

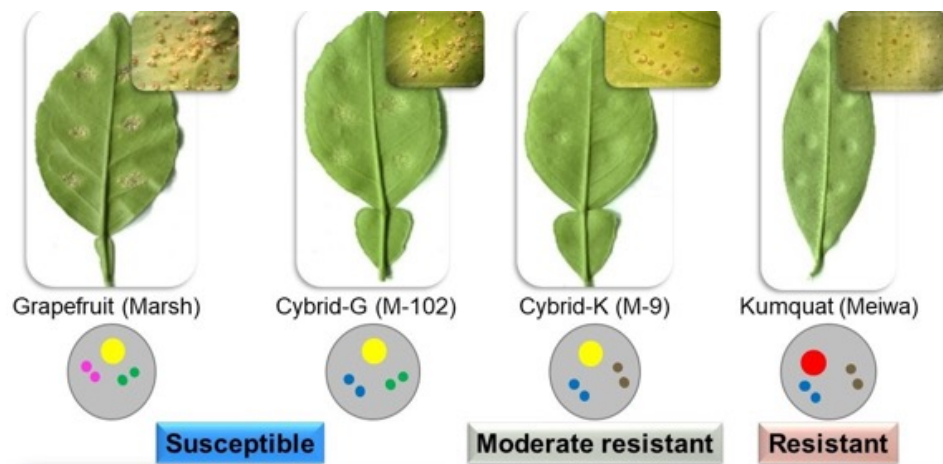
# Cybridization for Plant Improvement: Grapefruit Cybrids have Potential for Canker Improvement

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Cybridization, the combination of the cytoplasm (mitochondria and/or chloroplast genome) of one parent and the nuclear genome of the second parent without otherwise altering the nuclear genome and the desirable phenotypes of that parent, is a useful somatic hybridization approach for citrus improvement. These new combinations of nucleus and cytoplasm can reveal the cytoplasmic contribution to the phenotype and its potential for citrus cultivar improvement. Cybrids establish a precious model to understand the nuclear/cytoplasmic genome interactions at the molecular level unlike resistant transgenic plants that need a long time for evaluation and regulation. Additionally, there is a possibility that these plants



never become marketable. Plants produced using the cybridization technique are not regulated as GMOs. We developed putative cybrids by fusing protoplast of citrus canker highly resistant 'Meiwa' kumquat and protoplast of highly susceptible grapefruit 'Marsh', 'Flame', and 'N11-11' somaclone of 'Ruby Red'. Plants recovered from all three combinations displayed the typical grapefruit phenotype and validated somatic cybrids using mitochondrial, chloroplast, and EST-SSR molecular markers. Therefore, these novel cybrids will allow the study of the inheritance of resistance to Xcc in mitochondrial and chloroplast transfer. Cybrid clones showed a range of citrus canker resistance, but all grapefruit cybrids with

a kumquat chloroplast had a significantly lower number of lesions and Xcc population compared to grapefruit controls. However, cybrid clones with grapefruit chloroplast had a substantially higher number of lesions compared to clones with kumquat chloroplast. These cybrids have the potential to provide a high level of citrus canker resistance in commercial grapefruit orchards and act as a valuable model for understanding the contribution of chloroplast to plant disease resistance. Grapefruit with kumquat cytoplasm also has the potential for improved tolerance to HLB. Thus, this approach opens a new avenue for studying the role of organelles in plant disease resistance.

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