Arch. Tierz., Dummerstorf **50** (2007) 3, 322-323

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Assignment of the *TYK2* gene to equine chromosome 7q12-q13

(Brief report)

(Kartierung des *TYK2* Gens auf dem Pferdechromosom7q12-q13)

Background: Tyrosine kinase 2 (*TYK2*) is a member of the janus kinase gene family and encodes an 1187 amino acid protein. All four members of the janus kinase family *JAK1*, *JAK2*, *JAK3*, and *TYK2* associate with various cytokine receptors and mediate the signal transduction by tyrosine phosphorylation of downstream targets (YAMOAKA et al., 2004). Studies with *tyk2* deficient mice demonstrated impairment of interferon α/β signaling (KARAGHIOSOFF et al., 2003). Mutations in the murine *tyk2* gene are associated with increased susceptibility to infectious and autoimmune diseases (SHAW et al., 2003). The human *TYK2* gene consists of 25 exons spanning 30,003 bp on human chromosome 19p13.2 starting at 10,322,209 bp. The objective of this study was to determine the chromosomal location of *TYK2* in the horse by FISH and RH mapping.

Procedures:

BAC library screening/sequence analysis/chromosomal location: The equine BAC library CHORI-241 was screened as per standard protocols (http://bacpac.chori.org) with a heterologous ^{32}P-labelled insert of a human *TYK2* cDNA clone (IRALp962L0830) provided by the RZPD (http://www.rzpd.de/). An equine genomic BAC clone (CH241-352C1) with an insert of approximately 200 kb containing the *TYK2* gene was identified. BAC DNA was prepared from the BAC clone using the Qiagen plasmid midi kit (Qiagen, Hilden, Germany) and both BAC ends were sequenced. A BLASTN sequence comparison of the equine SP6 BAC end sequence (AccNo. AM113773) with the build 36.2 of the human genome sequence revealed a significant match (BLAST E-value 9.0e^{-51}) over 196 bp (identity = 87%) starting at 10,606,427 bp of HSA19p13.1, approximately 254 kb downstream of human *TYK2*, and matching with exon 11 and parts of the flanking introns of *SLC44A2 (solute carrier family 44, member 2)*. An internal BAC sequence for exon 18 of the human *TYK2* gene with a product size of 552 bp was amplified and verified the human *TYK2* gene.

Primer sequences/radiation hybrid (RH) mapping: Primers for PCR amplification of a 249 bp fragment were designed from the CH241-352C1 SP6 BAC end sequence using Primer3 software: F 5'-ATCACTGTAGGGGGCAGAGA-3' and R 5'-CTGGTGTC TCTGTGCAGGAA-3'. To confirm the cytogenetic assignment, the 5,000 rad TAMU equine radiation hybrid panel (CHOWDHARY et al., 2003) was used to map the equine *TYK2*.

Results: The equine genomic BAC clone CH241-352C1 containing the *TYK2* gene was located to ECA7q12-q13 by examination of 40 metaphase spreads (Fig.). The

sequence tagged site (STS) markers showed a retention frequency of 10.9% and the RH mapping revealed close linkage to *HTG33* (6.19 cR; LOD >3.0) which had been previously mapped on ECA7 (CHOWDHARY et al., 2003). The physical assignment of the equine *TYK2* and *SLC44A2* genes on ECA7q12-q13 is in latest agreement with equine-human comparative maps for ECA7 and HSA19 (PENEDO et al., 2005; PERROCHEAU et al., 2006) but does not agree with the previously published equine-human comparative map of the centromeric region of ECA7p, which showed conserved synteny to HSA11 (CHOWDHARY et al., 2003).

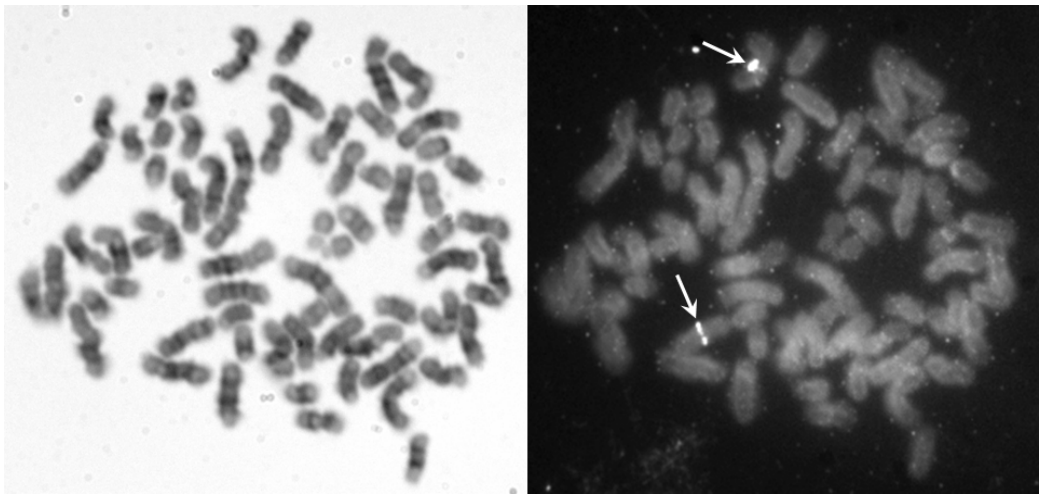

Fig.: Chromosomal assignment of the equine BAC containing *TYK2* by FISH analysis. G-banded metaphase spread before (left) and after (right) hybridization. Double signals indicated by arrows are visible on both equine chromosomes. (Chromosomale Zuordnung des Pferde BAC mit dem *TYK2* Gen mittels FISH. G-gebänderte Metaphase-Chromosomen vor (links) und nach (rechts) der Hybridisierung. Doppelsignale sind mit Pfeilen gezeichnet und auf beiden Chromosomen sichtbar).

References

CHOWDHARY, B.P.; RAUDSEPP, T.; KATA, S.R.; GOH, G.; MILLON, L.V.; ALLAN, V.; PIUMI, F.; GUERIN, G.; SWINBURNE, J.; BINNS, M.; LEAR, T.L.; MICKELSON, J.; MURRAY, J.; ANTCZAK, D.F.; WOMACK, J.E.; SKOW, L.C.:
The first-generation whole-genome radiation hybrid map in the horse identifies conserved segments in human and mouse genomes. Genome Research., **13** (2003), 742-751

KARAGHIOSOFF, M.; STEINBORN, R.; KOVARIK, P.; KRIEGSHAUSER, G.; BACCARINI, M.; DONABAUER, B.; REICHART, U.; KOLBE, T.; BOGDAN, C.; LEANDERSON, T.; LEVY, D.; DECKER, T.; MULLER, M.:
Central role for type 1 interferons and Tyk2 in lipopolysaccharide-induced endotoxin shock. Nature Immunology., **4** (2003), 471-477

PENEDO, M.C.T.; MILLON, L.V.; BERNOCO, D.; BAILEY, E.; BINNS, M.; CHOLEWINSKI, G.; ELLIS, N.; FLYNN, J.; GRALAK, B.; GUTHRIE, A.; HASEGAWA, T.; LINDGREN, G.; LYONS, L.A.; RØED, K.H.; SWINBURNE, J.E.; TOZAKI, T.:
International Equine Gene Mapping Workshop Report: a comprehensive linkage map constructed with data from new markers and by merging four mapping resources. Cytogenetic and Genome Research, **111** (2005), 5-15

SHAW, M. H.; BOYARTCHUK, V.; WONG, S.; KARAGHIOSOFF, M.; AGIMBEAU, J.; PELLEGRINI, S.; MULLER, M.; DIETRICH, W.F.; YAP, GS.:
A natural mutation in the Tyk2 pseudokinase domain underlies altered susceptibility of B10.Q/J mice to infection and autoimmunity. Proceedings of National Academy of Science USA, **100** (2003), 11594-11599

YAMAOKA, K.; SAHARINEN, P.; PESU, M.; HOLT, V.E. 3rd; SILVENNOINEN, O.; O'SHEA, J.J.:
The Janus kinases (Jaks). Genome Biology, **5** (2004), 253

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