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# **Selection for Individual Traits in the Early Generations of Potato Breeding Program Dedicated to Processing Chips**

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# **Abstract**

Potato (*Solanum tuberosum*) breeding and selection programs aim to develop new cultivars of superior and improved qualities. Early generation selection plays a critical role in the success of such a program. The major objective of this study was to determine the select ability of individual traits in the early generations of clones targeted for the potato chip processing industry. For this study, the cross between NDO1496-1 and A90359- 7 was used. Progenies of this cross were selected and the seeds grown in 50 x 35cm planter flats. Seedlings were then transplanted individually to diameter pots after 5 weeks. 175 clones were randomly selected for inclusion in the next year and the process repeated consecutively for four generations, with each generation being selected for desired traits – skin color, tuber shape, maturity, eye depth, specific gravity, and size and shape uniformity. Tuber inter-generation correlation, of examined traits, were positive and significant (P<0.01) except for specific gravity which differed due to environmental factors. The percentage distribution of vine maturity revealed that only 6.2% of the late maturity clones would be lost if the entire high vigor class of the first generation was eliminated. Eye depth and round tuber shape are priority traits for selection followed by specific gravity and color chip as described in the study.

**Keywords:** breeding, clonal selection, early generation, individual traits, potato

# **1. Introduction**

The potato (*Solanum tuberosum L.*) is emerging as the most important staple cash crop in the world with a high nutritional content and a relative low water footprint compared to other staple crops. The potato is rich in carbohydrates, micronutrients, dietary antioxidants, vitamins B, and C and protein content comparable to cereal grains (Burlingame, 2009). Since its introduction to Europe in the sixteenth century and its worldwide distribution, it has had an important contribution to food and nutrition security (He, 2012). Recently, potato has even become one of the largest food crops (Spooner et al., 2010) with a global average of 20 million hectares grown, producing more than 300 million tons annually.

The major objectives of all potato breeding and selection program is to develop new cultivars with increased yield, improved nutritional and health characteristics and resistance to diseases as well as supply growers and the potato industry with improved varieties which makes their task easier and profitable. In many of the developing countries, potatoes are used both in fresh form and as processed products such as French fries, chips, dehydrated powder or canned potatoes. Thus, while French fry producers generally prefer long-oval tubers of moderate size, chip producers like high dry matter potatoes of small round shape.

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The cultivated potato (*Solanum* spp.) with different ploidy level, is a highly heterozygous tetraploid species; 2n=fourX=48 chromosomes. The breeding of a new variety is a long involving process. As the first step, parents are selected for their potential to produce new desirable genotypes. True seed derived from the selected parents are then obtained and a large number of seedlings (genotypes), sometimes up to several hundred thousand annually are raised from the true potato seed and evaluated in the seedling (F1) or first clonal generation. A portion of these seedlings are selected and replanted in small pots for screening in the next clonal generation. Accurate testing of advanced clones can be started only after the population has been reduced to a manageable size. Superior lines are promoted into trials of various types (yield, disease resistance, quality management etc.) and assessed for merit as potential new varieties.

The breeding program used at Kangwon National University is rather typical in five distinct steps: evaluation of parents and production of crosses (year1-2), selection in early generations (year 3-4), evaluation of advanced clones (year 4-5), and introduction as a new variety (year 5-6). Early generation selection plays a critical role in the success of a breeding program. During this stage, more than 99% of the genotypes must be discarded by some easily employed method of screening. Early generation selection by potato breeders is often an intuitive process, based on visual estimates of commercial worth. This evaluation, often known a breeder's preference involves a complex series of visible traits that may be genetically unrelated. Hence, select ability of these characteristics is often unknown and individual traits are given equal weight during the selection process (Richardson et al., 1990). This can result in excessive or insufficient selection pressure and inefficiency in what has become a very expensive process.

Traits such as yield, specific gravity, plant appearance (Davies & Johnston, 1974; Tai, 1975; Brown et al., 1984, 1987; Tai & Young, 1984) and chip color from progenies (Neele & Louwes, 1989) have been particularly emphasized as selectable. Most important for the chip industry is an emphasis on chip color because if a desirable color is not present, then the relative importance of remaining traits is diminished. Quality traits important for cultivars used in potato chip manufacturing include high dry matter, low sugars and free from defects (Dale & Mackay, 1994). Chip defects can result from several external and internal problems including growth cracks, hollow heart, heat necrosis, mechanical injury, greening and tuber rot. This study was designed to determine the select ability of a large number of traits that individually contribute to overall appearance. The traits examined included those of tuber characteristics evaluated during the visual assessment in early generation selection. In addition, this study was limited to genotypes from a breeding population developed to produce cultivars with high specific gravity and good chip color.

# **2. Materials and Methods**

Out of the large number of crosses used for making selections in various potato-breeding programs, the cross of NDO1496-1 X A90359-7 was selected for this study. Progenies of this cross were randomly selected from combinations of parents with high specific gravity and good color chip tubers. The female parent 'NDO1496-1' had high solids, good yield, early variety and nice tubers with good chipping quality out of cold storage. However, it was susceptible to developing black spot from bruising. The male parent 'A90359-7' was selected with medium solids, good yield, and fine tuber type but sometimes irregular and fair chipping quality out of cold storage. 1,500 seeds of this cross were grown-at Chuncheon Korea on 3 April 1997-in seedling planter flats with 50 x 35 cm square cells per flat. The cells were filled with commercial horticulture soil and seeds sown in shallow (5cm deep) plastic trays in a glasshouse. Five weeks after, the seedlings from the true seeds were transplanted individually into 12cm diameter pots at the two-leaf stage containing compost. For the first five weeks, the seedlings were watered by a mist propagator and after transplanting, from below via a sand bed in the field. 175 clones from this cross, which produced sufficient tubers to supply seed for the following year, were randomly selected for inclusion in the study on 3 July 1997. Before harvest, these clones were visually scored and classified into very early (1), early (2), medium (3), late (4) and very late (5) maturity based on plant size and foliage cover, observed height and specific gravity.

Also before harvest, the haulms were removed and the water supply cut off to allow the soil to dry and then each pot was harvested individually. Seed tubers were stored at 20  $\circ$ C until sprouts appeared and then planted in the greenhouse at Namwon on 13 October 1997 through standard practice. The largest tubers from each of the 175 selected genotypes of the first clonal year together with tubers from Korea's three standard cultivars; Atlantic,

Superior and Jopoong were planted. At harvest, 175 progenies from this cross were again selected and evaluated for traits in tuber shape, eye depth, shape and size uniformity and specific gravity.

The clones were harvested on 2 February 1998 and stored at 20 OC until sprouts appeared and the tubers (F1C2) planted in the greenhouse in single hill plots with the three standard cultivars; Atlantic, Superior and Jopoong in the field on 7 May 1998 at Daekuanliong. Tubers were harvested on 20 August 1998 and again chipped at  $20^{\circ}$ C and planted at Nam-won together with the three standard cultivars; Atlantic, Superior and Jopoong in the greenhouse on 15 January 1999. Following harvest on 25 April 1999, tubers were once more evaluated on traits as the previous generation. A listing of the generations, location, soil type, planting and harvest dates are given in Table 1 below.



# **Table1: Planting locations of 175 clones of four generations; soil type, planting and harvest dates.**

As not all characteristics could be dealt with, this study was confined to characteristics considered most important to selection of traits in the early generations.

### **2.1 Features covered**

#### *2.1.1 Foliage type*

Scored annually around 20 July when plants began to flower. Classification was made in three groups:  $1=$ poor;  $2=$  medium;  $3=$  high

#### *2.1.2 Maturity*

The day when both leaves and stems had fully died were noted and classified into five groups. 1= very early;  $2=$  early;  $3=$  medium;  $4=$  late;  $5=$  very late

### *2.1.3Tuber shape*

Tuber shape is the ratio between the length and width of tubers. This was indicated by a figure, the so called length-index. The length-index = (length: width)  $X$  100 or l.i = (l : w)  $X$  100. Clones were grouped into five classes: 1= round; 2= round-oval; 3= oval; 4= oval-long; 5= long

# *2.1.4 Depth of eyes*

Also grouped into five classes:  $1=$  very deep eyes;  $5=$  very shallow eyes. The meaning of the interjacent classes is in proportion.

### 2.1.5 Flesh color

This was scored annually in only one tuber per clone and grouped into five classes:  $1=$  Red;  $2=$  white;  $3=$ buff;  $4=$  brown;  $5=$  dark brown

#### *2.1.6Under-water-weight*

This was determined as the weight of 5kgs of tubers weighed under water. From this the specific gravity can be calculated.

#### *2.1.7Size uniformity*

Scored annually in all tubers per clone and grouped into five classes:  $1=$  very poor;  $2=$  poor;  $3=$  medium;  $4=$ excellent; 5= very excellent

#### *2.1.8 Shape uniformity*

Scored annually in all tubers per clone and grouped into five:  $1=$  very poor;  $2=$  poor;  $3=$  medium;  $4=$ excellent; 5= very excellent

#### *2.1.9 Assessment of chip color*

For 175 clones, two color chip evaluations were done:  $1 -$  after three months storage at 10°C and  $2 -$  after three month's storage at 4<sup>o</sup>C and three weeks reconditioning at 20<sup>o</sup>C. Each chipping evaluation used uniform tubers free of defects. Ten slices were fired in oil at 180°C for 3-4 mins. Chip color was measured by one person subjectively by using the procedure outlined in the potato chip Food Association's *Fry Color Standards for Potatoes for Chipping* (1-5 scale:  $1 =$  light;  $5 =$  dark colored chip).

### *2.2 Statistical analyses*

Statistical procedures for a randomized complete block design were used to analyze specific gravity, eye depth, shape and chip processing. For each year, the genotype means were calculated and analyzed across the years by SAS general linear models procedure. Genotype effects were considered fixed while year effects were considered random. Mean separation was done by Duncan's multiple range test or the least significant different test. Vine maturity and tuber appearance rating data was analyzed by means of genotype observations in each of the 4 years in a two-factor factorial analysis of variance. Correlation coefficients were done between the overall trial means of each genotype for the agronomic traits (specific gravity, eye depth, and tuber shape) as well as maturity rating.

#### **3. Results**

The objective of this analysis was to determine if clones rejected (based on single traits) in the seedling generation were likely to be rejected again in later generations. For any single clone, four possibilities existed: 1) saved in the seedling generation then saved later, 2) save in the seedling generation then reject later, 3) reject in the seedling generation and then saved later, 4) rejected in the seedling generation then rejected again later. All possibilities were investigated in this study with the practical goal to determine which traits could confidently be used to reject clones in the early generations based on the tendency for those same clones to be rejected again in subsequent generations. Analysis of rejection patterns were based on negative selection criteria. For each trait, the level of acceptability was predetermined and any genotypes outside the acceptable range were considered to be rejected. Percentage of clones rejected for most traits fell in the range of 20% to 40% (Table 2). The range and coefficients of variation (CV) of tuber specific gravity for the three vigor groups in various generations are given in Table 3. The data shows a low variability for specific gravity in F1C1 generation. It indicates that low variability of these characters that was observed in the F1C1 generation was due to planting in the greenhouse leading high environmental effects.



Table 2: Mean performance scores for six traits of 175 clones grown as seedling generation, 1<sup>st</sup> clonal generation and 2nd clonal generation, and selected by visual discrimination.

1 Rated on a scale of 1 (round) – 5 (long).

2 Rated on a scale of 1 (early maturity) – 5 (late maturity).

3 Rated on a scale of 1 (deep) – 5 (shallow).

4 Rated on a scale of 1 (variable) – 5 (uniform).



### **Table 3: Range and coefficients of variation in different vigor groups in different generations**

An analysis of variance (Table 4) conducted with F1C2 material showed that there was significant genetic variability for all three vigor groups.



\*, \*\* Significant at p<0.05 and P<0.01, respectively

# **Table 4: Analysis of variance of different vigor groups in F1C2 generation**

The average performance of different vigor groups in three generations is presented in Table 5. The specific gravity of F1C1 generation differed significantly from other generations (P<0.05). The F1C1 produced low specific gravity while the F1 and F1C2 showed high specific gravity. The difference in specific gravity in F1C1 generation was mainly due to environmental factors.



 $X = \text{mean}$ :  $SE = \text{standard error}$ 

#### **Table 5: Mean performance of different F1 vigor groups in different generations**

The inter-generation correlation coefficients (Table 6) were calculated to examine the relationship of performance in different generations. All the correlation coefficients were positive and significant except for specific gravity in which the r-values between the F1 and F1C1 and between F1 and F1C2 were non-significant, indicating that selection for this character was not possible in the F1 generation. The correlation coefficient between specific gravity of the F1 and F1C1 ( $r=0.147$ ), F1 and F1C2 ( $r=0.131$ ) was low, indicating that no selection for specific gravity may be made in the F1 generation. Coefficients between specific gravity of the F1C1 and F1C2 (r=0.297) was not high enough to suggest a positive selection for good specific gravity in this generation. However, they did suggest that negative selection (rejection of poor specific gravity only) could be carried out in this generation. The correlation between tuber shapes of different generations was almost of the same magnitude as for specific gravity. The estimates of r (Table 6) between F1C1 and F1C2 were low and, therefore, negative selection at this stage can be recommended. Inter-generation correlation coefficients for eye depth was lower than those of tuber shape and specific gravity between the F1C1 and F2C2 generations (r=0.177) indicating that negative selection for this character could be carried out in the F1C1 generation.



\*, \*\* Significant at P<0.05 and P<0.01, respectively

#### **Table 6: Correlation coefficients (r) for tuber specific gravity, tuber shape and, eye depth in different generations**

To know how many good clones would be lost if seedlings of very high vigor were discarded, we worked out the distribution of F1C1 vine maturity in the five vigor groups (Table 7). The data shows that out of 20% of the genotypes that were observed in the late maturity (4=late maturity), 9.2% were very early maturity class (1=very early maturity), 6.9% from the poor group and 0.0% from very high group. Thus only 6.2% of the late maturity clones would be lost if the entire very high vigor class was eliminated in the F1 generation. Considering the extent of resources involved in carrying all of the seedlings to subsequent generations, it would be desirable to discard very high vigor seedlings at the time of transplanting in the field as there would be no significant loss of early maturity seedlings. In Europe and North America where the crop is under long photoperiods, high vigor of the seedlings is associated with late maturity. Hence, very vigorous clones are rejected in early testing stage in potato breeding programs. In contrast, the crop in India is grown largely under short-day conditions that restrict foliage growth and induce early tuberization and early maturity (Pushkarnath 1976). This makes even the very vigorous genotypes desirable for use under Indian conditions. Inter-generation correlation coefficients (Table 8) were calculated to examine the relationship of performance in different generations. Two of the correlation coefficients were positive and significant. The correlation coefficients between F1 vigor and F1 maturity (r=0.60) and between the F1 vigor and F1C1 maturity  $(r=0.59)$  were not high enough to suggest a positive selection for very early maturity in this generation. However, they did suggest that reject of very late maturity could be carried out in this generation.



Maturity 1=very early, 2=early, 3=medium, 4=late 5=very late

#### **Table 7: Percentage distribution of genotypes for F1 vigor groups and F1C1 maturity**



\*, \*\* Significant at P<0.05 and P<0.01, respectively

# **Table 8: Correlation coefficients (r) for F1 vigor in different generation's maturity**

#### **4. Discussion and conclusion**

Concern that genetic variability for specific gravity may be limited by selecting single-hill progenies for chipping is important. High specific gravity potatoes are desired by potato processors because of greater processing efficiency (high chip yield), less potato waste, less oil absorption, and a desirable hard crunch to chips (Smith, 1951; Talburt& Smith, 1967; Lulai& Orr, 1979; Eisenhauer, 1993). Since specific gravity and chip color directly affect the quality of marketable chips, the relationship between the two traits is important. Only one of the selections had a higher specific gravity than superior (1.083).

The rest of the selections had a specific gravity ranging from 1.037 to 1.126. A specific gravity of 1.083 is considered too low for efficient chip processing, and few selections had values this low. For example, Jopoong is generally low in specific gravity, yet it provides an indication of the specific gravity value manufacturers are willing to accept for processing if quality of chip color is good.

A specific gravity value not less than 1.083 is considered a reasonable selection minimum and 38% of the selections meet these criteria. Moreover, 18 selections had a specific gravity equal to or higher than Atlantic (1.096), showing that this breeding system does not exclude high specific gravity clones during selection, at least when diverse parents were used in the crosses.



Comparison of genotype and cultivars

**Fig.1. Comparison of 175 genotypes and three cultivars**

From the study, we can deduce that when breeding and selecting for the combination of both specific gravity and good chipping in a potato-breeding program, it seems appropriate to select first for chipping then for specific gravity. Certainly, if a genotype does not possess the desired chip color quality, then the magnitude of specific gravity is irrelevant. On the other hand, a good chipping genotype with a lower than desirable specific gravity may be processed provided that reliability in chip color quality compensate for the low specific gravity.

In this experiment, specific gravity data was recorded but no genotypes were discarded at the end of the three years. To examine the efficiency of selection at these stages, the relationship between the number of selected and rejected clones can be studied for the years taken in pairs. For example, the material in the first year (FCY) can be divided into two categories; selected and rejected depending on set criteria. The second year clones (SCY) can then be selected on the same or different criteria. From this, we can get the proportion of clones selected in the FCY. The ratio of these proportions will be termed as the selection ratio.





If the selection ratio is equal to zero, then there was no repeat selections made in the second year, or there were no clones selected in the second year that had been discarded in the first. A selection ratio less than 1.0 show that a higher proportion of clones were selected in the second years that were discarded in the first year than from those selected in the first year. Selection ration equal to 1.0 indicates that there was no association between the selections in the two years and a ratio greater than 1.0 show that selection was effective, with increasing values of selection ratio indicating efficiency.

The selection ratios of the 175 clones selected on specific gravity at different selection intensities in the seedling generation and second clonal years are shown in Table 9. Irrespective of the proportion retained in the second clonal year, there was a general similarity in the selection ratios as the percentage of the clones retained in the first clonal year were same. When 99% of the first year clones were retained, sampling variation played a large part and the selection ratios were either 0.0 or almost 1.0. The highest selection ratio was obtained by selecting 90% of the seedling generation genotypes and 25% of the first clonal year. Selection at this level may, however, not justify the effort involved in discarding only 10% of genotypes in the seedling generation. When large proportions of the first clonal year genotypes are discarded, the selection ratios are reduced to just above 1.0. From this, it would appear that moderate to high selection intensities would not give a significant improvement over random reduction. Similar results are seen for the selection intensities in the first and second clonal years.



Table 9: Selection ratios of 175 clones for total tuber weight at different selection intensities (i.e. % of clones retained). The upper figures show the ratios for selection in F1 followed by F1C1. The lower figures show selection in F1C1 followed by F1C2, while the lowest figures show selection in F1C1 followed by F1C2. The selection ratios suggest that selection for specific gravity was more effective between the first and second clonal years than between the seedling generation and first clonal year. The inefficiency of selection at the single plant stage was due, at least in part, to the inaccuracy of specific gravity assessment on a single plant basis at the level of replication used. Selection for specific gravity components appears to be more effective in the first clonal generation as selection at 75% level of intensity in the first clonal generation resulted in selected clones being almost 6 times more likely to be among the top 10% specific gravity in the second clonal year, than the discarded clones. A more intense selection level of specific gravity in the first year would appear to be counter-productive.

Visual evaluations were used to eliminate undesirable clones among the single-hill population. Caution is advised when selecting for desirable agronomic traits using visual evaluations (Tai, 1975, 1979; Tai & Young, 1984) and efficiency depends upon the breeder's ability to judge a number of traits simultaneously, and the heritability of the traits (Tai & Young, 1984). Tai (1975) reported that visual evaluations could discriminate clones of 'commercial' type, but correlations between visual scores on clones grown in single and 10 hill plots were low. 6% of the single-hill progenies were retained when visually evaluated. In general, selection was done for clones lacking obvious defects. Failure to select a higher percentage of progenies was related to maturity and the number of tubers per hill; six to seven tubers were needed to perform chip test and for replanting in the next year.

The use of diverse parents in crosses is important for maintaining genetic variability among progenies (Simmonds, 1979). The identification of good chipping clones in the early generations was successful; 22 of the 175 field selections produced light colored chips. The progeny population utilized in this study was partly responsible for the list of traits evaluated. The progeny family resulted from crossing of two parents with good color chip and high specific gravity and is the results of breeding goals to produce cultivars similar in appearance to Atlantic. The goal of this research was to individually evaluate visible and invisible traits and then determine which are selectable in early generations. The traits showed consistency of expression. A few of the traits appear to be selectable by a positive approach, some by using a negative approach, and others should probably not be considered in the early generations. In this study, a positive approach would be one in which only clones with acceptable or superior expression of a trait would be saved while a negative approach would be one in which only the clearly unacceptable clones are eliminated and all others are saved. Positive selection – Degree of chip color and severity of skin defects were sufficiently consistent in expression to be susceptible to positive selection. These traits had high correlation coefficients between traits and consistent rejection patterns. Although the rejection for degree of chip color was better when only bad chip color clones were eliminated, implying a need for negative selection, all other data showed the possibility of good response to positive selection.

The GxE interactions of tuber solids and sugar content have been shown to be similar than for yield (Storey & Davies, 1992; Vermeer, 1990), allowing general conclusions about breeding programs to be made for these traits. The same was assumed for chip color because it is closely associated with reducing sugar content. Since chip color is highly heritable (Stevenson & Cunningham, 1961; Accatino, 1973), highly significant chip color differences observed among genotypes, storage treatments and  $G \times E$  interaction were not surprising. It is well documented that genotypes do not respond similarly to cold chilling (Ewing et al., 1981) or reconditioning (Stevenson and Cunningham, 1961; Cunningham and Stevenson, 1963; Gould et al., 1979). It appears that these genotypes accumulated appreciable amounts of reducing sugar to produce dark chips within the first three months of storage and reducing sugars were sufficiently eliminated by reconditioning.

Negative selection – This study suggests that tuber shape, size uniformity and shape uniformity could be efficiently selected against using a negative approach. The strongest evidence for select ability for these traits comes from analysis of rejection patterns. Eye depth was expressed consistently through all analyses with a fairly good rejection pattern, indicating that negative selection should be effective for this trait. Gopal et al., (1992), reported the same conclusion. This may be because the progeny family did not have a wide range of expression for these traits. However, given the reflection pattern, only genotypes showing extreme problems for these traits should be eliminated in the early generations. Visible and invisible traits assessments are an important selection tool in the first, second, and sometimes the third clonal generations. For round-shaped, high specific gravity, and good chip color breeding populations, a breeder's preference scoring system can be constructed using the findings of this study. Clones may first be selected based on degree of eye depth and round tuber shape. Clones can then be eliminated based on the specific gravity and color chip; still others may be eliminated based on yield, tuber number, and average tuber weight. No preference should be based on uniformity of tuber shape or uniformity of tuber size in the early generations.

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