

Supplementary data

With the primers noted in Table A and 1ng of a cloned putative *GRAF*-promoter as the template, it was possible to generate different fragments of the 5'-flanking region of *GRAF*. After purification (QIAquick Spin Kits from Qiagen, Hilden, Germany) and restriction (*Hind*III and *Bgl*II, Boehringer Mannheim, Germany) using conventional methods the PCR-products were cloned into the *Hind*III/*Bgl*II digested pGL3-BasicVector (Promega, Mannheim, Germany).

Table A

Primer sequences for the generation of promoter fragments

Artificial restriction sites: Sense primers: AGATCT, *Bgl*II, #

Antisense primers: AAGCTT, *Hind*III, §

Name	First base	Nucleotide sequence (5'→3')	Last base	Orientation
<i>Prom up 1482</i>	-1482	§+#CACGTAGCTGAAGGAGGGCTGCTGGTTCC	-1454	Sense
<i>Prom up 981</i>	-981	§+#ACCGTGGGCGGGTTCTGAGCAGGAGTGGCC	-952	Sense
<i>Prom up 576</i>	-576	#GAGTTTGCCTGCGGACCTAGGCGTTGG	-560	Sense
<i>Prom up 507</i>	-507	#CTTTGATTGGATCAAGACCGCGGAAGGGCC	-478	Sense
<i>Prom up 242</i>	-242	#GTTTCCTGCTCGCGATCCGCTCCGTTGCC	-214	Sense
<i>Prom do 341</i>	-341	§TCGAGCGCCCTCTCACAGCAGCCTCGGCC	-369	Antisense
<i>Prom do 176</i>	-176	§GAGCTCGGACCCGGCCGAGGTGCTGCTGTTCC	-206	Antisense
<i>Prom do 158</i>	-158	§CGCAAGACTCCC GAAGCGAGCTCGGACC	-185	Antisense
<i>Prom do 108</i>	-108	§GGCTCCACAGGTGTGGCGCTGGCTCACTCC	-137	Antisense
<i>Prom do 75</i>	-75	§GCGGGACCCCGCCGGCTCCCCGACGGCC	-104	Antisense
<i>Prom do 52</i>	-52	§CCGCCGCCAGAGCACTCACGCGCGGGAC	-81	Antisense
<i>Prom do</i>	-1	§+#GTGCGCGCGGCGCCATCCGGGGGCGCGCC	-31	Antisense

Table B

Mutagenesis-primer for promoter constructs with deletion of the footprint regions

Name	First base	Nucleotide sequence (5'→3')	Last base	Orientation
<i>Prom s Fp1</i>	-483	GGGCCGGGCGTCCTCGGGCG	-464	Sense
<i>Prom a Fp1</i>	-494	TTGATCCAATCAAAGGGGCC	-513	Antisense
<i>Prom s Fp2</i>	-523	CCTGCCTCCAGGCCCTTTG	-504	Sense
<i>Prom a Fp2</i>	-554	CGCCTAGGTCCGCAGCGCAAAC	-575	Antisense

Table C

Buffers used in the Preparation of nuclear extracts and DNase I protection assays

Buffer name	Composition
Hypotonic buffer	10mM Hepes; 0.75mM spermidine; 0.15mM spermine; 0.1mM EDTA; 0.1mM EGTA; 1mM DTT; 10mM potassium chloride; 10mM benzamidine; 0.5mM PMSF
Restore buffer	68% sucrose; 50mM Hepes; 0.75mM spermidine; 0.15mM spermine; 10mM potassium chloride; 0.2mM EDTA; 1mM DTT
Resuspension buffer	20mM Hepes; 0.75mM spermidine; 0.15mM spermine; 0.2mM EDTA; 0.2mM EGTA; 2mM DTT; 25% glycerol; 10mM benzamidine; 0.5mM PMSF; mix 9 volumes of this buffer with 1 volume ammonium sulfate solution (4M)
Buffer D	20mM Hepes; 20% glycerol; 0.1M potassium chloride; 0.2mM EDTA; 0.5mM PMSF; 0.5mM DTT
Binding buffer	10mM Tris-HCl; 1mM EDTA; 0.1mM EGTA; 1mM DTT; 60mM potassium chloride; 5% glycerol
Stop buffer	0.5M sodium acetate; 30mM EDTA; 1%SDS; 0.2µg/ml glycogen

Figure legends

Supplementary Figure 1 Deletion analyses (**B**) of the *GRAF* promoter.

Left panel: The promoter segments (numbering relative to A (=+1) in the START-ATG codon). Right panel: Relative Firefly luciferase activity normalized against the promoting activity of SV40 (=100%). The data represent the mean \pm SD of three independent experiments.