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| Reviewer #1 (Comments to the Author (Required)):  Review constrained mixture model autoregulation The authors present a modeling framework that combines coronary wall biomechanics, the intramyocardial pump/cardiac elastance, vascular branching patterns and control of vascular activation by pressure, shear stress and metabolism in order to predict coronary pressure-flow relations. The model is mostly based on porcine data. The model predicts to a reasonable extent observations of pressure-flow relations and the effect of proximal stenosis and of vasodilators. While several studies have modeled aspects of coronary flow, the integration of vessel wall properties into network modeling is relatively novel. The motivation of including extensive wall modeling as well as networks rather than lumped models is the view that further extension towards growth and remodeling models require this step.  1. The work lacks a hypothesis. Related, it lacks new model predictions that could help designing new experiments and it fails to provide a vision on this.  Line 76-78:  This paper aims to develop a structurally-motivated computational framework to study coronary autoregulation, a physiological concept that fundamentally relies on functional changes of SMCs. The model, based on constrained mixture theory, provides a necessary tool which can be used to study baseline activation of SMCs and homeostatic stresses, while being informed by experimental pressure-diameter, tree morphometry, and hemodynamics data, for both subepicardial and subendocardial trees.  2. It also does not really improve insight into coronary autoregulation. Sure enough, model predictions are given on the P-Q relation and the state and load of individual segments, but the authors should indicate what can be learned from this simulation that was not already known.  3. I agree with the authors that a modeling framework like the one presented here will be needed for further extension towards growth/remodeling models. I see the main value of this work to be a demonstration of the way how to do this and an analysis of the problems that may be encountered in such work. However, the model, with 50 or so parameters, is hugely underdetermined and the predictions are rather limited. Moreover these predictions (e.g. the final P-Q relations) could have been made by much simpler models. The authors should place more stress on the general goal and be more explicit about the limitations of all the modeling choices.  Line 556-566:  Recent computational modeling endeavors in coronary physiology focused on capturing key features of short-term coronary autoregulation using lumped-parameter models (18, 64, 86). However, since these approaches lack structurally-motivated models for vascular tissue, they are not equipped for predicting long-term pathophysiologic responses of the coronary circulation. To address this need, this work aimed to produce a unified computational approach that could integrate vascular responses at the short time scales (e.g., autoregulation) as well as long time scales (e.g. vascular growth and remodeling) with integration of diverse data from morphometric, hemodynamics and structural measurements. This design goal resulted in the adoption of a structurally-motivated modeling framework, widely used in tissue growth and remodeling, such as the constrained mixture theory. The constrained mixture enables integration of microstructural properties and cellular level functions within the vessel wall in a nonlinear continuum mechanics framework.  4. While I understand the procedure, it is fully unclear how the model evolves towards a stable situation, and whether this steady state case is unique.  Line 345-346:  To ensure the uniqueness of the converged steady state, the initial conditions were randomly varied.  5. The work is based on a few experimental studies. It seems that these results are used as some form of ground truth, while clearly experimental studies come with their limitations. Thus, Kuo's lab produced unique data on biomechanics and reactivity of isolated coronary vessels, but these data were generated under very specific conditions (in vitro, young pigs, MOPS buffer, ...) and no doubt a different group would find different quantitative data when redoing this work (which would actually be a very good idea in general). My concern is that rigidly using such data as input might generate vessel wall models that are maybe less realistic. This could e.g. be the case for the mass fractions and zero-stress strains, which are extrapolated from the active P-d relations under the assumption that SMC intrinsic properties are equal between vessels. Moreover, the fit in fig 5 is limited. The authors should be more explicit in acknowledging this limitation.  Line 673-678:  First, the experimental measurements delineating the pressure-diameter relationships of myocardial coronary vessels are scarce. We used four available sets of pressure-diameter data for a wide range of subepicardial and subendocardial vessels from 400 μm to 20 μm. More pressure-diameter data in the coronary arterioles will enhance the accuracy and predictive capabilities of the model. Although this limitation may not be influential in low coronary pressure ranges due to small activation, it may affect the total flow in high coronary pressures.  6. Part of the adaptation model is based on MVO2. The authors need to demonstrate that actual terminal flow was sufficient. You could indicate in fig 9 the homeostatic total flow (terminal flow x number of terminal segments)  Line 267-270:  Based on blood flow velocity measurements in (8, 77, 81), a baseline flow rate of mm3/s was used for the subepicardial terminal arterioles. To determine the flow for the subendocardial tree, the total subendocardial to subepicardial flow ratio (ENDO/EPI) was assumed to be ~1.25 (22).  7. Related, how high was flow in absolute terms? I am not expecting a full quantitative comparison to experimental data but at least the authors should show that flow has a realistic order of magnitude.  8. On line 251 you mention flow as a soft boundary condition, implemented as a contribution to the cost function. It seems that in your simulation, you simultaneously use flow in the cost function and in the metabolic adaptation and shear effect. I find it very difficult to understand the consequences of this choice. In analogy to the much simpler original Murray law: one could make a model for diameter adaptation in a network based on either the cubic law (cost of volume and drag) or on the concept of shear stress, but using a combination of the two seems less logical. Homeostatic optimization is a consequence of the evolution of mechanisms such as shear stress sensing and should have no role in mechanistic models such as the current one. The authors should motivate their choice or, preferentially, provide a model that is based solely on (putative) mechanisms.  Extended Murray’s law.  9. Inlet and outlet coronary pressure are taken as 100 and 55 mmHg, respectively. Does the model of your endocardial tree include the transmural segment (with its resistance) or rather a local endocardial tree? How realistic is 100 mmHg inlet for the sub-endocardial tree? Chilian (Circ Res 1991) showed much lower pressures in 100 micron vessels in the sub-endocardium than sub-epicardium.  Line 615-617:  We must note that the inlet pressure for both subendocardial and subepicardial trees were assumed 100 mmHg based on the study by Mittal et al. (61) that showed the pressure does not significantly drop in vessels of 100-1000 μm.  10. In the epicardial stenosis application, you assume a fixed perfusion pressure distal to the stenosis. However, this pressure depends on the balance between the pressure loss (Poiseuille, Bernoulli) in the stenosis and the resistance of the microcirculation. Moreover, in this situation, the sub-endocardial and sub-epicardial circulation compete for flow. I think this needs combined simulation of both circulations and the stenosis, rather than a simple input pressure reduction. At least, this limitation should be discussed.  Line 509-511:  Although the pressure reductions across stenoses are triggered by different levels of epicardial occlusion, our choices of inlet pressures are motivated by the experimental study by Kanatsuka et al. (43).  11. For the P-Q relations you switch from constant output pressure to a combination of downstream resistance and capillary pressure. Why not employ this latter model in the first place? It is not trivial that this provides identical simulation outcomes for the first part of the work.  Line 431-433:  The value of these resistances was computed using the terminal arterioles pressure minus the venular pressure, divided by the terminal homeostatic flow in the coronary trees (viz., Rcoronary = P/qterm).  12. Eq. 3 on the myogenic response takes transmural pressure as stimulus. In classical work, Paul Johnson postulated the concept that the stimulus for the myogenic response is wall stress. A step upward in pressure increases the wall stress, and the response causes vasoconstriction, which again decreases the wall stress (Handbook of Physiology, around 1980). While it would be difficult to demonstrate in experiments that indeed wall stress is the stimulus, in my view this is still our best understanding of the myogenic response. At least, pressure should result in some local stress and/or deformation in order to be sensed. This means that the wall composition gets into the equation: A thicker wall with more SMC may sense less stress. While at first glance this may seem an unnecessary further complication, it does point out a fundamental point: future G/R models need to take such local stresses and the activation response into account. The authors should motivate their choice for P rather than stress as input, especially since other modelers use stress (Brings-Jacobsen et al, Am J Physiol Regul Integr Comp Physiol 294:R1379-R1389, 2008; VanBavel and Tuna, PLoS One . 2014 Jan 31;9(1):e86901.)  Line 625-631:  Our choice for the myogenic model was motivated by the ex-vivo experimental measurements in (53) where the transmural pressure is controls the myogenic response. Alternatively, other modeling studies have assumed that the circumferential wall stress () sensed by the SMCs is the signal for the myogenic control mechanism (36, 84), based on earlier studies (38). This theoretical discrepancy is circumvented in our model since the dependence of myogenic response on the vascular wall thickness , vessel diameter , and the transmural pressure is implicitly integrated in the mechanical equilibrium equation.  13. Maybe I missed it, but code seems missing.  Reviewer #2 (Comments to the Author (Required)):  Gharahi et al. present an interesting paper on autoregulation in the coronary circulation. I have a few remarks.  Major: 1. I was quite surprised to find so little mention of 'time' in this manuscript. Clearly, autoregulation is dynamic, and the different responses (myogenic, shear, ...) very likely have different time constants, which, potentially, also vary along the arterial tree. What's more, all boundary conditions are also strongly time-dependent (the heart beats...). This is a major limitation, to which only three general sentences in the Limitations section are devoted. a. How do the authors plan to address this limitation in the future? b. The authors hint at using this framework for growth and remodeling (G&R) simulations, but are the modeled steady-state responses representative of what happens in vivo? c. Will a (long-term) G&R simulation yield representative results if the short (pulsatility) and medium (autoregulation) time scales are not taken into account?  I suggest to add dynamics to the modeling presented. Alternatively, perhaps simulations using smaller, more 'conceptual' models from literature that do include time can be used to show that steady-state responses indeed are representative of the dynamic case.  If this is really beyond scope, please: d. Add the term "steady state" to the title, abstract, and introduction sections so it is clearer for the reader what to expect. e. Take this into account in Discussion lines 567-569 where time scales are discussed. f. Discuss in more detail how the steady-state assumption affects results, and how time behavior could be implemented. I would also separate the short time scale between something like 'very short' (pulsatility), 'medium' (autoregulation), and 'long' (G&R).  2. The APS strongly promotes transparent reporting. Please share the source code of your model.  3. No mention of 'implementation' is made. What language what used? Specific solvers?  Line 354-355:  All the steps of the model were implemented in MATLAB (Mathwork, Natick, MA).   4. Many parameters are obtained through fitting. However, the cost or error functions used in the manuscript are not explicitly described. Please add those to the Appendix.  Lines 778-784:  Parameter estimation  The parameter estimations in sections 2.2 and 2.4 are performed by minimizing the following error function:  where and are theoretical and experimental values of the quantity . In section 2.2, the quantity is the diameter of vessels in passive and myogenic responses, and in section 2.4, the flow in the coronary arteries (q) over the flow when coronary pressure is 100 mmHg. Specific constraints for the optimization problem are stated in the text.  5. (related to previous point) Sensitivity analyses are performed by assessing how the error varies with a change in parameters. However, it is unclear at this point what the 'error' exactly is.  6. The term 'tension' is frequently used (e.g., but not limited to, Eq. 1), however, this is ambiguous. It seems that T (and T\_act) refers to a Cauchy stress, but S to a quantity involving mass (since division of S by rho leads to a T; Eq. 1). However, both T and S are referred to as 'tension'. I suggest to refer to all T's in the manuscript as Cauchy stresses.  Line 139-141:  The active tension () exerted by SMCs, was defined as   |  |  |  | | --- | --- | --- | |  |  | (1) |   where S is the active stress, is the density of arterial tissue, area density of SMCs, and are stretches corresponding to the maximum and zero active tension, respectively, and is the vessel circumferential stretch.  7. Related to Minor #1: S\_max (Table 1) is given in MPa. From Eq. 2 I deduce that S\_p, then, also has units of MPa. Next, Eq. 2 suggests that S also is in MPa. Then, if that S (in MPa) feeds into Eq. 1, T\_act would be in MPa/(kg/m^3), or 10^6\*Nm/kg, which seems odd. Also, comparing the strain energy functions in equations A10 and A11: A10 are in Pa/kg, but A11 is in MPa/kg \* m^3. Shouldn't rho\_wall (Eqs 1 and A11) be a mass rather than a mass density? Please check/clarify.  Clarified in previous comment.  8. Some equations contain 'hard-coded', nondimensionless constants, which makes their solution unit-dependent. E.g. Eq. 12: the "250" must be µm (to match D), and the 0.02 must be in 1/µm (so that the exponent is dimensionless). I suggest using symbols for those constants, and defining those below/above the equation. This also holds for equations C1-C4.  Line 318-321:  Based on the experimental observations (13, 43, 51, 52), the diameter-dependent weights and representing the magnitude of each mechanism in the total stimulation are prescribed (Fig. 2)   |  |  |  | | --- | --- | --- | |  | and . | (12) |   where and are 0.02 and 0.013 1/μm, and and are 250 and 50 μm, respectively.  9. Table 4 and 5: please indicate that the sensitivities are in %.  Fixed  Reviewer #3 (Comments to the Author (Required)):  Review of MS H-00519-2020  Constrained Mixture Theory Model to Study Autoregulation in the Coronary Circulation By Hamidreza Gharahi, Daniel A. Beard, C. Alberto Figueroa and Seungik Baek  The overall impression is that this is a solid piece of work - the authors have done a lot to align the model with experimental data although a large number of parameters still needed to be estimated. Some of the model assumptions can of course be debated (please see below)  There are two issues that perhaps should be considered: 1) Is this kind of paper is well suited for the audience of AJP heart and Circ. It is a rather large and complex model (including the appendix) which would perhaps be more suitable for a journal like "Bulletin of Mathematical biology" or similar. Many of the formulations requires rather extensive background knowledge in mathematics if the reader should be able the judge the results - for instance (line 192-194) "The downhill simplex method was used to estimate the parameters of the constrained mixture model (72) by minimizing the mean squared error"........etc  2) I think that the paper to some degree misses a central scientific question to be addressed - some hitherto unexplained observation that could be addressed and explained with the model. In some way this is only the first step (although a very large one), but the reader is to some degree left with the feeling that now we have a model the reproduces quite well several basic experimental observations, but then nothing more happens (although the authors points to future directions).  Line 79-81:  This paper aims to develop a structurally-motivated computational framework to study coronary autoregulation, a physiological concept that fundamentally relies on functional changes of SMCs. The model, based on constrained mixture theory, provides a necessary tool which can be used to study baseline activation of SMCs and homeostatic stresses, while being informed by experimental pressure-diameter, tree morphometry, and hemodynamics data, for both subepicardial and subendocardial trees.  Below I have some more specific comments to the text  3) Introduction: Tension is an adequate measure in thin-walled structures, however a majority of the coronary vessels have a substantial wall:lumen ratio which makes wall stress a more adequate measure. What is the reason for not using wall stress consistently instead of tension? (wall stress is also used and it is confusing with the two very different measures being used side by side). Please explain  Changed the name of from active SMC tension to active SMC stress, since its actually stress not tension. It first appears in eq. 1. Similarly and are defined as is a pressure-dependent stress and the total maximum active stress  4) Introduction: Other substances than NO are being produced in variable amounts by the endothelium in response to variation in local shear stress (e.g. thromboxane, endothelin) - are any of these of significance in the coronary circulation?  ???  5) line 73: A short explanation of the concept "Constrained mixture theory models" when it is first encountered, would be nice for readers unacquainted with the field.  Line 74-77:  Constrained mixture theory models have been widely applied to describe the nonlinear mechanical behavior of arterial tissue, accounting for the contributions of the main load-bearing constituents (e.g., collagen, elastin and SMCs) (25, 72). The core idea of the constrained mixture theory is that the vascular wall constituents are constrained to deform together but may have individual mechanical properties and stresses.  6) line 79-80: "1) study of adaptations following drops in epicardial pressure": It is uclear at this point if the sentence refers to short- or longterm (structural) adaptation.  Fixed.  7) L112-113: Is this correct? As far as I remember (I may be wrong though) Murray was concerned with the amount of blood and its metabolism (not tissue metabolism), but not with the vascular wall (has been included in other later formulations)  Extension of Murray’s law  Fig. 1 very clear and informative figure  L128-129: which diameter - the wall has a certain thickness so T will depend on which diameter is chosen  Internal diameter  L134-135 Is it T or S which is the active tension generated by smc's? - not clear from the text  L135: S is an unfortunate letter as it is normally used for stress rather than tension  The definition of S has been fixed. S is stress and T is the tension. So corresponds to the state of activation of SMCs, T corresponds to the tension the exert.  L161 - 163: It appears from the text that this is an assumption. However if the material properties of e.g. Elastin is the same in vessels of different size wouldn't it be a more logical assumption that pre-stretch should be the same, to match the optimal operation rage of the material?  Optimal range of material changes given the homeostatic pressure. (not in paper)  It seems somewhat inconsistent first to base network optimization on the amount of wall material (line 218-219) and then afterwards using a thin wall approximation (line 231) which assumes the wall has no thickness. This may be an ok approximation but seems a bit odd.  Thin wall assumption does not assume no thickness. (not in paper)  Line 261-262 - since the myocardium is primarily perfused in the diastole this seems a bit high - perhaps 80 mm Hg would be better, however it is unlikely to change the conclusions  Line 615-617:  We must note that the inlet pressure for both subendocardial and subepicardial trees were assumed 100 mmHg based on the study by Mittal et al. (60) that showed the pressure does not significantly drop in vessels of 100-1000 μm.  Consequences of Eq. 15 are very difficult to grasp but the formulation is probably reasonable. Apart from that, I think the formulations in sec. 2.4 are reasonable.  Line 204 - 207 "A sensitivity index 𝑋 was defined as the percentage change in model error 𝐸 associated with introducing a 10% change in the parameter values one at a time 𝑋 = 𝐸(𝜃) |𝐸(𝜃 + 0.1𝜃) − 𝐸(𝜃)|, (4)where 𝜃 is the estimated parameter value, and 𝐸(𝜃) is its associated error, defined as the difference between predicted and measured diameters."  I am not sure I understand this formulation. Sensitivity is not the size of an error. The results should change when you change a given (relevant) parameter. This is not an error - this is the correct behavior. If nothing changes when you change a relevant parameter then this parameter has no role in the model and should be removed. Sensitivity is about how much the results change upon a given (percent) change in a parameter. Please explain.  We present the sensitivity of identification of adjustable model parameters by using the sensitivity index (X). (Not in text)  Line 360 - about the mass-fractions. These appear to be estimated based on passive distension and active myogenic behavior. I think that this can be a reasonable approach, but these fractions are anatomical features of the vessels. From the discussion section it appears that anatomical data do exist, (it should be possible to e.g. get an idea of the fraction of smooth muscle tissue from simple histological sections)  Fig. 8 - is the high baseline activation level in small arteries realistic? If they are almost maximally activated already at normal physiological pressure they would be unable to react with contraction to an increase in pressure - i.e. they would not show any myogenic response when pressure is raised - or have I missed something (A can only go to 1 right?).  The activation level presented here is the myogenic activation level activation level, and not the total activation of SMCs. Therefore, if A~1 that means the vasodilation from metabolic and shear-dependent mechanisms are small and the SMC response is modulated by myogenic response.  Line 148-150:  Therefore, we have assumed the following functional relationship for S:   |  |  |  | | --- | --- | --- | |  |  | (2) |   where is the myogenic activation level (), determined by wall shear stress () and metabolic activity () mechanisms; and is a pressure-dependent stress, and it represents the myogenic control.  Fig 10. Regarding the activation level: It appears that the activation level of small arteries is extremely high throughout the pressure range. Again that seems odd- why doesn't an activation level close to 100% lead to complete closure of the vessel an thereby to complete cessation of network flow? (Except perhaps at very high pressure levels). Please explain.  Same as above  Fig. 11. Panel E and F. It appears as if the pressure increase transmits throughout the network at all pressures. Isn't it normally so, that increasing myogenic contraction will cause an increased pressure dissipation so that the closer you come to the capillary bed the more "normal" will the pressure be, despite increased perfusion pressure (maybe some of this effect is seen for the small arterioles (green curve) - but not really for the other vessels). Please explain  ???  Fig. 12: would it be possible to compare this figure to some experimental data points plotted in the figure? I guess the figure is only valid during diastole - since normally you would think that it is the endocardial flow, which is most severely restricted in when the heart has to produce a high mean arterial pressure.  Physiologically relevant results in the same ranges could not be found. So, we sufficed to explaining them in the text.  The discussion is informative and thorough regarding model limitations, but could be expanded on the wider physiological perspectives of the model. |