Immunofluoresence microscopy sample preparations and stainings

1. infect cells as described for invasion assay
2. fix cells on coverslips for 25min @ RT with 4% EM grade paraformaldehyde
3. wash well with PBS
4. block 15 min with serum or BSA i.e. 1% normal goat serum if using secondary from goat
5. add primary antibody diluted in PBS + %BSA + 0.1% sapponin +1% normal goat serum (1/ 200 anti-FLAG monoclonal made in mouse)
6. incubate in humid dark chamber 1-3 @ RT or as long as O/N @ 4^0C
7. wash with PBS
8. incubate with diluted secondary antibody (dil in same as primary) ie 1/200 goat anti-mouse-TRITC
9. wash w/ PBS
10. can stain plasma membrane with DiD @ RT for 15 min
    2µl stock+ 250 µl DMSO + 1.648 ml PBS
11. wash and mount with antiquench media
12. seal all edges of coverslip with nail polish