Structure and functional analysis of cT-DNAs in the genus Nicotiana

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The genus Nicotiana contains at least six different species (Nicotiana tabacum, tomentosiformis, tomentosa, otophora, kawakamii and glauca) with cellular T-DNAs. These DNAs are believed to derive from natural infection of Nicotiana ancestors by Agrobacterium rhizogenes strains, followed by spontaneous regeneration of hairy roots. The paternal tobacco ancestor Nicotiana tomentosiformis contains 4 different cT-DNAs (TA to TD), apparently acquired by four successive transformations (1). An important question is whether these cT-DNAs are expressed and could modify plant growth in some way. The TB region contains a mas2' gene which encodes the synthesis of the Amadori-type opine desoxyfructosyl-glutamine (DFG) in hairy roots. We have shown that the TB-mas2' gene is highly expressed in root tips of some tobacco cultivars and leads to detectable amounts of DFG (2). A unique cT-DNA was found in Nicotiana otophora, and called TE. I will report on the mapping of the TE region and the study of one of its potential ORFs, coding for a 6b-like gene. 6b genes have so far only been found in A. tumefaciens and A. vitis and belong to the plast gene family. The T-6b and AB-6b genes from A. vitis cause large changes in tobacco development probably due to uptake and retention of sucrose (3). We have expressed the TE-6b gene from N. otophora in tobacco under 35S promoter control and obtained an unusual phenotype that is not typical for 6b genes. I will describe this phenotype in more detail and discuss how it might be related to the classical 6b phenotype and to what extent it may play a role in the growth and development of *N. otophora*.

- (1) Chen K, Dorlhac de Borne F, Szegedi E, Otten L (2014) Deep sequencing of the ancestral tobacco species *Nicotiana tomentosiformis* reveals multiple T-D NA inserts and a complex evolutionary history of natural transformation in the genus *Nicotiana*. Plant J. 80:669-682.
- (2) Chen K, Dorlhac de Borne F, Julio E, Obszynski J, Pale P, Otten L (2016) Root-specific expression of opine genes and opine accumulation in some cultivars of the naturally occurring GMO *Nicotiana tabacum*. Plant J. doi: 10.1111/tpj.13196. [Epub ahead of print].
- (3) Chen K, Otten L (2016) Morphological analysis of the *6b* oncogene-induced enation syndrome. Planta 243:131-148.

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