Characterizing the Novel Fruit Surface Morphology of 'Marina' Peach Using Scanning Electron Microscopy

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Abstract. Scanning electron microscopy (SEM) was used to compare the novel surface morphology of 'Marina' peach [plant introduction (PI) 133984] to a normal peach ('Contender') and a nectarine ('Sunglo'). Samples were collected before, during, and after anthesis. Compared to 'Contender', 'Marina' showed different trichome structure, lower trichome density, and delayed initiation of trichomes on the gynoecium. No pubescence was observed on 'Sunglo' nectarine at any sampling date. Trichomes were present on the flower bud scales of all three cultivars. Arrangement and structure of trichomes on flower bud scales of 'Marina' differed from those on 'Contender' and 'Sunglo'.

Presence or absence of fruit pubescence, controlled by the *G* locus, is used for differentiating two botanical varieties of *Prunus persica* (L.) Batsch: *P. persica* var. *persica* (peach) and *P. persica* var. *nucipersica* (Suckow) C.K. Schneid. [syn. var. *nectarina* (Ait. f.) Maxim.] (nectarine). Homozygous (*gg*) trees produce fruit that lack pubescence and are called nectarines. The peach allele is completely dominant to the nectarine allele (Conners, 1920). Many trichomes, part of the epidermal cell layer (Dorsey and Potter, 1932), form to produce the fruit pubescence. Examination of the peach plant introduction (PI) and variety collection at North Carolina State Univ. showed PI 133984 ('Marina' peach) had a novel fruit surface morphology unlike all other peaches and nectarines in the germplasm collection. 'Marina' originated as a bud sport from 'Duke of York' peach (Brooks and Olmo, 1972) and was introduced into the United States from South Africa in 1939 (U.S. Dept. of Agriculture, 1950). 'Duke of York' resulted from a cross of 'Early Rivers' nectarine with 'Alexander' peach (Hedrick, 1917). The objective of this study was to compare the novel fruit surface morphology of 'Marina' peach to a standard peach and nectarine at different developmental stages using scanning electron microscopy (SEM).

Materials and Methods

Gynoecia (pistils) or fruit of 'Marina' peach, 'Contender' peach, and 'Sunglo' nectarine were collected at various intervals during ontogeny. The earliest samples were collected about 8 weeks before anthesis and the last sample was collected at fruit maturity. Samples were fixed in a 2% glutaraldehyde solution buffered with potassium phosphate to pH 7.3. After fixation,

samples were rinsed with buffer solution and dehydrated in a graded ethanol series (50%, 70%, 90%, and 100% three times). Samples were critical-point dried with CO₂ in a Tousimis Samdri PVT-3B critical-point drier at a pressure of 1200 to 1450 psi. Samples were mounted on aluminum SEM stubs using silver-conducting paint and sputter-coated with 25 nm of a gold-palladium mixture at three equatorial angles and one polar angle using a Technics Hummer V sputter-coater. Sputter-coating thickness was measured with an Anatech Ltd digital thickness monitor in the fast position. Samples were observed on a Philips 505-T scanning electron microscope at various magnifications and tilt angles with a kV setting of 10 and the spot size set on 50. Micrographs were created with Polaroid type 55 film.

Results

Gynoecia collected about 8 weeks before anthesis from all three cultivars were glabrous (Fig. 1 a–c). Samples collected about 4 weeks before anthesis showed that 'Marina' and 'Sunglo' were glabrous, but 'Contender' had initiated trichome development, as evidenced by small circular protrusions on the ovary (Fig. 2 a–c). At anthesis, 'Marina' was glabrous to the unaided eye, but SEM showed initiation of trichome development near the juncture of the ovary and style (Fig. 3b). The sample collected from 'Contender' at anthesis showed the ovary was fully pubescent (Fig. 3a), and the ovary of 'Sunglo' remained totally glabrous (Fig. 3c). Observation of 'Sunglo' nectarine at subsequent sampling dates showed no pubescence on the gynoecium or fruit surface (Figs. 4c, 5, 6d, and 7c). Pistils of 'Marina' collected at shuck split (2–3 weeks after anthesis) had both pubescent and glabrous sectors (Fig. 4b). The glabrous and pubescent sectors showed no distinct pattern on the ovary, and the pattern of trichome distribution varied from fruit to fruit. Trichome development was initiated on the basipetal portion of the style and was absent on the stigmatic end (Fig. 5). Gynoecia of 'Contender' were fully pubescent on the ovary and most of the style, but the stigma and stigmatic end of the style remained glabrous (Figs. 4a and 5). Trichomes on 'Marina' and 'Contender' were unicellular. Trichome density on 'Contender' appeared greatest about 8 weeks after anthesis (Fig. 6a). 'Marina' fruit collected at this time had two distinct sector types on the fruit surface, designated smooth and rough. Smooth sectors were characterized by low trichome density and atypical trichome morphology (Fig. 6c). The atypical trichomes had a peg-like base and many were

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Fig. 1. Electron micrographs of gynoecia collected about 8 weeks before anthesis show lack of pubescence on 'Contender' peach (A), 'Marina' peach (B), and 'Sunglo' nectarine (C). Magnification is 94 $\times$ (A), 98 $\times$ (B), and 86 $\times$ (C).

Fig. 2. Electron micrographs of gynoecia collected about 4 weeks before anthesis show trichome initiation on 'Contender' (A), but 'Marina' (B) and 'Sunglo' (C) are glabrous. Magnification is 90 $\times$ (A), 102 $\times$ (B), and 98 $\times$ (C).

Fig. 3. Electron micrographs (41 $\times$) of gynoecia collected at anthesis show that 'Contender' (A) is fully pubescent and 'Marina' (B) has initiated trichome production. 'Sunglo' (C) is still glabrous.

Fig. 4. Electron micrographs (18 $\times$) of gynoecia collected 2 to 3 weeks after anthesis show 'Contender' (A) is pubescent, 'Sunglo' (C) is glabrous, and 'Marina' (B) has pubescent and glabrous sectors.

Fig. 5. Light micrograph (9 $\times$) comparing 'Contender', 'Marina', and 'Sunglo' gynoceia (left to right) collected 2 to 3 weeks after anthesis, showing pubescent and glabrous sectors on 'Marina'.

folded over or broken off at the base. Additionally, a few flat, ribbon-like trichomes were observed on smooth sectors of 'Marina'. Smooth sectors on these fruit probably corresponded to the glabrous sectors seen on the fruit examined 2 to 3 weeks after anthesis. Rough sectors on 'Marina' (Fig. 6b) had a greater trichome density than smooth sectors (Fig. 6c) but a lower trichome density than 'Contender' (Fig. 6a). Trichomes on rough sectors had abnormalities identical to those on smooth sectors. At fruit maturity, trichome density on 'Marina' and 'Contender' was less than that of samples collected at 8 weeks after anthesis (Fig. 7 a and b). At maturity, trichomes were easily removed from fruit of 'Marina' with slight hand pressure, unlike those of 'Contender'.

Flower bud scales from all cultivars collected about 8 weeks before anthesis were pubescent (Fig. 8 a–c). Trichomes on 'Marina' flower bud scales were abnormal compared to those on 'Contender' and 'Sunglo'. Trichome abnormalities found on flower bud scales of 'Marina' were similar to the abnormalities found on fruit. Flower bud scales from 'Contender' and 'Sunglo' had trichomes that were

Fig. 6. Electron micrographs of fruit surface 8 weeks after anthesis show 'Contender' (A) with a high trichome density compared to a rough (B) and smooth (C) sector on 'Marina'. 'Sunglo' (D) is glabrous. Magnification = 370 $\times$, except B = 155 $\times$.

Fig. 7. Electron micrographs (267 $\times$) of fruit surface at maturity show a reduced trichome density on 'Contender' (A) and 'Marina' (B). 'Sunglo' (C) is glabrous.

normal and identical to each other. Stomata were present on the floral bud scales and fruit surfaces of all three varieties.

Discussion

SEM showed that trichomes on the gynoecia, fruit, and flower bud scale surfaces of 'Marina' were abnormal compared to 'Contender' peach and to flower bud scales from 'Sunglo' nectarine. Our results agree with those of Dorsey and Potter (1932), and show that all rudimentary gynoecia of peach were glabrous in early winter. Gynoecia and fruit surfaces of 'Sunglo' were glabrous at every sampling date and had many stomata. Delayed initiation of trichomes on the gynoecia, reduced trichome density, abnormal trichome morphology, the presence of pubescent and glabrous sectors on young fruit, the near absence of trichomes on the style, the presence of trichomes easily removed with slight hand pressure, and the presence of rough and smooth sectors on the mature fruit distinguished 'Marina' from 'Contender'.

The genetic and histogenic basis of the phenotype of 'Marina' has not been determined. According to Brooks and Olmo (1972), 'Marina' arose as a bud sport from 'Duke of York' (heterozygous *Gg*) peach. It is possible that 'Marina' is a periclinal chimera. Dermen and Stewart (1973) determined the relative contributions of the three histogenic layers (denoted as L-I, L-II, and L-III) to the developing and mature peach fruit. Using cytochimeras, they determined that all three histogenic layers contribute to the ovary of the fruit, but that the contribution of the L-III histogenic layer was restricted to the ovary and the basal region of the style. The L-III layer was absent in most of the style and in all of the stigma. The lack of L-III derived tissue in the stigmatic end of the style and the absence of pubescence on the stigmatic end of the style of 'Marina' suggests a possible role of the L-III in its phenotype. Based on studies of chimeric peach-nectarine fruit and nectarine bud sports, Dermen (1960) has suggested that the L-III may influence the formation of pubescence on the L-I derived epidermis in peach fruit. 'Marina' may be an appropriate clone to further investigate this possibility. 'Marina' has been self-pollinated and crossed to peach and nectarine cultivars to determine if the unique phenotype is heritable. These crosses will also provide information on the genotypic composition of the L-II histogenic layer of 'Marina'.

'Marina' is not unique in having a distinct trichome morphology on the fruit surface and flower bud scales. Okie and Prince (1982) reported a novel fruit epidermis on 3 out of 70 F_1 seedlings obtained from a cross of 'Pekin' peach $\times$ 'Durbin' nectarine. The three seedlings lacked the long, unicellular trichomes typically present on normal peaches, but instead were covered with very short, multicellular trichomes. Additionally, these three seedlings had glabrous flower bud scales unlike typical peach clones. Subsequent unpublished studies by Okie (personal communication) indicated that this character is controlled by a single recessive gene that is not allelic to *G*. Wen et al. (1995) compared the characteristics of peaches and the nectarine bud sports that arose on them. They concluded that each bud sport differed from the parent clones and from the other bud sports. Our results suggest the possibility that additional alleles may exist at the *G* locus in peach. Alternatively, our results support the observations of Fogle and Faust (1975) and Wen et al. (1995) and suggest that there may be genes that modify the expression of the *G* locus. In contrast to normal nectarines, which have pubescent flower bud scales and glabrous fruit, 'Marina' has aberrant trichome morphology on both the fruit and flower bud scales. This suggests that 'Marina' may possess a novel allele at the *G* locus that controls both trichome morphology

Fig. 8. Electron micrographs of flower bud scales collected about 8 weeks before anthesis show normal trichomes on 'Contender' (A) and 'Sunglo' (C) but aberrant trichomes on 'Marina' (B). Magnification is 86 $\times$ (A) and 76 $\times$ (B and C).

and organ specificity of expression. Additionally, the phenotype conferred by these different alleles may be influenced by the specific histogenic layers containing the mutant allele.

Our results agree with those of Fogle and Faust (1975) and confirm the lack of pubescence on nectarine fruit. Early in development, 'Marina' was glabrous similar to a nectarine. Later developmental stages showed that the phenotype of 'Marina' was intermediate between that of a peach and nectarine. Unlike a normal peach, the pubescence on 'Marina' was easily rubbed off with only slight hand pressure. This characteristic may have economic value, allowing the pubescence to be removed with only light pressure during postharvest handling. Mechanical removal of peach pubescence before shipping is a common postharvest practice. Dorsey and Potter (1932) proposed that brushed peach fruit was more susceptible than nonbrushed peach fruit to brown rot [*Monilinia fructicola* (Wint.) Honey] infection. Smith (1936) reported that the major avenue of infection was through broken trichomes. Field observation of fruit on 'Marina' showed it was highly susceptible to infection by the brown rot fungus. The basis of this susceptibility is unknown, but the broken trichomes on its fruit surface may provide an entrance for this pathogen.

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