

Supplementary Information - Simvastatin attenuates endothelial dysfunction in a coronary artery-on-a-chip

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Note S1 - Python Script – Pump Control

```
1. import numpy as np
2. import pandas as pd
3.
4. # Define pixelPerMicron as a constant
5. pixelPerMicron = 1.204
6.
7. # Read parameters from separate CSV file
8. parameters_df = pd.read_csv("/Users/jsin6148/Downloads/parameters.csv")
9.
10. # Loop through each row in the parameters file, skipping the first row
11. for index, row in parameters_df.iloc[22:].iterrows():
12.     filename = row['filename']
13.     bifX = float(row['bifX'])
14.     bifY = float(row['bifY'])
```

```

15.     bifMean = float(row['bifMean'])
16.
17.     # Provide the full path to the filename
18.     full_path_to_file = "/Users/XYZ/Downloads/" + filename
19.
20.     print("Processing file: {filename}")
21.
22.     # Read the CSV file into a NumPy array
23.     data = np.loadtxt(full_path_to_file)
24.
25.     # Specify parameters
26.     boxSize = 200
27.     analyseAreaSize = round(boxSize * pixelPerMicron)
28.     minDistance = 1140
29.     xPosition = 0
30.     yPosition = 0
31.
32.     # Iterate over the data using array slicing
33.     for r in range(0, data.shape[0] - (analyseAreaSize - 1), 10):
34.         for c in range(0, data.shape[1] - (analyseAreaSize - 1), 10):
35.             # Extract the 2x2 grid
36.             analyseArea = data[r:r + analyseAreaSize, c:c + analyseAreaSize]
37.
38.             # Calculate mean intensity
39.             meanIntensity = np.mean(analyseArea)
40.
41.             # Check thresholds
42.             if meanIntensity > bifMean * 2: # Assuming fold-change threshold is 2
43.                 # Calculate distance
44.                 xDistance = bifX - c
45.                 yDistance = bifY - r
46.                 hypotenuse = (np.sqrt(xDistance ** 2 + yDistance ** 2)) / pixelPerMicron
47.
48.                 if hypotenuse < minDistance and xDistance != 0 and yDistance != 0:
49.                     minDistance = hypotenuse

```

```
50.             xPosition = c
51.             yPosition = r
52.
53.     # Print results for each file
54.     print("Final min distance:", minDistance, "microns")
55.     print("This occurs at x:", xPosition, "and y:", yPosition)
56.
57. print("All files processed.")
58.
```

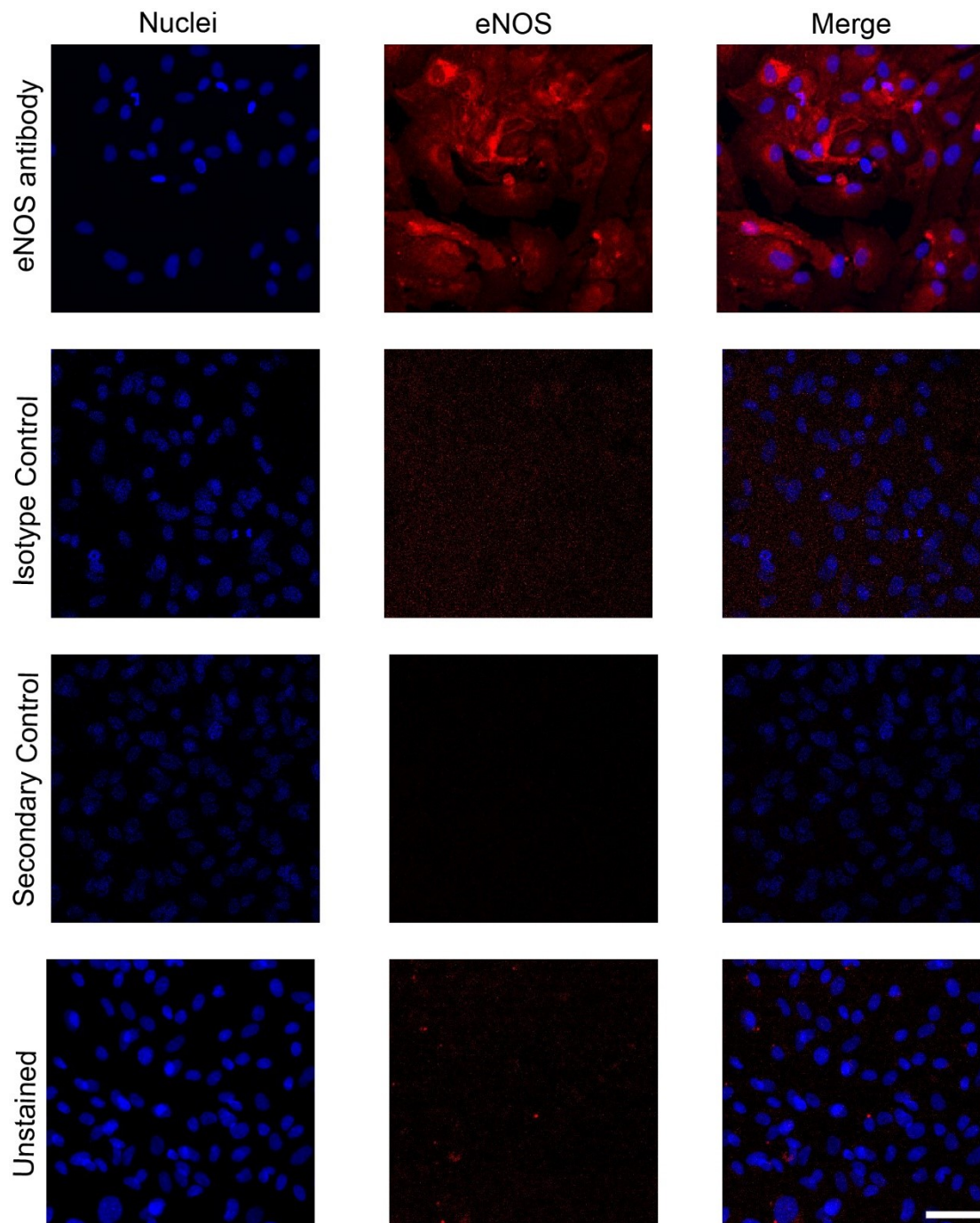


Figure S1- Isotype validation of eNOS immunofluorescence in HCAECs. Representative confocal micrographs of HCAECs labelled with eNOS rabbit anti-human polyclonal antibody showing eNOS signals, rabbit IgG isotype control showing low background signals, secondary goat anti-rabbit IgG alone showing low background signal, and the unstained group showing minimal fluorescence. Nuclei were stained with Hoescht. The scale bar is 100 μm .

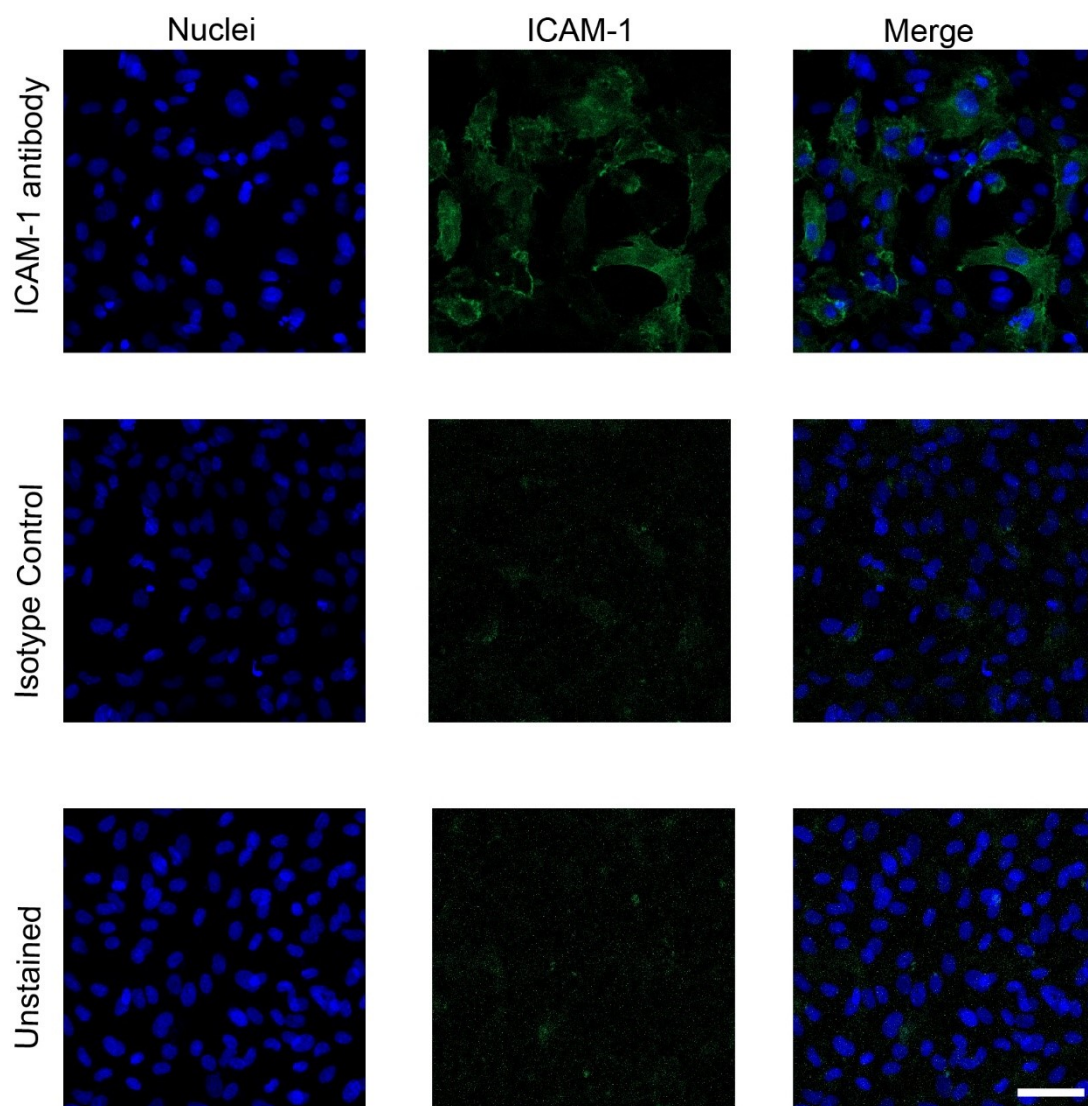


Figure S2- Isotype validation of ICAM-1 immunofluorescence in HCAECs. Representative confocal micrographs of HCAECs labelled with CD54 (ICAM-1) monoclonal antibody showing ICAM-1 fluorescence, mouse IgG1 kappa isotype control showing low background signals, and the unstained group showing minimal fluorescence. Nuclei were stained with Hoescht. The scale bar is 100 μ m.