

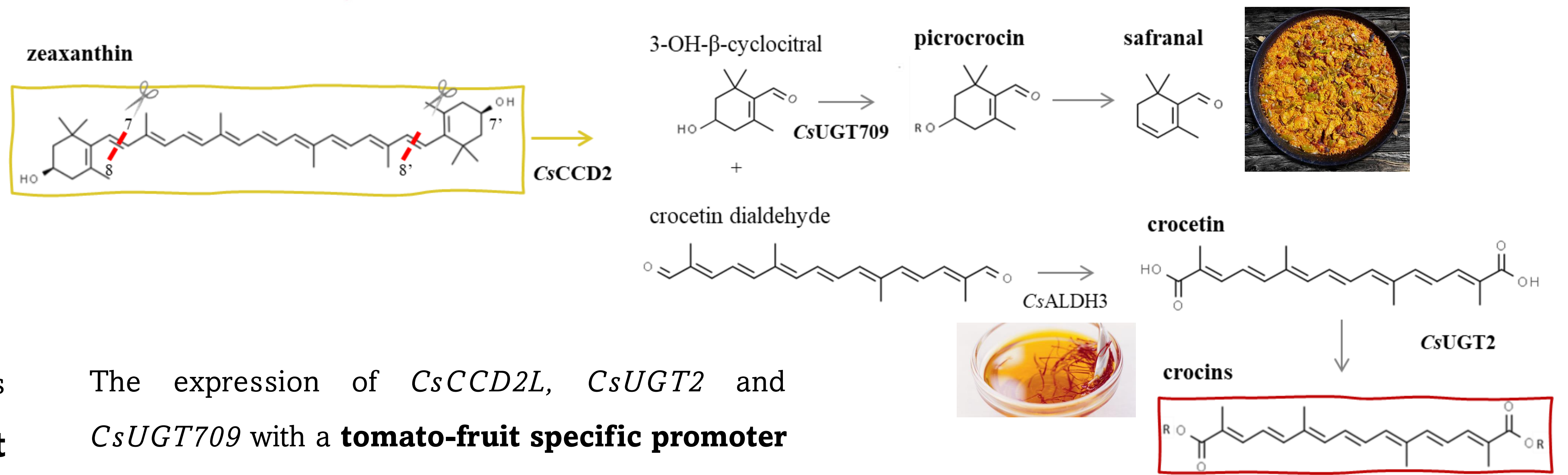
A biotechnological tomato platform to produce high levels of saffron apocarotenoids

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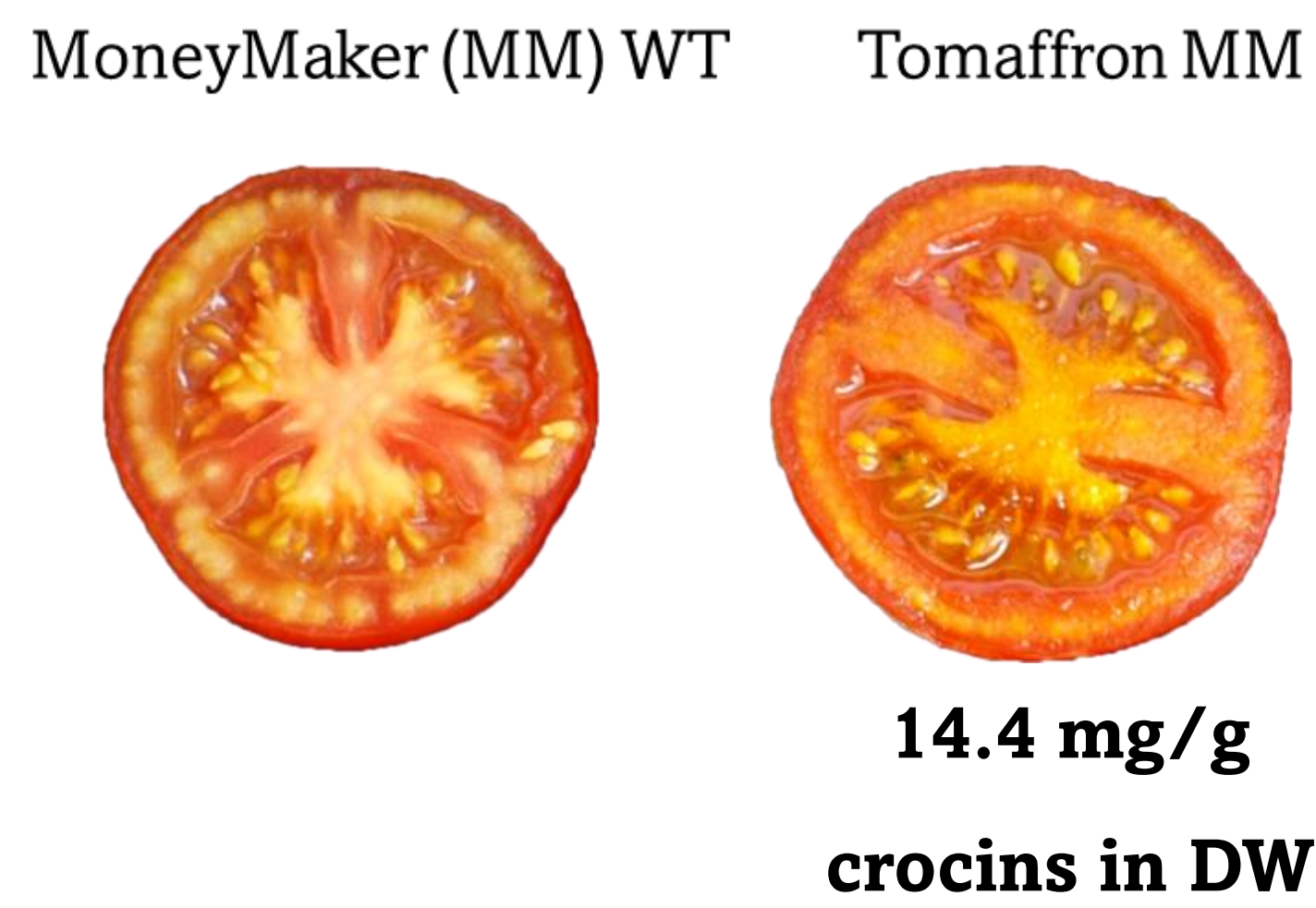
8.000-10.000 \$/kg



Saffron apocarotenoids are the responsible for the **organoleptic** and **medical** properties of the spice



The expression of *CsCCD2L*, *CsUGT2* and *CsUGT709* with a **tomato-fruit specific promoter** (E8) resulted in **TOMAFFRON**



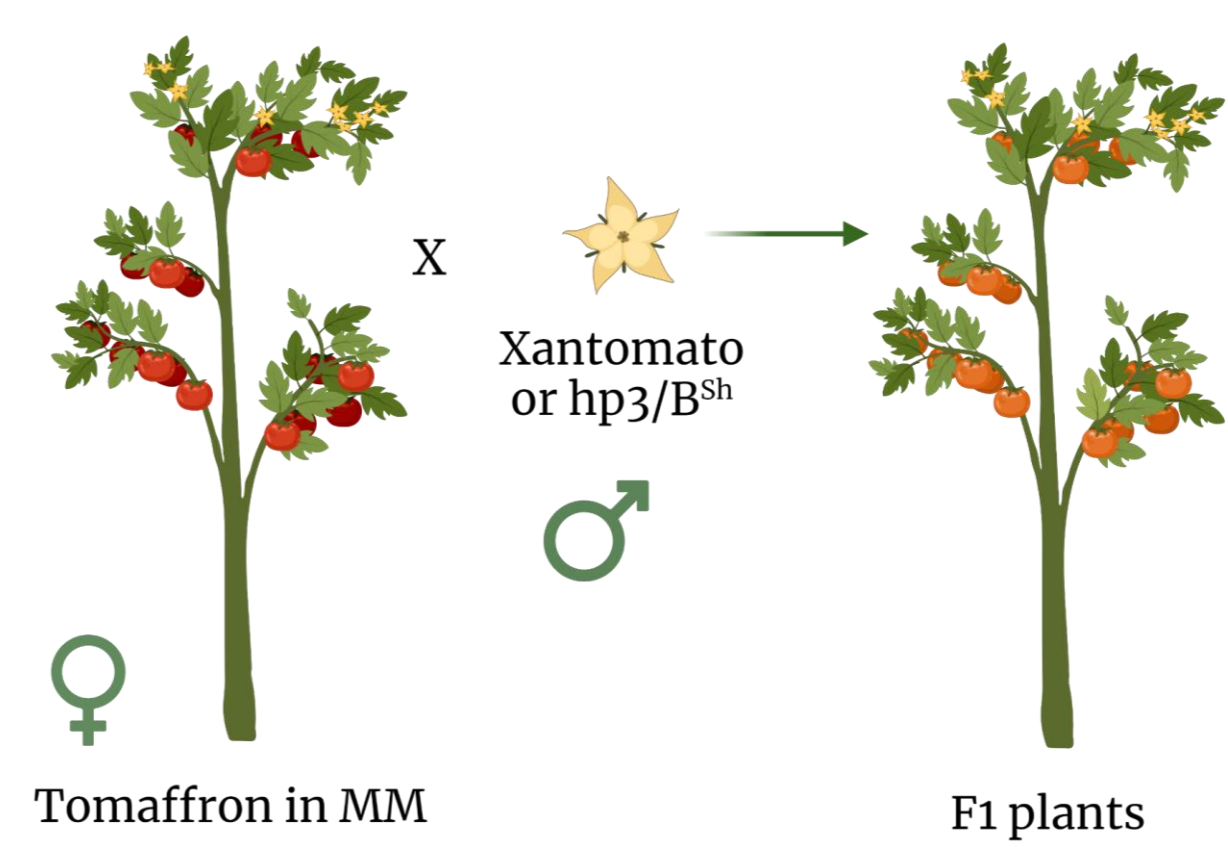
Tomato fruit is the heterologous platform accumulating **the highest levels of crocins**, despite the traces amount of the direct precursor, zeaxanthin

Introducing the saffron cassette in Xantomato

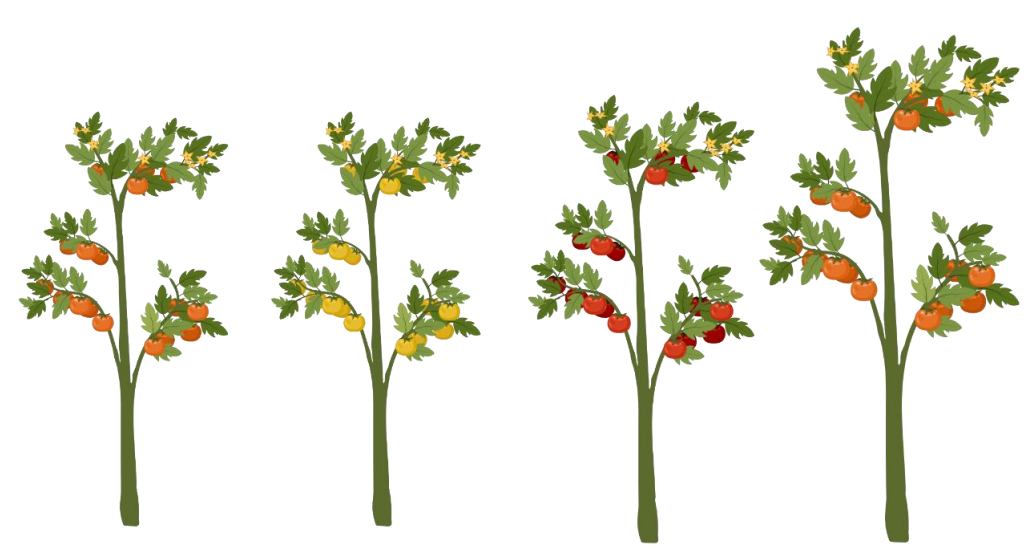
HOW DO WE IMPROVE IT?

Xantomato carries four mutations (*hp3*, *hp2^{dg}*, *B^{Sh}*, and green stripe) that result in fruits with high levels of zeaxanthin

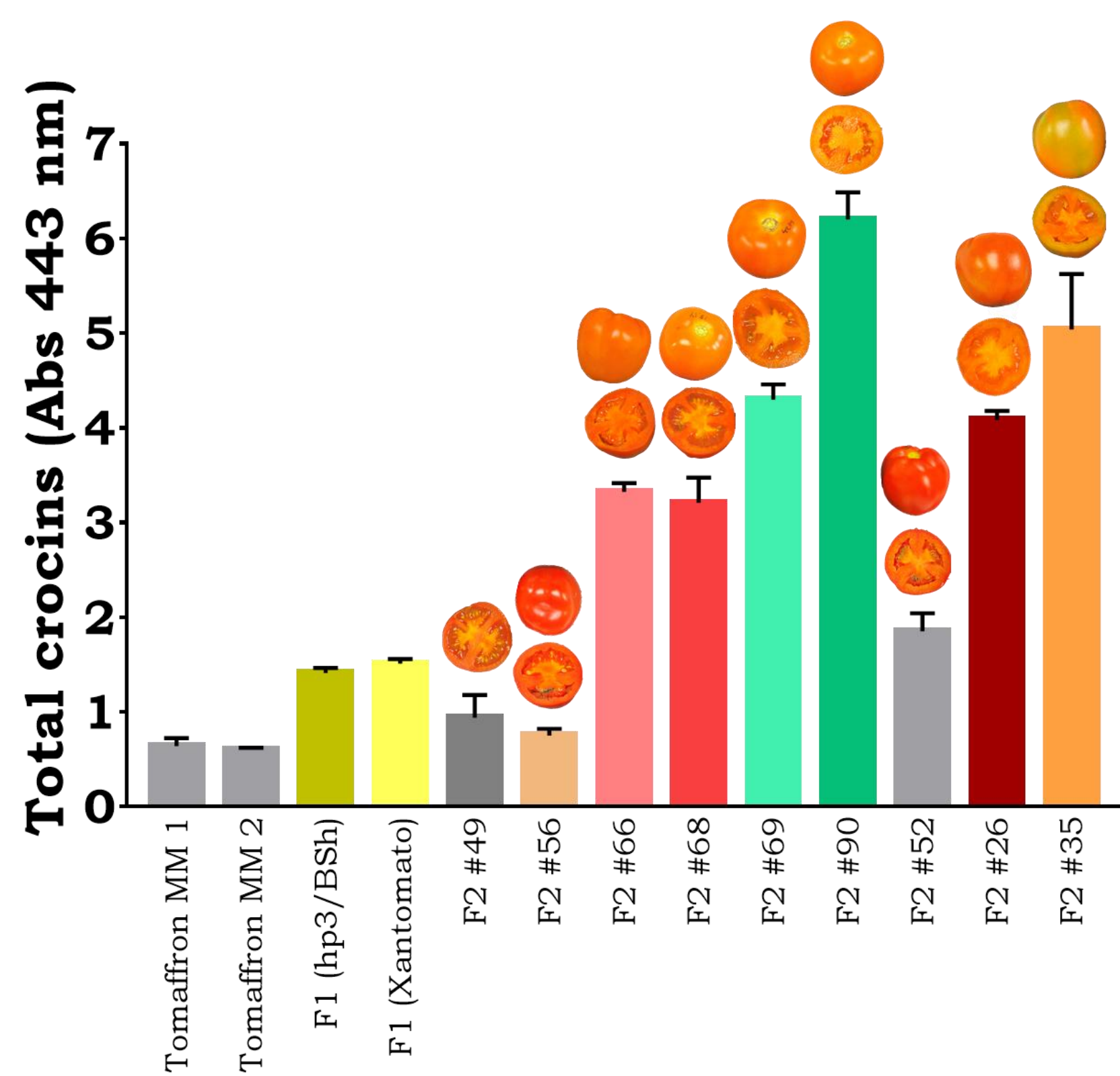
Conventional breeding approach



The F1 fruits from the crosses with Xantomato and the intermediate mutant *hp3/B^{Sh}* accumulated **2 times more crocins** than transgenic MM

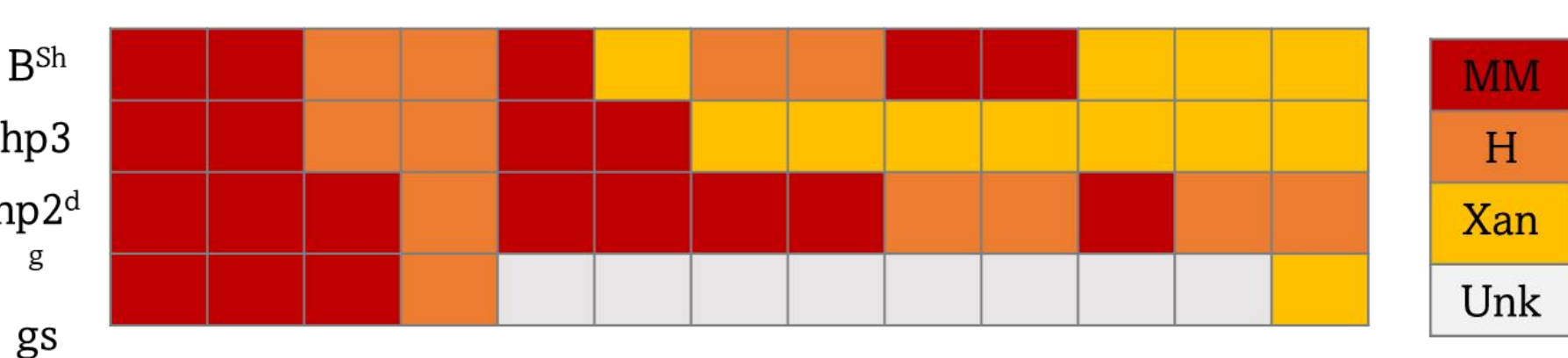


Population of F2 plants carrying different combination of the mutations

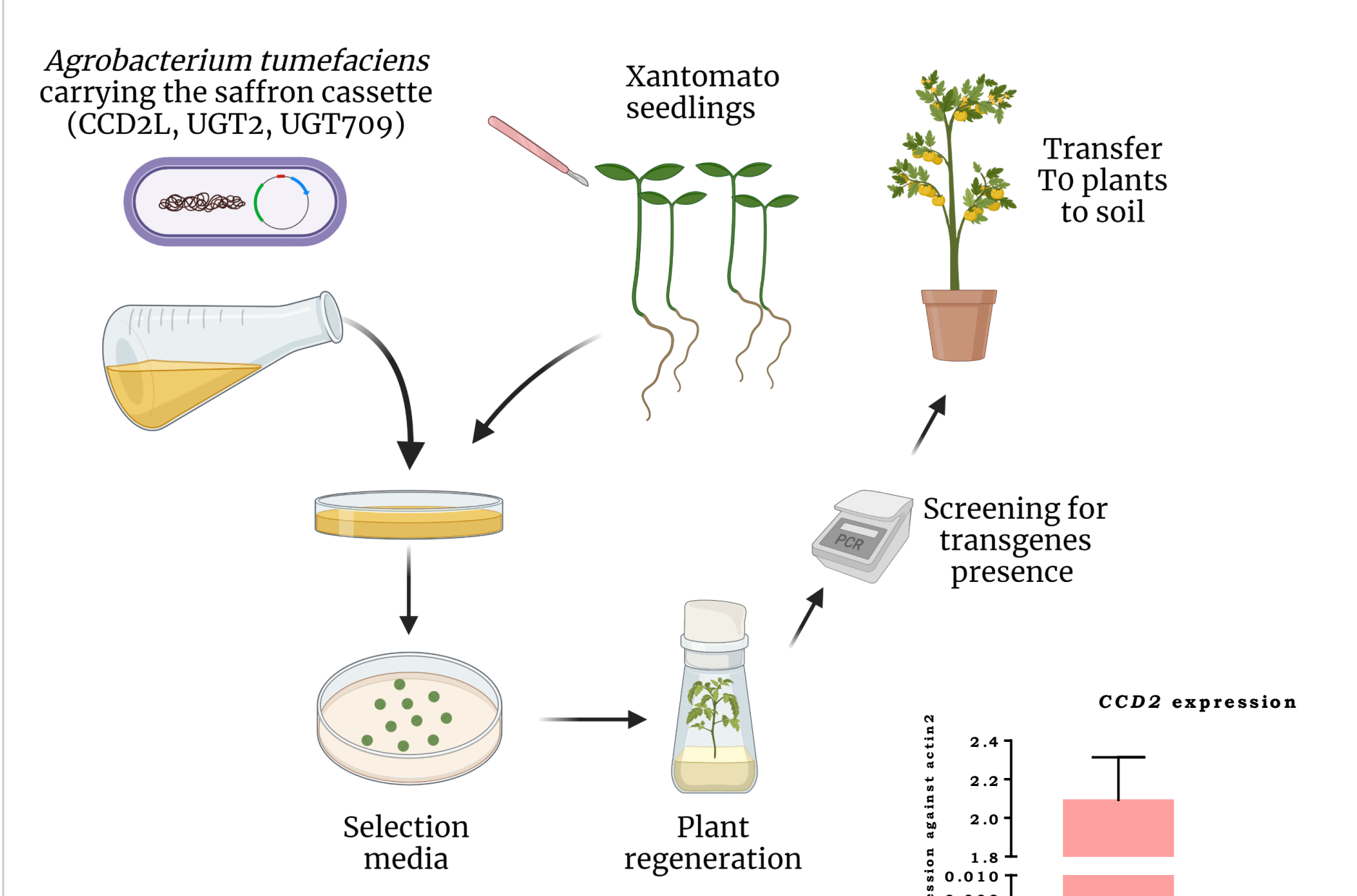


Fruits from **F2 #90** accumulated **10 times more crocins** than tomaffron MM, followed by **F2 #35** fruits accumulating **8 times more crocins**

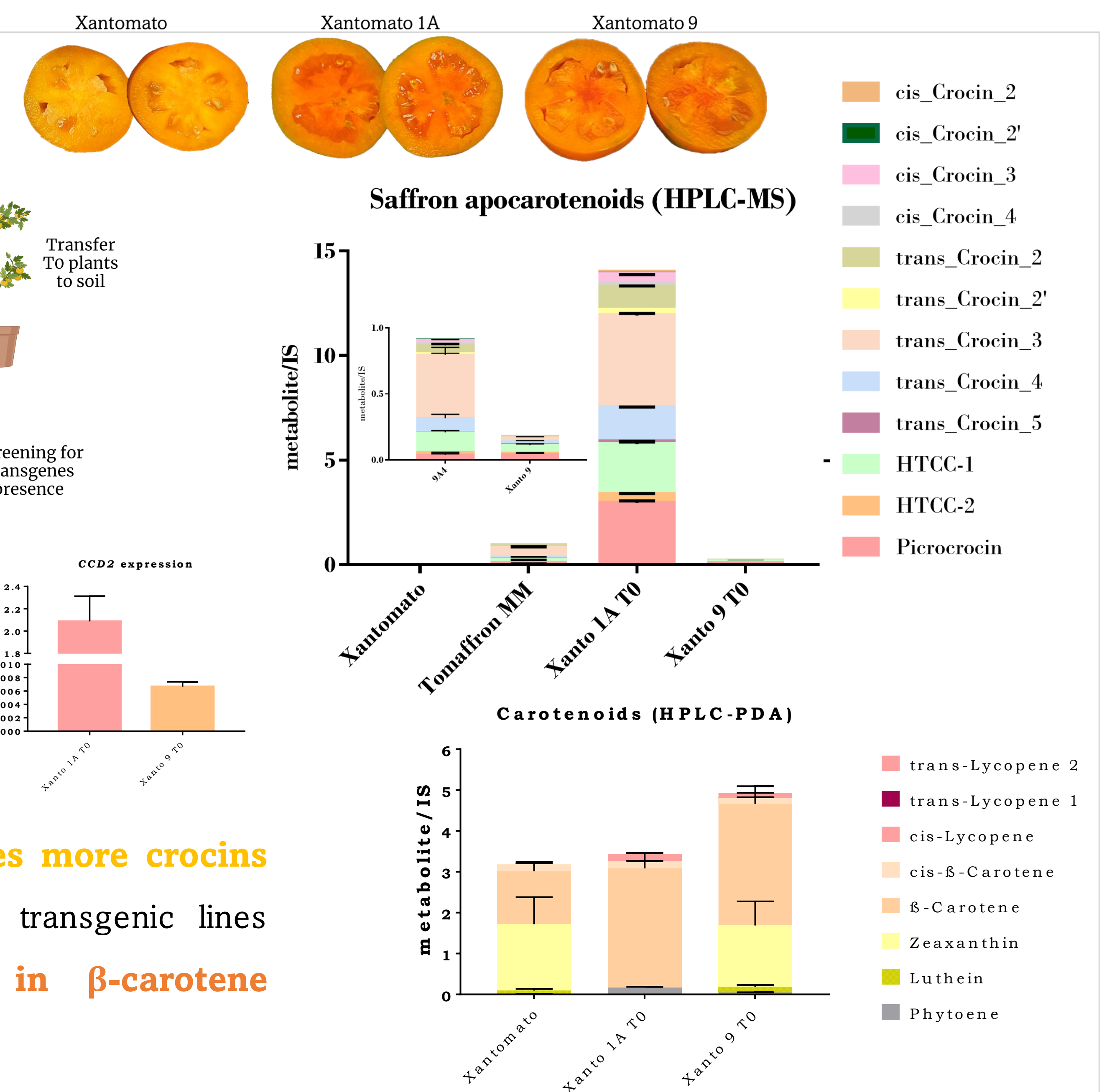
Tomaffron MM F2 #90



Biotechnological approach

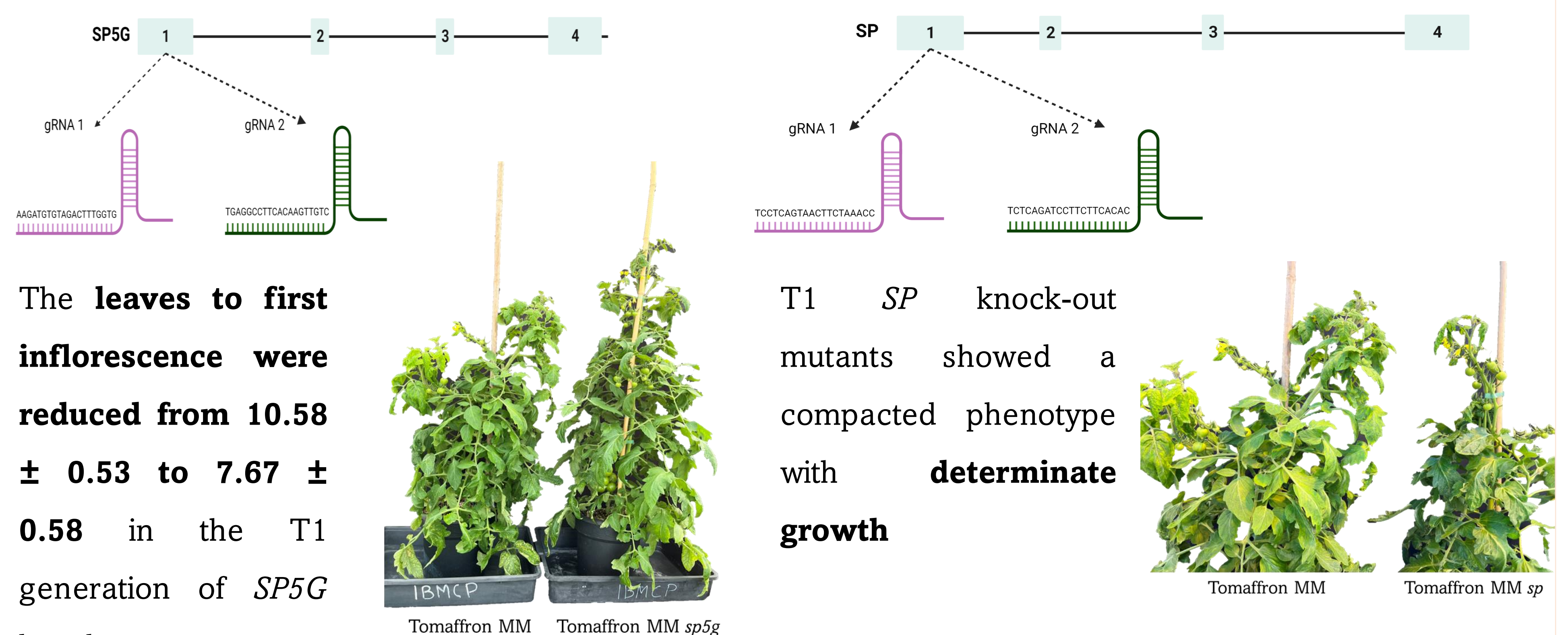


Xanto #1A accumulates **12 times more crocins** than transgenic MM, and both transgenic lines showed a **2-fold increase in β -carotene** compared with Xantomato



Modifying the chassis of the plant

Targeting *SELF-PRUNING* (*SP*) and *SP5G* genes through CRISPR/Cas9 in **tomaffron MM** to accelerate the flowering and concentrate in time the harvesting of the fruits



The **leaves to first inflorescence** were **reduced from 10.58 ± 0.53 to 7.67 ± 0.58** in the T1 generation of *SP5G* knock-out mutants

T1 *SP* knock-out mutants showed a compacted phenotype with **determinate growth**

Crosses between *SP5G* and *SP* T1 mutants were performed