KLK8 (kallikrein-related peptidase 8)

Yves Courty

Centre d’Etude des Pathologies Respiratoires, INSERM U1100 - EA6305, Faculte de Medecine, 10 bvd Tonnelle, 37032 Tours cedex, France (YC)

Published in Atlas Database: July 2012
Online updated version: http://AtlasGeneticsOncology.org/Genes/KLK8ID41088ch19q13.html
DOI: 10.4267/2042/48467
This work is licensed under a Creative Commons Attribution-Noncommercial-No Derivative Works 2.0 France Licence.
© 2013 Atlas of Genetics and Cytogenetics in Oncology and Haematology

Identity
Other names: HNP, NP, NRPN, PRSS19, TADG14
HGNC (Hugo): KLK8
Location: 19q13.41
Local order: Telomere to centromere.

Note
This gene is one of the fifteen kallikrein subfamily members located in a cluster on chromosome 19. Kallikreins are a subgroup of serine proteases having diverse physiological functions.

DNA/RNA

Description
The KLK8 gene is approximately 7.8 kb in length, consisting of 8 exons (5 of them are coding exons) and 7 introns.

Transcription
Human KLK8 was originally cloned (Yoshida et al., 1998) as the human ortholog of the mouse brain protease neuropsin (Chen et al., 1995). Using Northern blot and RT-PCR analyses, it has been shown that KLK8 is expressed mainly in breast, cervix, esophagus, skin, ovary, testis, salivary glands and vagina. Adrenal, brain, colon, heart, kidney, lung, muscle and prostate also express KLK8 mRNA at medium to low levels. The transcription start site of KLK8 appears tissuespecific (Lu et al., 2009). Eight alternatively spliced variants have been identified for the KLK8 gene. These variants differ in the number and length of the 5’ untranslated exons and/or coding exons. The splice variants are predicted to encode 6 protein isoforms.

Type 1 and Type 2 transcripts differ in their coding exon 2 sequence. Type 2 includes extra 45 amino acids at the N-terminus of the coding exon 2.
Thus, Type 1 and Type 2 KLK8 mRNA variants produce 2zymogens that differ only in their propeptide sequences. Type 2 variant is absent in nonhuman primates, and is thus a human-specific splice form (Li et al., 2004; Lu et al., 2007). Type 1 mRNA is predominantly expressed in the pancreas and Type 2 mRNA in adult brain and hippocampus. Type 2 KLK8 is also abundantly expressed in fetal brain, placenta and in human embryonic stem cells, suggesting a potential role in embryogenesis (Mitsui et al., 1999; Lu et al., 2009).
The Type 3 mRNA variant includes coding exons 1, 4, and 5 and encodes a truncated form of the KLK8 protein (Magklara et al., 2001). Type 4 variant lacks coding exons 2, 3, 4. It encodes a putative protein of 32 amino acid residues that contains the KLK8 signal peptide and another peptide that is not related to KLK8 (Magklara et al., 2001).
Type 3 and Type 4 mRNAs are abundant in many normal tissues (brain, pancreas, skin) and are overproduced in ovarian and lung cancers (Magklara et al., 2001; Planque et al., 2010). Coding exon 2 is missing in Type 5 mRNA whereas Type 6 mRNA lacks coding exon 3. For these both variants, the alternative splicing creates a stop codon that prematurely terminates translation. Type 5 and Type 6 mRNAs were detected in lung cancer cell lines and tissues (Sher et al., 2006; Planque et al., 2010).

Pseudogene
None identified.
KLK8 (kallikrein-related peptidase 8) 

The KLK8 gene comprises 8 exons (dark color, classic numerals) and 7 introns. Shown here are the 6 alternative transcripts predicted to encode protein variants (only the coding exons are depicted). Yellow boxes: non-coding sequences; blue and green boxes: coding sequences. Sequences coding for identical amino acids are indicated in green whereas blue designates sequences generating distinct amino acids.

**Protein**

**Description**

The canonical KLK8 protein encoded by Type 1 mRNA has a secretion signal (pre-) peptide (28 amino acids), followed by an activation (pro-) peptide (4 amino acids) and the mature chain (228 amino acids) with 1 potential N-linked glycosylation site. The catalytic triad of His73, Asp120, Ser212 (relative to Met = 1) is conserved and is essential for proteolytic activity. After synthesis as a KLK8 precursor, the signal peptide is then cleaved and pro-KLK8 (zymogen) is subsequently secreted from the cell. Upon activation, the propeptide is removed to generate the mature active enzyme. Type 2 KLK8 has an insert of 45 amino acids between Ala23 and Gly24 at the C-terminus of the leader sequence of canonical KLK8. Therefore, this isoform has larger signal peptide and propeptide and has been produced intact as recombinant protein (Lu et al., 2009). Beside the canonical KLK8 protein, only the predicted peptide encoded by KLK8 Type 4 mRNA has been yet detected in vivo. This form was identified by mass spectrometry in bronchoalveolar lavage fluid (Oumeraci et al., 2011).

**Expression**

KLK8 protein has been detected in a wide range of tissues at low (10 to 100 ng/g, adrenal, cervix, heart, kidney, liver, ovary, salivary gland, vagina) to high (1 µg to 10 µg/g, breast, esophagus, skin, tensil) levels (Shaw and Diamandis, 2007). KLK8 has also been detected in body fluid, such as milk, amniotic fluid, cerebrospinal fluid, seminal plasma, serum, saliva and sweat (Kishi et al., 2003; Shaw and Diamandis, 2007; Eissa et al., 2011).

Age at the first full term pregnancy (FFTP) influences secretion of KLK8 protein in breast milk. Indeed in a recent study, a significant increase in KLK8 expression was observed from the onset of lactation to breast weaning depending on FFTP age (26) (Qin et al., 2012).

**Localisation**

KLK8 is a secreted protein and is localized intracellularly to the cytoplasm. In epidermis, KLK8 is localized within the trans-Golgi network, lamellar granules and intercellular spaces between the stratum granulosum and stratum corneum (Ishida-Yamamoto et al., 2004). Diffuse cytoplasmic staining was observed for KLK8 in the secretory segment in eccrine sweat glands and in the intradermal sensory nerve (Komatsu et al., 2005). KLK8 is present in relatively high levels in ductal cells, as well as in non-ductal cells, of normal salivary gland tissues and benign and malignant salivary gland tumors (Darling et al., 2008).

In brain, KLK8 is expressed in the cell body of oligodendrocytes (He et al., 2001).
KLK8 (kallikrein-related peptidase 8)

Schematic structure of the KLK8 Type1 protein. The amino acid numbering for the residues of the catalytic triad (His70, Asp120, Ser212) are relative to the full-length protein starting from Met1.

Function
KLK8 is a serine protease which exhibits trypsin-like activity with strong preference for Arg over Lys in the P1 position (Kishi et al., 2006; Eissa et al., 2011). KLK8 activity is inhibited by general serine protease inhibitors such as a2-antiplasmin, protein C inhibitor and PI-6 (Proteinase inhibitor 6) (Scott et al., 2007). Several potential substrates have been identified for KLK8 in human or mouse including extracellular matrix components (Single-chain tPA, fibronectin, gelatin, collagen type IV, fibrinogen) (Rajapakse et al., 2005), cell adhesion molecules (L1cam) and membranous receptors (EphB2, PAR2) (Matsumoto-Miyai et al., 2003; Nakamura et al., 2006; Attwood et al., 2011; Ramachandran et al., 2012), antimicrobial peptides (LL-37) and zymogens of kallikrein-related peptidases (proKLK1 and proKLK11) (Eissa et al., 2011). The physiological functions of KLK8 are not fully understood. Accumulating evidence has suggested pivotal roles for KLK8 in development, maturation and cognitive functions. KLK8 induces neurite outgrowth and fasciculation of cultured hippocampal neurons. Plays a role in the formation and maturation of orphan and small synaptic boutons in the Schaffer-collateral pathway, regulates Schaffer-collateral long-term potentiation in the hippocampus and is required for memory acquisition and synaptic plasticity (Komai et al., 2000; Oka et al., 2002; Nakamura et al., 2006; Terayama et al., 2007; Horii et al., 2008; Yoshida, 2010; Ishikawa et al., 2011; Shiosaka and Ishikawa, 2011). KLK8 has also been involved in skin desquamation and wound healing and in keratinocyte proliferation (Inoue et al., 1998; Kirihara et al., 2003; Kishihe et al., 2007; Yoshida, 2010; Eissa et al., 2011; Kishihe et al., 2012). It has been shown that KLK8 is differentially expressed in a number of malignancies, including ovarian, cervical, head and neck, breast and salivary gland cancers (Kishi et al., 2003; Cane et al., 2004; Borgono et al., 2006; Darling et al., 2008; Liu et al., 2008; Kountourakis et al., 2009), but the mechanisms of its involvement in these cancer have yet to be determined. In lung cancer, KLK8 suppress tumor cell invasiveness in vitro and in vivo (Sher et al., 2006).

Homology
The human KLK8 protein sequence shares 40-70% homology with other members of the human tissue kallikreins, and 70% identity with that of the mouse orthologue.

Mutations
Note
Genomewide DNA linkage analysis identified a susceptibility locus for intracranial aneurysm (IA) on chromosome 19q13 in the Finnish population. Two SNPs located in the intronic region of KLK8 were found significantly associated with IA (Weinsheimer et al., 2007). A significant allelic association between several KLK8 SNPs and bipolar disorder has recently been reported (Izumi et al., 2008).

No germinal or somatic mutations are identified to be associated with cancer so far.

Implicated in
Carcinomas
Note
Several carcinomas (ovarian, cervical, oral, salivary glands and lung cancers) show high expression of the KLK8 gene. Depending on the cancer type, KLK8 acts as tumor promoting or tumor suppressing factor.

Ovarian cancer
Disease
Expression of KLK8 was not detected on the surface epithelium of normal ovaries by immunohistochemistry. In contrast, KLK8 protein was detected in ovarian carcinomas with a significantly higher detection rate of KLK8 expression in early stage disease compared to advanced stage disease (Shigemasa et al., 2004). Other analyses using sandwich-type immunoassays found KLK8 protein in
cancer tissue extracts, serum and ascites fluid of ovarian cancer patients (Kishi et al., 2003; Shigemasa et al., 2004; Borgono et al., 2006).

**Prognosis**

It had been proposed that KLK8 is an independent marker of favorable prognosis in ovarian cancer at both the mRNA and protein levels. For example, KLK8 mRNA levels were found associated with longer disease-free survival (DFS) (Magklara et al., 2001; Shigemasa et al., 2004). The tissue concentration of KLK8 was also described as an independent marker of favorable prognosis in ovarian cancer. Patients with KLK8-positive tumors had a significantly longer DFS and overall survival than KLK8-negative patients (Borgono et al., 2006). Higher ascites fluid KLK8 concentration was also associated with better ovarian cancer DFS (Kishi et al., 2003). Using another approach, Kountourakis et al. showed significant correlations between tumour mask KLK8 protein expression levels and clinicopathological variables, including grade, residual disease and clinical response to chemotherapy. There was also a significant correlation between KLK8 tumour mask expression and five years progression-free survival (Kountourakis et al., 2009).

**Breast cancer**

**Disease**

KLK8 is downregulated in breast cancer tissues and cell lines (Yousef et al., 2004). On the other hand, KLK8, along with several other kallikrein genes, could be primarily up-regulated by 17β-estradiol and, to a lesser degree, by other steroid hormones in hormone receptor-positive breast cancer cell lines MCF-7 and T-47D, suggesting a coordinated kallikrein expression as part of a complex regulatory mechanism that controls the expression of these genes and also their downstream physiological function (Paliouras and Diamandis, 2007).

**Cervical cancer**

**Disease**

At the mRNA level, KLK8 was found to be highly expressed in 82% primary cervical cancer cell lines and in 87% established cervical cancer cell lines. In addition, immunohistochemistry staining of paraffin-embedded cervical cancer specimens showed KLK8 expression in tumor cells and its absence on normal cervical epithelial cells (Cane et al., 2004).

**Bladder cancer**

**Disease**

Reverse transcription-polymerase chain reaction analysis of 42 primary bladder tumor samples revealed an higher expression level of KLK8 mRNA in invasive tumors than in superficial tumors (Shinoda et al., 2007).

**Salivary gland cancers**

**Disease**

The KLK8 immunoreactivity was determined in normal salivary gland tissue and in malignant salivary gland tumors. In general, all the tumors showed a relatively high overall staining for both ductal and non-ductal cells, particularly mucoepidermoid carcinomas and adenocarcinoma NOS (Darling et al., 2008).

**Oral squamous cell carcinoma**

**Disease**

Comparison of oral squamous carcinoma (OSC) cell lines with either overexpression or silencing of uPAR revealed that the more aggressive phenotype is associated with a co-overexpression of KLK5, KLK7, KLK8 and KLK10. Furthermore, immunohistochemical analysis demonstrated strong reactivity for KLKs 5, 7, 8 and 10 in both orthotopic murine tumors and human OSC tissues. These results suggest that KLK8 along with other KLKs is involved in malignant progression of oral squamous cell carcinoma (Pettus et al., 2009).
skin of psoriasis vulgaris, seborrheic keratosis, lichen planus, and squamous cell carcinoma patients, compared with normal and basal cell carcinoma skin, suggested that human KLK8 is involved in keratinocyte differentiation and skin barrier formation (Kuwae et al., 2002; Shingaki et al., 2010; Shingaki et al., 2012). KLK8 protein overexpression was also detected in psoriasis, atopic dermatitis, and peeling skin syndrome skin tissues (Komatsu et al., 2006; Komatsu et al., 2007a; Komatsu et al., 2007b). KLK8 together with KLK5, KLK6, KLK7, KLK10 and KLK12 was upregulated in normal human keratinocytes following Sp1 silencing. Moreover, thymic stromal lymphopoietin (TSLP), an epithelial-derived Th(H)2-promoting cytokine, was induced in Sp1-silenced keratinocytes because of elevated KLK activity. This observation suggests that KLKs may contribute to Th(H)2 immune responses in the skin by inducing TSLP (Bin et al., 2011).

**Brain diseases**

**Note**

Under non-pathological conditions, KLK8 protein is localized mainly to the neurons of the cerebral cortex and hippocampus. Immunohistochemistry for KLK8 also demonstrated signals in cerebellum (The Human Protein Atlas). A variety of transcriptional controls through both physiological and nonphysiological activity, such as long-term potentiation, chemically induced plasticity, kindling epileptogenesis, and experimental encephalitis, have been shown to positively regulate Klk8 gene expression in mice (Momota et al., 1998; Komai et al., 2000; He et al., 2001; Ishikawa et al., 2011). Increased anxiety like response was also observed in Klk8/neuropsin-deficient mice (Horii et al., 2008). Recently, Attwood et al. (Attwood et al., 2011) have shown that Klk8/neuropsin is involved in stress-related plasticity in the amygdala by the cleavage of EphB2 during stress and the reduction of EphB2-NMDA binding. So currently, accumulating evidence supports pivotal roles of Klk8 in the early phase of synaptic plasticity, late associativity, and behavioral memory (Shiosaka and Ishikawa, 2011). Further studies are required to determine if KLK8 is involved in human brain diseases, however an overexpression of KLK8 has yet been observed in Alzheimer's disease hippocampus (Shimizu-Oka et al., 2001).

**References**


KLK8 (kallikrein-related peptidase 8) Courty Y


Lu ZX, Peng J, Su B. A human-specific mutation leads to the origin of a novel splice form of neuropin (KLK8), a gene involved in learning and memory. Hum Mutat. 2007 Oct;28(10):978-84


Planque C, Choi YH, Guyetant S, Heuzé-Vourc’h N, Briolilas L, Courty Y. Alternative splicing variant of kalliKrein-related


This article should be referenced as such: