

SuppFigure legend

SuppFigure 1. Structure of MO-TES391 and KIAA1864 transcripts.

A. Schematic drawing of the reverse strand of BAC clone AC027281 (Chr.16) and structure of MO-TES391 exons 1 to 12 (introns are shown in grey).

In silico alignment with BAC clones containing one of the two copies of the *hydin* gene located on chromosomes 1q21.1 and 16q22.2 (AC092369 and AC027281, respectively) revealed that 54 bp of the central part of the MO-TES391 sequence were missing in AC092369, but present in AC027281, strongly suggesting that MO-TES391 is encoded by the *hydin* gene located on chr. 16.

B. Comparison of MO-TES391 (exons 1-12) with HYDIN-012 (exons 1-20) and KIAA1864 (exons 1-20). Identical regions are connected with dotted lines and marked in grey. I-III indicate the fragments subcloned from MO-TES391 in Supp. Fig. 2. CDS= coding sequence (putative); nucleotide N° are indicated.

In silico analysis: MO-TES391 sequence (1893 bp) shows strong homology with 5 human *hydin* transcripts registered at the Ensembl and NCBI databases. Three of those, NM_001270974.1, HYDIN-001 and HYDIN-201 are putative protein coding sequences comprising nearly all described 86 *hydin* exons, and include MO-TES391 sequence bp 1-1660. Two 3' exons (exons 11-12) are however unique to MO-TES391, suggesting alternative splicing. Additionally, 352 bp of the central part of MO-TES391 (exons 6 and 7, bp 820-1173) are lacking in the two further transcripts HYDIN-011 and -012. AB058767 (mRNA for KIAA1864 hypothetical protein, 4599bp) and AK027571 (mRNA for FLJ14665 hypothetical protein, 1876bp) share long overlapping sequences with MO-TES391 (1311 and 1006 bp respectively) with >99.9% of identity, but both lack exons 6 and 7 as well.

SuppFigure 2. Mapping of major immunogenic region of MO-TES391.

A. Testing of serological reactivity against MO-TES391 fragments in SMARTA2. MO-TES391 fragments (I-III) were amplified by PCR using plasmid pBK-CMV/MO-TES391 as template and oligonucleotides listed in Supp. Table 2. PCR amplicons were digested with MfeI and XhoI and cloned into pET-17(b) using EcoRI/XhoI cloning sites. Negative control BSA (1 μ g); Background control lysate of host cells (100 ng of total protein); Positive control 1 rabbit-anti-human IgG (10 ng); Positive control 2 total human IgG (10 ng); Position of fragments I-III is shown in Supp. Fig. 1; Either sera from CRC patients (CC1-7) or rabbit polyclonal antibodies raised against the full-length recombinant MO-TES391 (right control array) were used.

B. Frequency of serological reactions against the three MO-TES391 fragments in selected CRC patients with anti-MO-TES391 reactivity (n=30).