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Chapter 1 : ADD YOUR PAGE TITLE

If there has been a failure of due process in this, it begins with the apparent absence of any clear process of respecting, recording and processing complaints by students of the school, or even.

Research Article Metabolism Free access Fong,1 and Stephen G. Young, or Anne P. Fong ; sgyoung mednet. Young ; abeigneux mednet. Find articles by He, C. Find articles by Hu, X. Find articles by Jung, R. Find articles by Larsson, M. Find articles by Tu, Y. Find articles by Duarte-Vogel, S. Find articles by Kim, P. Find articles by Sandoval, N. Find articles by Price, T. Find articles by Allan, C. Find articles by Raney, B. Find articles by Jiang, H. Find articles by Bensadoun, A. Find articles by Walzem, R. Find articles by Kuo, R. Find articles by Beigneux, A. Find articles by Fong, L. Find articles by Young, S. August 9, ; Accepted: September 11, Abstract In mammals, GPIHBP1 is absolutely essential for transporting lipoprotein lipase LPL to the lumen of capillaries, where it hydrolyzes the triglycerides in triglyceride-rich lipoproteins. In all lower vertebrate species e. The obvious question is whether the LPL in lower vertebrates is able to reach the capillary lumen. Using purified antibodies against chicken LPL, we showed that LPL is present on capillary endothelial cells of chicken heart and adipose tissue, colocalizing with von Willebrand factor. When the antibodies against chicken LPL were injected intravenously into chickens, they bound to LPL on the luminal surface of capillaries in heart and adipose tissue. LPL was released rapidly from chicken hearts with an infusion of heparin, consistent with LPL being located inside blood vessels. GPIHBP1 and LPL are equally important for intravascular lipolysis; a deficiency of either protein markedly impairs intravascular triglyceride hydrolysis and leads to severe hypertriglyceridemia chylomicronemia 5 â€” All Ly6 proteins have at least one approximately 80â€”amino acid Ly6 domain with 8 or 10 cysteines, all arranged in a characteristic spacing pattern and all disulfide bonded, creating a 3-fingered structural domain GPIHBP1 is unique among Ly6 proteins in having a disordered acidic domain at its amino terminus, with 21 of 26 consecutive residues in the human protein being aspartate or glutamate Recent work by Mysling et al. However, when one examines the genomes of lower vertebrates e. LPL is highly conserved in every lower vertebrate species e. The function of LPL in plasma triglyceride metabolism is conserved in lower vertebrates. When goat polyclonal antibodies against cLPL were injected intravenously into chickens, they inactivated LPL, blocking TRL processing and resulting in hypertriglyceridemia It is conceivable that TRL processing in lower vertebrates might be fundamentally different, with TRLs being transported across capillaries and undergoing processing by the LPL within the interstitial spaces. This possibility may not be farfetched, given that TRLs produced by the chicken intestine are said to move into capillaries by vesicular transport The first step was to assess the specificity of the antibodies, which had been purified on a cLPL immunoaffinity column generated with LPL purified from chicken adipose tissue. We examined, by Western blotting, the ability of the immunopurified antibodies to bind to LPL in crude homogenates of chicken tissues. We were confident that the kDa band in the heart extracts was a cLPL breakdown fragment, but to be certain about that conclusion we repeated the Western blot studies with IgGs that had been further purified on a second cLPL immunoaffinity column generated with a cLPL polypeptide produced in E. To determine if the purified IgG fraction would be useful for immunohistochemistry studies, we first tested their ability to bind to CHO cells that had been transfected with an expression vector for V5-tagged cLPL. Regardless of whether the cells were fixed with methanol or paraformaldehyde, the purified IgGs against cLPL bound avidly and specifically to transfected cells, colocalizing with an antibody against the V5 tag Figure 2. The antibodies bound specifically to cLPL from E. In heart, the anti-cLPL antibodies also bound to a smaller fragment of approximately 38 kDa. Immunocytochemistry studies were performed on fixed and permeabilized cells with a goat antibody against cLPL and a mouse monoclonal antibody against the V5 tag. Binding of the primary antibodies was detected with an Alexa Fluor â€”conjugated donkey antiâ€”goat IgG red and an Alexa Fluor â€”conjugated donkey antiâ€”mouse IgG green. As experimental controls, we included transfected cells that had been incubated with secondary antibodies alone and cells that had been incubated with the anti-cLPL antibody alone. A Immunocytochemistry studies on cells that had been fixed with methanol. B Studies of cells fixed with

paraformaldehyde PFA. In immunohistochemistry studies, we found that much of the LPL in the chicken heart is associated with capillary endothelial cells, colocalizing with an endothelial cell marker von Willebrand factor; vWF Figure 3A. There was no binding of nonimmune goat IgGs or the secondary antibody alone to capillaries in chicken tissues Figure 3A. The presence of cLPL on capillaries of heart and adipose tissue was confirmed in a second experiment Figure 3B. In a third experiment, we also observed colocalization of vWF and LPL Supplemental Figure 1 ; supplemental material available online with this article; https: There was occasional LPL staining that did not coincide with vWF staining, more so than in comparable experiments in mouse tissues, raising the possibility that a fraction of the cLPL in tissues remained within the interstitial spaces Figure 3 and Supplemental Figure 1. Figure 3 LPL in chicken tissues is associated with capillaries. As controls, we examined a section incubated with secondary antibodies alone and a section stained with the vWF antibody and nonimmune goat IgG. To determine if cLPL is located along the luminal surface of capillaries, 9-day-old chickens were injected intravenously with an Alexa Fluor â€”labeled goat IgG against cLPL, a fluorescein-labeled Lens culinaris agglutinin a lectin , and an Alexa Fluor â€”labeled nonimmune goat IgG. After perfusion-fixation, white adipose tissue WAT , heart, liver, and cerebellum were harvested for immunohistochemistry. The lectin also bound to the luminal surface of large blood vessels, whereas IgGs against cLPL did not. Also, there was no binding of cLPL antibodies to capillaries of the cerebellum Figure 4. Only trace amounts of the nonimmune goat IgG were detectable in capillaries of chicken heart, WAT, or cerebellum, demonstrating that the perfusion of tissues was adequate Figure 4. As expected, the nonimmune goat IgGs were bound by immunoglobulin receptors in the liver Figure 4. Figure 4 Chicken lipoprotein lipase cLPL is located along the luminal surface of capillaries. A 9-day-old chicken was given an intravenous injection of 0. The lectin bound to endothelial cells of capillaries and larger blood vessels; the goat IgG against cLPL bound to capillaries but not larger blood vessels arrows. The nonimmune goat IgG did not bind to blood vessels of the heart, WAT, or cerebellum indicating an effective perfusion but as expected did bind to macrophages in the liver. LPL was released rapidly from isolated chicken hearts by perfusing the hearts with heparin Figure 5A. The rapid release of LPL from isolated chicken hearts during the heparin perfusion was consistent with results with isolated mouse hearts. LPL was released rapidly from hearts of wild-type mice with heparin, but LPL release from hearts of Gpihbp1-deficient mice where the LPL is mislocalized to the interstitial spaces was reduced in amount and substantially delayed Figure 5D. We also examined the release of LPL into the plasma compartment after giving chickens an intravenous injection of heparin. Figure 5 Chicken lipoprotein lipase cLPL can be released from tissues with heparin. As a control, a chicken heart was perfused with saline only Saline. For these studies, we pooled fractions 3â€”5 from 2 of the chickens Heparin-1 and Heparin We then performed LPL activity assays. The 4 data points represent duplicate lipase assays on the fractions from 2 chickens. Heparin-mediated release of mLPL from the heart of a Gpihbp1-deficient mouse was delayed white bars. Similar results were observed in 2 other pairs of wild-type and Gpihbp1-deficient mice. Six fractions were collected, and cLPL mass and activity were measured. The 4 data points represent lipase assays, performed in duplicate, on the fractions from 2 chickens. The location of cLPL within tissues i. Immunocytochemistry studies were performed on permeabilized and nonpermeabilized cells with a goat antibody against the S-protein tag red and a mouse antibody against the V5 tag green. Introducing the corresponding mutation into cLPL p. Immunocytochemistry studies were performed on permeabilized and nonpermeabilized cells with a goat antibody against the S-protein tag red and a mouse monoclonal antibody against the V5 tag green. Table 1 The GPIHBP1 locus on human chromosome 8 and the syntenic region on chicken chromosome 2 The expression patterns of the 4 chicken Ly6E-like proteins did not closely resemble the pattern of GPIHBP1 in mammals high in heart, adipose tissue, lung; medium levels in skeletal muscle; low levels in liver; absent from the brain. ENSGALG was expressed at high levels in the skin, medium to low levels in the Harderian gland, and very low levels in the heart. ENSGALG was expressed at high levels in skin, Harderian gland, and thymus; medium levels in thyroid and gizzard; and low levels in the heart. Figure 9 Testing the ability of chicken lipoprotein lipase cLPL to bind to chicken Ly6E-like proteins on the surface of transfected cells. The Iso-Seq data were generated from adult chicken brain, Hamburger-Hamilton HH stage 26 whole embryos, and pooled embryonic heart samples HH stages 18â€”20,

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25, and The short-read RNA-seq data set represented a high-depth search of the mapped exome. The Iso-Seq data set allowed us to identify unmapped chicken transcripts i. Discussion In mammals, GPIHBP1 plays 3 crucial roles in intravascular triglyceride metabolism â€" shuttling LPL to its site of action in the capillary lumen, facilitating the margination of lipoproteins along capillaries, and stabilizing the structure and activity of LPL 2 â€" 4. However, GPIHBP1 â€" the protein that is so essential for intravascular lipolysis in mammals â€" has never been identified in the chicken or any other lower vertebrate species. The results were clear: LPL in chickens does reach the capillary lumen. First, by immunohistochemistry, LPL is present on capillaries of chicken heart and adipose tissue, colocalizing with vWF an endothelial cell marker. Second, a goat IgG against cLPL, when injected intravenously into chickens, binds to LPL on the luminal surface of capillary endothelial cells, whereas a nonimmune chicken IgG does not.

Chapter 2 : Former Utah mayor who stepped down to serve in Afghanistan killed in action Saturday | TheB

Mitochondrial DNA analyses of the red rock lobsterJasus edwardsii supports an apparent absence of population subdivision throughout Australasia.

Irreversible Covalent Inhibition Given what you already know about protein structure, it should be easy to figure how to inhibit an enzyme. Since structure mediates function, anything that would significantly change the structure of an enzyme would inhibit the activity of the enzyme. Hence extremes of pH and high temperature, all of which can denature the enzyme, would inhibit the enzyme in a irreversible fashion, unless it could refold properly. Alternatively we could add a small molecule which interacts noncovalently with the enzyme to either change its conformation or directly prevent substrate binding. Finally, we could covalently modify certain side chains, that if they are essential to enzymatic activity, would irreversibly inhibit the enzyme. We discussed previously the types of reagents that would chemically modify specific side chains that might be critical for enzymatic activity. For example, iodoacetamide might abolish enzyme activity if a Cys side chain is required for activity. These reagents will usually modify several side chains, however, and determining which is critical for binding or catalytic conversion of the substrate can be difficult. One way would be to protect the active site with a saturating quantities of a ligand which binds reversibly at the active site. Then the chemical modification can be performed at varying reaction times. The critical side chain would be protected from the chemical modification, but the extent of protection would depend on the Kd, concentration of the protecting ligand. Competitive Inhibition Reversible Competitive inhibition occurs when substrate S and inhibitor I both bind to the same site on the enzyme. In effect, they compete for the active site and bind in a mutually exclusive fashion. This is illustrated in the chemical equations and molecular cartoon below. There is another type of inhibition that would give the same kinetic data. If S and I bound to different sites, and S bound to E and produced a conformational change in E such that I could not bind and vice versa , then the binding of S and I would be mutually exclusive. This is called allosteric competitive inhibition. Inhibition studies are usually done at several fixed and non-saturating concentrations of I and varying S concentrations. The key kinetic parameters to understand are Vm and Km. Let us assume for ease of equation derivation that I binds reversibly, and with rapid equilibrium to E, with a dissociation constant Kis. Kis is also named Kic where the subscript "c" stands for competitive inhibition constant. A look at the top mechanism shows that even in the presence of I, as S increases to infinity, all E is converted to ES. That is, there is no free E to which I could bind. Vm is not changed. However, the apparent Km, Kmapp, will change. The double reciprocal plot Lineweaver Burk plot offers a great way to visualize the inhibition. In the presence of I, Vm does not change, but Km appears to increase. These intersecting plots are the hallmark of competitive inhibition. Note that in the first three inhibition models discussed in this section, the Lineweaver-Burk plots are linear in the presence and absence of inhibitor. This suggests that plots of v vs S in each case would be hyperbolic and conform to the usual form of the Michaelis Menton equation, each with potentially different apparent Vm and Km values. An equation, shown in the figure above, can be derived which shows the effect of the competitive inhibitor on the velocity of the reaction. This shows that the apparent Km does increase as we predicted. Kis is the inhibitor dissociation constant in which the inhibitor affects the slope of the double reciprocal plot. Hence, plots of Y vs log L for a series of binding reactions of increasingly higher Kd lower affinity would reveal a series of identical sigmoidal curves shifted progressively to the right, as shown below. The same would be true of vo vs S in the presence of different concentration of a competitive inhibitor, for initial flux, Jo vs ligand outside, in the presence of a competitive inhibitor, or ML vs L or Y vs L in the presence of a competitive inhibitor. In addition, as the curves above show, multiple complete plots of v0 vs lnS at varying fixed inhibitor concentration or for variant enzyme forms different isoforms, site-specific mutants over a broad range of lnS can be made which facilitates comparisons of the experimental kinetics under these different conditions. This is especially true if Km values differ widely. Consider the activity of an enzyme. If a competitive inhibitor is added, the activity of the enzyme would drop until at saturating infinite I, no activity would remain. Graphs showing this are shown below. It also describes how good an enzyme is in

differentiating between different substrates. If has enzyme encounters two substrates, one can be considered to be a competitive inhibitor of the other. One can hypothesize that on binding S, a conformational change in E occurs which presents a binding site for I. Inhibition occurs since ESI can not form product. It is a dead end complex which has only one fate, to return to ES. Let us assume for ease of equation derivation that I binds reversibly to ES with a dissociation constant Kii. Kii is also named Kiu, where the subscript "u" stands for the uncompetitive inhibition constant. A look at the top mechanism shows that in the presence of I, as S increases to infinity, not all of E is converted to ES. That is, there is a finite amount of ESI, even at infinite S. Under these conditions, the apparent Vm, Vmapp is less than the real Vm without inhibitor. In addition, the apparent Km, Kmapp, will change. In the presence of I, both Vm and Km decrease. It turns out that they change to the same extent. Therefore the plots will consist of a series of parallel lines, which is the hallmark of uncompetitive inhibition. An equation, shown in the diagram above, can be derived which shows the effect of the uncompetitive inhibitor on the velocity of the reaction. This shows that the apparent Km and Vm do decrease as we predicted. Kii is the inhibitor dissociation constant in which the inhibitor affects the intercept of the double reciprocal plot. Note that if I is zero, Km and Vm are unchanged.

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Chapter 3 : pseudoabsence - Wiktionary

Lipoprotein lipase reaches the capillary lumen in chickens despite an apparent absence of GPIHBP1 Cuiwen He, , Loren G. Fong, Stephen G. Young.

I see that there is the repeated assertion that Mr Jackson is being denied due process on this issue because of the publicity surrounding it. Her only action was to inform Jackson that the accusation was made against him. I understand that Mr Jackson, learning of those allegations proceeded to contact several of his past students saying that he had learnt that someone influential was seeking to find out the extent of what they might have told someone, and to convince them not to testify in any investigation some of whom had confided in me directly, and who had confided in others who then relayed that information to me. I advised them to record any future contact with him, whether via Facebook, text message or phone call. I contacted Chief Education Officer, Mr Marcel Hutson and advised him of the situation on Thursday, 16th, and he immediately booked to meet with me on Monday, 20th, since he would be out for the weekend. On Friday, the 17th, I put up a general status on social media about investigating a predatory business teacher at a senior secondary school and it was others who associated Mr Jackson with that status. That is something that should speak for itself. Several people even angrily exhorted me to name the person I was referring to, something I refused to do until I had handed in my letter to the CEO on Monday. On Friday afternoon, I was contacted by Jerome Khan, purporting to be an unofficial mediator between myself and Mr Jackson on what he claimed to be a personal dispute that escalated online. My response to this was that while my primary intention is to get Jackson out of the school, I believe that he had committed multiple offences under the Sexual Offences Act and I could not guarantee that legal steps would not be taken against him. He texted the e-mail that same afternoon, proof I needed to corroborate my narrative should the need arise. I want to make it clear that both officers respected my submission and acted swiftly in response. Both needed to be commended on this. Considering that the school administration had a history of suppressing complaints against Mr Jackson, and considering that he continued to have access to students, and considering that the protocol seemed to be to inform him of an accusation before any actual investigation, I decided to release my letter to Mr Hutson to the press, precisely to ensure that there was public awareness of a dangerous problem in a public school that was systematically not being addressed. If I am proven wrong in this, I will publicly resign my post as a public officer. It is critical that the press and everyone dealing with this issue understand that obfuscation only benefits those who are in fact guilty of something, whether the commission of an act or the covering up of it. It is curious that even now Jackson did not go to the union, choosing instead to host a press conference with three attorneys, during which he claimed that he took a leave of absence when it was subsequently learnt that he was in fact sent on administrative leave. The task for investigators is to find out what happened to these complaints, some going as far back as How were they treated and by whom? If there has been a failure of due process in this, it begins with the apparent absence of any clear process of respecting, recording and processing complaints by students of the school, or even parents, against teachers with predatory behaviour. The question is, why?

Chapter 4 : Absente - Wikipedia

Apparent absence of stroke and ischaemic heart disease in a traditional Melanesian island: a clinical study in Kitava. Lindeberg S(1), Lundh B. Author information: (1)Primary Health Care Centre, SjÃ¶bo, Sweden.

Chapter 5 : Mac Miller Dead: Rapper Dies of Apparent Overdose at 26 â€“ Variety

For IBM/Lenovo ThinkPad laptop enthusiasts. Blog posts are encouraged. If your post does not appear in the new queue, it's because of the stupid spam filter. Use the link to 'message the moderators' below to tell them.

Chapter 6 : Apparent Absence of Microbial Life inside an Alkaline Slag Dump

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Characterization of an HIV-1 isolate displaying an apparent absence of virion-associated reverse transcriptase activity.

Chapter 7 : Wikipedia:Conflict of interest - Wikipedia

In the current study, our goal was to determine if LPL in chickens, despite an apparent absence of GPIHBP1, could reach the capillary lumen. The results were clear: LPL in chickens does reach the capillary lumen.