

A gene for the major cytoplasmic tRNA^{Tyr} from *Nicotiana rustica* contains a 13 nucleotides long intron

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Tobacco leaf nuclear DNA was isolated from pure nuclei via protoplasts (1). A λ library prepared by Eco RI-digestion, ligation of the fragments into a λ vector and in vitro packaging (2), was screened with a 5'-³²P-labeled synthetic oligodeoxynucleotide complementary to nts. 18-37 of tRNA^{Tyr} from tobacco leaves (3). A hybridizing Eco RI-fragment of about 3 kbp from a recombinant λ phage, subcloned in pUC19, yielded the recombinant plasmid pNt1. The sequence of the tRNA^{Tyr} gene and of its flanking regions are shown below (non-coding strand; gene boxed; intron underlined):

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TTCTCAAAT CTTTATTCTT -60 AACTAATA CGAATGTGA -40 CGACTTCTAT ATAGGTATGA -20 GATTAGAGTA
      1          21          41
TCTCGCAAGA CCGACCTTAG CTCAGTTGGT AGAGCGGAGG ACTGTAGTGG TACTGCTGAG ATCCTTAGGT
61          81          101          121
CACTGGTTCG AATCCGGTAG GTCGGATTTTT GTTTTTCTGT GTTCTTTGCT TTTTCATTTC ATCATCAAAA
    
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This is the first nuclear plant tRNA gene for which a corresponding tRNA is known: its sequence is colinear with one of the two major cytoplasmic tRNAs^{Tyr} of *Nicotiana* (3). Special features of this gene are a 13 nucleotides long intron and a (T)₄G(T)₅ transcription termination signal immediately following the tDNA. This gene is efficiently transcribed and correctly spliced in a HeLa cell nuclear extract (van Tol et al., in preparation).

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- (2) Maniatis, T., Fritsch, E. F. and Sambrook, J. *Molecular Cloning. A Laboratory Manual.* Cold Spring Harbor Laboratory, 1982.
- (3) Beier, H., Barciszewska, M., Krupp, G., Mitnacht, R. and Gross, H. J. (1984) *EMBO J.* **3**, 351-356.