + s^2_{GLY} + s^2_e, \quad [2]$$

$$\text{Dataset } EC_{CHIP}\text{: } s^2 = s^2_L + s^2_Y + s^2_{LY} + s^2_G + s^2_{GL} + s^2_{GY} + s^2_{GLY} + s^2_{C(GLY)} + s^2_e, \quad [3]$$

$$\text{Dataset WC: } s^2_{\bar{\chi}} = s^2_{GL}/L + s^2_{C(GL)}/CL + s^2_{T(CGL)}/CLT + s^2_e/CHLT, \quad [4]$$

$$\text{Dataset } EC_{ALL}\text{: } s^2_{\bar{\chi}} = s^2_{GY}/Y + s^2_{GL}/L + s^2_{GLY}/LY + s^2_e/CLY, \quad [5]$$

$$\text{Dataset } EC_{CHIP}\text{: } s^2_{\bar{\chi}} = s^2_{GY}/Y + s^2_{GL}/L + s^2_{GLY}/LY + s^2_{C(GLY)}/CLY + s^2_e/CChLY, \quad [6]$$

where C is number of clonal propagules, Ch is number of chips from a tuber, H is number of tuber halves, L is number of locations, T is number of tubers, and Y is number of years.

Sixty-four sampling units were arbitrarily used as the base in all resource allocation modeling experiments because it reflects a realistic evaluation of two chips each from two tubers from each of four clonal propagules grown at two locations over 2 yr. Therefore, the product of variables used in the denominator of Eq. [4], [5], and [6] was set to 64. The number of replications (years or locations) and samples (clonal propagules, tubers, tuber halves, or chips) used in modeling experiments examines the full range of genotype mean variance (Table 1; Supplemental Tables S1 and S2). For instance, analysis of 64 tubers from a single propagule grown for 1 yr at one location reflects one method to maximize sampling that is focused on the precision component of the variance of a genotype mean by eliminating environmental effects. On the other hand, analysis of one tuber from one propagule grown at 64 locations in 1 yr is a way to maximize replication and address the precision component of the variance of a genotype mean by examining varietal stability over diverse geography. While conducting an experiment with extreme sampling or replication is highly impractical, these extremes were included in analysis and discussion to demonstrate the full range of the variance of a genotype mean.

RESULTS AND DISCUSSION

Sources of Variation and Their Relative Contribution

The genotypic component accounted for the largest portion of variance for all variables in the EC_{ALL} dataset (Table 2), for the variables chip color, fructose, and glucose in the WC dataset (Supplemental Table S3), and for chip color in the EC_{CHIP} data set (Supplemental Table S4). This indicates that selection of superior genotypes is possible in a breeding program focused on improving chip color, tuber sugar composition, and tuber dry matter content. Environmental variance due to years, locations, and location × year was minimal or zero for all variables in all datasets except a major contribution of the year component to sucrose variation in dataset EC_{ALL}. This experiment was conducted in hoophouses that gave control of some environmental conditions. Environmental variance is likely to increase in field-based studies if different locations have more extreme differences in growing conditions that favor one location over another. Genotype × environmental variance explained from 8 to 23% of total variance and was a secondary source of variance for all variables in all datasets when compared to other sources of

Table 1. Modifying the number of locations (L), clonal propagules (C), tubers (T), and tuber halves (H) changed the variance of a genotypic mean ($s^2_{\bar{x}}$) for the variables of chip color, tuber sugar concentrations, and tuber percent dry matter based on variance estimates in the WC (within clonal propagule) dataset. For each hypothetical experiment, L × C × T × H was always equal to 64 samples.

Hypothetical experiment				Samples	$s^2_{\bar{x}}$				
L	C	T	H		Chip	Fructose	Glucose	Sucrose	Dry matter
1	1	32	2	64	1.23	78.3	71.0	214.4	7.96
1	1	64	1	64	1.22	77.2	69.5	212.1	7.87
1	2	16	2	64	0.96	54.1	57.4	154.6	5.15
1	2	32	1	64	0.95	53.0	55.9	152.3	5.06
1	4	8	2	64	0.83	42.0	50.6	124.6	3.74
1	4	16	1	64	0.82	40.9	49.1	122.3	3.65
1	8	4	2	64	0.76	35.9	47.2	109.7	3.04
1	8	8	1	64	0.75	34.8	45.7	107.4	2.95
1	16	2	2	64	0.73	32.9	45.5	102.2	2.69
1	16	4	1	64	0.71	31.8	44.0	99.9	2.60
1	32	1	2	64	0.71	31.4	44.7	98.5	2.51
1	32	2	1	64	0.70	30.3	43.2	96.2	2.42
1	64	1	1	64	0.69	29.5	42.8	94.3	2.33
2	1	16	2	64	0.63	40.4	37.1	109.7	4.08
2	1	32	1	64	0.62	39.3	35.6	107.4	3.99
2	2	8	2	64	0.50	28.3	30.3	79.8	2.67
2	2	16	1	64	0.48	27.2	28.9	77.5	2.58
2	4	4	2	64	0.43	22.2	26.9	64.8	1.97
2	4	8	1	64	0.42	21.1	25.5	62.5	1.88
2	8	2	2	64	0.40	19.2	25.2	57.4	1.62
2	8	4	1	64	0.38	18.1	23.8	55.1	1.53
2	16	1	2	64	0.38	17.7	24.4	53.6	1.44
2	16	2	1	64	0.37	16.6	22.9	51.3	1.35
2	32	1	1	64	0.36	15.8	22.5	49.5	1.26
4	1	8	2	64	0.33	21.4	20.2	57.4	2.14
4	1	16	1	64	0.32	20.3	18.7	55.1	2.05
4	2	4	2	64	0.27	15.3	16.8	42.4	1.43
4	2	8	1	64	0.25	14.3	15.3	40.1	1.34
4	4	2	2	64	0.23	12.3	15.1	34.9	1.08
4	4	4	1	64	0.22	11.2	13.6	32.6	0.99
4	8	1	2	64	0.22	10.8	14.2	31.2	0.91
4	8	2	1	64	0.20	9.7	12.8	28.9	0.82
4	16	1	1	64	0.19	9.0	12.3	27.0	0.73
8	1	4	2	64	0.18	11.9	11.7	31.2	1.17
8	1	8	1	64	0.17	10.8	10.2	28.9	1.08
8	2	2	2	64	0.15	8.9	10.0	23.7	0.82
8	2	4	1	64	0.14	7.8	8.5	21.4	0.73
8	4	1	2	64	0.13	7.4	9.2	20.0	0.64
8	4	2	1	64	0.12	6.3	7.7	17.7	0.55
8	8	1	1	64	0.11	5.5	7.3	15.8	0.46
16	1	2	2	64	0.11	7.2	7.5	18.1	0.68
16	1	4	1	64	0.09	6.1	6.0	15.8	0.59
16	2	1	2	64	0.09	5.7	6.6	14.4	0.51
16	2	2	1	64	0.08	4.6	5.2	12.1	0.42
16	4	1	1	64	0.07	3.8	4.7	10.2	0.33
32	1	1	2	64	0.07	4.8	5.4	11.6	0.44
32	1	2	1	64	0.06	3.7	3.9	9.3	0.35
32	2	1	1	64	0.05	3.0	3.5	7.4	0.26
64	1	1	1	64	0.04	2.5	2.8	6.0	0.23

variance (Table 2; Supplemental Tables S3 and S4). Accuracy of the genotype mean was high because observations were similar in multiple independent environments (Table 2). This suggests the specific location and year of future

Table 2. Variance estimates for the variables of chip color, tuber sugar concentrations, and tuber percent dry matter in the EC_{ALL} (environment and clonal propagule for all years and genotypes) dataset.

Covariance parameter		Chip color	Fructose	Glucose	Sucrose	Dry matter
s^2_L	Location	0.0	0.0	0.0	0.0	0.0
s^2_Y	Year	0.0	1.3	6.4	357.8	0.0
s^2_{LY}	Location × year	0.0	0.4	0.0	0.0	0.1
s^2_G	Genotype	1.7	137.4	109.0	518.7	30.1
s^2_{GL}	Genotype × location	0.0	9.8	7.6	10.2	0.0
s^2_{GY}	Genotype × year	0.2	12.7	12.7	145.5	0.0
s^2_{GLY}	Genotype × location × year	0.4	13.7	9.6	85.6	6.3
s^2	Residual includes clonal propagule	1.1	99.1	101.6	263.1	12.4

experiments is expected to minimally impact analysis of chip color, tuber sugar concentrations, and tuber percent dry matter in cases such as this where phenotypic variation in quantified parameters was large. This situation is common in potato breeding programs during the first few years of selection, and hence these results are widely applicable to potato breeding.

Sampling error from clonal propagules, tubers, halves, or chips contributed a large portion of variance for all tuber traits analyzed and decreased the precision of the variance of a genotype mean. This was evident in the large residual estimates for chip color, tuber sugar concentrations, and tuber percent dry matter in dataset EC_{ALL} (Table 2) and also in the high estimates of tuber within-propagule variance for all traits in the WC dataset (Supplemental Table S3). Decreasing the size of the experimental unit to four propagules would decrease block size, permit additional blocking within locations and years, and theoretically decrease sampling error. In a previous study, coefficients of genetic variation and heritability for tuber yield were stabilized with as few as two clones per plot and three replications (Diniz et al., 2006). However, in practice, smaller experimental unit size may be problematic when evaluating highly stoloniferous wild species because more space is necessary to separate experimental units. The number of tubers available for analysis or planting in the following season will also decrease with smaller experimental unit size. The sampling variance component due to tubers within a clonal propagule exceeded the variance estimate of the propagule within a genotype for all traits in the WC dataset (Supplemental Table S3), which highlighted the effect of micro-environment or within-plant variation in development on each tuber. Tuberization of wild *Solanum* species was encouraged by growth in 10 cm diameter pots but this may have introduced inconsistency in the soil environment during tuberization and bulking in this experiment. Heterogeneity of tubers within a hill in the field exists, however, so this source of variation must be tolerated with the knowledge that it decreases precision. In the end, this study demonstrated that analysis of a single tuber from a propagule may decrease accuracy of measuring the genotype mean if other modes of replication are not included in the experimental design.

Experimental Design to Minimize the Variance of a Genotype Mean

Variance of genotype means for all traits decreased as sampling decreased and experimental replication increased (Table 1; Supplemental Tables S1 and S2). A surface area graph of results from Supplemental Table S2 for chip color (Fig. 1) demonstrates the curvilinear decrease in variance as the number of chips or clonal propagules evaluated (sampling) was reduced and the number of trialing environments (replications) increased. These results are consistent with previous studies focused on potato tuber yield where increasing sampling within a plot was much less effective that increasing the number of plots (Bisognin et al., 2006; Caligari et al., 1985). Figure 1 also highlights the need to balance experimental replicates between years and locations. This analysis demonstrated no benefit of examining chip color, tuber sugar concentrations, or tuber percent dry matter on an intra-tuber level, so emphasis should be placed in examining more tubers or clonal propagules for these traits when experimental replication is not feasible (Table 1).

A relatively large variance estimate of a genotype mean might be acceptable at earlier generations of selection when the goal is to cull grossly inferior genotypes, but low variance estimates of a genotype mean are required at later generations of selection to identify superior genotypes. Note, however, that higher economic cost is incurred along with the greater scientific benefits of increased experimental replication. The use of many environments or blocks for replication purposes is impractical at early or intermediate stages in the potato breeding pipeline as it is for other crops when many genotypes need to be screened rapidly. However, maximum experimental replication (years, locations, or blocks) is an objective at pre-commercial release phases of cultivar development. When compared to total sampling or total replication variance, increasing the number of environments to two

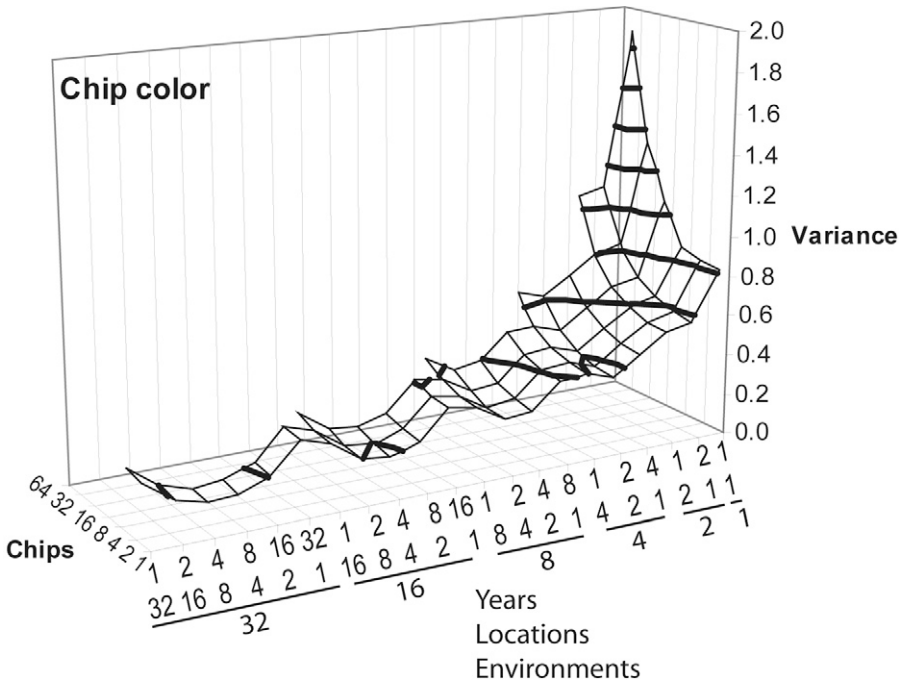

Figure 1. Allocation of experimental resources via number of locations, years, clonal propagules, and chips from each clonal propagule modified the variance of genotype mean chip color. A constant number (64) of sampling units was used for each simulation. The number of clonal propagules (not shown on graph axes) varied depending on the number of locations, years, and chips. Variance of a genotypic mean ($s^2_{\bar{x}}$) for the variable of chip color were based on variance estimates from the EC_{CHIP} (environment, clonal propagule, and chip score) dataset.

locations in 2 yr resulted in a 65% reduction in the variance of genotype means when averaged over all traits. Hence, a moderate increase in effort through replication results in a substantial increase in the ability to identity superior genotypes. Use of 4 yr and four locations seems plausible or slightly excessive as replication factors in breeding programs with an intermediate quantity of genotypes to evaluate but resulted in a 90% average decrease in the calculated variances for genotypic means over all traits when compared to the difference of total sampling to total replication variance (Fig. 1).

Applicability to Other Breeding Systems

The approach described is generally applicable to breeding other crops. Estimates of variance for individual parameters in the model are based on analysis of phenotypic data for genotypes grown over multiple locations, years, and clonal propagules. As such, the approach avoids the need to estimate heritability for each of the parameters of interest. The best models for resource allocation will be constructed from data sets where variability attributable to genotype, location, and sampling is comparable with that obtained in a breeding program (Vermeer, 1990). The datasets utilized here were obtained from *Solanum* species grown at two locations 300 km apart over 3 yr. Environmental conditions were typical of those expected in the Upper Midwest of the United States, and hence, variance estimates for location and year are likely to be reasonable for that geographic area. These estimates may be very imprecise, however, when used for geographic regions with distinctly different climate patterns or for species with different sensitivity to environment. Likewise, estimates for variance attributable to genotype were based on the wide phenotypic range found in the three datasets used by us. The relative contribution of genotype and environment to variation in traits of interest depends, of course, on the extent of genotypic variation in the breeding population. In the case of smooth bromegrass (*Bromus inermis* Leyss.), for example, genetic gain for neutral detergent fiber concentration was maximized by basing selection on multiple harvests within a year rather than replication of genotypes or locations (Stendal and Casler, 2006).

Extensive variation in chip color and tuber sugar composition is common in early generations of potato breeding programs, and therefore, the variance estimates used here for modeling resource allocation are reasonable. Models derived from datasets in which genotypic variation is smaller than that observed for potato are likely to place an even higher emphasis on replication across environments and years than found here, since environment is expected to make an even larger contribution to phenotype in this situation. Simulations by Moreau et al. (2000) showed that unreplicated trials are optimal for traits sensitive to genotype × environment interactions. An example of this was observed for sugarcane (*Saccharum* spp.), where decreased replication at a location resulted in no loss of precision because there was little variability between

replicates. Variability between locations was large, however, and increasing the number of locations was recommended (Brown and Glaz, 2001).

CONCLUSIONS

These results demonstrate the influence of replication in reducing the environmental component of phenotypic variance for the traits of chip color, tuber sugar concentrations, and tuber percent dry matter to better allow the potato breeder to correctly identify superior genotypes. The feasibility of replicating several hundred thousand genotypes early in the potato breeding program is debatable, but replication is recommended as soon as possible. Tai and Young (1984) reported that moderate selection intensity in combination with small experimental unit size (4-hill plots) permitted replication over years and improved breeding efficiency of the Canadian potato breeding program. In the future, progeny testing of parents and utilization of molecular markers as they become available are expected to decrease the initial population sizes in breeding programs, allowing earlier replication and more efficient potato breeding.

Supplemental Information Available

Supplemental material is available at http://www.crops.org/publications/cs. Included are tables describing variance of genotypic means in hypothetical experiments based on the EC_{ALL} (environment and clonal propagule for all years and genotypes) (Supplemental Table S1) and EC_{CHIP} (environment, clonal propagule, and chip score) (Supplemental Table S2) datasets and variance estimates for the variables chip color, sugar concentration, and tuber dry matter for the WC (within clonal propagule) (Supplemental Table S3) and chip color for the EC_{CHIP} datasets.

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