

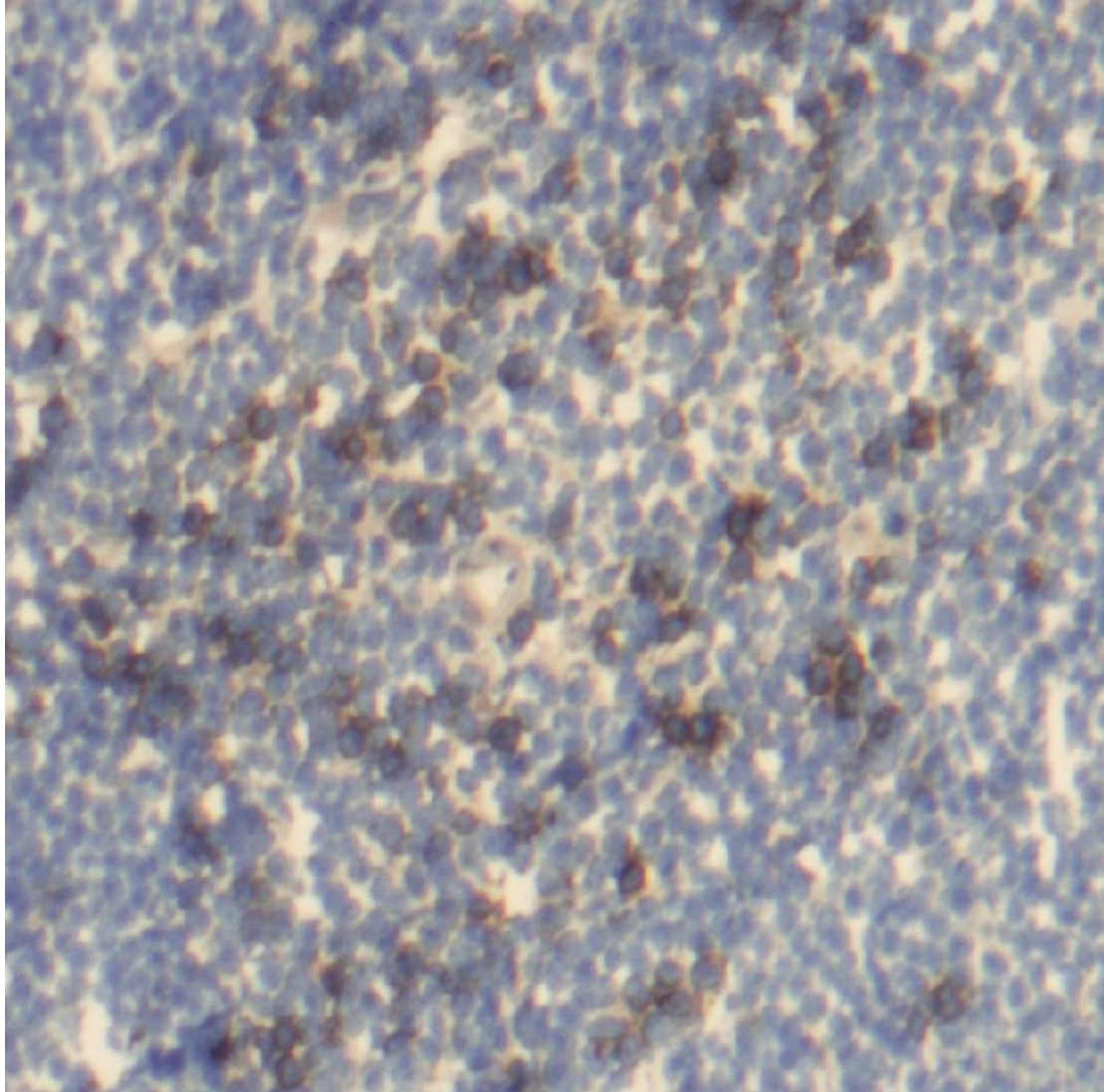
Dextramer™ staining of antigen specific T-cells on frozen tissue sections - Chromogenic detection

Protocol for acetone fixed cryosections.

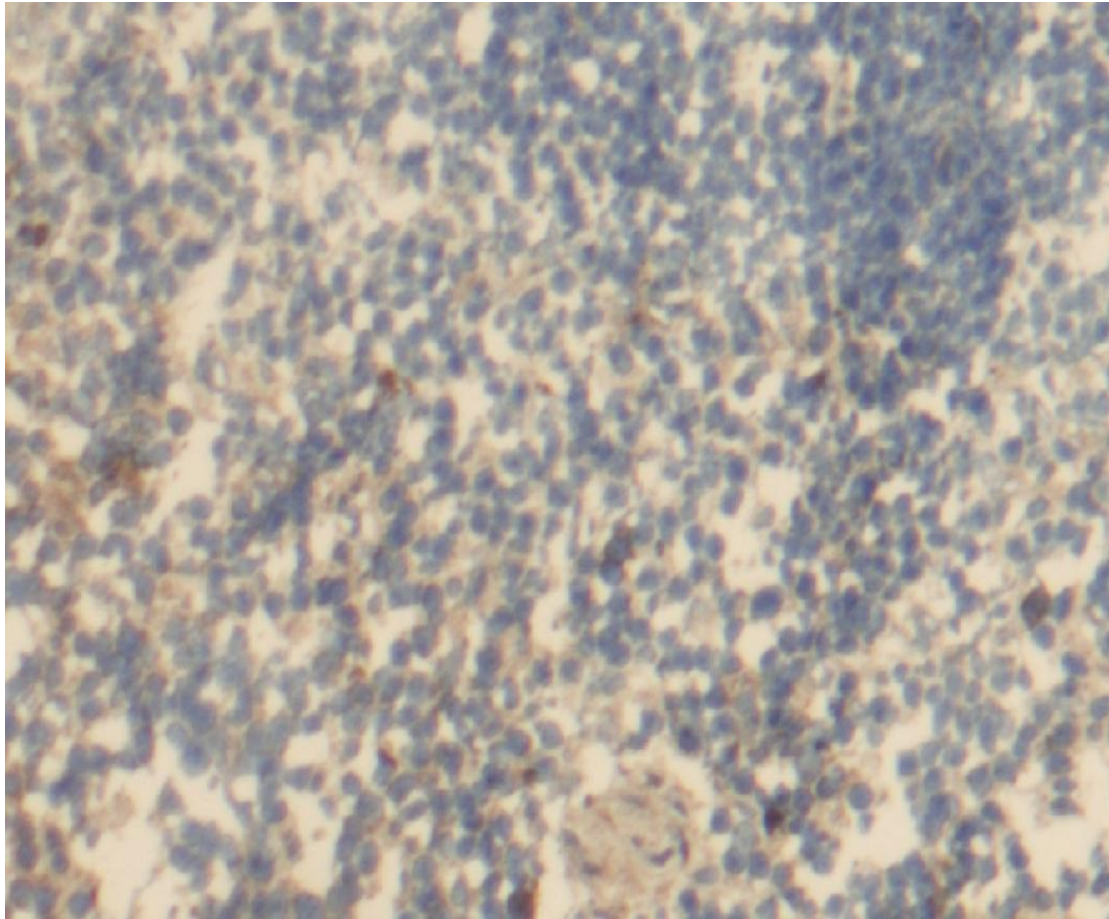
1. Equilibrate the cryo-frozen tissue to -20°C in the cryostat. Cut 5µm sections and then dry sections on slides at room temperature. Store sections frozen until use, -20°C
2. Equilibrate frozen sections to RT. Fix with acetone for 5 minutes.
3. Immediately after fixation transfer slides to TBS buffer, 10 minutes.
4. Incubate with FITC conjugated MHC Dextramer™ at appropriate dilution 1:40-1:80 for 30 minutes at room temperature
5. Decant solutions and gently tap slides against filter paper, submerge in TBS (wash buffer, 50 mM Tris-HCL pH7,6, 150 mM NaCl)
6. Decant and wash for 10 minutes in wash buffer
7. Incubate with rabbit polyclonal anti-FITC antibody (e.g. Dako P5100) at 1:100 dilution in TBS for 30 minutes at room temperature
8. Repeat step 5 and 6
9. Incubate with Envision anti-Rabbit HRP (Dako K4003) for 30 minutes at room temperature
10. Repeat step 5 and 6
11. Develop with DAB+ (Dako K3468) for 10 minutes in fume hood
12. Rinse slides in tap-water for 5 minutes
13. Counter stain with hematoxylin (Dako S3309) for 2 minutes
14. Repeat step 12, mount slides

Example

Dextramer™ staining of transgenic mouse spleen



Mouse spleen section positive stained for LCMVgp33 specific T-cells with H-2D^b(KAVYNFATC)/ FITC Dextramer™ (Immudex code JA2160-FITC). Visualized using rabbit anti-FITC/Envision anti-rabbit HRP (Dako P5100/K4003).



Mouse spleen section stained with negative LCMVnp396 Dextramer™ control H-2D^b(FQPQNGQFI)/ FITC Dextramer™ (Immudex code JA2160-FITC). Visualized using rabbit anti-FITC/Envision anti-rabbit HRP (Dako P5100/K4003).

Products and support

If you need information concerning Immudex products, have a technical enquiry, or are interested in customer-defined Dextramers and/or collaboration with Immudex, please contact us on E-mail customer@immudex.com, or phone **+45 20 437 665**

If you need ordering information, please contact us on E-mail ordering@immudex.com, or phone **+45 29 134 224**