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## ECTOPIC EXPRESSION OF ATMYB90 IN TOMATO RESULTS IN INCREASED ANTHOCYANIN ACCUMULATION AND DEFECTIVE POLLEN DEVELOPMENT.

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*MYB90, anthocyanins, pollen development*

Anthocyanins are naturally occurring secondary metabolites, responsible for the red/purple pigmentation of many plant organs. At the transcriptional level, anthocyanin biosynthesis is mainly regulated by well-characterized bHLH and MYB transcription factors (TFs). We produced tomato plants (cv. Micro-Tom) ectopically expressing the Arabidopsis *AtMYB90/ Production of Anthocyanin Pigment2* (*PAP2*) gene under the control of the constitutive *CaMV35S* promoter. Three independent *35S:MYB90* lines (L1, L2, and L3) disclosing high levels of transgene expression were selected for further analyses. At the seedling stage, the transgenic lines revealed intense pigmentation of leaves and petioles. Consistently, L1, L2, and L3 tissues disclosed increased expression of key endogenous anthocyanin biosynthetic genes, including *Phenylalanine Ammonia-Lyase* (*PAL*), *Chalcone Synthase* (*CHS* ), *Dihydroflavonol 4-Reductase* (*DFR*), and A*nthocyanidin Synthase* (*ANS*), compared with the wild type. At anthesis, the *35S:MYB90* flowers showed intense red pigmentation of the stamens. Anthocyanin accumulation occurred in the external epidermis and in the hairs located on the anthers' internal surface. We did not detect pigment accumulation in immature or mature fruits from the transgenic lines. At maturity, L1, L2, and L3 plants yielded a slightly reduced number of tomatoes compared with the wild type. Similarly, fruit size and fruit weight were partially reduced in the transgenic lines. Most interestingly, fruits from the *35S:MYB90* plants displayed a significant reduction in the number of mature seeds, compared with the control plants. On average, the wild type yielded 16.5 ± 0.9 seeds per fruit, whereas seeds production was limited to 3.5 ± 0.4 seeds/fruit in L1, to 3.6 ± 0.5 in L2, and to 2.8 ± 0.4 in the L3 line (mean ± S.E., $p<0.001$, ANOVA). Microscopic examination revealed that anthers from the *35S:MYB90*

lines produced a significantly increased number of small or stunted pollen grains compared with the wild type. Alexander staining indicated that such defective pollen grains were largely non-viable. Scanning electron micrographs confirmed that wild type pollen grains were turgid and oval-shaped, whereas the majority of transgenic pollen grains were small and collapsed. As a whole, these findings uncover a possible role for MYB90 in pollen development and fertility, which has not been previously reported in tomato or other plant species.