

Gene Section

Review

RACGAP1 (Rac GTPase activating protein 1)

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Abstract

Review on RACGAP1, with data on DNA, on the protein encoded, and where the gene is implicated.

Keywords: RACGAP1; meningioma; colorectal cancer; melanoma; Hepatocellular carcinoma; gastric cancer

Identity

Other names: CYK4, HsCYK-4, ID-GAP, MgcRacGAP, KIAA1478, ID-GAP, RCGAP1 (Toure A et al., 1998; Toure A et al., 2001; Burkard ME et al., 2009)

HGNC (Hugo): RACGAP1

Location: 12q13.12

Local order

According to NCBI Map Viewer, genes locating in the region next to RACGAP1 in centromere to telomere direction on 12q13 are: TMBIM6 (12q13.12), transmembrane BAX inhibitor motif containing 6; FAIM2 (12q13), Fas apoptotic inhibitory molecule 2; AQP2 (12q13.12), aquaporin 2 (collecting duct); AQP5 (12q13), aquaporin-5; RACGAP1; ASIC1 (12q12), acid sensing ion channel subunit ; SMARCD1 (12q13-q14), SWI/SNF related, matrix associated, actin dependent regulator of chromatin, subfamily d, membre 1; GPD1 (12q13.12), glycérol-3-phosphate dehydrogenase 1; CERS5 (12q13.12), ceramide synthase 5.

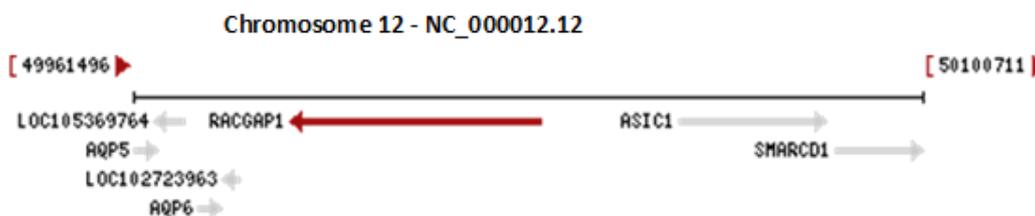
DNA/RNA

Description

According to NCBI Map Viewer, RACGAP1 maps to NC_000012.12 in the region between 49961496 and 50100711 on the minus strand and spans 56,214 bases. The total number of exons is 22. As reported by SABiosciences' database, the transcription factor binding sites, in the RACGAP1 gene promoter, are: STAT3, FOXO1, GATA1, TBP, FOXD3, FOXO4, SRY, REPIN1 (AP-4).

Transcription

RACGAP1 gene has 12 alternative transcript variants, which encode 4 isoforms of Rac GTPase activating protein 1. The locus of the reference variant (variant 1) is in NM_013277 and it has 3361bp. Variant 2 lacks an exon and contains an alternate exon in the 5' UTR, while variant 3 lacks two exons and uses an alternate splice site in the 5' UTR, variant 4 lacks two exons in the 5' UTR, variant 5 lacks two exons and contains an alternate exon in the 5' UTR and variant 6 lacks two exons and uses two alternate splice sites in the 5' UTR. Variants 7, 8 and 9 lack two exons and use an alternate splice site in the 5' UTR. All the transcript variants from 1 to 9 encode for isoform a. The 10th variant lacks three exons in the 5' region and initiates translation at a downstream start codon compared to variant 1. Its encoded isoform b has a shorter N-terminus than isoform a.



A schematic illustration of RACGAP1 gene The RACGAP1 gene is shown with red bar and it is flanked by AQP6 and AQP5 genes on the left side and ASIC1 and SMARCD1 genes on the right side. RACGAP1's location is on 12q13.12, and it has 22 exons (NCBI).

The 11th variant lacks two exons, contains an alternate exon and uses an alternate splice site in the 5' region. It initiates translation at an alternate start codon. Its encoded isoform c has a longer N-terminus than isoform a.

Lastly, variant 12 lacks three exons in the 5' region and initiates translation at an alternate downstream start codon. The encoded isoform d has a distinct N-terminus and is shorter than isoform a. Transcription is regulated by STAT3, FOXO1, GATA-1, TBP, FOXD3, FOXO4, SRY and AP-4 binding sites (NCBI database).

Pseudogene

FKSG42 is a RACGAP1 pseudogene as reported by HGNC. It is located in 12q12 chromosome and it has 1 exon.

Protein

RACGAP1 protein plays an important role in the Rho GTPase activation cycle. In fact, it binds activated forms of Rho GTPases and stimulates GTP hydrolysis. Thus, it inhibits Rho-mediated signals. Regarding RACGAP1 protein function, it has a regulatory role in the starting of cytokinesis, regulates spermatogenesis and proliferation of neuronal cells, as well as it controls cell growth and differentiation of hematopoietic cells (Boguski M.S. et al., 1993; Hamanaka R. et al., 1994). Lately, it was found that high levels of RACGAP1 is correlated with a worse prognosis in patients with high grade meningiomas (Brennan IM et al., 2007), while Wang SM et al. showed that RACGAP1 upregulation is associated with relapse of hepatocellular cancer (Wang SM et al., 2011).

Description

According to Swiss Prot database the protein contains 632 amino acids and its molecular mass is 71027 Da. About protein's structure, it is a heterotetramer of two of RACGAP1 and KIF23 ID: 41068> (Kinesin Family membre 23) molecules. It is found in the centralspindlin complex, a motor complex required for the assembly of the mitotic spindle (Nature Reviews Molecular Cell Biology, 2009). It is associated with alpha-, beta- and gamma-tubulin and microtubules. The protein interacts via

its Rho-GAP domain with RND2 (Rho family GTPase 2) and PRC1 (Protein Regulator of Cytokinesis). Its GAP activity towards CDC42 (Cell Division Cycle42 effector protein) is inhibited by interaction with PRC1 in vitro, which may be important for maintaining a normal spindle morphology.

Furthermore, RACGAP1 associates with AURKB (Aurora Kinase B) during M phase and interacts with SLC26A8 (Solute Carrier family 26 member 8) via its N-terminus and RAB11FIP3 (RAB11 Family Interacting Protein 3).

During anaphase and cytokinesis, the protein binds to ECT2 (Epithelial Cell Transforming 2) in a microtubule-dependent manner and it is enhanced by phosphorylation by PLK1 (Polo-like Kinase 1). Lastly, it interacts directly with KIF23.



The Crystal Structure Of The Human Rac GTPase Activating Protein 1 (RACGAP1) (PROTEIN DATA BANK)

Expression

It is highly expressed in the testis, thymus, placenta, gastrointestinal tract, urinary tract, uterus, cervix, skin, tonsil, nasopharynx, bone marrow and lymph nodes.

It is found at lower levels in spleen and peripheral blood lymphocytes. In testis, RACGAP1 expression is restricted to germ cells with the highest levels of expression found in spermatocytes.

Its expression is monitored during the cell cycle and it peaks at G2/M phase (Toure A et al., 1998; Toure A et al., 2001; Naud N et al., 2003; Zhang P et al., 2015).

Localisation

RACGAP1 colocalizes with RND2 in Golgi-derived proacrosomal vesicles and the acrosome. During interphase, it is localized to the nucleus and cytoplasm along with microtubules, while in anaphase it is organized to the central spindle and in telophase and cytokinesis to the midbody. Colocalization of RACGAP1 with RhoA occurs at the myosin contractile ring during cytokinesis and with ECT2 at the mitotic spindles during anaphase/metaphase, at the cleavage furrow during telophase, and to the midbody at the end of cytokinesis. Lastly, it neighbours with CDC42 to spindle microtubules from prometaphase to telophase (Mishima M et al., 2002).

Function

MgcRacGAP is a component of the central spindle complex that serves as a microtubule-dependent and Rho-mediated signaling, required for the myosin contractile ring formation during the cytokinesis. To specify, it helps for a proper attachment of the midbody to the cell membrane during cytokinesis. GAP1 plays also key role in controlling cell growth and differentiation of hematopoietic cells, adipocytes and myoblasts. It may regulate the cell proliferation in the nervous system as well as the sulfate transport in male germ cells. In addition, MgcRacGAP, by inhibiting the activity of RAC1 and CDC42, which act on the microtubule and actin cytoskeleton, controls cell proliferation, adhesion and motility. It may play a role in regulating cortical activity through RHOA-mediated signals during cytokinesis. At last, it is found to be essential for the early stages of embryogenesis (Toure A et al., 1998; Kawashima T et al., 2000; Hirose K et al., 2001; Yuce O et al., 2005).

Homology

According to NCBI, the RACGAP1 gene is conserved in chimpanzee, Rhesus monkey, dog, cow, mouse, rat, chicken, zebrafish, fruit fly, mosquito, and frog.

Mutations

Note

The mammalian Polo-like kinase PLK1 (Hamanaka R et al., 1994) was shown recently to be an essential activator of RhoA (Brennan IM et al., 2007; Burkard ME et al., 2007; Petronczki et al., 2007; Santamaria et al., 2007). Inhibition of PLK1 prevents ECT2 association with HsCyk-4 (alias of RACGAP1) and blocks its recruitment to the central spindle (Brennan IM et al., 2007; Burkard ME et al., 2007; Petronczki et al., 2007; Santamaria et al., 2007), proposing that PLK1 might serve as a stimulatory kinase. Benjamin A. Wolfe et al., examined the mechanism by which PLK1 stimulates association between ECT2 and

HsCyk-4 during anaphase to trigger the cytokinesis (Benjamin A. Wolfe et al., 2009). In the meanwhile, Lowery DM and colleagues found that the N terminus (Nt) of HsCyk-4 contains seven consensus motifs for PLK1 phosphorylation (Lowery DM et al., 2005). Benjamin et al., trying to determine which sites are phosphorylated by PLK1, confirmed that only peptides containing amino acids 139-174 of HsCyk-4 did so. Within this region, they took up four hypothetical PLK1 phosphorylation sites (Ser149, Ser157, Ser164, and Ser170), which are well conserved across species, and they altered them. Mutation of any of these four residues was insufficient on its own to prevent PLK1-stimulated binding. The results show that, in vitro phosphorylation of HsCyk-4-4A (amino acids 111-188) by PLK1 was dramatically reduced. Conversely, they mutated these four serine residues to Asp (HsCyk-4-4D) so as to mimic the phosphorylated state, and generated a form of HsCyk-4 able to associate with ECT2-BRCT even in the absence of PLK1 phosphorylation. Based on these results they conclude that PLK1 phosphorylates multiple serine residues in the N terminus of HsCyk-4 in vitro, thereby stimulating its association with ECT2.

Implicated in

Meningioma

Disease

Meningiomas are a diverse set of tumors arising from meninges, the membranous layers covering the brain and the spinal cord (Cushing H, 1922). Meningiomas specifically derive from the arachnoid granulations of the arachnoid matter (Buetow MP et al., 1991). These tumors are usually benign in nature and slowly growing; however, a small percentage are malignant (Goldsmith BJ et al., 1994). The classification of meningiomas is based upon the WHO classification system: Grade I (Benign) 90% - meningothelial, fibrous, transitional, psammomatous, angioblastic, Grade II (Atypical) 7% - chordoid, clear cell, atypical (includes brain invasion), Grade III (Anaplastic/malignant) 2% - papillary, rhabdoid, anaplastic (most aggressive). Most meningiomas produce no symptoms throughout a person's life, and if accidentally found, require no treatment other than periodic observation. Typically, symptomatic meningiomas are treated with either radiosurgery or conventional surgery.

Prognosis

Ke HL et al. did a research on how RACGAP1 expression is correlated in the meningiomas (Ke HL et al., 2013). The results showed a higher RACGAP1 expression in Grade III meningioma comparing to Grade I. Higher levels of RACGAP1

mRNA were significantly correlated with tumor size, higher Simpson grade, histological type and clinical course ($P < 0.05$). Additionally, patients with high levels of RACGAP1 mRNA had a significantly worse survival than the lower level ones ($P = 0.008$). Upon these results, the researchers suggested that RACGAP1 may be used as a potential predictive biomarker for disease aggressiveness and patients' prognosis.

Hepatocellular carcinoma

Disease

Hepatocellular carcinoma (HCC), also called malignant hepatoma, is the most common type of liver cancer. Most cases of HCC are secondary to either a viral hepatitis infection (hepatitis B or C) or cirrhosis. The clinical manifestations of HCC are usually jaundice, ascites, blood clotting abnormalities, loss of appetite, unintentional weight loss, abdominal pain, especially in the right upper quadrant, nausea, vomiting, or tiredness. Prognosis and treatment options for HCC depend on staging but also on other factors such tumor grade, Child-Pugh score and overall patient's performance status and comorbidities. High-grade tumors will have a poor prognosis, while low-grade tumors may go unnoticed for many years (Naud N et al., 2003)

Prognosis

It has been proved that relapse of Hepatocellular Carcinoma (HCC) increases with higher HBV viral load (Yu SJ et al., 2014). Wang SM et al., proved that high RACGAP1 expression associates with high risk of relapse of HBV-positive HCC patients (Wang SM et al., 2011). According to their research, silencing of RACGAP1 in Hep3B and MHCC97-H, in cancer cells with high endogenous RACGAP1 expression, inhibited cell migration and invasion. This outcome suggests that RACGAP1 besides being an independent informative prognostic biomarker, it could also be a potential molecular target for designing therapeutic strategies for HCC.

Colorectal cancer

Disease

Colorectal cancer is the development of cancer in the large bowel. Signs and symptoms may include melena (black stools), a change in bowel habits, pain, weight loss, cachexia, or persistent fatigue. Treatments available for colorectal cancer include surgery, radiation therapy, chemotherapy, targeted therapy or combination of above depending on the disease site and stage. Cancers confined to the wall of the colon are usually curable with surgery while cancer that has spread distantly are usually incurable. In very advanced stages management might simply focus on quality of life and symptoms control (National Cancer Institute, 2014).

Prognosis

RACGAP1 plays a key role in regulating various cellular functions including cytokinesis, transformation, invasive migration and metastasis. In the study of Imaoka H et al., the function and clinical significance of RACGAP1 expression in colorectal cancer (CRC) was investigated (Imaoka H et al., 2015).

Reduced RACGAP1 expression by siRNA in CRC cell lines showed significantly decreased cellular proliferation, migration and invasion whereas increased expression of RACGAP1 was highly associated with worse prognosis.

In multivariate analyses, the researchers found that high levels of RACGAP1 was an independent prognostic biomarker for lymph node metastasis and worse disease-free and relapse free survival in CRC.

Gastric cancer

Disease

Stomach cancer, also known as gastric cancer, develops from the lining of the stomach. Early symptoms may include heartburn, upper abdominal pain, nausea and loss of appetite. Later signs and symptoms may include weight loss, vomiting, difficulty swallowing, and melena.

The cancer may spread from the stomach to other parts of the body, particularly the lymph nodes, liver, lungs, peritoneum or bones etc. Worldwide, stomach cancer is the fifth leading cause of cancer and the third leading cause of death from cancer. Regarding its management, this disease is not usually curable unless it is found at a very early stage.

Unfortunately, the majority of patients are diagnosed when the disease has already become advanced. Treatment options include surgery, chemotherapy, radiation therapy (National Cancer Institute, 2014) and lately biological therapy and immunotherapy.

Prognosis

Saigusa S et al. studied the clinical significance of RACGAP1 expression at the invasive form of gastric cancer (Saigusa S et al., 2015). Patients with positive RACGAP1 expression at the invasive form, had significantly poorer prognosis than those without expression ($P < 0.0001$). In the multivariate analysis, lymph node metastasis ($P = 0.0106$), distant metastasis ($P = 0.0012$) and positive RACGAP1 expression ($P = 0.001$) were independent prognostic factors. Based on these results, researchers conclude that RACGAP1 expression may play a crucial role in the progression of gastric cancer.

Melanoma

Disease

Melanoma is a type of cancer that develops from the melanine-containing skin cells, known as melanocytes. Early signs of melanoma are changes to the shape or color of an existing mole or, in the case of nodular melanoma, the appearance of a new

lump anywhere on the skin. At later stages, the mole may itch, ulcerate or bleed (National Cancer Institute, 2015). Melanomas are usually caused by DNA damage resulting from long-standing exposure to ultraviolet (UV) light from the sun. Confirmation of the clinical diagnosis is based on a skin biopsy. Treatment of melanoma may include surgery, chemotherapy, immunotherapy and radiotherapy, depending again on disease and patients' stage and status.

Prognosis

Melanoma cell migration across vascular endothelial cells is crucial for tumor metastasis. According to Zhang P et al., RACGAP1-driven focal adhesion formation, promotes melanoma cells to migrate through vascular endothelium by causing adherens junction disassembly (Zhang P et al., 2015). After, depletion of RACGAP1 with RACGAP1-targeting siRNA or overexpression of RACGAP1 mutant (T249A), melanoma cell transendothelial migration and disorganization of adherens junctions was reduced. In addition, RACGAP1 promoted the activation of RhoA, FAK, PXN (paxillin) and triggered focal adhesion formation and cytoskeletal rearrangement. By overexpressing FRNK (FAK-related non-kinase) in endothelium, they proved that RACGAP1 damages the endothelial barrier and facilitates melanoma transmigration in a focal adhesion-dependent manner.

References

- Cancer biology (4th ed.). Oxford: Oxford University Press. p. 223. Oxford: Oxford University Press. p. 223. Sim, edited by Fiona; McKee, Martin (2011).
- . Nature Reviews Molecular Cell Biology 2009; 10, 441
- Boguski MS, McCormick F. Proteins regulating Ras and its relatives Nature 1993 Dec 16;366(6456):643-54
- Brennan IM, Peters U, Kapoor TM, Straight AF. Polo-like kinase controls vertebrate spindle elongation and cytokinesis PLoS One 2007 May 2;2(5):e409
- Buetow MP, Buetow PC, Smirniotopoulos JG. Typical, atypical, and misleading features in meningioma Radiographics 1991 Nov;11(6):1087-106
- Burkard ME, Maciejowski J, Rodriguez-Bravo V, Repka M, Lowery DM, Clauser KR, Zhang C, Shokat KM, Carr SA, Yaffe MB, Jallepalli PV. Plk1 self-organization and priming phosphorylation of HsCYK-4 at the spindle midzone regulate the onset of division in human cells PLoS Biol 2009 May 5;7(5):e1000111
- Cushing H. The meningiomas (dural endotheliomas): their source, and favoured seats of origin Brain 1922; 45 (2): 282-316.
- Goldsmith BJ, Wara WM, Wilson CB, Larson DA. Postoperative irradiation for subtotally resected meningiomas A retrospective analysis of 140 patients treated from 1967 to 1990 J Neurosurg
- Hamanaka R, Maloid S, Smith MR, O'Connell CD, Longo DL, Ferris DK. Cloning and characterization of human and murine homologues of the Drosophila polo serine-threonine kinase Cell Growth Differ 1994 Mar;5(3):249-57
- Hirose K, Kawashima T, Iwamoto I, Nosaka T, Kitamura T. MgcRacGAP is involved in cytokinesis through associating with mitotic spindle and midbody J Biol Chem 2001 Feb 23;276(8):5821-8
- Imaoka H, Toiyama Y, Saigusa S, Kawamura M, Kawamoto A, Okugawa Y, Hiro J, Tanaka K, Inoue Y, Mohri Y, Kusunoki M. RacGAP1 expression, increasing tumor malignant potential, as a predictive biomarker for lymph node metastasis and poor prognosis in colorectal cancer Carcinogenesis 2015 Mar;36(3):346-54
- Kawashima T, Hirose K, Satoh T, Kaneko A, Ikeda Y, Kaziro Y, Nosaka T, Kitamura T. MgcRacGAP is involved in the control of growth and differentiation of hematopoietic cells Blood 2000 Sep 15;96(6):2116-24
- Ke HL, Ke RH, Li ST, Li B, Lu HT, Wang XQ. Expression of RACGAP1 in high grade meningiomas: a potential role in cancer progression J Neurooncol 2013 Jun;113(2):327-32
- Kumar V, Fausto N, Abbas A (editors). Robbins Cotran Pathologic Basis of Disease (7th ed.) 2003, Saunders. pp. 914-917
- Lamarche N, Hall A. GAPs for rho-related GTPases Trends Genet 1994 Dec;10(12):436-40
- Lee JS, Kamijo K, Ohara N, Kitamura T, Miki T. MgcRacGAP regulates cortical activity through RhoA during cytokinesis Exp Cell Res 2004 Feb 15;293(2):275-82
- Lowery DM, Lim D, Yaffe MB. Structure and function of Polo-like kinases Oncogene 2005 Jan 10;24(2):248-59
- Matsuura A, Lee HH. Crystal structure of GTPase-activating domain from human MgcRacGAP Biochem Biophys Res Commun 2013 Jun 7;435(3):367-72
- Mishima M, Kaitna S, Glotzer M. Central spindle assembly and cytokinesis require a kinesin-like protein/RhoGAP complex with microtubule bundling activity Dev Cell 2002 Jan;2(1):41-54
- Naud N, Touré A, Liu J, Pineau C, Morin L, Dorseuil O, Escalier D, Chardin P, Gacon G. Rho family GTPase Rnd2 interacts and co-localizes with MgcRacGAP in male germ cells Biochem J 2003 May 15;372(Pt 1):105-12
- Niia F, Tatsumoto T, Lee KS, Miki T. Phosphorylation of the cytokinesis regulator ECT2 at G2/M phase stimulates association of the mitotic kinase Plk1 and accumulation of GTP-bound RhoA Oncogene 2006 Feb 9;25(6):827-37
- Petronczki M, Glotzer M, Kraut N, Peters JM. Polo-like kinase 1 triggers the initiation of cytokinesis in human cells by promoting recruitment of the RhoGEF Ect2 to the central spindle Dev Cell 2007 May;12(5):713-25
- Saigusa S, Tanaka K, Mohri Y, Ohi M, Shimura T, Kitajima T, Kondo S, Okugawa Y, Toiyama Y, Inoue Y, Kusunoki M. Clinical significance of RacGAP1 expression at the invasive front of gastric cancer Gastric Cancer 2015 Jan;18(1):84-92
- Santamaria A, Neef R, Eberspächer U, Eis K, Husemann M, Mumberg D, Prechtel S, Schulze V, Siemeister G, Wortmann L, Barr FA, Nigg EA. Use of the novel Plk1 inhibitor ZK-thiazolidinone to elucidate functions of Plk1 in early and late stages of mitosis Mol Biol Cell 2007 Oct;18(10):4024-36
- Touré A, Dorseuil O, Morin L, Timmons P, Jégou B, Reibel L, Gacon G. MgcRacGAP, a new human GTPase-activating protein for Rac and Cdc42 similar to Drosophila rotundRacGAP gene product, is expressed in male germ cells J Biol Chem 1998 Mar 13;273(11):6019-23

Toure A, Morin L, Pineau C, Becq F, Dorseuil O, Gacon G. Tat1, a novel sulfate transporter specifically expressed in human male germ cells and potentially linked to rhoGTPase signaling *J Biol Chem* 2001 Jun 8;276(23):20309-15

Wang SM, Ooi LL, Hui KM. Upregulation of Rac GTPase-activating protein 1 is significantly associated with the early recurrence of human hepatocellular carcinoma *Clin Cancer Res* 2011 Sep 15;17(18):6040-51

Wolfe BA, Takaki T, Petronczki M, Glotzer M. Polo-like kinase 1 directs assembly of the HsCyk-4 RhoGAP/Ect2 RhoGEF complex to initiate cleavage furrow formation *PLoS Biol* 2009 May 5;7(5):e1000110

Yüce O, Piekny A, Glotzer M. An ECT2-centralspindlin complex regulates the localization and function of RhoA *J Cell Biol* 2005 Aug 15;170(4):571-82

Yu SJ, Kim YJ. Hepatitis B viral load affects prognosis of hepatocellular carcinoma *World J Gastroenterol* 2014 Sep 14;20(34):12039-44

Zhang P, Bai H, Fu C, Chen F, Zeng P, Wu C, Ye Q, Dong C, Song Y, Song E. RacGAP1-driven focal adhesion formation promotes melanoma transendothelial migration through mediating adherens junction disassembly *Biochem Biophys Res Commun* 2015 Mar 27;459(1):1-9

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