

Evidence for the effect of serotonergic and dopaminergic gene variants on stress levels in horses participating in dressage and harness racing

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Table S1. Description of BDNF, COMT, HTR1A, SLC6A4 and TPH2 polymorphisms analyzed and allelic frequencies of the SNP under study comparing different stress levels for Pura Raza Español (PRE) and Spanish Trotter Horse (STH) populations.

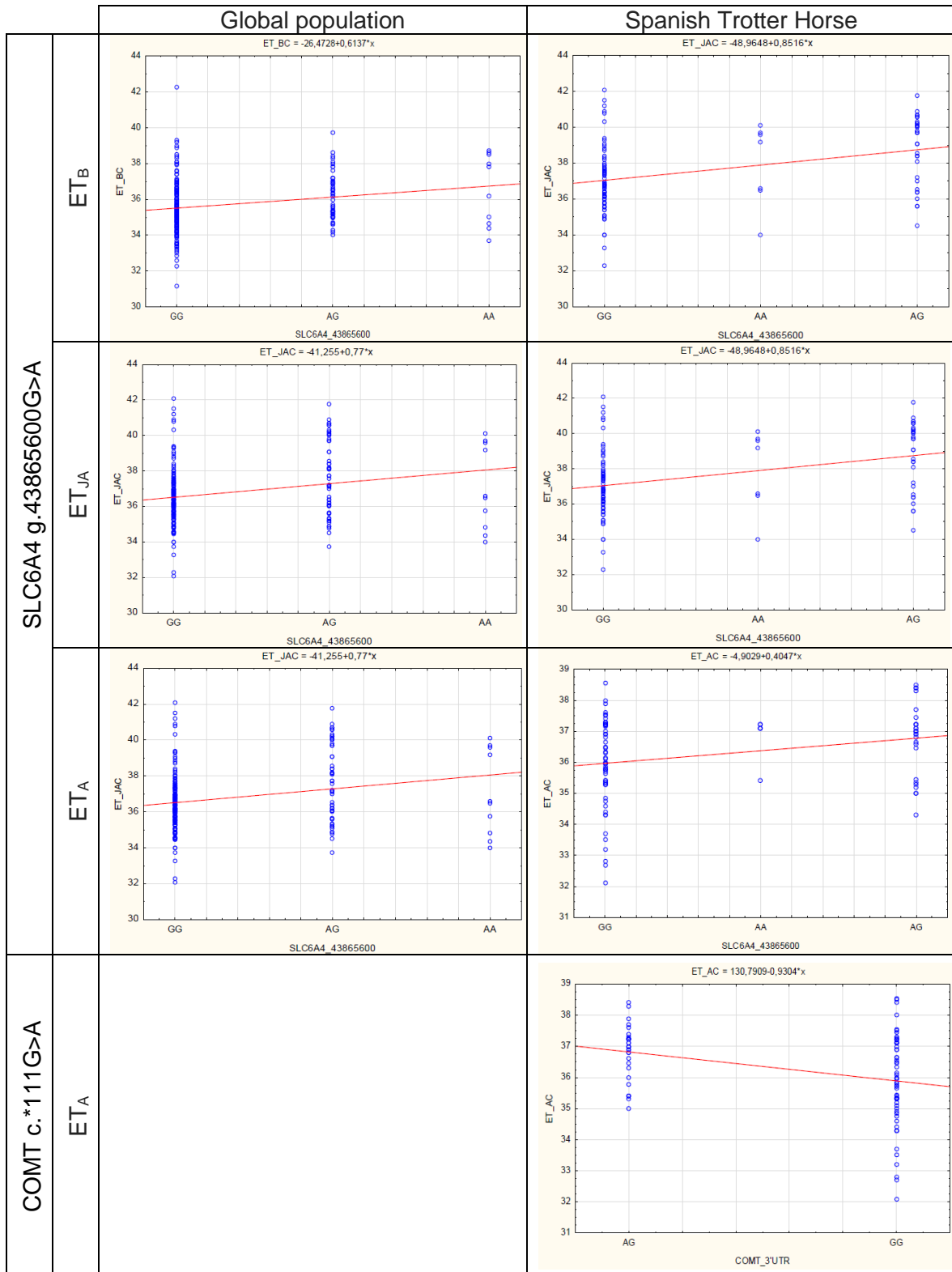
Gene (Target variant)	Variant Name	Polym. (DNA)	Polym. (cDNA)	Position (Chr.)	Reference		PRE			STH		
							Low stressed (32.1-36.0°C)	High stressed (36.0-39.4°C)	Total	Low stressed (32.2-37.7°C)	High stressed (37.8-42.1°C)	Total
COMT-3'UTR (A)	COMT ^{3'UTR}	g.989906 G>A	c.*111G>A	Exon 4 (ECA8)	(Dall'Olio <i>et al.</i> 2009)	G	0.51	0.54	0.53	0.87	0.77	0.82
						A	0.49	0.46	0.47	0.13	0.23	0.18
BDNF (A)	BDNF ^{A87A}	g.94041447 G>A	c.261G>A	Exon 1 (ECA7)	-	G	0.64	0.61	0.62	0.78	0.84	0.81
						A	0.36	0.39	0.38	0.22	0.16	0.19
HTR1A (C)	HTR1A ^{E257D}	g.9478637 G>C	c.771G>C	Exon 1 (ECA21)	Hori <i>et al.</i> (2016)	G	0.88	0.94	0.91	0.88	0.87	0.88
						C	0.12	0.06	0.09	0.12	0.13	0.12
TPH2 (G)	TPH2 ^{H447R}	g.671744 A>G	c.1340A>G	Exon 11 (ECA28)	-	A	0.91	0.94	0.92	0.92	0.93	0.92
						G	0.09	0.06	0.08	0.08	0.07	0.08
SLC6A4 (A)	rs68968521	g.43865600 G>A	-	Intron 10 (ECA11)	-	G	0.86	0.90	0.84	0.88	0.70	0.83
						A	0.14	0.10	0.16	0.12	0.30	0.17
SLC6A4 (T)	g.727C>T*	g.43865368 C>T	-	Intron 10 (ECA11)	-	C	0.79	0.75	0.77	0.71	0.77	0.74
						T	0.21	0.25	0.23	0.29	0.23	0.26
SLC6A4 (T)	g.1708C>T*	g.43864387 C>T	-	Intron 10 (ECA11)	-	C	0.93	0.95	0.94	0.91	0.97	0.94
						T	0.07	0.05	0.06	0.09	0.03	0.06
SLC6A4 (C)	rs68968520	g.43864288 A>C	-	Intron 10 (ECA11)	-	A	0.62	0.60	0.61	0.36	0.38	0.37
						C	0.38	0.40	0.39	0.64	0.62	0.63
SLC6A4 (AC)	SLC6A4 ^{T539A-I}	g.AG43864 196_7TC	c.AC1615 -1616GT	Exon 11 (ECA11)	Momozawa <i>et al.</i> (2006)	GT	0.24	0.27	0.25	0.37	0.25	0.31
						AC	0.76	0.73	0.75	0.63	0.75	0.69
SLC6A4 (A)	rs396986545	g.43864091 C>A	-	Intron 11 (ECA11)	-	C	0.72	0.74	0.76	0.97	0.98	0.98
						A	0.28	0.26	0.24	0.03	0.02	0.02

The Low and High stressed groups were made according to ET_{JA} (eye temperature measured just after the competition) values. * Two new SNP variants described for the first time in *Equus caballus* (Genbank ref: MF043946-MF043951)

Table S2. Description of the PCR primers at BDNF, COMT, HTR1A, SLC6A4 and TPH2 polymorphisms analyzed in the study indicated.

Gene	Genbank accession	PCR primers (5' – 3')
COMT (ECA8)	AB17828	F: TGACAGTCTGGATGTGGGACTAG R: TGTGAGAAAGGGATGCTCATGG
TPH2 (ECA28)	AB264323	F: CGAAGACCACGTGCTTGCCAG R: CAGATGCAGTTTGGTTAGGGA
HTR1A (ECA21)	AB264325	F: CGACTACGTGAACAAGAGGACACC R: CGCGGGCTACTCCTTTATCATC
BDNF (ECA7)	AB264324	F: GCACTTAGAACAGTACCTGGCACACAGC R: CTGACAATGCTTTCTGGTCCTCTGGG
SLC6A4 (ECA11)	NC_009154	F: ATCCAGAGGCCCTTTTGAGTTT R: AACAGGAACATTCTCGCCTCTT F: ATTCTCTGTCGAGTGGGTTTAC R: CAGGGGAGCGATGTGATAAAGA

Figure S1. Scatter plot of the associated variants: g.43865600G>A of SLC6A4 gene and c.*111G>A of COMT gene in the global population and the Spanish Trotter Horse.



ET_B, ET_{JA} and ET_A= eye temperature measured approx. 2 hours before, just after and approx. 2 hours after the competition, respectively.

Figure S2. Interaction network between the serotonin transporter (5HTT) protein (SLC6A4 gene) and catechol O-methyltransferase isoform S-COMT protein (COMT gene) in *Equus caballus*. From string-db.org

