Preface to a review of plant breeding sponsored by the California Avocado Commission

Since 2010 there has been an on-going process of critical review and change to production research funded by the California Avocado Commission (CAC). A number of changes to the system and process of production research have occurred that has set strategic goals and addressed weaknesses in the system. The most notable changes have been to improve the accountability of the research contracts and to place the research efforts in a multi-year context with well-defined objectives and milestones to be met as the research projects are conducted. As a continuation of the review of production research the areas of significant activity that are of strategic interest for the long term progress of the California Avocado industry are being critically examined.

Plant breeding is a sensible very long term strategic activity for CAC to advance the industry and is accordingly an important activity that CAC has sponsored for several decades in partnership with the University of California, Riverside. To this author's knowledge there is no record of a comprehensive review of the plant breeding program that has looked at the goals and objectives of the program, benchmarks of success, a cost benefit analysis and an identification of the barriers to the development of new varieties for commercial use. The fact is, major changes in the industry due to a change in the varietal mix have been rare as a new variety needs to overcome a number of significant challenges to become part of the mainstream varietal mix. To understand these challenges the plant breeding strategic component parts can be defined as: deciding why a new variety is needed, what that variety should have as traits, making the new variety, testing the new variety, outreach on the new variety, and commercial development of the new variety.

The discussion on current and future plant breeding activity has focused on making new varieties. The real impediments to the plant breeding program are not the making of new varieties. The limitations are first to identify why a new variety is needed and then a plan with milestones and benchmarks developed so that the industry understands progress on the new variety and the costs and benefits likely to accrue to the California avocado industry. The second, the long testing period of new varieties which is needed no matter how fast or selective the generation of new varieties. The long testing period is not overcome by new genetic technology which allows more selective techniques to be applied so that wastage in evaluating unsuitable selections can be reduced. Third, in the commercialization of a new variety where there appears to be little effort by CAC to develop the necessary retail pull in demand for a new fruit variety that will lead to enhanced profitability for the grower. CAC's active involvement in the commercialization of a new variety is needed so that handlers will aggressively support the planting of a new variety giving the critical mass necessary to establish and maintain its commercial success. At present there is no call from the major retailers for avocado varieties with different characteristics. This means that to bring a new variety to the retail shelf requires CAC to commit considerable resources for outreach and marketing. To date without CAC support the commercialization of new varieties has largely been unsuccessful in bringing new varieties to the California avocado market.

That the commercialization of new varieties appears to have been poorly managed may be due to CAC having no intellectual property rights to new varieties. Intellectual property rights are forgone as CAC does not fully fund the plant breeding program. The reason the Commission has been disengaged from

the commercialization of new varieties is likely to be that this could be seen as interfering in commercial arrangements between nurseries and UC. CAC may want to seek an alternative arrangement, given the financial limitation of the UC system and the need for increased involvement from CAC.

Without direction from the CAC Board the type of new varieties being bred has been from an interpretation of the industry needs which may be different to the CAC Board's view. Further the new varieties obligate CAC to fund activities that may not be high priority to the CAC Board. The consequence is the CAC Board has a difficult challenge to extricate from future funding commitments should the Commission goals and objectives change. The argument is presented that CAC must continue funding plant breeding, virtually forever, because of past commitments as there is no CAC Board plan for plant breeding. Currently CAC has proposals that, if accepted, would very significantly (about double) CAC investment in plant breeding and genetics research. The Commission does not have unlimited funds and there has always been tension in apportioning funding across and within the various items in the Commissions overall budget. Therefore, not only the amount of funds but also the opportunity cost of committing to long term spending on plant breeding and genetics needs careful consideration. Before committing to long term funding of plant breeding a plan that incorporates benchmarks and performance measures through to the retail shelf that allows tracking of progress and change in activity is therefore essential for prudent management of such an important program. Further the plan should have a process for determining the types of new varieties needed, how new technology can be used for plant breeding and for commercializing new varieties. The plan needs to integrate researchers, handlers, nurseries, retailers, UC and CAC. Without such a plan there is the potential for CAC to provide large sums of money and to achieve little real change in the California Avocado industry.

The current direction of breeding new fruit types appears to be focused on characteristics like superior taste and appearance, early or late maturity and so on. The traits of pest and disease resistance are not high priorities. There is also a program of developing and evaluating new rootstocks that has been running in parallel with the breeding of new fruit varieties. A better use of resources would be combination these program to utilize the diverse skills of the researchers. Developing new rootstock varieties appears to be more straightforward than for a new fruit type as the commercialization of a rootstock does not have the challenge of creating a market niche with retailers. To create new varieties of rootstocks or fruit types the same breeding methods are used and further testing of rootstocks is needed by combining them with fruit scions so that the interactions between the two are understood. With a more immediate use for rootstocks much of the plant breeding focus should be on developing rootstocks with new fruit varieties as a lesser activity.

To make the best use of the potential opportunities new genetic technology the Commission should seek to create partnerships with the wider avocado plant breeding community, both domestically and internationally. The Florida based USDA avocado plant breeding program is particularly interesting because of the significant work they've already completed with genetic markers.

Plant breeding is needed as a long term strategic activity to advance the California avocado industry, but we should recognize that plant breeding is essentially a high risk investment with a high payoff potential. The funding from CAC for plant breeding needs to be prudent in that it is not excessive and nor is it too

little and it is essential that the funding is recognized as a long term requirement that should be consistent in the amount. The Commission has been deficient in better communicating its objectives for plant breeding within the former Production Research Committee structure and to the researchers directly. Therefore, the CAC Board should develop a comprehensive plan with goals and objectives set by the CAC Board. The plan should address the limitations of the current program: in establishing the market driven reasons for a new variety, having a realistic set of varietal traits sought in a new variety, compressing the elapsed time for the testing and evaluation period, improving the outreach to growers for new varieties to explain the financial reasons for planting, creating critical mass for a new variety and obtaining the support of handlers, nurseries and retailers for planting and selling the new variety. Careful consideration needs to be given to improvements in the plant breeding process and how new genetic technology can be used in a cost and results effective manner. Finally, it is clear is that the CAC Board should be more engaged in the area of plant breeding to further the Commission's goals for the California avocado industry.

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EXECUTIVE SUMMARY

A review of the results of the CAC sponsored plant breeding program reveals that a small number of potentially commercially viable rootstocks or fruiting cultivars have been developed during its 55 year history. Very few have been accepted by the California avocado industry with only the California bred or selected Lamb Hass fruiting scion and Toro Canyon rootstock currently being planted in commercially significant numbers. While in principle a plant breeding program could solve some dire issues for California avocado growers, the path to a commercially successful outcome is complex and fraught with numerous roadblocks. Plant breeders need clear direction and prioritization from the industry, as the development period for rootstocks or new fruiting varieties is at least a decade, over which time consumer preferences and grower needs may change significantly. A commercially successful varietal development program must anticipate and incorporate end user demand and retailer preferences (market pull), grower needs and the input of handlers and nurseries. Ultimately, commercial success for new varieties that are different than Hass will require substantial marketing efforts by CAC and an integrated approach in which nurseries and handlers promote and support the planting of new varieties.

Rootstock development is a lengthy process as well, but it is clear that rootstocks which are salt tolerant and/or disease resistant, or that provide dwarfing characteristics would have the greatest immediate utility by the grower community and enhance profitability. The impediments to the development of new varieties whether they are rootstocks or new fruit types has largely been resource limitation and technological inertia. New advances in technology especially in the field of genetics show promise to break the logjam of creating new varieties by more efficient selection processes but does not remove the need for a long testing and development period. The promises made over new genetic technology are largely unfulfilled despite more than 20 years of CAC support for genetic research and will continue to require very significant funding levels just to establish the basis of desirable genetic changes when creating new varieties. Before embarking on what could be a long and expensive research path, CAC should understand the limitations of genetic research for avocado plant breeding. Further, roadblocks to commercial success include a lack of control of the genetic material and the process for releasing the new varieties or rootstocks to the grower community both national and international, and the lack of adequate CAC support for the commercialization process through nurseries. There is further risk in having only one site for the germplasm collection and for much of the breeding program where the quality of water has declined and the site is at risk for development. A site change from Irvine may be an appropriate consideration as part of this review. Finally, the industry will need to develop extensive communication and information sharing processes for the newly released plant material to provide adequate information for growers to make investment decisions on planting new varieties.

AVOCADO PLANT BREEDING REVIEW

The following is an analysis of the Avocado Plant Breeding Program by the CAC Research Program Director. This document describes a review of CAC funded plant breeding activities for rootstocks, fruiting cultivars and their horticultural evaluation. It attempts to be a frank appraisal of the historical record of avocado plant breeding and looks at CAC sponsored avocado plant

breeding not just from a technical perspective but also attempts to look at plant breeding as an investment. This review is one person's interpretation of the California avocado plant breeding program and the opinions expressed within this document may not be in agreement with other industry stakeholders. As such this review takes the opportunity to be provocative and to ask what may be "awkward" questions of the avocado plant breeding program from an industry strategic perspective.

The California Avocado Commission sponsored plant breeding program is a long standing technical activity that has been a serious research effort since 1956. Over the past 55 years considerable effort and money has been expended searching for improved avocado cultivars that will be beneficial to California avocado growers. With such a long running and strongly entrenched research program there is a danger that this review will be seen as an attempt to knock down what some may consider a "sacred cow" of the Commission research sponsorship. This view is wrong. The Commission has undertaken over the past two years to look at its technical investment and make sure all Commission investment is aligned with applying solutions that will meet the strategic needs set by the CAC Board and the Commissions mission statement. Over 55 years the make-up and nature of the Commission has changed as have CAC staff and Commissioners. Much institutional memory is lost and the reasons for why things are done the way they are is no longer clear. Also over time programs can drift from their original purpose or goals such that the program becomes increasingly meaningful only for a minority of the industry. For this reason it is necessary to periodically review and reassess activity that has significant funding implications.

Avocado plant breeding and genetic projects form a significant and continual commitment for CAC production research funding. In 2011 the root rot resistant rootstock breeding program reached a twenty-year mark. Consideration is being given to another long term funding commitment for a further potential 20 years. Therefore, it appears reasonable that now is an appropriate time to critically review the plant breeding program before CAC begins another extended term of plant breeding research. As preparation for this review an avocado plant breeding planning meeting successfully discussed with researchers the goals and objectives of their respective research programs. The topics discussed in the meeting and the questions and issues raised by CAC stakeholders have given guidance on the content addressed in this review that can then be described in concept proposals. In response to CAC's 2011 call for proposals and in determining continued support for ongoing research projects CAC received proposals on plant breeding and genetics totaling more than $640,000 (the annual proposal requests from Smith, Douhan and Dandekar) per annum when added to the $190,000 that is needed for scion breeding and evaluation totals over $830,000 per annum. The concept proposals received by CAC on plant breeding and genetics were not moving the CAC sponsored plant breeding program in a direction indicated by the CAC technical strategic imperatives nor were the proposals pitched within a fully integrated approach. For example, scion breeding and rootstocks are separate programs despite overlap in their testing and evaluation requirements..

Given the potential for plant breeding to advance the California avocado industry there is a desire by CAC to increase the understanding of the current plant breeding approach, to exchange information and to examine plant breeding with regard to it being a strategy in moving the industry forward to achieve the Commissions broad goals. In particular, a much greater understanding of the plant breeding program fundamentals and how they relate to business goals is required. New genetic methods are being proposed as having the potential to increase the efficiency of developing new cultivars and potentially reducing the time needed to bring new cultivars to the industry. Therefore, when considering the application of genetic technology to avocado plant breeding an important question is: What does it mean to go down the genetic path?

BACKGROUND

Typically, plant breeding is a strategy used by plant-based industries when there is a problem that is too expensive to solve using conventional treatments or that appears to have no treatment options. A good example is when root rot was first recognized as caused by *Phytophthora cinnamomi* the science of biochemistry and genetics was very primitive compared to today. Likewise it was recognized that seedlings could by chance have very desirable characteristics, especially with respect to pest and disease resistance, without the need to understand the mechanisms involved in resistance. Additionally opportunities were likely to be seen in developing the avocado market in the USA around Hass-like early or late maturing cultivars as there were no imports. The need for locally grown avocado cultivars that could supply the shoulders of the Hass season has largely been superseded by Hass imports meeting the demand for a year round supply of avocados.

Avocado plant breeding is best considered as an investment in the future of the California avocado industry. When the plant breeding program was initiated there must have been consideration given to the costs and return on investment but there is no written evaluation available. The University of California appears to have initially borne most of the financial burden of a scientific plant breeding program and over time the industry has slowly increased its contribution to the program but still does not fund the full cost of plant breeding. The commercial payoff in developing a new avocado variety with very strongly desired commercial characteristics can, in principle, be very large. However, who benefits financially from a new cultivar is often nebulous with the financial gains not necessarily flowing back to the patent holder or developer of the new cultivar or even to the major financial contributors who funded the plant breeding effort. There does not appear to have ever been an evaluation of the California avocado industry investment in plant breeding and the direct or indirect financial returns on the growers contribution from assessments. Similarly there does not appear to have been a periodic re-examination of the goals for avocado plant breeding with the exception perhaps of setting what traits are desirable. It would appear that the CAC Board has not involved itself in setting the broad goals for avocado plant breeding and in developing commercialization plans for new avocado cultivars for the California industry. Examining the business case for avocado plant breeding and setting the industry goals and objectives will then be very helpful in determining the level of investment in plant breeding needed from the industry to achieve the technical imperatives set by the CAC Board on January 2011.

As almost all commercial avocado varieties had resulted from chance selections when the breeding program commenced, it is likely that a logical, well-organized scientific approach to breeding new cultivars was expected to result in cultivars without the negative traits of chance seedling selections and be expected to be a faster process with less inherent risk of failure. The ability of the California avocado industry to tap into chance selections is now very low as few avocado growers plant avocado seeds to see what trees develop in the hope that they will hit the jackpot with a new variety. The CAC sponsored plant breeding program is the major effort in California on variety improvement.

Before 1937, there was no scientific avocado plant breeding program, instead private individuals and enthusiasts made selection from chance seedlings. The first scientific avocado plant breeding was started in 1937 at UC Riverside and in 1939 at UCLA. These initial programs were small and did not yield any new cultivars. Avocado plant breeding really started in 1956 at UC, Riverside by Dr B.O. Berg who in the first 20 years planted and screened around 15,000 seedlings. From this period came Gwen, Whitsell and Esther with commercial potential. This represented about 1 potential new cultivar per 5,000 seedlings tested. In the second 20 year period around 60,000 seedlings were tested and these resulted in the new cultivars Sir Prize, Harvest, GEM and Lamb Hass. Of these Lamb Hass and GEM appear to have the strongest commercial potential although not necessarily in California (prior to the introduction of Lamb Hass, the most successful commercial varieties were all chance seedlings). This represented a ratio of seedling to new cultivar of 1 per 15,000 seedlings tested. However, Lamb Hass is only slowly increasing in production and GEM has only really been adopted in other avocado growing countries. Over 40 years 75,000 seedlings yielded seven new varieties requiring large acreage and millions of dollars in man hours, grower research spending, UC contributions and lost revenue to growers (Guy Witney presentation to the joint Australia New Zealand Conference, 2005). When Dr. B.O. Bergh retired in the mid 1990's the plant breeding program was nearly stopped due to the perception that plant breeding would not be supported by the California avocado industry. Dr Mary Lu Arpaia essentially rescued the program and restarted plant breeding after about 15 years of no new seedling plantings. Therefore, the clock for scion plant breeding has been reset and the scion breeding program can be considered to be only about 15 years old. This is barely sufficient time for new fruit cultivars to be developed.

The current plant breeding program appears to have four parts:

- Conventional scion breeding: selection and evaluation of improved fruit varieties (this includes new varieties from foreign breeding programs) and rootstocks
- Conventional root rot resistant rootstock breeding: selection and evaluation of improved rootstocks (this includes new rootstocks from foreign breeding programs) with resistance to *Phytophthora cinnamomi* and latterly other traits like salt tolerance, dwarfing etc.
- Application of molecular markers to avocado improvement
- Germplasm preservation

AVOCADO TRAIT OBJECTIVES

The table below lists the traits Dr B.O. Berg was using in evaluating seedlings for suitability as new avocado cultivars. From reading the California Avocado Society Yearbooks these traits are an expansion of the characteristics the CAS Variety Committee used. What is not clear is whether the traits were included because this was what the market wanted through feedback from retailers and major stakeholders other than growers or handlers.

(B.O. Berg, 1976 Proceedings of the First International Tropical Fruit Short Course)

Fruit quality	
Medium size	Thick ovate shape
Uniformity	Pulp
Skin	Proper softening
Medium thickness	Appetizing color
Readily peelable	Absence of fibers
Insect, disease resistance	Pleasing flavor
Free from blemishes	Long shelf life
Attractive color	Slow oxidation
Long on the tree storage	Chilling tolerance
Seed	High oil content
Small	High nutritional value
Tight in its cavity	
Shoot qualities	
Spreading habit	Tolerant of chlorosis
Easy to propagate	Tolerant of other stresses
Strong grower	Short fruit maturation period
Tolerant of pests and diseases	Precocious
Tolerant of wind	Regular bearing
Tolerant of cold	Wide adaptability
Tolerant of heat	Heavy bearer
Tolerant of salinity	
Rootstock qualities	
Conducive to high quality fruit	Easily grafted
Conducive to healthy, productive trees	Tolerant to *Phytophthora* and other organisms
Free from sun-blotch	Tolerant of salinity
Dwarfing or semi-dwarfing	Tolerant of chlorosis
Genetically uniform	Tolerant of drought
Hardy and vigorous	Tolerant of other adverse soil conditions
Easily propagated	

The list of traits is quite extensive and detailed. It would appear that to get a single new avocado variety with all these traits would be a near impossible task. It has proven very difficult to just breed a root rot resistant rootstock that has only one really strong trait. It may be better to limit the traits to a maximum of the 5 most important and so long as other desirable traits are not wholly negative a new cultivar with the desirable traits will be very valuable. Therefore, putting priorities on the traits would be useful in helping guide plant breeders to those traits of greatest value to the California avocado industry.

GENETIC RESEARCH

For many commercial fruit crops a mixture of classical plant breeding and genetic engineering are used in plant breeding programs. These two general approaches to plant breeding are used by researchers who are trying to develop a new variety with specific characteristics and are used in the scientific avocado breeding programs the world over. New methods that more deliberately seek to change specific genes of a plant are generally referred to as genetic engineering. The understanding and technology of genetic engineering has increased a great deal in the past decade as has the genetic information on avocados, most notably the genome of avocado has been sequenced. The potential now exists for novel methods of generating new varieties to be applied to avocado plant breeding. Plants genetically modified with genes from distantly related organism are often referred to as transgenic plants, while plants genetically modified with genes from closely related organisms are referred to as cisgenetic plants. In both situations genes need to be inserted into the host plant using genetic engineering methods. With genetic engineering only the genes of interest are changed allowing a more precise creation of new varieties and is considered to speed up the classical plant breeding process in making new varieties. The time needed to evaluate a new variety remains the same, however.

The genes that influence a desirable trait can be as few as one or involve many different genes. The new techniques are tools that can more efficiently screen seedlings for the desirable traits as long as the genes of interest have been identified and positively associated with the desired traits of a new cultivar. One such method is known as marker assisted selection. The markers used are not the genes themselves but a marker associated with a quantitative trait locus (QTL) in a genome. Marker assisted selection is used when traits are hard to measure, have low inheritance or are only expressed once the plant is mature. In the case of avocados any selection for desirable characteristics of the fruit has to wait until fruit appear on the tree. With marker assisted selection these traits can be assessed when the seedlings are newly emerged well before the seedling flowers. Marker assisted selection is not perfect as there is a reliance on having markers that are tightly linked to the gene of interest requiring QTL mapping of the genome to occur first. To develop QTL maps a cross between two contrasting varieties is needed then the plants grown on so that the traits can be analyzed and assessed. This requires significant research before markers can be applied.

The genetics research on avocados is still, despite many decades of development, still fundamentally at the stage of creating a foundation for better understanding of the functions in the tree. The utilization of the increased genome information is still a long way off, possibly 15 years (Dr Mary Lu Arpaia comment at the Avocado Plant Breeding Planning Meeting) and more conventional breeding methods will still need to be employed. New genetic techniques would appear to have most merit if part of a bigger plan for breeding where sufficient resources can be supplied so that goals and objectives of the breeding program are well described for the plant breeder to try and achieve. The Commission then needs to commit to the program for the long term.

PROGRAM ACTIVITY

There are a number of different techniques available today for plant breeding from simply selecting interesting new plants for propagation to sophisticated genetic engineering methods. The traditional method of plant breeding for fruit crops has been to allow fruit to result from natural crosses between varieties and then screening large numbers of seeds for interesting traits. This method is referred to as classical plant breeding and has been the main method used in the UC Riverside plant breeding of new avocado varieties. Classical plant breeding is generally a hit and miss method requiring an element of luck and large numbers of seeds for screening to obtain a new desirable variety. To confirm that a new variety has commercially useful characteristics there is a long period, ten to fifteen years or more, of testing required. The testing has to demonstrate the horticultural performance of the new variety in the field is consistently superior to other varieties and that the genetics of the new variety is stable through a number of a propagation cycles.

Avocados are a long lived tree, a commercial life of greater than 80 years is possible, where the seedlings have a relatively long juvenile stage, potentially up to 14 years, before fruit are produced. An avocado plant breeding program using traditional techniques for fruiting cultivars will therefore be long term potentially requiring years before the fruit produced by trees can be evaluated. It is almost certain that at least 10 years and possibly 20 years could elapse between the first seedling and a final commercial cultivar with good investment potential. Because breeding avocados is long term the funding requirements are also long term. The approximate total spending on plant breeding and associated research (genetics, somatic hybridization and evaluation) by the California Avocado Commission for the development of rootstocks and fruiting cultivars from 1991 to 2010 has been over $5,6000,000 ($280,000 per year average, in 2011 a further $440,000 for the scion breeding, rootstock breeding and marker assisted selection was funded) from a total research spending of about $16,000,000 in the same period.

PLANT BREEDING ACTIVITY SPONSORED BY CAC

Enhancement of avocado productivity. Plant improvement: selection and evaluation of improved varieties and rootstocks.

This research program has been led by Dr Arpaia since 1997 and essentially continues the program run for several decades by Dr Bergh. The stated goal of the avocado scion breeding program in the 2009-2010 research progress report is: "to help maintain and enhance the California avocado industry by introducing consistently heavier producing, high quality avocado varieties, better pollenizer varieties and to test improved rootstock hybrids". To achieve the goal there will be continued evaluation of new material generated by traditional selection techniques, collaboration with other researchers as they develop refined techniques to increase the efficiency of selection and introduction of new material from other improvement programs. The program has changed slowly over time since 1997 where there are now tiers or stages of evaluation. Tier 1 is the first evaluation of fruit from seedlings, Tier 2 is the evaluation of fruit from trees propagated from the most promising selections from Tier 1. The Tier 2 trees are based at the South Coast Research and Extension Center (SCREC). The trees from the Tier 2

evaluation selected for Tier 3 evaluation are propagated onto suitable rootstock and then planted at locations statewide from San Diego to Tulare county.

A rootstock trial planted in 1999 using Lamb Hass and Hass is nearly complete with the rootstocks Zentmeyer, Dusa and Evstro having the greatest cumulative yields under root rot free conditions. A new rootstock trial is planned with more scions and rootstock varieties.

Genetic research has focused on developing molecular markers for alternate bearing that can be used to select for trees with low biennial bearing traits. A mapping population of Bacon and Hass seedlings is being established to generate a genetic linkage map to identify key horticultural traits of precocity, productivity, fruit quality, tree architecture and cold/heat tolerance.

Marker Assisted Selection and Linking Candidate Genes to Biochemical Phenotypes in Avocado

In a number of research projects led by Professor Michael Clegg and latterly Dr Harley Smith considerable effort has being going into establishing genetic maps of valuable traits and a population of trees that can be used for quantitative genetic analysis. In 2001, four replicates of each of 200 open pollinated Gwen seedlings on Duke 7 rootstocks were planted. The objective of the research is to ultimately allow a program of marker assisted selection of desirable traits when evaluating seedlings in the early stages of the plant breeding program. The value of having a large number of trees of many genetic types is that the influence of the genes can be separated from the influence of the environment or location. Initially four traits were selected, tree height and diameter, diameter of the stem, fruit productivity of the trees and flowering data. This research came up with a pool of 127 SSR markers. By 2007 a new class of molecular markers called SNPs were being developed to assess cultivar origins and had been used to identify the racial composition of many avocado cultivars and set up the genetic research for the next project looking at the genes controlling nutritional composition of avocado fruit. The amount of a number of nutritional compositional compounds were found to be strongly linked to genes although the environment also had a strong influence. The project of linking candidate genes to biochemical phenotypes in avocados was successful indentifying markers for putative health-related nutrients that has been used over the past couple of years to select seedlings with the genes that are related to high levels of health-related nutrients.

Screening and Evaluation of New Rootstocks with Resistance to Phytophthora cinnamomi

This program has had several different program leaders over the years with the most recent Professor John Menge who retired about 2005 when the program was passed onto Dr Greg Douhan.

The objective of this program has been to collect, select, breed and develop avocado germplasm that is tolerant to *Phytophthora cinnamomi* root rot of avocado. In recent years six rootstocks, Thomas, Dusa, Toro Canyon, Uzi, Steddom and Zentmeyer have been released or are in the process of release. A further six rootstock selections have strong promise, Brandon,

Eddie, Anita, Johnson, VC801 and VC207. The breeding program has proceeded by inoculating avocado seedlings with *Phytophthora cinnamomi* under greenhouse conditions. Plants are screened for up to two years in the greenhouse under heavy and continuous Phytophthora pressure. The plants are then clonally propagated and saved as advanced germplasm at the South Coast Experimental Field Station in Irvine. The plants are allowed to grow for several years until enough budwood can be collected to produce clonal rootstock trees. It takes a full year to produce the trees and then they are tested under field conditions using Hass as the scion in location where *Phytophthora cinnamomi* is known to be a problem. From the initial seedling screening stage to being planted for the first time can take up to five years. The trees then start to yield in three or four years time and it takes more time and testing in further locations to determine if the rootstocks are acceptable. Since 1989, 58,500 seedlings have been screened, an average of 2,785 per year, and the program currently is testing 42 rootstock selections in 18 field trials. Most of these selections were progeny from 9 maternal cultivars including Duke 7, Thomas, G6, Spencer, Duke 9, UC2001, Barr Duke, Toro Canyon and PP40. The rootstock program is now currently establishing a 'second generation' of advanced selections to move to field trials that were originally collected from the better forming rootstocks. There is also advanced selections from some of the VC maternal parents, which are all salt tolerant selections to begin the process of incorporating salt tolerance and root rot resistance in a single rootstock.

Other activity in the root rot tolerant rootstock breeding program has been to investigate the genetic diversity of *Phytophthora cinnamomi* isolates associated with root rot in California. There are two different 'clonal' genotypes present in California one of which is suspected to be a recent import into the Southern growing area. The different isolates will be used in the greenhouse screening so that the seedlings are challenged by the real diversity of *Phytophthora cinnamomi* in California.

GENERAL PLANT BREEDING PROCESS AND COMMENT ON CAC INVOLVEMENT
Once it is decided that new cultivars with desired traits is an appropriate solution to an industry goal or problem, there are a number of general steps in the plant breeding process.

1. Identify traits of interest - the details of the discussion where the traits were derived is not known. It is likely they were developed from discussion between the plant breeder and members of the California Avocado Society when the plant breeding program was initiated. And was based on previous characteristics developed by the CAS varieties committee.

2. Generate new plants - the method of open pollination between different cultivars is used to get new seedlings for evaluation and is well explained in the research proposals and progress reports.

3. Identify if the plants have the traits of interest - the plants are grown until they produce fruit and meet some of the traits of interest. This is also well explained in the research proposals and progress reports.

4. Evaluate the characteristics of the new plants - the plants are propagated to determine of the promising traits exhibited are stable and likely to be present if the new plants are several propagation cycles distant from the original mother tree. This step also allows for negative traits to appear. This activity is well explained in the research proposals and progress reports.

5. Test horticulture performance - the plants are grown to evaluate yield potential and alternate bearing and horticultural characteristics in a controlled setting. This is well explained in the research proposals and progress reports.

6. Field test new varieties - the new plants are tested on groves in different soils and environmental conditions for productivity and performance under more typical grower management. While this is explained in the research proposals the quality of tree management is very variable depending on the care supplied by the grower and could be improved and more use could be made of recording the mistakes made in growing the new variety.

7. Release for commercial use - CAC appears to have little involvement in the decisions regarding release of new cultivars for commercial use.

8. Commercial development - there is only a very limited support from the Commission in assisting the development of new varieties on groves. As a result it would appear that California avocado acreage planted in new varieties is very small. More collaboration is required with nurseries, handlers and retailers to identify the information needed for business decisions by growers.

The Commission generally has most input into the steps of the general plant breeding process through the requests for funding for plant breeding projects. The overall involvement by the Commission in each of the steps in the process does not appear to be very strong with Commission support for the commercial development of a new variety appearing to be largely absent. For best results in bringing a new avocado variety into the industry the Commission needs to have more input and partnership within the plant breeding effort in identifying traits of interest and the broader goals of the plant breeding effort. To assist in the horticulture performance evaluation the knowledge of which that will assist with the commercialization of new cultivars.

INVESTMENT IN PLANT BREEDING AND ASSOCIATED RESEARCH PROJECTS

The investment by the Commission in the past 20 years on plant breeding and associated projects has been substantial in total but relatively modest when the average funding per year is considered (see table and figure below). The plant breeding program has had modest success with a small number of new avocado cultivars released commercially and a small number of those cultivars planted in significant numbers by California avocado growers. Two rootstocks, Toro Canyon and Dusa, have had commercial success. However, Toro Canyon was not bred and is a chance selection while Dusa is also a chance selection from South Africa. The California plant breeding program successfully evaluated of these two rootstocks suitability for California conditions and soils allowing commercial release. The fruiting cultivar Lamb Hass has

been planted in reasonable numbers and has in recent years produced 11,000,000 to 15,000,000 pounds per year for good prices. This is the most successful fruiting cultivar from the California plant breeding program. Based on the very limited numbers of new avocado cultivars that are commercial successes, when judged by the criteria of being planted by California avocado growers, and the time and investment in the CAC sponsored plant breeding program, the potential for delivery of new avocado cultivars from plant breeding that will be a commercial success appears to be very low. The plant breeding program as a research effort is therefore a high risk strategy to solve an industry problem or as a strategy for meeting an industry goal.

STRENGTHS AND WEAKNESSES OF THE CURRENT PLANT BREEDING PROGRAM

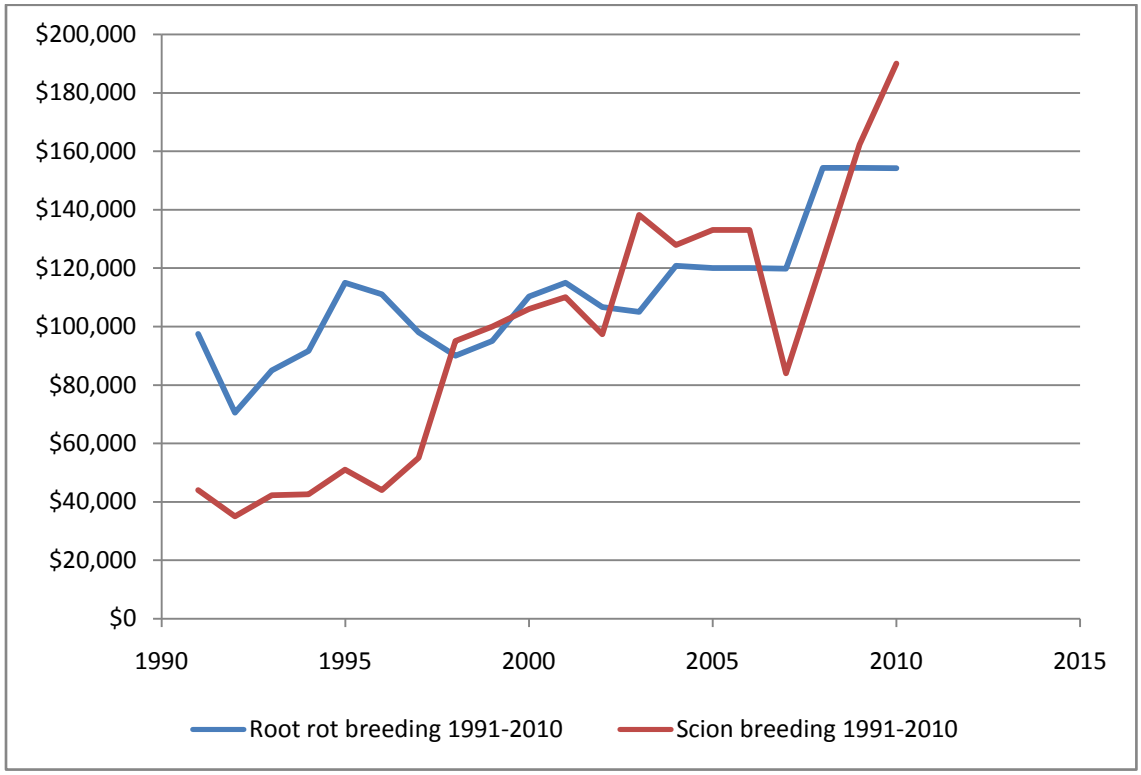
No research program is perfect and all have a range of strengths and weaknesses. The current plant breeding program has a number of strengths and weaknesses listed in the table below.

Strengths	Weaknesses
Large cultivar breeding base	CAC no control over intellectual property
Established plantings (good breeding capital)	No trained plant breeder
Horticultural evaluation of California bred and imported cultivars is strong	Fragmented activity (no team approach) rootstocks and scions separate
Cost reasonable ($5.6 million in 20 years)	No CAC commercialization assistance plan
Could modernize quickly using up to date genetic technology	Breeding objectives not reviewed or verified by industry
Produced seven new cultivars in 20 years	Two rootstock breeding programs
Access for foreign breeding programs	Lack of prioritization of numerous cultivar traits
Produced one commercially viable fruit (Lamb Hass)	Lack of focus on salinity and water
Produced commercially viable rootstocks	CAC no financial stake in the results
	Potential for cost inflation
	Germplasm at one site
	One institution provides all research

INVESTMENT IN PLANT BREEDING AND ASSOCIATED RESEARCH PROJECTS 1991-2010

Project	Investigator	Years	Funding
Screening/Evaluation (rootstocks)	Menge/Coffey/Douhan	1991-2010	$2,233,859
Avocado Breeding, Productivity, Improv.	Arpaia	1994-2010	$1,292,116
Variety Breeding	Bergh	1991-1993	$121,200
Clonal Trial-Rootstock Prod Trial	Arpaia	1991-1997	$85,400
Persea species Collection	Scora	1994-1996	$7,900
Outcrossing in Avocados	Clegg	1993-1996	$144,000
Avo Rootstock Dev by Somatic Hybr	Litz - UF	1996-2002	$352,091
Avocado tissue culture rejuvenation	Hardison	1995	$5,000
Avocado Germplasm preservation	Arpaia	1996	$10,000
Genetic markers in avocado improvement	Clegg	1997	$40,000
Somatic Embryogen/Persea amer.	Litz - UF	1995	$15,000
Molecular genetics	Clegg	1991-1994	$134,000
Germplasm Acquisition & Bio	Scora	1992-1993	$5,200
Avocado Germplasm Preservation	Arpaia	1998	$10,000
Development of Molecular Markers, Microsat	Clegg	1998-2010	$817,588
Rootstocks vs Root Rot & Salinity	Zilberstaine - Israel	1998-2001	$60,000
Rootstock Screening & Salinity Mgmt of Trees	Crowley/Arpaia	2006	$63,000
Implementation of Molecular Markers	Smith	2010	$103,000
Lamb Hass Maturity/Fruit Quality	Arpaia	1999-2001	$178,600
		Total:	**$5,677,954**

YEARLY INVESTMENT ON SCION BREEDING AND ROOT ROT RESISTANT ROOTSTOCKS



In the past twenty years the costs of plant breeding have steadily risen as inflation erodes the value of money and the University has a declining funding base declining more when the economic cycle moves from expansion to recession. As the plant breeding program uses long-lived trees that have ongoing costs for maintenance new money is required when new research projects are undertaken rather than a redirection of funds. It is potentially very difficult to change the direction of a plant breeding program if change is needed.

With any long term investment like plant breeding much can change over the lifetime of the program. And it is essential that such a program be designed to allow for changing needs over time. When setting goals and objectives for a plant breeding program there is a risk that the market conditions the traits of interest are intended to meet will change substantially over the lifetime of the breeding program. Therefore, setting broad goals may be the best option as after 20 years the original reasons for developing the new cultivar may no longer be valid. Periodic review of the plant breeding goals, more than just the traits of interest, has merit in terms of limiting wasted investment on new cultivars that may have little perceived value to growers by the time they are ready for release.

NEW GENETIC TECHNOLOGY

Genetic engineering techniques have been proposed that allow either a very targeted selection of traits of interest or for the new plants to be altered to have the traits of interest without the need for random pollination of different varieties. Genetic engineering new avocado cultivars may have potential to deliver new cultivars for evaluation. This may take out some of the element of luck in finding an ideal new cultivar but does not relieve the bottleneck in the time required to test new varieties. The greatest issue with genetically altered avocado plants is having a reliable and efficient regeneration protocol following gene transfer, i.e. getting viable plants once the genes have been altered in cell cultures. The most research progress currently is in the regeneration trees for rootstocks. Avocados have proved to be difficult to tissue culture such that commercial production of trees does not use tissue culture techniques. This remains the greatest constraint to genetic engineering of avocado trees.

The technology to alter genes in plant cells is available. In order to change the genetics of the plants knowledge of which genes control important traits is required. In the marker assisted selection projects much of the cost has been in establishing which markers are the correct ones to assess as relating to the traits of interest. Fortunately for the California avocado industry the sequencing of the avocado genome is near completion in Mexico. Much of this genome will become publicly available in the next few years greatly expanding the potential knowledge base on avocado genetics. However, even with the genome sequenced there will be considerable additional research needed to understand the genetics and mechanisms within the avocado tree that may be associated or directly related to desirable traits. It is expected that the cost of additional research on genetics will be high and the reduction in the uncertainty of delivering new commercially desirable cultivars is unknown. The potential consumer and customer resistance to genetically engineered avocado cultivars remains unknown.

QUESTIONS RAISED

In writing this assessment of the CAC plant breeding program (including research projects on genetics designed to assist the plant breeding program) the author is left pondering a number of challenging questions:

Why should the California Avocado Commission invest in plant breeding?

1. What are the compelling reasons for CAC to invest in avocado plant breeding?
2. How can avocado plant breeding help to achieve the strategic technical imperatives set by the CAC Board at the January, 2011 Board meeting?
3. What should be the goals of the plant breeding program?

What level of investment does a plant breeding program need?

4. What would be the business case for investment in avocado plant breeding?
5. What is the opportunity cost of a significant investment in avocado plant breeding?
6. Can funding for the plant breeding program be leveraged from other funding sources?
7. How can the breeding capital be maintained without becoming cost prohibitive?

How could a plant breeding program be organized and conducted?

8. Is there scope for the plant breeding program to be conducted by a range of research providers?
9. What is the potential for new commercially viable avocado cultivars to be released within the next 5 years to California avocado growers?
10. Should CAC have ownership rights to new commercially viable cultivars released to California avocado growers?
11. Should the plant breeding on rootstocks and fruiting cultivars be combined into one project?
12. Should CAC assist with the commercialization of new cultivars?
13. Should CAC also have a role in the importation and evaluation of new avocado cultivars from around the world?

What personnel and activities are needed?

14. What range of technical expertise would be required to move the plant breeding program forward should change be desirable?
15. Would a team approach of an integrated program incorporating a plant breeder, plant physiologist, horticulturist, plant pathologist, entomologist and soil scientist be an advantage to achieving the goals of a plant breeding program?
16. What new technologies are proposed to improve the plant breeding program and what are their limitations and resource requirements?

What can be done to measure success?

17. When should review of the plant breeding program be conducted as meeting the goals and objectives?
18. What are the limitations of the plant breeding program and potential solutions?

CONCLUSIONS

In 2011, over $640,000 of requests to fund plant breeding and genetics research projects was received by the Commission. Were all of the funding requests to be granted and added to the funding already approved for the scion plant improvement project there would be more than a doubling, to about $830,000 per year, of the Commissions current investment in plant breeding and associated research. In order to evaluate such a potentially large investment a greater understanding of the current plant breeding program and an evaluation of its successes and limitations is needed. The increased investment has a considerable opportunity cost in that the money spent on plant breeding and genetics would not be spent on research into what some may consider issues of greater importance and impact on California avocado growers, such as water use efficiency.

The Commission has determined that it has the strategic technical imperatives of: Increasing average per acre production, Maintaining a premium quality product, Effective grower education and Maintaining critical industry mass. In principle, plant breeding can be helpful in increasing production, maintaining a premium product and critical industry mass through new cultivars suited to new more marginal growing areas. In practice, the plant breeding program over time has been marginal in its delivery of new cultivars to advance the California avocado industry. This is the reason as to why an investment in avocado plant breeding would be viewed as high risk with only a relatively small chance of return on investment. However, should the right cultivar be found and its development supported by the Commission the financial payoff to the wider industry could be very large.

The reasons for the plant breeding program failing to meet expectations are summarized below:

- There is a long lag time in evaluating the horticultural characteristics of interest resulting in a very extended evaluation period.
- Lack of clear Commission set goals for the plant breeding program which align with grower and consumer needs.
- The Commission despite sponsoring plant breeding research has no direct financial stake in new cultivars or in the decision to release new cultivars to growers.
- The Commission has not had much involvement in the product development phase in supporting new avocado cultivars as part of a strategy to meet the technical imperatives.
- Two parallel rootstock programs appear to be wasteful of talented researchers and their resources.
- Overall more plant stock, i.e. trees, and better tools are needed to enhance the productivity of the plant breeding program.
- Lack of industry support for new cultivars – Sir Prize, Harvest, etc

The tools to improve the plant breeding process that show the most promise, currently, are the new genetic technologies. Unfortunately, the genetic tools are still in a development phase and are likely to take many years before significant gains in efficiency are made within the plant breeding program. It will take time to identify the genes of interest associated with specific traits

and how their manipulation can be utilized in the development of new cultivars. The genetic work may allow the faster development of new cultivars but there will remain an extended period of testing to be sure the traits are expressed in the field. A further unknown is the acceptability of genetically engineered or enhanced plants to the market.

Rootstocks appear to have the greatest significant commercial value being more straightforward to market to growers and do not have the complication of needing to develop a critical mass and markets for a new fruit variety. Further rootstocks with desirable traits like root rot resistance, salt tolerance and dwarfing characteristics would have more immediate value and payoff to growers than a new fruit cultivar. An added bonus is that rootstocks lend themselves better to the process of genetic engineering as the characteristics engineered into the new plant are unlikely to transfer easily to the scion, i.e. the fruit from grafted trees with engineered rootstocks are not themselves genetically engineered.

What appears to be needed to advance the plant breeding program is dialogue with the plant breeding researchers where some of the questions raised in this document can be answered or discussed. From a purely business investment point of view plant breeding is a long term high risk activity and much of that risk will not be removed through the application of new genetic techniques as considerable understanding of the genetics will take time and a lot of additional funding. The Commission needs to decide what it sees as the goals of a plant breeding program and communicate these to the plant breeding researchers.