DOWNLOAD PDF DISEASES INVOLVING THE GOLGI CALCIUM PUMP J. VANOEVELEN . [ET AL.]

Chapter 1 : - NLM Catalog Result

CHAPTER 14 DISEASES INVOLVING THE GOLGI CALCIUM PUMP J. VANOEVELEN, L. DODE, L. RAEYMAEKERS, F. WUYTACK AND L. MISSIAEN Laboratory of Physiology, KULeuven Campus Gasthuisberg O&N1, Herestraat

Four-vessel occlusion rats were used as animal models of cerebral ischemia. This variation was similar to the alteration of calcium in separated Golgi vesicles. The key target of neuroprotection after the onset of ischemic stroke: Neural Regen Res ; ZPH designed this study. LHL wrote the paper. All authors approved the final version of the paper. Introduction The function of the Golgi apparatus in secretion and protein modification has long been known and is one of the earliest discoveries in the cell. These functions are important for biochemical reactions in neuronal cells, but it was not known whether the Golgi apparatus also played a role in cell signaling. In , Krino first elucidated the phenomenon of delayed neuronal death following ischemia in the gerbil hippocampus. Cell death at the site of ischemia and its penumbra is recognized as the predominant cause of cerebral IRI Ferrer and Planas, It is well known that calcium overload is the vital signal for cell death and is also the prominent event in cerebral IRI Uematsu et al. As previously reviewed Li et al. These include calcium channels, calcium pumps, calcium binding proteins and other calcium regulating proteins. Based on these determinations, we can accurately explain calcium alterations in Golgi apparatus stress related to cerebral IRI, which gives further insight into possible treatments for ischemic stroke. Rats were divided into four groups: Five subgroups [3 hours R3 h , 6 hours R6 h , 24 hours R24 h , 3 days R3 d and 7 days R7 d] were set in the reperfusion group Li et al. Rats in the control group were fed at room temperature and had no treatments. Rats of the sham operation group were only subjected to the operation so as to expose the vertebral arteries and carotid arteries. Rats in the ischemia group were executed to occlude the carotid arteries for 20 minutes. The blood fluid of carotid arteries was recovered in the reperfusion sub groups at different time points. Establishment of animal models Rats from ischemia group and reperfusion groups were used to establish animal models according to the method of four-vessel occlusion supplied by Pulsineli et al. Subsequently, the first operation was carried out to assure complete occlusion of the vertebral arteries by electrocauterization through the alar foramina of the first cervical vertebra. Twenty-four hours later, the second operation was taken to control the occlusion and reperfusion of the parallel common carotid arteries. Animals from the ischemia group received four-vessel occlusion and were sacrificed immediately. Animals from the reperfusion groups received four-vessel occlusion, then reperfusion for 3, 6, 24 hours and 3, 7 days, and then were sacrificed as soon as possible. Whole brains of these rats in various groups and subgroups were peeled out immediately. The temporal lobe and hippocampus were embedded in wax, sliced into coronal sections, and then subjected to immuno-detection. Direct immunostaining procedures in separated neuronal cells were performed as previously described in Short Protocol of Molecular Biology Ausubel et al. Sections were incubated with direct fluorescent monoclonal antibody of SPCA1 in 10 mM phosphate buffer mouse monoclonal, 1: The total extracted proteins were resolved by sodium dodecyl sulfate-polyacrylamide gel electrophoresis. After being washed with 0. To reduce differences among animals, sample loading on sodium dodecyl sulfate-polyacrylamide gel electrophoresis, and variability due to staining detection, western blot assay was performed for each reperfusion time point per animal in triplicate. The results were documented by gel imaging system Tannon Inc. The nerve cell was prepared according to the method of Dildy et al. The brain was cut into tiny pieces and digested in 0. The upper supernatant was discarded and the yellow precipitate in the remaining upper layer was mildly retrieved by a glass stick and resuspended in 5-mL dextran-homogenized medium. The suspension was set to mL 1. Golgi vesicles were present in the upper layer of the gradient suspension. The real-time fluorescence F was detected at nm. The maximum fluorescence F max was determined after adding Triton X and the minimum fluorescence F min was measured after appending 20 mM ethylenebis oxyethylenenitrilo tetraacetic acid, followed by the manipulation of Triton. In addition, before formal determination, fluorescent wave scanning was taken to verify whether the maximum

excited wavelength could reach nm. Calcium concentration was calculated by the following formula: The least significance difference test was used for post hoc testing. Although calcium overload remained to the time of reperfusion for 7 days, there was an obvious alleviation in late reperfusion [Figure 1] A. In the late phase of reperfusion, from 6 to 24 hours, the calcium content of Golgi vesicles was elevated to a much higher level than control or sham operation group. Over reperfusion for 7 days, it recovered to the original state [Figure 1] B and [Figure 2]. Figure 2 Data of fluorescent density of SPCA1 primary antibody during ischemia and reperfusion as detected by fluorescence immunohistochemistry. When ischemia and early reperfusion occurred, the fluorescent density of SPCA1 primary antibodies was much weaker than the control, in particular, the density was at its lowest point during reperfusion [Figure 4] C and D. Although the level of fluorescent density increased rapidly, it recovered to the normal level slowly [Figure 4] F-H. No fluorescent signals are detected. B, C SPCA1 expression in the brain, particularly in the cortex and hippocampus neurons, at 6 hours after reperfusion. SPCA1-immunoreactive cells show blue fluorescence. Click here to view Figure 4 Fluorescent immunoreactivity of SPCA1 during ischemia and reperfusion immunofluorescence, fluorescence microscope. SPCA1 expression was analyzed by fluorescence in situ immunohistochemistry. Neuronal cells expressing SPCA1 are marked by blue fluorescence. Protein expression of SPCA1 manifested a low-high-low tendency during cerebral ischemia and reperfusion [Figure 5]. The lowest state occurred in the phase of reperfusion and the highest state occurred in the adjacent 6 hours. In I and R3 h, an obvious decline occurred. In R6 h, a dramatic increase appeared. After R24 h, the expression decreased very slowly.

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Chapter 2 : Golgi apparatus - Ganfyd

Abstract. Secretory-pathway C a 2+-transport ATPases (SPCA) provide the Golgi apparatus with Ca 2+ and Mn 2+ needed for the normal functioning of this organelle. Loss of one functional copy of the human SPCA1 gene (ATP2C1) causes Hailey-Hailey disease, a rare skin disorder characterized by recurrent blisters and erosions in the flexural areas.

Find articles by Desma M. Faddy Find articles by Helen M. Kenny Find articles by Paraic A. Roberts-Thomson Find articles by Sarah J. Monteith Find articles by Gregory R. This article has been cited by other articles in PMC. Abstract Calcium signaling is a key regulator of pathways important in tumor progression, such as proliferation and apoptosis. Most studies assessing altered calcium homeostasis in cancer cells have focused on alterations mediated through changes in cytoplasmic free calcium levels. Here, we show that basal-like breast cancers are characterized by an alteration in the secretory pathway calcium ATPase 1 SPCA1 , a calcium pump localized to the Golgi. Inhibition of SPCA1 in MDA-MB cells produced pronounced changes in cell proliferation and morphology in three-dimensional culture, without alterations in sensitivity to endoplasmic reticulum stress induction or changes in global calcium signaling. Instead, the effects of SPCA1 inhibition in MDA-MB cells reside in altered regulation of calcium-dependent enzymes located in the secretory pathway, such as proprotein convertases. These studies identify for the first time a calcium transporter associated with the basal-like breast cancer subtype. The pronounced effects of SPCA1 inhibition on the processing of IGF1R in MDA-MB cells independent of alterations in global calcium signaling also demonstrate that some calcium transporters can regulate the processing of proteins important in tumor progression without major alterations in cytosolic calcium signaling. Inhibitors of SPCA1 may offer an alternative strategy to direct inhibitors of IGF1R and attenuate the processing of other proprotein convertase substrates important in basal breast cancers. Breast Cancer, Calcium, Calcium ATPase, Calcium Transport, Golgi Introduction Calcium levels within a cell control a variety of cellular processes relevant to tumorigenesis such as proliferation, migration, and apoptosis 1 , 2. Although cellular calcium homeostasis is precisely controlled, there is an emerging appreciation that it is remodeled during cancer with downstream consequences on cellular function 3. Deregulation of calcium homeostasis may arise via changes in calcium-transporting proteins such as channels and pumps whose expression can be up- or down-regulated in cancers 2. Within the Golgi reside calcium-regulated enzymes, such as the proprotein convertases that have been described as master switches in tumorigenesis 6 , and whose substrates include insulin-like growth factor receptor IGF1R , 4 a protein linked epidemiologically, clinically, and experimentally to breast cancer 7. The identification of regulators of proprotein convertases, such as regulators of Golgi luminal calcium levels, would be important therapeutically as they may offer an alternative means to globally control these pathways. One regulator of Golgi luminal calcium levels is the secretory pathway calcium ATPase 1 SPCA1 , an active transporter of calcium into the secretory pathway 8 , â€” Hailey Hailey disease is characterized only by symptoms involving stratified squamous epithelium that arise due to loss of cell to cell adhesion acantholysis Within the cell, SPCA1 inhibition appears to alter the cell surface levels of exogenously expressed proteins suggesting a functional consequence of SPCA1 in protein trafficking Physiologically, SPCAs appear to have important roles in the mammary gland and likely contribute to the secretion of calcium into milk 16 , â€” Although the Golgi apparatus does not appear to be a major site of calcium storage, in most cells it is suggested to have a more prominent role in specialized secretory cells, such as those of the mammary gland Despite this important physiological role, there is a paucity of data regarding the pathophysiological consequences of SPCA1 in the breast. There have been very few studies addressing the consequences of SPCA1 inhibition on cytosolic calcium changes produced by cell stimuli. SPCA1 silencing in HeLa cells has only relatively minor effects on cytosolic calcium responses produced by histamine 20 ; however, in spermatozoa, SPCA1 appears to play a major role in the regulation of cytosolic calcium transients Cytosolic calcium levels have demonstrated effects on cellular functions, including proliferation, gene expression, and

contractility 1. However, the consequences of altered Golgi luminal calcium levels in mammalian cells are relatively unexplored. The Golgi is responsible for the post-translational modification of proteins prior to secretion, for example glycosylation of proteins Calcium levels within the Golgi lumen are likely to regulate other calcium-dependent enzymes, including the proprotein convertases. Indeed, in the yeast Yarrowia lipolytica, there is one report of YlPMR1 secretory pathway calcium ATPase homolog regulating the processing of the proprotein convertase substrate alkaline extracellular protease Proprotein convertase substrates have a significant role in cancer 6 , and as yet there are no studies investigating the link between SPCA1, calcium-regulated proprotein convertases, and cancer. Insulin-like growth factor receptor IGF1R is a proprotein convertase substrate important for mammary gland development and milk production 26 , Expression of IGF1R is increased in breast cancers and is associated with cancer initiation, evasion of apoptosis, motility, proliferation, and resistance to cancer therapy 7 , Suggesting a possible association with breast cancers of the poorest prognosis, IGF1R expression is significantly higher in those tumors from women with BRCA1 mutations compared with those tumors from women without BRCA1 mutations Mice overexpressing IGF1R and constitutively active Kras develop mammary gland tumors of the basal-like subtype Basal-like breast tumors are classified as such on the basis of their gene expression profile Often, basal-like breast tumors have alterations in BRCA1, and they are associated with a poor outcome and a paucity of therapeutic options Here, we studied the possible association of SPCA1 with different breast cancer subtypes and the functional consequences of loss of SPCA1 in a breast cancer cell line. Our results identify SPCA1 as the first calcium transporter associated with basal-like breast cancers. SPCA1 modulation and regulation of Golgi calcium transport may provide a unique opportunity for therapeutic targeting of this important pathway in breast cancer. This work redefines the potential role of calcium transporters in tumorigenesis by identifying critical roles in cancer-relevant pathways that occur independently of changes in global calcium signaling. Cell lines were routinely tested for mycoplasma contamination and monitored for morphological characteristics. Microarray Analysis SPCA1 expression was assessed in breast cancer clinical samples and cell lines in a dataset obtained from Rosetta Inpharmatics consisting of the microarray profiles of primary breast tumors 33 and an Affymetrix microarray dataset of breast cancer cell lines classified as basal-like or luminal as described previously 34 , In both cases, the median level of SPCA1 was compared between the sample groups. RNA was isolated at the time points depicted in the figures, and protein was isolated at 72 h post-siRNA treatment. The secondary antibody was goat anti-rabbit HRP conjugate 1: Line analysis was performed using the Quantity One Analysis software. Cells were transfected, and h post-transfection cells were treated with increasing concentrations of tunicamycin for 48 h. Viability was assessed using the CellTiter 96 Aqueous One Cell proliferation assay Promega as described previously Cells were allowed to attach for 6 h prior to treatment with siRNA and then cultured for 96 h with feeding every 2 days. At 24 and 96 h post-transfection, cells were separated from MatrigelTM by incubation in Dispase Cells were equilibrated for 15 min to room temperature. Statistical Analysis Unless otherwise stated, data analyses were performed using GraphPad Prism version 4. SPCA1 levels were significantly elevated in clinical samples classified molecularly as belonging to the basal subtype compared with all other subtypes, ERRB2, luminal A, luminal B, and normal Fig. The association between SPCA1 and the basal subtype was further strengthened by the significant elevation of SPCA1 levels in breast cancer cell lines classified as basal-like compared with those classified as luminal Fig. SPCA1 levels were also elevated with increasing tumor grade Fig.

Chapter 3 : Answer: Can you identify this condition? - Europe PMC Article - Europe PMC

Diseases involving the Golgi calcium pump Since the transcriptional activation and regulation of the ATP2C2 promoter is so far unknown, the restricted tissue and cellular expression pattern of ATP2C2 cannot be explained at the moment.

These authors contributed equally to this work. ZPH designed this study. LHL wrote the paper. All authors approved the final version of the paper. Accepted Mar This article has been cited by other articles in PMC. Four-vessel occlusion rats were used as animal models of cerebral ischemia. This variation was similar to the alteration of calcium in separated Golgi vesicles. These functions are important for biochemical reactions in neuronal cells, but it was not known whether the Golgi apparatus also played a role in cell signaling. In , Krino first elucidated the phenomenon of delayed neuronal death following ischemia in the gerbil hippocampus. Cell death at the site of ischemia and its penumbra is recognized as the predominant cause of cerebral IRI Ferrer and Planas, It is well known that calcium overload is the vital signal for cell death and is also the prominent event in cerebral IRI Uematsu et al. As previously reviewed Li et al. These include calcium channels, calcium pumps, calcium binding proteins and other calcium regulating proteins. Based on these determinations, we can accurately explain calcium alterations in Golgi apparatus stress related to cerebral IRI, which gives further insight into possible treatments for ischemic stroke. Rats were divided into four groups: Five subgroups [3 hours R3 h , 6 hours R6 h , 24 hours R24 h , 3 days R3 d and 7 days R7 d] were set in the reperfusion group Li et al. Rats in the control group were fed at room temperature and had no treatments. Rats of the sham operation group were only subjected to the operation so as to expose the vertebral arteries and carotid arteries. Rats in the ischemia group were executed to occlude the carotid arteries for 20 minutes. The blood fluid of carotid arteries was recovered in the reperfusion sub groups at different time points. Establishment of animal models Rats from ischemia group and reperfusion groups were used to establish animal models according to the method of four-vessel occlusion supplied by Pulsineli et al. Subsequently, the first operation was carried out to assure complete occlusion of the vertebral arteries by electrocauterization through the alar foramina of the first cervical vertebra. Twenty-four hours later, the second operation was taken to control the occlusion and reperfusion of the parallel common carotid arteries. Animals from the ischemia group received four-vessel occlusion and were sacrificed immediately. Animals from the reperfusion groups received four-vessel occlusion, then reperfusion for 3, 6, 24 hours and 3, 7 days, and then were sacrificed as soon as possible. Whole brains of these rats in various groups and subgroups were peeled out immediately. The temporal lobe and hippocampus were embedded in wax, sliced into coronal sections, and then subjected to immuno-detection. Direct immunostaining procedures in separated neuronal cells were performed as previously described in Short Protocol of Molecular Biology Ausubel et al. Sections were incubated with direct fluorescent monoclonal antibody of SPCA1 in 10 mM phosphate buffer mouse monoclonal, 1: The total extracted proteins were resolved by sodium dodecyl sulfate-polyacrylamide gel electrophoresis. After being washed with 0. To reduce differences among animals, sample loading on sodium dodecyl sulfate-polyacrylamide gel electrophoresis, and variability due to staining detection, western blot assay was performed for each reperfusion time point per animal in triplicate. The results were documented by gel imaging system Tannon Inc. The nerve cell was prepared according to the method of Dildy et al. The brain was cut into tiny pieces and digested in 0. The upper supernatant was discarded and the yellow precipitate in the remaining upper layer was mildly retrieved by a glass stick and resuspended in 5-mL dextran-homogenized medium. The suspension was set to mL 1. Golgi vesicles were present in the upper layer of the gradient suspension. The real-time fluorescence F was detected at nm. The maximum fluorescence Fmax was determined after adding Triton X and the minimum fluorescence Fmin was measured after appending 20 mM ethylenebis oxyethylenenitrilo tetraacetic acid, followed by the manipulation of Triton. In addition, before formal determination, fluorescent wave scanning was taken to verify whether the maximum excited wavelength could reach nm. Calcium concentration was calculated by the following formula: The least

significance difference test was used for post hoc testing. Although calcium overload remained to the time of reperfusion for 7 days, there was an obvious alleviation in late reperfusion Figure 1A.

Chapter 4 : Diseases involving the Golgi calcium pump | leo raeymaekers - blog.quintoapp.com

Diseases involving the Golgi calcium pump Since the transcriptional activation and regulation of the ATP2C2 promoter is so far unknown, the restricted tissue and cellular expression pattern of.

Previous Section Next Section Introduction Calcium levels within a cell control a variety of cellular processes relevant to tumorigenesis such as proliferation, migration, and apoptosis 1 , 2. Although cellular calcium homeostasis is precisely controlled, there is an emerging appreciation that it is remodeled during cancer with downstream consequences on cellular function 3. Deregulation of calcium homeostasis may arise via changes in calcium-transporting proteins such as channels and pumps whose expression can be up- or down-regulated in cancers 2. Within the Golgi reside calcium-regulated enzymes, such as the proprotein convertases that have been described as master switches in tumorigenesis 6 , and whose substrates include insulin-like growth factor receptor IGF1R , 4 a protein linked epidemiologically, clinically, and experimentally to breast cancer 7. The identification of regulators of proprotein convertases, such as regulators of Golgi luminal calcium levels, would be important therapeutically as they may offer an alternative means to globally control these pathways. One regulator of Golgi luminal calcium levels is the secretory pathway calcium ATPase 1 SPCA1 , an active transporter of calcium into the secretory pathway 8 , â€” , Hailey Hailey disease is characterized only by symptoms involving stratified squamous epithelium that arise due to loss of cell to cell adhesion acantholysis Within the cell, SPCA1 inhibition appears to alter the cell surface levels of exogenously expressed proteins suggesting a functional consequence of SPCA1 in protein trafficking Physiologically, SPCAs appear to have important roles in the mammary gland and likely contribute to the secretion of calcium into milk 16 , â€” , Although the Golgi apparatus does not appear to be a major site of calcium storage, in most cells it is suggested to have a more prominent role in specialized secretory cells, such as those of the mammary gland Despite this important physiological role, there is a paucity of data regarding the pathophysiological consequences of SPCA1 in the breast. There have been very few studies addressing the consequences of SPCA1 inhibition on cytosolic calcium changes produced by cell stimuli. SPCA1 silencing in HeLa cells has only relatively minor effects on cytosolic calcium responses produced by histamine 20 ; however, in spermatozoa, SPCA1 appears to play a major role in the regulation of cytosolic calcium transients Cytosolic calcium levels have demonstrated effects on cellular functions, including proliferation, gene expression, and contractility 1. However, the consequences of altered Golgi luminal calcium levels in mammalian cells are relatively unexplored. The Golgi is responsible for the post-translational modification of proteins prior to secretion, for example glycosylation of proteins Calcium levels within the Golgi lumen are likely to regulate other calcium-dependent enzymes, including the proprotein convertases. Indeed, in the yeast Yarrowia lipolytica, there is one report of YlPMR1 secretory pathway calcium ATPase homolog regulating the processing of the proprotein convertase substrate alkaline extracellular protease Proprotein convertase substrates have a significant role in cancer 6 , and as yet there are no studies investigating the link between SPCA1, calcium-regulated proprotein convertases, and cancer. Insulin-like growth factor receptor IGF1R is a proprotein convertase substrate important for mammary gland development and milk production 26 , Expression of IGF1R is increased in breast cancers and is associated with cancer initiation, evasion of apoptosis, motility, proliferation, and resistance to cancer therapy 7 , Suggesting a possible association with breast cancers of the poorest prognosis, IGF1R expression is significantly higher in those tumors from women with BRCA1 mutations compared with those tumors from women without BRCA1 mutations Mice overexpressing IGF1R and constitutively active Kras develop mammary gland tumors of the basal-like subtype Basal-like breast tumors are classified as such on the basis of their gene expression profile Often, basal-like breast tumors have alterations in BRCA1, and they are associated with a poor outcome and a paucity of therapeutic options Here, we studied the possible association of SPCA1 with different breast cancer subtypes and the functional consequences of loss of SPCA1 in a breast cancer cell line. Our results identify

SPCA1 as the first calcium transporter associated with basal-like breast cancers. SPCA1 modulation and regulation of Golgi calcium transport may provide a unique opportunity for therapeutic targeting of this important pathway in breast cancer. This work redefines the potential role of calcium transporters in tumorigenesis by identifying critical roles in cancer-relevant pathways that occur independently of changes in global calcium signaling. Cell lines were routinely tested for mycoplasma contamination and monitored for morphological characteristics. Microarray Analysis SPCA1 expression was assessed in breast cancer clinical samples and cell lines in a dataset obtained from Rosetta Inpharmatics consisting of the microarray profiles of primary breast tumors 33 and an Affymetrix microarray dataset of breast cancer cell lines classified as basal-like or luminal as described previously 34 , In both cases, the median level of SPCA1 was compared between the sample groups. RNA was isolated at the time points depicted in the figures, and protein was isolated at 72 h post-siRNA treatment. The secondary antibody was goat anti-rabbit HRP conjugate 1: Line analysis was performed using the Quantity One Analysis software. Cells were transfected, and h post-transfection cells were treated with increasing concentrations of tunicamycin for 48 h. Viability was assessed using the CellTiter 96 Aqueous One Cell proliferation assay Promega as described previously Cells were allowed to attach for 6 h prior to treatment with siRNA and then cultured for 96 h with feeding every 2 days. At 24 and 96 h post-transfection, cells were separated from MatrigelTM by incubation in Dispase Cells were equilibrated for 15 min to room temperature. Statistical Analysis Unless otherwise stated, data analyses were performed using GraphPad Prism version 4. SPCA1 levels were significantly elevated in clinical samples classified molecularly as belonging to the basal subtype compared with all other subtypes, ERRB2, luminal A, luminal B, and normal Fig. The association between SPCA1 and the basal subtype was further strengthened by the significant elevation of SPCA1 levels in breast cancer cell lines classified as basal-like compared with those classified as luminal Fig. SPCA1 levels were also elevated with increasing tumor grade Fig.

Chapter 5 : Papers with the keyword SPCA1 (Page 4) | Read by QxMD

J. Vanoevelen, L. Raeymaekers, J.B. Parys, et blog.quintoapp.comol trisphosphate producing agonists do not mobilize the thapsigargin-insensitive part of the endoplasmic-reticulum and Golgi Ca 2+ store Cell Calcium, 35 (), pp. -

Here, we will review the properties and functional role of the SPCAs. SPCA1 is expressed in all cells Vanoevelen et al. The P2 -subfamily Figure 1. There are no introns in the coding region, like for most genes in S. AAA comprises of amino acids and has a molecular weight of kDa. Its transcripts are alternatively spliced and, like it is often the case in C. CAC with amino acids. AC contains only 3 intervening sequences separating the 4 exons indicating that several introns have been lost during evolution. The first mammalian member of the SPCA family was cloned from rat but could not be characterized functionally at that time Gunteski-Hamblin et al. ATP2C1 is localized on the q arm of chromosome 3 at position The gene consists of 28 exons. YY1 and Sp1 factors, when overexpressed, transcriptionally activate the ATP2C1 promoter in normal human keratinocytes. Gene structure of ATP2C1 and alternative splicing. Shows the structure of the ATP2C1 gene. Exons are represented by boxes, with wide boxes depicting the open reading frame. The thin horizontal line represents the position of introns. Diagonal lines illustrate the splicing patterns generating isoforms ATP2C1a-d. Structure of the different ATP2C1 splice variants: Isoform a results from the splicing of exon 26 to exon 27; b. Isoform b is the product of cryptic splicing at D1 ; c. Exon 26 splices directly to exon 28 to produce isoform c; d. Activation of splicing at cryptic site D2 produces isoform d contains exon 28 and the partial portions of exon 27 up to the D1 splice-donor site. Transcript ATP2C1d contains exon 28 and the partial portions of exon 27 up to the D2 splice-donor site. ATP2C2 is found at cytogenetic position q Corresponding SPCA2 genes are also present in birds, amphibians and mammals, but not in fish. ATP2C2 consists of 27 exons. Diseases involving the Golgi calcium pump Since the transcriptional activation and regulation of the ATP2C2 promoter is so far unknown, the restricted tissue and cellular expression pattern of ATP2C2 cannot be explained at the moment. The phosphorylated residues are Asp S. The latter 4 amino-acid-long loops must move upwards to contact the catalytic site during dephosphorylation of the phosphoenzyme intermediate E 2 -P. This motif modulates ion transport in S. This function in the yeast SPCA depends on critical packing interactions at the cytoplasmic interface between the side chains of Gln in M6 and Val in M4 Mandal et al. It contains a dileucine L L motif, which may be a retrieval signal from the plasma membrane Xiang et al. These two C-terminal motifs could be involved in localization and shuttling of SPCA2 between the stacks of the trans- Golgi network, vesicles derived from this network, and even the plasma membrane. Results on the relative expression level in various tissues are still conflicting. Expression levels, both at the mRNA and at the protein level, in rat brain and testis were much higher than in other tissues like lung and liver Wootton et al. Such relatively high expression in brain and testis was not observed at the mRNA level in humans Vanoevelen et al. The cell-type dependent expression of SPCA1 in various animal species thus deserves further study. SPCA1 is highly expressed in human epidermal keratinocytes Hu et al. The epidermis consists of undifferentiated keratinocytes in the basal layer that develop into the differentiated cells of the more superficial spinous, granular and cornified layers Eckert et al. They correlated the preferential localization of SPCA1 in the undifferentiated cells of the basal layer with the induction of differentiation markers upon suppression of SPCA1 expression. It should however be remarked that these results on the preferential localization of SPCA1 in the undifferentiated cells of the basal layer contrast with the immunohistochemical data of Porgpermdee et al. Its mRNA is also found to be enriched in trachea, salivary gland, thyroid, keratinocytes, prostate, mammary gland, brain and testis Vanoevelen et al. SPCA2 levels in the mammary gland increase during lactation Dmitriev et al. Together with the observation that SPCA2 is mainly expressed in the mucus-secreting goblet cells in human colon Vanoevelen et al. These findings on overexpressed pumps were later confirmed for the endogenous SPCA1 in cultured cells and tissues. Intruigingly, the different splice variants of the Drosophila orthologue SPoCk all reside in a different

subcellular compartment. This contrasts with the situation in human where, with the notable exception of SPCA1c which is an inactive isoform retained in the endoplasmic reticulum for degradation, SPCA1a, -b, -c all are sent to the Golgi apparatus Dode et al. The cholesterol concentration is low in the endoplasmic reticulum and increases in the distal direction of the secretory pathway. Membranes of the trans-Golgi compartments are therefore thicker and more rigid. A localization in the trans-Golgi implicates that SPCA1 may also appear in the more distal parts of the secretory pathway. SPCA1 in human spermatozoa is localized in an area behind the nucleus which extends into the midpiece Harper et al. There are only few reports on the subcellular localization of SPCA2 and no clear picture emerges from them. SPCA2 in human goblet cells colocalizes with Golgi markers in a compact structure near the apical pole of the nucleus Vanoevelen et al. These observations contrast with those on cultured mouse hippocampal neurons, which show a punctate distribution of SPCA2 in the cell body and in the dendrites, with a partial colocalization with the trans-Golgi marker TGN38 Xiang et al. SPCA2 in hippocampal neurons may therefore be localized in post-Golgi segments of the secretory pathway. This represents the first evidence for functional differences between the human SPCA1 isoforms. The physiological significance of this finding, and, implicitly, of the SPCA1 isoform diversity remains however to be further elucidated. Since this isoform has a truncated transmembrane segment M10, it could lead to an improperly folded protein that might be sensitive to enhanced cellular degradation. SPCA1c cannot be phosphorylated, and, therefore, cannot couple the energy from the energy-donating ATP molecule to the vectorial ion transport. SPCA pumps have some unique catalytic adaptations, optimizing them to function in the Golgi and more down-stream compartments. Human spermatozoa may represent a notable exception. The disease was first described by the Hailey dermatologist brothers Hailey and Hailey, The symptoms typically arise after puberty or after the third or fourth decade. It is a blistering skin disease with vesicles and itchy erosions. Pain and an unpleasant smell become dominant symptoms as fissures and maceration develop. The distribution of cutaneous lesions is usually symmetrical. They mainly occur in the bodily folds, particularly the groin and axillar regions. Mucosal surfaces are rarely involved. The disease has a fluctuating course with remissions and exacerbations triggered by friction, sweating, heat, stress, infection, ultraviolet radiation or tissue damage like in allergic contact dermatitis. Many patients have fingernails with asymptomatic longitudinal white lines. The skin lesions may develop into squamous cell carcinomas Chun et al. Sporadic diseases associated with HHD are affective disorder Korner et al. HHD patients also display a higher frequency of contact allergies Reitamo et al. Histologically, the disease is characterized by a loss of adhesion between suprabasal keratinocytes acantholysis and abnormal keratinisation dyskeratosis of the epidermis. In HHD, acantholysis is the most prominent histological feature while in the clinically related Darier disease OMIM , dyskeratosis is much more apparent. Ultrastructural analysis of acantholytic cells reveals perinuclear aggregates of keratin filaments that have retracted from desmosomes Harada et al. Close to mutations are known Van Baelen et al. There are no genotype-phenotype Vanoevelen et al. They occur in a mosaic form along the Blaschko lines. These unilateral patterned skin lesions corre- spond to clones of keratinocytes descending during embryogenesis from a common precursor that as a result of a somatic mutation had lost the only remaining wild-type ATP2C1 allele. The exacerbated areas are therefore superimposed on the ordinary symmetrical germ-line transmitted HHD phenotype. More than half of the investigated mutants resulted in low levels of protein expression despite normal mRNA levels and a correct targeting of the pump to the Golgi compartments e. Other mutants resulted in a lack of ion transport. Expression of the latter mutant in yeast restored the cellular phenotype of a yeast strain lacking PMR1 Ton and Rao, It is therefore unclear how this mutant leads to the HHD defect. This mutation may represent a benign polymorphism. The actual defect might be associated with the non-coding parts of ATP2C1. Physiopathology of HHD The precise link between altered expression or functionality of SPCA1 and the development of skin lesions has not yet been established. Secondly, we do not know whether the main defect in ion homeostasis is located in the lumen of the secretory pathway or in the cytosol. If this were the case, the similarities between Darier disease and HHD would be easily explained. In contrast, Porgpermdee et al.

Although HHD patients with affective disorder have been described Korner et al. This is intuitively not expected for a mutated protein that is ubiquitously expressed. Additional factors might also be involved, like, amongst others, the disturbed glyco- sylation state of components involved in keratinocyte-specific cell-to-cell contacts, or modifications of specific keratins. Alternatively, compensatory mechanisms that protect non-cutaneous tissues may be lacking in keratinocytes. Indeed, the skin of HHD patients is typically symptom- free during the first decades of life and even thereafter the symptoms are only seen in certain areas of the skin in response to sweating, local warming or friction. The symptoms in HHD patients can be elicited in a matter of minutes by locally rubbing the skin. This effect was also confirmed in vitro. Functional knockdown of ATP2C1 in human keratinocytes renders them defective in post-translational processing and Vanoevelen et al.

Chapter 6 : SPCA manganese - PubMed Result

The plasma membrane calcium ATPase and disease. Tempel, B.L (et al.) Pages Diseases involving the Golgi calcium pump. VANOEVELEN, J. (et al.) Pages

Chapter 7 : Golgi Ion Pumps | Hailey-Hailey Disease Society

There is an increasing number of scientific papers dealing with our understanding of Golgi Ion Pumps. Hailey-Hailey Disease appears to be a debilitating manifestation of an underfunctioning Calcium/Manganese ion pump, SPCA1 (ATP2C1).