

Supplement File 1 – QuPath script for automated assessment of immunohistochemical staining with anti-MPO, anti-CD31 and anti-CD68 antibodies

Script for anti-MPO

```
setImageType('BRIGHTFIELD_H_DAB');

setColorDeconvolutionStains({'Name': "H-DAB default", "Stain 1": "Hematoxylin", "Values 1": "0.65111 0.70119 0.29049 ", "Stain 2": "DAB", "Values 2": "0.26917 0.56824 0.77759 ", "Background": "255 255 255 "});

selectAnnotations();

runPlugin('qupath.imagej.detect.nuclei.PositiveCellDetection', {"detectionImageBrightfield": "Hematoxylin OD", "requestedPixelSizeMicrons": 0.5, "backgroundRadiusMicrons": 8.0, "medianRadiusMicrons": 0.0, "sigmaMicrons": 1.5, "minAreaMicrons": 10.0, "maxAreaMicrons": 400.0, "threshold": 0.1, "maxBackground": 2.0, "watershedPostProcess": true, "excludeDAB": true, "cellExpansionMicrons": 5.0, "includeNuclei": true, "smoothBoundaries": true, "makeMeasurements": true, "thresholdCompartment": "Cytoplasm: DAB OD mean", "thresholdPositive1": 0.2, "thresholdPositive2": 0.4, "thresholdPositive3": 0.6, "singleThreshold": true});
```

Script for anti-CD31 and anti-CD68

```
setImageType('BRIGHTFIELD_H_DAB');

setColorDeconvolutionStains({'Name': "H-DAB default", "Stain 1": "Hematoxylin", "Values 1": "0.65111 0.70119 0.29049 ", "Stain 2": "DAB", "Values 2": "0.26917 0.56824 0.77759 ", "Background": "255 255 255 "});

selectAnnotations();

runPlugin('qupath.imagej.detect.nuclei.PositiveCellDetection', {"detectionImageBrightfield": "Hematoxylin OD", "requestedPixelSizeMicrons": 0.5, "backgroundRadiusMicrons": 8.0, "medianRadiusMicrons": 0.0, "sigmaMicrons": 1.5, "minAreaMicrons": 10.0, "maxAreaMicrons": 400.0, "threshold": 0.1, "maxBackground": 2.0, "watershedPostProcess": true, "excludeDAB": true, "cellExpansionMicrons": 5.0, "includeNuclei": true, "smoothBoundaries": true, "makeMeasurements": true, "thresholdCompartment": "Cell: DAB OD mean", "thresholdPositive1": 0.2, "thresholdPositive2": 0.4, "thresholdPositive3": 0.6, "singleThreshold": true});
```