

1 **Legends to Videos**

2 All videos are Z series of fresh tissue from CSF1R-FRed x *Csf1r*-EGFP mice without fixation. Each
3 movie involves 1 μ m steps from the surface and the depth is shown. High resolution versions of
4 these videos can be downloaded from <https://doi.org/10.14264/8c6d4ba>.

5 **Video 1. Liver.** Shows the transition from ramified subcapsular macrophage populations spread
6 in the plane of the surface (0-25 μ m) to underlying Kupffer cells.

7 **Video 2. Brain cortex.** Shows the transition from meningeal macrophages spread in the plane of
8 the surface (0-15 μ m) to underlying microglia (15-37 μ m). The second half of the video shows a 3D
9 reconstruction of microglial processes.

10 **Video 3. Heart.** Shows transition from epicardial macrophage populations spread in the plane of
11 the surface (0-20 μ m) to macrophages spread along muscle fibers in the myocardium.

12 **Video 4. Colon.** Shows transition from macrophages spread on the serosal surface (0-35 μ m) to
13 macrophages spread long longitudinal and circular muscle fibres (35-70 μ m) to macrophages
14 surrounding villi in the submucosa and mucosa.

15 **Video 5. Lung.** Shows transition from subcapsular macrophages on the pleural surface (0-20 μ m)
16 to underlying alveolar and interstitial macrophage populations.

17 **Video 6. Kidney.** Shows transition from subcapsular macrophages spread in the plane of the
18 surface to underlying interstitial macrophages surrounding tubules in the cortex (note that there
19 is red autofluorescence in the tubules).

20 **Video 7. Skeletal muscle (zygomaticus major).** Oblique series distinct morphologies of
21 macrophages of the epimysium and transition to elongated macrophages spread along muscle
22 fibers.

23 **Video 8. Abdominal wall-peritoneal side.** Shows change in polarisation of macrophage
24 populations aligned with changes in muscle fiber orientation.

25 **Video 9. Abdominal wall-skin side.** Shows change in polarisation of macrophage populations
26 aligned with changes in muscle fiber orientation.

27 **Video 10. Tendon (zygomaticus major).** Shows transition from macrophages on the surface to
28 tendon macrophages spread longitudinally along connective tissue fibers.

29 **Video 11. Pleural wall-ribs.** Shows macrophages aligned along intercostal muscle fibers and
30 transition (10-45 μm) to dense periosteal macrophage population on the surface of ribs.

31 **Video 12. Bladder-detrusor muscle.** Shows macrophages spread longitudinally along muscle
32 fibres at all depths (note muscle is spontaneously contracting).

33 **Video 13. Adrenal gland.** Shows transition from subcapsular macrophages to populations of the
34 zona glomerulosa. Note the tissue surface is not flat and subcapsular populations are progressively
35 visualized on the edge of the field.

36 **Video 14. Eye. Sclera and choroid.** Shows transition from the sclera (0-25 μm) to the choroid
37 layers of the eye. Note that the tissue is not flat and scleral populations are progressively visualized
38 on the edge of the field.

39 **Video 15. Seminal vesicle.** Shows the transition from a subcapsular macrophage population (0-
40 20 μm) through a fibromuscular layer to an underlying glandular structure region (45m-80 μm)
41 where macrophages outline the surface of glands and ducts.

42 **Video 16. Epididymis.** Shows the transition from a subcapsular macrophage population (0-20 μm)
43 to dense interstitial macrophage populations aligned with the surface of seminiferous tubules.

44 **Video 17. Van deferens.** Shows the transition from a serosal/subcapsular macrophage
45 populations of the outer layer (0-20 μm) to populations aligned with the middle (longitudinal)
46 muscle layer (20-40 μm) and then at right angles, to the circular muscle layer (note muscle is
47 spontaneously contracting).

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