

# Generation of cisgenic apple (*Malus x domestica* BORKH.) with a biotic resistance to apple scab caused by *Venturia inaequalis*

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The cultivated apple (*Malus x domestica* Borkh.) is one of the economically most important fruit crop worldwide. Among hundreds of existing apple cultivars only a handful of them are favoured by consumers due to their appearance, quality, flavour and storability. These cultivars are all susceptible to different plant diseases like apple scab caused by *Venturia inaequalis*. The introduction of the natural occurring scab resistance gene *HcrVf2* from *Malus floribunda 821* by classical breeding is time consuming and expensive, because of self-incompatibility, heterozygosity and a long juvenile period of 5 to 12 years. Genetic engineering offers the opportunity to overcome these limitations. The introduction of *HcrVf2* into the genome of existing cultivars via *Agrobacterium*-mediated plant transformation requires an efficient selection as realized by the *neomycin phosphotransferase II* marker gene in apple. Such marker genes conferring resistances to antibiotics are not accepted by the con-

sumer and need to be eliminated. A vector was developed allowing the site-specific excision of all unwanted DNA sequences after selection, mediated by a heat-shock inducible expression of the FLP recombinase. The vector contains beside *HcrVf2* under its own regulatory elements a recombination cassette flanked by direct repeated *flp* recognition target sites. The recombination cassette comprises of two marker genes *nptII* and *dao1*, both driven by a *CaMV 35S* promoter, as well as the *flp* recombinase gene under control of the heat-inducible *Gmhsp17.5-E* promoter. The second marker gene *dao1* codes for the DAAO protein, which converts the non-toxic amino acids D-valin and D-isoleucin to plant toxic products. A further selection of gene modified cells in which the FRT-flanked box was removed by recombination is possible. Using this vector gene modified lines were produced and investigated by PCR, RT-PCR. First results will be presented.