Supplementary Information

A microfluidic model to study the effect of arrhythmic flows on endothelial cells

Austin Lai^{1,2}, Adam Hawke³, Mokhaled Mohammed³, Peter Thurgood^{2,3}, Gianmarco Concilia³,

Karlheinz Peter^{2,4}, Khashayar Khoshmanesh^{2,3,*}, Sara Baratchi^{1,2,4,*}

¹ School of Health and Biomedical Sciences, RMIT University, Bundoora, Victoria

² Baker Heart and Diabetes Institute, Melbourne, Victoria

³ School of Engineering, RMIT University, Melbourne, Victoria

⁴ Baker Department of Cardiometabolic Health, University of Melbourne, Melbourne, Victoria

Corresponding authors:

Khashayar Khoshmanesh, Khashayar.Khoshmanesh@rmit.edu.au

Sara

Baratchi,

Sara.Baratchi@Baker.edu.au



Supplementary Figure 1. Varying the amplitude and frequency of pulsatile flow using PWM signal:

(a) The amplitude of the pulsatile flow can be modulated by varying the PWM signal's duty cycle.(b) The frequency of the pulsatile flow can be modulated by varying the time between each burst of the PWM signal



Supplementary Figure 2: Photos of the experimental set up used for this study showing the

(1) microfluidic channels, (2) tubing, (3) piezo1 electric pump and (4) reservoir.



Supplementary Figure 3. Arrhythmic flows do not affect cytoskeleton remodelling of endothelial cells. Summary graph showing the frequency of the orientation angle of endothelial stress fibers under 20 or 4 dyne/cm² shear stress and variation in frequency, amplitude, or both. Data is representative of four independent experiments and data is analysed using two-way ANOVA.



Supplementary Figure 4: β -catenin expression is regulated by the frequency and amplitude of pulsatile flow. Box plot graphs showing the expression level of β -catenin in endothelial cells cultured exposed to arrhythmic flows at 4 or 20 dyne/cm². * P < 0.05, ** P < 0.01, *** P < 0.001, **** P < 0.0001.



Supplementary Figure 5: ICAM-1 expression is regulated by the frequency and amplitude of pulsatile flow. Box plot graphs showing the expression level of β -catenin in endothelial cells cultured exposed to arrhythmic flows at 4 or 20 dyne/cm². * P < 0.05, ** P < 0.01, *** P < 0.001, **** P < 0.0001.