Gene Section
Review

ASCL1 (achaete-scute homolog 1 or achaete-scute complex homolog 1)

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Identity

Hugo: ASCL1
Other names: ASH1; HASH1; MASH1
Location: 12q23.2
Note: Renault B et al performed fluorescent in situ hybridisation using YAC 896h that contains ASCL1 and other chromosome 12 specific markers such as IGF1, PAH and TRA1 to localise ASCL1. They established using genetic markers that ASCL1 localises distal to Phenylalanine hydroxylase (PAH) and proximal to tumor rejection antigen (TRA1) on chromosome 12q22-q23 cytogenic interval. (Beatrice Renault et al. Genomics 30, 81-83 (1995)).
LINEAGE: Eukaryota, Metazoa, Chordata, Craniata, Vertebrata, Euteleostomi, Mammalia, Eutheria, Euarchontoglires, Primates, Haplorrhini, Catarrhini, Hominidae, Homo. NCBI GI#: g119618109; g22658430; g20455478; g13111927; g12803079; g55743094; g306460; g13325212.

DNA/RNA

Description

2 exons spanning 2824bp although only exon1 codes for the protein. Orientation '+' strand. Exon 1 101875594-101876910 (1317 bp), Exon 2 101877270-101878417 (1148 bp), Intron 1-2 101876911-101877269 (359 bp). Gene id= 429 (KEGG). Refseq: NM_004316.2.
HASH1/ASCL1 promoter has two independent transcription start sites of which proximal INR element (YYANYY consensus binding site) plays a predominant role. The general enhancer has several Sp1 binding sites. Tissue restricted expression control comes from the repressor regions present in the distal (over 13.5kb upstream) and proximal 5' flanking region. The proximal repressor is shown to have a class C element site to which HES1 can bind and regulate HASH1/ASCL1 expression.
Variant: 158--158 Glu to Gly (E to G) Var_013179 in P50553.

Transcription

2.46 Kb mRNA, coding sequence 711 bp.

Protein

Note: This gene encodes a member of the basic helix-loop-helix (bHLH) family of transcription factors. The protein activates transcription by binding to the E box (5'-CANNTG-3'). Dimerization with other bHLH proteins is required for efficient DNA binding. This protein plays a role in the neuronal commitment and differentiation and in the generation of olfactory and autonomic neurons. It is highly expressed in medullary thyroid cancer (MTC) and small cell lung cancer (SCLC) and may be a useful marker for these cancers. The proximal coding region of the cDNA contains a striking 14-copy repeat of the triplet CAG that exhibits polymorphism in human genomic DNA The presence of a CAG repeat in the gene suggests that it could play a role in tumor formation.
Protein size: 237 aa, 25.45kDa. Refseq: NP_004307.2.

Description

The amino terminal of the protein contains poly alanine and poly glutamine repeat rich region. The polyglutamine length polymorphism in ASCL1 has been postulated that it could influence predispositions to Parkinson's disease. The carboxy terminus of the protein contains the basic helix loop helix domain, which is important for interaction with other transcription factors.
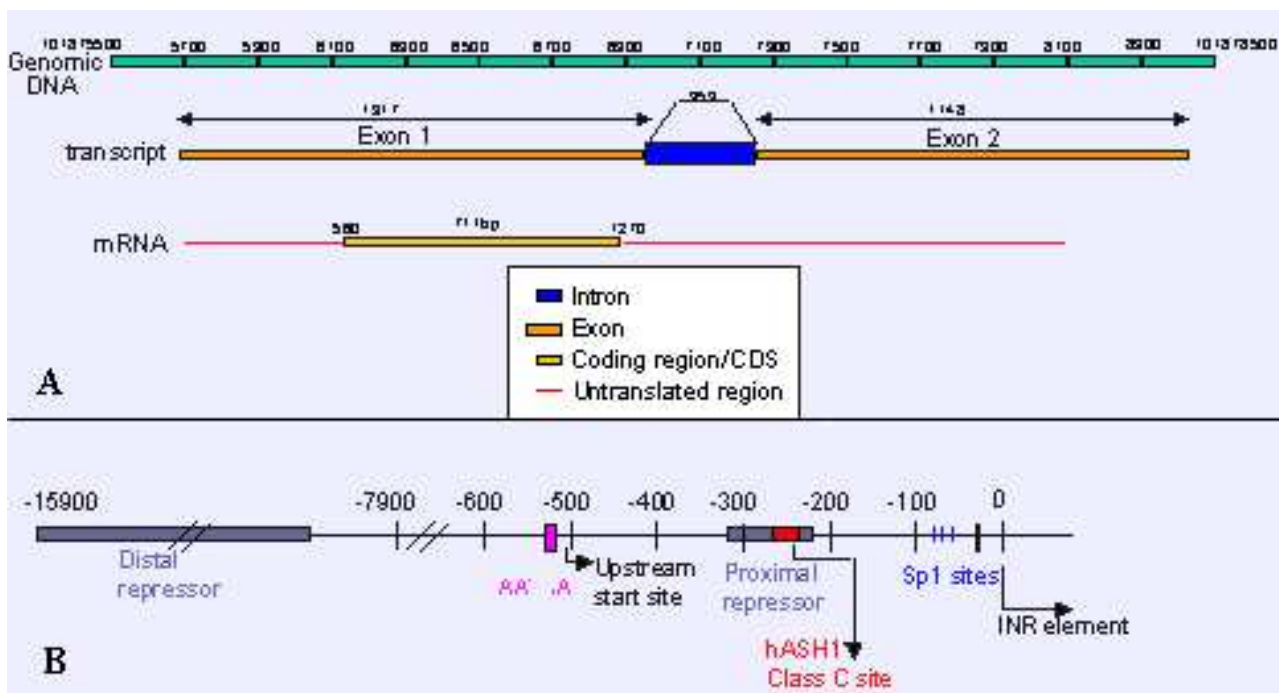
A: Schematic representation of the ASCL1 genomic DNA depicting exons, introns, transcript and mRNA with untranslated and coding region. B: Schematic diagram showing HASH1/ASCL1 promoter with proximal and distal repressors, HASH1/ASCL1 class C site, enhancer Sp1 binding sites and transcription initiation sites (TATA box and INR (initiator) element).

Expression

The gene product is expressed basically nuclear and is expressed in tissues like brain, lung and nervous system.

Function

ASCL1 functions as a bHLH transcription factor that binds to E-box whose canonical sequence is 5'CANNTG 3'. It also acts as protein binding, transcription factor, and in cell differentiation. MASH1/ASCL1 is expressed during development of rat retina and interacts specifically with an E-box identified in the promoter for the opsin gene during rod photoreceptor differentiation. NOTCH1 and its downstream signal transducer HES1 regulates the transcription of HASH1/ASCL1 and is very instrumental in neuronal developmental pathways particularly dictating neuroendocrine differentiation in various organs.

Interacting partners: E1A binding protein p300, Ubiquilin 1, Bone morphogenetic protein 2 (BMP2), Trancription factor 3 (TF3), Transcription factor 4 (TF4), Myocyte specific Enhancer factor 2A (MSEF2A), Neurogenin.

Homology

Mus musculus Ascl1 achaete-scute complex homolog-like 1 (Drosophila) NM_008553.2; Rattus norvegicus Ascl1 achaete-scute complex homolog-like 1 (Drosophila) NM_022384.1; Canis familiaris similar to achaete-scute homolog 1 LOC482628; Danio Rerio achaete scute homolog A asha NM_131219.1.

Mutations

Note: Congenital central hypoventilation syndrome is a rare disorder of the chemical control of breathing. The ASCL1--PHOX2A--PHOX2B developmental cascade was proposed as a candidate pathway for this disorder, as well as for Haddad syndrome, because the cascade controls the development of neurons with a definitive or transient noradrenergic phenotype. De Pontual et al. identified heterozygosity for mutations in the ASCL1 gene in 2 patients with CCHS and 1 patient with Haddad syndrome. The authors also developed an in vitro model of noradrenergic differentiation in neuronal progenitors derived from the mouse vagal neural crest. All Ascl1 mutant alleles result in impaired noradrenergic neuronal development when over expressed from adenoviral constructs.

Schematic representation of ASCL1 protein depicting various motifs such as poly alanine and poly glutamine repeats, a basic motif and a bHLH DNA binding motif.

ALLELIC VARIANTS

1. ASCL1 C52A, PRO18THR
In a patient with CCHS, de Pontual et al identified heterozygosity for a 52C-A transversion in the ASCL1 gene, translating in a pro18-to-thr substitution. The patient was also heterozygous for a polyalanine expansion mutation in PHOX2B, which is known to induce CCHS.

2. ASCL1, 15-BP DEL, NT111
Heterozygosity for a 15-bp deletin (111-115 Del 15nt) in the ASCL1 gene was reported in a patient with CCHS by de Pontual et al. The mutation was predicted to result in loss of 5 of 13 alanine residues (ala37-ala41) in a polyalanine tract.

3. ASCL1, 24-BP DEL, NT108
Another heterozygosity for of 24-bp deletion (108-131del24nt) in the ASCL1 gene was identified in a patient with Haddad syndrome (209880), de Pontual et al The mutation was predicted to result in loss of 8 of 13 alanine residues (ala36-ala43) in a polyalanine tract. Genetic association with Parkinson's disease has been shown by Ide M et al (PMID=16021468) and that with sudden infant death syndrome or (SIDS) has been shown by Weese-Mayer DE et al (PMID= 15240857).

Implicated in

Note: Upon characterization of expression of ASCL1 in several human cancer cell lines and tumors it is found that ASCL1 is highly expressed in and has been implicated to impart neuroendocrine behaviour to various NE- tumors e.g gastrointestinal NE carcinoma (NEC), Pheochromocytomas, olfactory neuroblastomas or esthesioneuroblastoma, pulmonary and thyroid carcinoids, Medullary thyroid cancer (MTC), small cell lung cell cancer (SCLC) and recently in prostate small cell carcinoma. Apart from this, ASCL1 protein is also shown to be highly upregulated in progressive secondary glioblastoma (GBM). Notch signalling down regulates ASCL1 levels but its expression is shown to be very minimal or non-existent in neuroendocrine tumors and hence inhibition of ASCL1 expression that has been implicated to impart neuroendocrine behaviour could be a therapeutic target to suppress tumor growth.

Neuroendocrine tumors

Note: Neuroendocrine tumors originate from cells that are capable of amine precursor (such as dopa and 5-hydroxytryptophan) uptake and decarboxylation (APUD cells). As a result, these tumors have high intracellular levels of carboxyl groups and nonspecific esterase, which are used as a neuroendocrine marker. These tumors have NE- phenotype characterized by expression of ACTH, vasopressin, calcitonin gene related peptide (CALCA/CGRP), Gastrin releasing peptide and secretory proteins like synaptophysin and chromogranins, serotonin. HASH1/ASCL1 appears to be cardinal/ hallmark feature of each of these tumor types. Clinically Chromogranin A is most commonly used as a marker to identify NE- tumors.

Olfactory neuroblastomas or Esthesioneuroblastoma (ENB)

Note: paranasal sinus nasal cavity esthesioneuroblastoma or esthesioneurocytoma, esthesioneuroma, and esthesioneuroepithelioma.

Disease

Esthesioneuroblastoma (ENB) is a rare tumor arising out of the nasal vault from cells of the developing sympathetic nervous system. When neuroblastoma cells are induced to differentiate, as indicated by neuronal morphology and upregulation of neuronal marker genes, the HASH-1/ASCL1 expression is rapidly downregulated with a concomitant, transient upregulation of HES-1. ENB is generally a slow-growing tumor with a high 5-year survival (81%). Recurrences usually occur within the first 2 years. However, late recurrences are common, and hence follow-up must be done for a prolonged time.

Prognosis

HASH1/ASCL1 is expressed in immature olfactory neurons and is critical for their development. Mhawech et al found distinct expression of HASH1/ASCL1 in all estersioneuroblastoma samples as compared to the poorly differentiated tumors that were negative. They also report inverse correlation between grades of esthesioneuroblastoma and HASH1/ASCL1 mRNA levels and propose that HASH1/ASCL1 could be used as useful tool to distinguish estersioneuroblastoma from poorly differentiated tumors of sinonasal region.

Pulmonary and thyroid carcinoids

Note: Carcinoid tumors or carcinoids.

Disease

These tumors originate in hormone-producing cells of the gastrointestinal (GI) tract (i.e., esophagus, stomach, small intestine, colon), the respiratory tract (i.e., lungs, trachea, bronchi), the hepatobiliary system (i.e., pancreas, gallbladder, liver), and the reproductive glands (i.e., testes, ovaries). Carcinoids are classified as slow growing neuroendocrine tumors. They develop in peptide- and amine-producing cells, which release hormones like serotonin in response to signals from the nervous system. Excessive amounts of these hormones cause a condition called carcinoid syndrome in approximately 10% of patients with carcinoid tumors.

Prognosis

Multiple endocrine neoplasia type 1 (MEN1) is a genetic disorder that increases the risk for neuroendocrine tumors, including carcinoids. Gastrointestinal conditions (e.g., peptic ulcer disease, pernicious anemia, atrophic gastritis, Zollinger-Ellison syndrome) increase the risk for carcinoid tumors of the

GI tract. Carcinoid patients also have an increased risk for Cushing's syndrome. Atypical Carcinoids (fast growing and potentially metastatic) express high levels of HASH1/ASCL1 (although lower than NECs) and is associated with poor prognosis for survival.

RAF-1 activation is detrimental to tumorigenesis in carcinoid cells. Raf1 activation in an estrogen inducible system in pancreatic carcinoid cell line (BON) and in pulmonary cell lines leads to marked reduction in NE-markers such as 5-HT, chromogranin A, and synaptophysin and HASH1/ASCL1 has been observed.

Medullary thyroid cancer (MTC)

Disease

Medullary thyroid cancer is a neuroendocrine tumor derived from the parafollicular calcitonin producing C cells of thyroid and accounts for about 3% of thyroid cancers.

Inheritance: About 20% of have an inherited form of the disease and familial MTC are transmitted in an autosomal dominant fashion involving mutations in the RET proto-oncogene. So far surgery remains only curative treatment modality.

Prognosis

The classical tumor marker and the secreted hormone is calcitonin which is tightly regulated by Notch signalling and HASH1/ASCL1 levels. It has also been shown that by activating RAF-1 signalling mediated by MEK induction leads to complete suppression of ASCL1 and mRNA protein which is frequently upregulated in MTC. HASH1/ASCL1 over expression is linked to poor prognostic value.

Small cell lung cancer (SCLC)

Note: Oat cell carcinoma.

Disease

SCLC cells are small and round to fusiform with scant cytoplasm.SCLC tumors are poorly differentiated neuroendocrine tumors as compared to bronchoid carcinoid tumors and is an aggressive and highly metastatic tumor, accounting to about 20% death from lung cancer. Owing to its NE-phenotype, these tumors secrete chromogranin A, GRP and calcitonin in addition to over expressed HASH1/ASCL1.

Genetically c-myc has shown to be over expressed by gene amplification and retinoblastoma (Rb) is frequently mutated in SCLC. P53 and PTEN also show aberrant expression. Loss of chromosome 3 sequences appears to occur frequently at the very earliest stages of neoplastic transformation. Losses at the short arms of chromosome 3 and 17 and the long arm of 5 are seen consistently in almost all SCLC patients. Although to date, there are no known examples of amplification or rearrangement of the HASH1/ASCL1 gene.

Prognosis

HASH1/ASCL1 is associated with significantly reduced survival in small cell lung carcinoma patients and has adverse prognostic association.

Pheochromocytomas

Note: Chromaffin tumors.

Disease

Because pheochromocytomas arise from chromaffin cells, they are occasionally called chromaffin tumors. Pheochromocytomas are found in the adrenal medulla. The adrenal medulla normally secretes two hormones, called norepinephrine and epinephrine(also known as adrenaline). Pheochromocytomas cause the adrenal medulla to secrete too much adrenaline and often causes the adrenal glands to make excess of hormones called catechol-amines which in turn causes high blood pressure and other symptoms.

Atleast in rat pheochromocytoma cell line PC12, MASH1/ASCL1 is readily detected which can be further induced by NGF treatment. Whether HASH1/ASCL1 also is over expressed in human cancers needs careful examination and distinction between tumor types.

Inheritance: About 10-25% of this cancer can be familial and mutations in genes e.g. VHL, RET, NF1, SDHB and SDHD are implicated.

Prognosis

Pheochromocytoma can be potentially fatal, but it is relatively uncommon (2-8 cases per million people annually). As with other neuroendocrine tumors, high levels of HASH1/ASCL1 expression seen in pheochromocytomas correlate with poor prognosis. Activation of MEK1 / MEK2 - ERK1 / ERK2 is necessary for differentiation of pheochromocytoma (PC12) cells and leads to decreased cell proliferation.

Cytogenetics

Allelic losses at Chromosome 1p, 3p, 17p and 22q have been reported in sporadic and familial forms of pheochromocytomas.

Gastrointestinal neuroendocrine carcinoma (NEC)

Disease

Gastrointestinal NECs are defined as small cell carcinoma, morphologically similar to the small cell carcinoma of the lung. Gastrointestinal NE carcinoma (NEC) are extremely aggressive, but its pathophysiologic features remain poorly understood. Shida et al assessed HASH1/ASCL1 expression in human NECs by quantitative RT-PCR and in situ hybridisation and showed marked upregulation of HASH1/ASCL1 mRNA in NECs which was weak in

carcinoid tumors and scarcely expressed in adenocarcinomas and normal mucosa.

Prognosis

Levels of HASH1/ASCL1 can be used as a more sensitive and specific marker than conventional pan-endocrine markers for clinical diagnosis of gastrointestinal tumors to differentiate among gastrointestinal tumors particularly between carcinoids, adenocarcinomas and neuroendocrine gastrointestinal tumors in addition to other NE markers.

Astrocytoma (secondary glioblastoma (GBM))

Note: Glioblastoma.

Disease

Astrocytoma is the most common type of brain cancer arising from the astrocytes affecting cerebral hemispheres in adults and the brain stem in children accounting to almost 60% of brain tumors.

According to the world health organization, astrocytomas are classified in to four grades:
1. Grade I or pilocytic astrocytoma (PA);
2. Grade II diffused astrocytoma (DA);
3. Grade III Anaplastic astrocyoma (AA);
4. Grade IV Glioblastoma multiforme (GBM).

GBM can further be classified as being primary or secondary based on the genetic mutations, age at occurrence, tendency of progression and clinical course. Familial clustering of gliomas is frequently observed associated with defined inherited tumor syndrome incuding the Li-Fraumeni syndrome, Turcot syndrome, and the NF1 syndrome. Several genes have been associated in distinguishing one or the other form of GBM notably among which are P53, MDM2, EGFR, CDK4. LOH on chromosome 9,10, 13, 17,19, 22 frequently occur in GBMs.

Prognosis

The median survival time for a GBM individual is about 12 months and age at the time of occurrence plays a significant prognostic factor. Recently reported methylation at the O6- methyl guanine DNA methyl-transferase (MGMT) promoter has been shown to confer favourable prognostic value in terms of response to chemotherapy and longer survival. ASCL1 is highly upregulated in secondary GBMs as compared to the primary GBMs and can be thus ascribed as a distinguishing marker between the two. Concomitantly, there is repression of NOTCH1 signalling and HES1 expression in the secondary GBM. It is observed that primary GBM patients show rapid tumor progression and poor prognosis.

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