

Supplementary data 1: Genotyping

DNA was isolated from EDTA blood samples. Genotyping of the Ex1f-VNTR was carried out at the department of Human Genetics of the Radboud University Nijmegen Medical Centre. PCR was performed with 1 µl 10x PCR buffer II (Applied Biosystems, Nieuwerkerk a/d IJssel, The Netherlands), 1 µl of 25 mM MgCl₂, 1 µl dNTPs (2.5 mM of each dNTP; GE Healthcare, Zeist, The Netherlands), 0.50 µl forward and reverse primer together (10 pmol/µl of each primer, VIC[®]-labeled 5'-CCCTGCGTGGCTACTACTACATT-3' and 5'-CTGGGCTCCAAAGCATAACAT-3' with 'PIG'-tail; Applied Biosystems) and 0.08 µl AmpliTaq Gold polymerase (Applied Biosystems). The cycling conditions were: 12 min 95 °C, 10 amplification cycles of 30 sec at 94°C, 54°C for 30 sec and 72°C for 1 min, followed by 22 cycles of 30 sec at 89°C, 54°C for 30 sec and 1 min at 72°C, followed by a final 10 min at 72°C. After the PCR, fragment length analysis was performed on the ABI prism 3730 Genetic Analyser (Applied Biosystems) and results were analyzed with GeneMapper[®] Software, version 4.0 (Applied Biosystems). After testing for Hardy-Weinberg equilibrium^{1, 2}, the resulting genotypes were converted to short (S) and long (L) alleles (see table below). The frequency of the dichotomous *NOS1* ex1f-VNTR genotype in Caucasian populations is as follows: SS=21.0%; SL=51.4%; LL=27.6% (n=7847 population-based subjects from Germany, Italy, Norway, Sweden, Estonia, Spain, The Netherlands and Austria) (Prof. Dr. A. Reif, personal communication).

NOS1 Ex1f-VNTR^a

Short alleles			Long alleles		
<i>Allele size</i>	<i>Reif</i>	<i>freq</i>	<i>Allele size</i>	<i>Reif</i>	<i>freq</i>
138	-	1	178	198	2
161	182	36	180	200	40
163	184	9	182	202	31
169	188	1	184	204	82
172	192	57	186	206	5
174	194	7			
176	196	1			

^aDisplayed are the size of the alleles found in the current study sample (*allele size*), the corresponding allele size from the study of Reif et al. 2009 (*Reif*) and frequency of these alleles in the current study sample (*freq*).

References

1. Raymond, M. & Rousset, F. GENEPOP (version 1.2): population genetics software for exact tests and ecumenicism. *Journal of heredity* **86**, 248-249 (1995).
2. Rousset, F. GENEPOP '007: a complete re-implementation of the GENEPOP software for Windows and Linux. *Molecular Ecology Resources* **8**, 103-106 (2008).