

Performance of a tetraploid avocado rootstock 'Canacado VC-320'

L. Winer^a

Department of Fruit Crops Extension, Israel Ministry of Agriculture.

Abstract

In 2008 a commercial group developed an avocado rootstock colchicine-induced autotetraploid. In this paper, we present systematic data collected over seven years from 'Hass' avocado trees grafted onto this tetraploid rootstock as compared with four other common rootstocks in Israel. 'Hass' trees grafted onto the tetraploid rootstock 'Canacado VC-320' are significantly more productive than the other vegetative clone rootstocks included in the present study. The cumulative yield of 'Hass' avocado grafted on 'Canacado VC-320' rootstock over 5 consecutive seasons was 65.8 t ha⁻¹ and the range of yield of the other vegetative clones over the same period was 37.6 to 52.9 t ha⁻¹. 'Hass' avocados on tetraploid rootstock were also more productive than grafted onto the 'Degania 117' seedling rootstock. The tetraploid rootstock 'Canacado VC-320' also showed less sensitivity than other rootstocks/scion combinations to branch dieback related to *Botryosphaeria* infection.

Keywords: *Botryosphaeria*, 'Hass', colchicine, polyploidy

INTRODUCTION

There is considerable genetic diversity among the various species in the avocado genus, particularly regarding fruit skin type and color, flesh color, and flavor, as well as tree sensitivity to different soil conditions (Cui, 1993; Ferguson and Huang, 2007). This diversity provides many opportunities for developing new types of avocado fruit and rootstocks. Hybridization between different types of avocado has been applied in research programs all over the world. Another strategy for improving avocados is manipulating plant ploidy levels in vitro via chromosome doubling by using antimetabolic agents that disrupt mitosis to improve avocado rootstock (Wu et al., 2011). From the late 1930s to the early 1950s, colchicine was used to double the chromosome number of numerous crop species (Blakeslee and Avery, 1937; Eigsti, 1938, 1992). Dermen from the USDA led this effort by inducing polyploidy in apples (Dermen, 1952; USDA, 1956), cranberries (Dermen and Bain, 1944), grapes (Dermen, 1954; USDA, 1955), peaches (Dermen, 1947a), pears (Dermen, 1974b) and strawberries (Dermen and Darrow, 1938). Chromosome doubling of diploids was achieved by in vitro colchicine treatment of somatic tissues from mature vines combined with the use of flow cytometry to identify the autotetraploid plants produced (Wu et al., 2011). Although induction of autotetraploid is a procedure that has been used for at least 80 years, there seems to have been little systematic evaluation of fruit quality and morphology or of the mature cropping potential of the induced tetraploids. Recently, Saleh has suggested that tetraploid citrus rootstocks are more tolerant of salt stress than their corresponding diploid (Saleh et al., 2008).

Researchers in Israel have invested numerous efforts to overcome inflorescence and branch dieback related to *Botryosphaeria* fungus contamination (Sheinberg and Simanski, 2017).

In this paper, we present systematic data collected in Israel over seven years from 'Hass' avocado trees grafted onto a commercial autotetraploid avocado rootstock colchicine-induced in comparison with four other common rootstocks (Ben-Ya'acov, 1976).

^aE-mail: winerleo@gmail.com



MATERIALS AND METHODS

Plant material

Budwood from a colchicine-induced autotetraploid provided by the Kaima company named 'Canacado VC-320' was grafted onto rootstock 'Degania 117' to provide plant material for vegetative proliferation. This process was performed at Haskelberg Nursery, according to a method based on the principles developed by E.F. Frolich from the University of California Los Angeles, who developed an etiolation method that was modified and patented by Brocaw Nurseries in California. According to this method, an avocado seed is grown in a container to serve as a nurse for the future rootstock. Once the seedling is large enough for grafting, a cleft graft is made, and the graft is left to develop. The young plant is placed in a dark room, and the shoot is permitted to grow chlorophyll-free for a few weeks. A loosely clamped metal ring is placed just above the bud union. The etiolate tissue is treated with IBA to induce rooting. A portion of the new stem adjacent to the bud union is covered with sterile rooting media in a second container, after which it is removed from the dark chamber. When the adventitious roots develop successfully, the nurse seed is severed from the rooting stem, and the newly rooted shoot is given time to develop under humid conditions. Once the plant hardens off, it is grafted onto the 'Hass' cultivar and is allowed to develop for 2 to 3 months. This double-grafted plant is later transplanted to a larger container and then grown for several more months. The ring placed on the base of the etiolate tissue slowly constricts the growing plant, eventually severing the nurse seed. The same method was used for the production of three other West Indian vegetative rootstocks from Dr. Avraham Ben Yaakov's collection in Israel: 'VC-28', 'VC-52' and 'VC-65'. All of the vegetative proliferated rootstocks were grafted using the 'Hass' variety 'Degania 117', a seed proliferated rootstock from the Ben Yaakov collection was also used in the present research. Those rootstocks, all of West Indian origin, were characterized by Dr. Ben Yaakov according to their sensitivity to lime, salt conditions, and productivity. All of the rootstocks can grow with good production in conditions of maximum 20% lime and water salinity of 250 mg L⁻¹ chloride of irrigation water.

Plantation location

The experimental plot was planted in a light soil farm in the Sharon region in the center of Israel on 21 August 2011.

Plantation plot

Twenty-five plants of each rootstock grafted with the 'Hass' cultivar were planted in a plot with loamy soil. Each rootstock was planted in 5 replicates of 5 plants per replicate and randomly distributed in the spacing of 6×3 m. Ettinger cultivar was planted as a pollinizer in a ratio of 1:9. The experimental plot was drip-irrigated with wastewater with an average of 200 mg L⁻¹ chloride.

Tree development

The rootstock and scion diameter were measured immediately after planting on 22 August 2011 to record the standard of the plants. Eighty-eight weeks after planting, on 1 May 2013, the diameter of the scion was measured again to record the relative development of the trees.

Botryosphaeria sensitivity

Branch dieback related to *Botryosphaeria* contamination was determined in two different surveys. The first survey was performed about four years after planting (15 June 2015), and the second survey was performed 7.5 years after planting (28 December 2018). The branch dieback index was determined visually according to the following scale: 0=no dieback, 1=low index, 2=medium index, 3=high index. The percentage of plants with index of dieback higher than 2 was calculated.

Yield production

The yield of every replicate was collected separately, and the mean accumulated yield of every group of rootstocks was determined over five seasons (from 2014 to 2018). Variance analysis was used to determine statistical differences between the mean yield of each rootstock/scion combination.

RESULTS AND DISCUSSION

Plant development

The 'Canacado VC-320' plants planted on 21 December 2011 was significantly less developed than the other rootstocks planted on the experimental plantation. The rootstock means diameter of 'Canacado VC-320' was 12.6 mm on the day of planting (21 August 2011). The mean diameter of all other rootstocks in the experimental plantation ranged from 14.2 to 15.1 mm (Figure 1). The scion on 'Canacado VC-320' plants was also less developed than in all four other rootstock scion combinations planted in the experimental plantation. The scion of 'Canacado VC-320' plants had a mean diameter of 10.9 mm, and the diameter of the other rootstock scion combination ranged from 12.6 to 14 mm on the day of planting (Figure 1). The diameter of the scion was measured again 88 weeks after planting (1 May 2013). Our records indicate that the scion of 'Canacado VC-320' still had a development gap showing a mean scion diameter of 325 mm, which is significantly less than the mean diameter of all other combinations that showed a range between 336 to 345 mm (Figure 2).

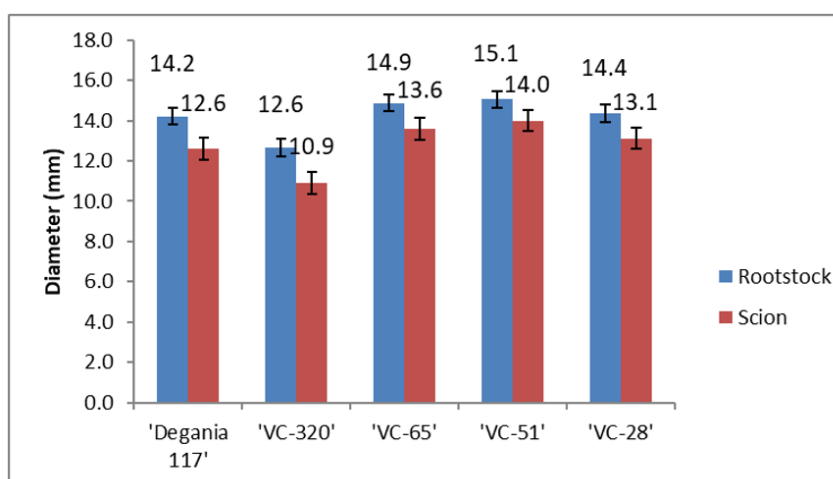


Figure 1. Rootstock and scion diameter on the day of planting. The ranges represent the calculated standard error of the mean.

Botryosphaeria sensitivity

The susceptibility of a 'Hass' avocado grafted onto tetraploid 'Canacado VC-320' rootstock was significantly lower compared with 'Hass' plants grafted onto other rootstocks. In the 2015 survey, the relative number of trees grafted onto 'Canacado VC-320' scion that showed medium and high index of dieback was only 9.1% compared with higher proportions in the trees of 'VC-51', 'VC-65', 'VC-28' and 'Degania 117', where the proportion of trees highly infected was 16.7, 44.4, 34.8, and 47.8%, respectively (Figure 3).

In the 2018 survey, we determined a much more intensive proportion of dieback as a consequence of botryosphaeria infection. The less sensitive trees were those grafted onto 'Canacado VC-320', with 26.1% of trees showing a high index of branch dieback.

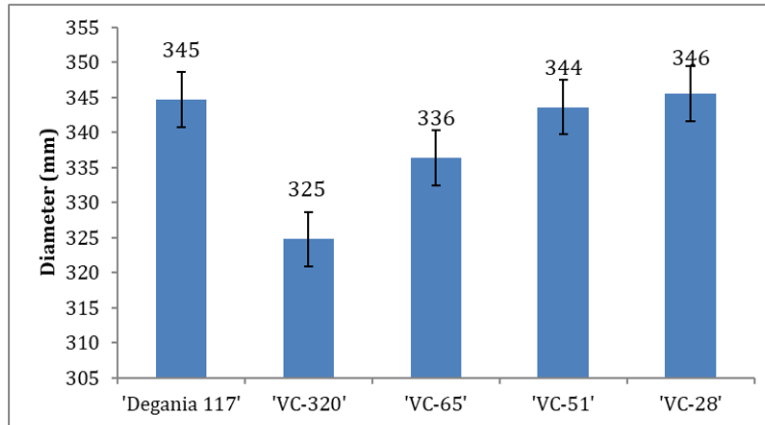


Figure 2. Scion diameter 88 weeks after planting. The ranges represent the standard error of the mean.

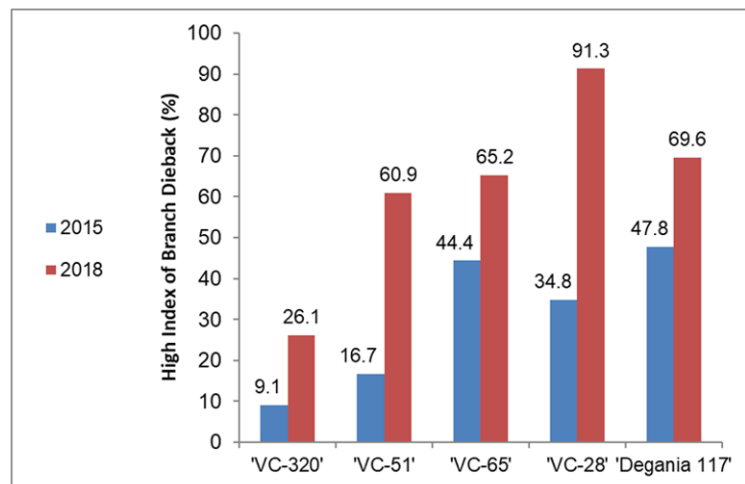


Figure 3. The percentage of plants with a high index of branch dieback related to *Botryosphaeria* was determined in 2015 and 2018.

The other trees grafted onto 'VC-51', 'VC-65', 'VC-28', and 'Degania 117' demonstrated a proportion of trees with a high index of branch dieback of 60.9, 65.2, 91.3, and 69.6%, respectively (Figure 3).

Yield

The crop from 'Hass' avocado trees grafted onto 'Canacado VC-320' was significantly higher compared to other scions/rootstock combinations. The first yield in 2014 of trees grafted onto 'Canacado VC-320' was more than 2.5 times higher than the other three combinations (Figure 4). The second crop collected from trees grafted onto VC-320 in 2015 (9.32 t ha^{-1}) was 1.5 times higher compared to the crop from trees grafted onto 'Degania 117' (6.47 t ha^{-1}) (Figure 4). In 2016 the crop of trees grafted onto 'Canacado VC-320' (11.96 t ha^{-1}) was similar to that of the trees grafted onto 'VC-65' (12.25 t ha^{-1}) (Figure 4). Also, in 2017, the crop of 'Canacado VC-320' and 'VC-65' were similar (10.54 and 10.30 t ha^{-1} , respectively) (Figure 4). In 2018 there were favorable conditions for a large "on" crop in Israel. In the experimental parcel, we also saw a high yield ranging from 19 to 31 t ha^{-1} . In those conditions as well, we saw a slight advantage with 'Hass' trees grafted onto 'Canacado VC-320', with the highest yield of 31.47 t ha^{-1} (Figure 4). The cumulative yield over five seasons was significantly higher with 'Hass' trees grafted onto 'Canacado VC-320' with 65.8 t ha^{-1} (Figure 5). The second

best mean cumulative yield was for 'Hass' trees grafted onto the seedling rootstock 'Degania 117' and the vegetative clone 'VC-65' with 51.8 and 52.9 t ha⁻¹, respectively (Figure 5). The third best mean cumulative yield was with the vegetative clone trees of 'VC-51' and 'VC-28', with 37.6 and 41.3 t ha⁻¹, respectively (Figure 5).

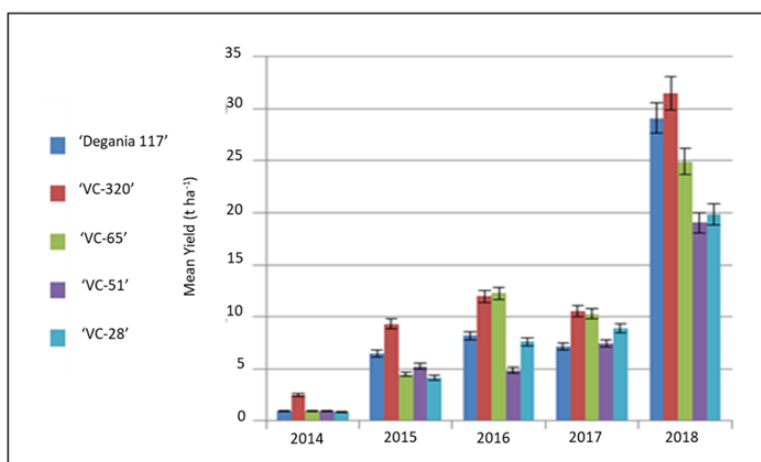


Figure 4. The mean yield of five seasons (2014/15, 2015/16, 2016/17, 2017/18, and 2018/19). The ranges indicate the standard error of the mean.

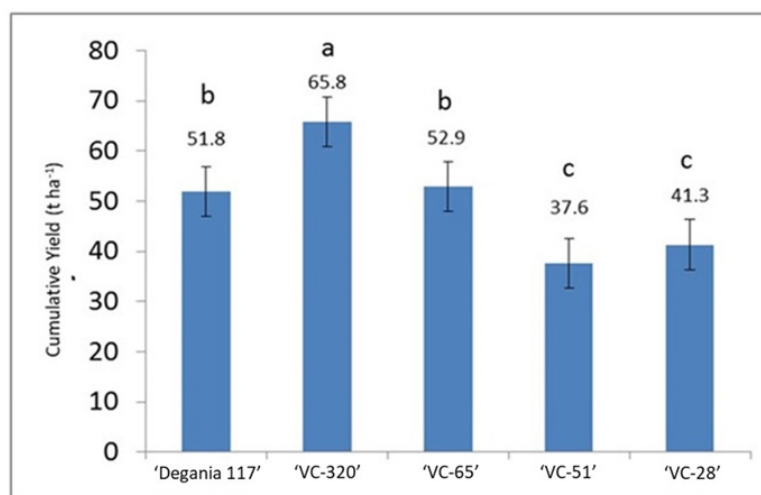


Figure 5. The mean cumulative yield of five seasons (2014/15, 2015/16, 2016/17, 2017/18, and 2018/19). The ranges indicate the standard error of the mean.

CONCLUSIONS

'Hass' trees grafted onto the tetraploid rootstock 'Canacado VC-320' are significantly more productive than the other vegetative clone rootstocks included in the present study. The tetraploid rootstock was also more productive than 'Hass' avocados grafted onto the 'Degania 117' seedling rootstock. The tetraploid rootstock 'Canacado VC-320' also showed less sensitivity than other rootstock/scion combinations to branch dieback related to botryosphaeria infection.

ACKNOWLEDGEMENTS

I am very grateful to Prof. Dani Steinberg from the Department of Plant Protection and Pathology and Mr. Eli Simanski for their assistance with the surveys for determining

botryosphaeria contamination in the experimental plantation in 2015 and 2018.

Literature cited

- Ben-Ya'acov, A. (1976). Avocado rootstocks in use in Israel. California Avocado Society 1975–76. Yearbook 59, 66–68.
- Blakeslee, A.F., and Avery, A.G. (1937). Methods of inducing doubling of chromosomes in plants: by treatment with colchicine. *J. Hered.* 28 (12), 393–411 <https://doi.org/10.1093/oxfordjournals.jhered.a104294>.
- Cui, Z.X. (1993). *Zhongguo Mihoutao [Actinidia in China]* (Jinnan: Shandong Science and Technology Publisher).
- Dermen, H. (1947a). Inducing polyploidy in peach varieties. *J. Hered.* 38 (3), 77–82 <https://doi.org/10.1093/oxfordjournals.jhered.a105695>. PubMed
- Dermen, H. (1947b). Polyploid pears. *J. Hered.* 38 (6), 189–192 <https://doi.org/10.1093/oxfordjournals.jhered.a105726>.
- Dermen, H. (1952). Polyploidy in the apple found seven years after colchicine treatment. *J. Hered.* 43 (1), 7–8 <https://doi.org/10.1093/oxfordjournals.jhered.a106264>.
- Dermen, H. (1954). Cochploidy in grapes. *J. Hered.* 45 (4), 159–172 <https://doi.org/10.1093/oxfordjournals.jhered.a106467>.
- Dermen, H., and Bain, H.F. (1944). A general cytohistological study of colchicine polyploidy in cranberry. *Am. J. Bot.* 31 (8), 451–463 <https://doi.org/10.1002/j.1537-2197.1944.tb08056.x>.
- Dermen, H., and Darrow, G.M. (1938). Colchicine-induced tetraploid and 16-ploid strawberries. *Proc. Am. Soc. Hortic. Sci.* 36, 300–301.
- Eigsti, O.J. (1938). A cytological study of colchicine effects in the induction of polyploidy in plants. *Proc. Natl. Acad. Sci. USA* 24 (2), 56–63 <https://doi.org/10.1073/pnas.24.2.56>. PubMed
- Eigsti, O.J. (1992). Introduction/H Kihara. In *Classic Papers in Horticultural Science*, J. Janick, ed. (Englewood Cliffs, N.J.: Prentice Hall), p.554–556.
- Ferguson, A.R., and Huang, H. (2007). Genetic resources of kiwifruit: domestication and breeding. *Hortic. Rev. (Am. Soc. Hortic. Sci.)* 33, 1–121 <https://doi.org/10.1002/9780470168011.ch1>.
- Saleh, B., Allario, T., Dambier, D., Ollitrault, P., and Morillon, R. (2008). Tetraploid citrus rootstocks are more tolerant to salt stress than diploid. *C. R. Biol.* 331 (9), 703–710 <https://doi.org/10.1016/j.crvi.2008.06.007>. PubMed
- Sheinberg, D., and Simanski, E. (2017). Confrontation with botryosphaeria in avocado orchards in Israel. Research report submitted to Israel Avocado Committee (in Hebrew).
- USDA U.S. Department of Agriculture. (1955). Big grapes coming. *Agr. Res.* (December) (Washington, D.C.: U.S. Dept. Agr.).
- USDA U.S. Department of Agriculture. (1956). Big apple with a big future. *Agr. Res.* (January) (Washington, D.C.: U.S. Dept. Agr.).
- Wu, J.H., Ferguson, A.R., and Murray, B.G. (2011). Manipulation of ploidy for kiwifruit breeding: in vitro chromosome doubling in diploid *Actinidia chinensis* Planch. *Plant Cell Tissue Organ Cult.* 106 (3), 503–511 <https://doi.org/10.1007/s11240-011-9949-z>.