# Polyembryony in Mango (Mangifera indica L.) Is Controlled by a Single Dominant Gene

# Y. Aron, H. Czosnek<sup>1</sup>, and S. Gazit

Faculty of Agricultural, Food and Environmental Quality Sciences, The Hebrew University of Jerusalem, Rehovot 76100, Israel

## C. Degani

Agricultural Research Organization, The Volcani Center, Bet Dagan 50250, Israel

Additional index words. genetics, inheritance, plant breeding

Abstract. The segregation pattern of individuals originating from selfing of several monoembryonic cultivars and one polyembryonic line indicated that polyembryony in mango was of genetic nature. All the plants originating from monoembryonic cultivars bore monoembryonic fruits. A one-monoembryonic to three-polyembryonic segregation pattern was observed among individuals originated from the polyembryonic line, indicating that polyembryony in mango is under the control of a single dominant gene.

Polyembryony, the formation of several embryos in one ovule, was reported in 59 families, 138 genera, and 239 species (Tisserat et al., 1979). Several fruit trees, including mango, have polyembryonic genotypes. Of these embryos, one is sexual and the rest are vegetative, emerging from the nucellus tissue that envelops the embryo sac. The vegetative, or nucellar embryos, and the plants developing from them are clones of the mother plant, regardless of the pollen parent genotype. Thus, polyembryonic plants are used extensively as rootstocks in mango and other species. Polyembryonic mango cultivars are more productive than monoembryonic cultivars, perhaps because of a stronger stimulus for development of the young fruit by the vegetative embryos (Campbell, 1961; Knight, 1970; Sturrock, 1967). Several theoretical models of the genetics of polyembryony in mango have been proposed, but have not been tested experimentally. Sturrock (1968) proposed that polyembryony is controlled by a single recessive gene, and that the monoembryonic cultivar Hayden is an outcome of a cross between the homozygous dominant monoembryonic cultivar Mulgoba and the polyembryonic 'Turpentine'. If so, the gene controlling polyembryony in 'Hayden' is heterozygous. In addition, Sturrock assumed that the monoembryonic Edward resulted from a cross between the monoembryonic 'Paheri' and the polyembryonic 'Carabo', and that the gene controlling polyembryony in 'Edward' is heterozygous.

Received for publication 19 Nov. 1997. Accepted for publication 9 July 1998. The cost of publishing this paper was defrayed in part by the payment of page charges. Under postal regulations, this paper therefore must be hereby marked *advertisement* solely to indicate this fact.

Henryk Czosnek; to whom all reprint requests should be addressed. Tel: 972 8 9481249; fax: 972 8 9468265; e-mail: czosnek@agri.huji.ac.il

According to Oppenheimer (1955), the monoembryonic 'Maya' also originated from a polyembryonic cultivar. The aim of our paper is to report segregation to mono- and polyembryony in mango populations derived from selfing of the monoembryonic cultivars Edward, Hayden, and Maya (which are presumably heterozygous for the gene controlling polyembryony), a polyembryonic selection 13/1, and its monoembryonic descendant 21/6.

#### **Materials and Methods**

Trees of 'Edward', 'Hayden', and selections 13/1 and 21/6 growing at the Hebrew Univ. and Habsor experimental farms were enclosed in net (50 mesh) cages before flowering in 1992 and 1993. A small beehive was placed in each of the cages at the beginning of flowering. Seeds from ripe fruits from these trees were planted in 750-mL plastic pots. The emerging seedlings were examined for five enzymic loci to assure that they were products of selfing, and, in 13/1, to select seedlings resulting from sexual crosses. [For details of the allozyme analysis, see Aron et al. (1997)]. We also examined a number of young trees originating from selfing of 'Hayden' and 'Maya' in the mango breeding program of Drs. Degani and Lavi of the Volcani Institute. Scions from seedlings that resulted from selfing in the monoembryonic cultivars and from fertilization in the polyembryonic line were grafted onto mature trees to shorten the time required for fruiting. First fruits were harvested in the next season and the number of embryos per seed was examined. In an attempt to find association between the polyembryonic gene and DNA markers, the segregating population was further analyzed for rapidly amplified polymorphic DNA (RAPD) markers together with 11 monoembryonic and 64 polyembryonic cultivars and selections, including 13/1, and the sexual seedlings derived

from the polyembryonic selection 13/1. Extraction of DNA and the RAPD analysis were performed according to Schnell et al. (1995) using 251 primers (kits A to M; Operon Technologies, Alameda, Calif.).

#### **Results and Discussion**

During the 1992 season, only a small number of fruits developed on caged trees, but much better yield was obtained the next year. The total number of fruits obtained from the caged trees varied from 13 to 274 in 1992 and 1993. Allozyme analysis showed that all cultivars and selections other than 'Edward' were homozygous at one to three loci (Table 1). Origin from selfing was determined if the seedlings were homozygous to the same relevant locus and segregated for other loci. We found no seedlings with unexpected profiles, and assumed that all were derived from selfing.

Sexual seedlings derived from the polyembryonic selection 13/1 were examined to determine if they were homozygous to any of the loci *Idh*, *Tpi*, and *Pgm-1*, to which 13/1 is heterozygous. Based on these three loci, ≈88% of the seedlings were sexual. Two hundred plants originating from selfing of the monoembryonic cultivars and selections and 35 sexual derivatives of the polyembryonic 13/1 were grafted on old mango trees in Spring 1993 and 1994, and bore fruits 2 years later.

All the selfed derivatives of monoembry-onic parents bore monoembryonic fruits (Table 2). Segregation was observed among the 13/1 derivatives; 24 individuals had polyembry-onic fruits, and the other 11 had monoembry-onic fruits. This is a monohybrid segregation pattern ( $X^2 = 0.771$ , P = 0.7-0.8), indicating that polyembryony in 13/1 is governed by a single dominant gene, and that 13/1 is heterozygous for that gene. These results contradict Sturrok's assumption (1968) that polyembryony in mango is genetically recessive. According to Campbell (1992), 'Edward' originated from a natural cross between 'Hayden'

Table 1. Allozyme profile of mango cultivars and selections grown in cages.

Cultivar/	Enzyme locus				
selection	Aco	Pgi-2	Idh	Tpi	Pgm-1
Edward	ac	ab	ac	ab	ac
21/6	cc	aa	cc	ab	ac
13/1	cc	· aa	ac	ab	ac
Maya	cc	ab	cc	aa	ac
Hayden	cc	ab	ac	ab	aa

Table 2. Segregation for polyembryony in populations of mango cultivars and selections originated by selfing.

Cultivar/ selection	Monoembryonic plants	Polyembryonic plants
Edward	37	0
Hayden*	19	0
Maya*	9	0
21/6	135	0
13/1	11	24

\*Including individuals from the mango breeding program, Volcani Institute.

## BREEDING, CULTIVARS, ROOTSTOCKS, & GERMPLASM RESOURCES

(female) and the polyembryonic 'Carabao' (male). If so, 'Carabao' should be heterozygous for the polyembryonic gene, because no segregation to mono- and polyembryony was noted in the selfed population of the cultivar 'Edward'. Moreover, the involvement of 'Carabao' in the origin of 'Edward' has been dismissed on the basis of the allozyme profile (Degani et al., 1990) and DNA fingerprints (Adato et al., 1995). The dominant nature of polyembryony in mango, as indicated in this study, is similar to that reported for citrus. The difference may be in the number of genes involved. The involvement of a single gene was reported in citrus by Parleviet and Cameron (1959) and supported by Iwamasa et al. (1967). Two complementary genes were reported in tetraploid citrus (Cameron and Soost, 1980). On the other hand, Deidda and Chessa (1982) proposed the involvement of three dominant genes.

Of the primers used, 226 were informative but none of them showed any association with poly- or monoembryony. Recently, Lopez-Valenzuela et al. (1997) reported, following examination of nine polyembryonic and six monoembryonic mango cultivars, an association between a RAPD marker originating from the primer OPM12 and polyembryony. We

employed this specific primer as well, but no association was found in the segregating population or in the bulk of cultivars, which included some of those employed by Lopez-Valenzuela et al. (1997). The discrepancy between these two results may be due to the small and fortuitous number of cultivars used by Lopez-Valenzuela et al. (1997).

#### Literature Cited

- Adato, A., D. Sharon, U. Lavi, J. Hillel, and S. Gazit. 1995. Application of DNA fingerprints for identification and genetic analysis of mango (*Mangifera indica*) genotypes. J. Amer. Soc. Hort. Sci. 116:322–326.
- Aron, Y., H. Czosnek, S. Gazit, and H. Degani. 1997. Segregation distortion and linkage of isozyme loci in mango. HortScience 32:918– 920.
- Cameron, J.W. and R.K. Soost. 1980. Mono- and polyembryony among tetraploid *Citrus* hybrids. HortScience 15:730–731.
- Campbell, C.W. 1961. Comparison of yields of polyembryonic and monoembryonic mangos. Proc. Fla. State Hort. Soc. 74:363–365.
- Campbell, R.J. 1992. A guide to mango in Florida. Fairchild Tropical Garden, Miami.
- Degani, C., R. El-Batsri, and S. Gazit. 1990. Enzyme polymorphism in mango. J. Amer. Soc. Hort. Sci. 115:844–847.

- Deidda, P. and I. Chessa. 1982. A three-gene hypothesis for inheritance of nucellar embryony in citrus. Revista della Ortoflorofrutticoltura Italiana 66:431–437.
- Iwamasa, M., I. Ueno, and M. Nishiuri. 1967. Inheritance of nucellar embryony in citrus. Bul. Hort. Res. Sta. Okitsu, Japan, Series B 1–8.
- Knight, R.J. 1970. Polyembryonic mangos, their utilized potential. Proc. Trop. Reg. Amer. Soc. Hort. Sci. 14:145–155.
- Lopez-Valenzuela, J.A., O. Martinez, and O. Parades-Lopez. 1997. Geographic differentiation and embryo type identification in *Mangifera indica* L. cultivars using RAPD markers. HortScience 32:1105–1108.
- Oppenheimer, H. 1955. Growing subtropical fruit trees (in Hebrew). Am Oved Publ.
- Parleviet, J.E. and J.W. Cameron. 1959. Evidence on the inheritance of nucellar embryony in *Citrus*, Proc. Amer. Soc. Hort. Sci. 74:252–260.
- Schnell, R.J., C.M. Ronning, and R.J. Knight. 1995. Identification of cultivars and validation of genetic relationships in *Mangifera indica* L. using RAPD markers. Theor. Appl. Genet. 90:269–274
- Sturrock, T.T. 1967. Nucellar embryos of mango. Proc. Fla. State Hort. Soc. 80:350–354.
- Sturrock, T.T. 1968. Genetics of mango polyembryony. Proc. Fla. State Hort. Soc. 81:311–314.
- Tisserat, B., E.B. Esen, and T. Murashige. 1979.
  Somatic embryogenesis in angiosperms. Hort.
  Rev. 1:1–72.