

SUPPLEMENTAL VIDEO LEGENDS

Video 1: Lytic granules in control DMSO-treated NK cells converge upon the MTOC after CD28, or CD11a triggering. Time-lapse video of lytic granule dynamics in a DMSO-treated YTS GFP-tubulin cell on anti-CD28- (left) or anti-CD11a- (right) coated imaging chambers (Figure 2). Green fluorescence demonstrates GFP-tubulin and red, LysoTracker-loaded acidified lysosomes. The MTOC was defined as the focus of GFP intensity. Images were obtained at a rate of 6 frames per minute and at least one image per 60s is shown from T = 0min to 15min.

Video 2: PP2 inhibits NK cell lytic granule convergence after CD28, or CD11a triggering. Time-lapse video of lytic granule dynamics in a PP2-treated YTS GFP-tubulin cell on anti-CD28- (left) or anti-CD11a- (right) coated imaging chambers (Figure 2). Green fluorescence represents GFP-tubulin and red, LysoTracker-loaded acidified lysosomes. The MTOC is defined as the focus of GFP intensity. Images were obtained at a rate of 6 frames per minute and at least one image per 60s is shown from T = 0min to 15min.

Video 3: LY294002 does not block NK cell lytic granule convergence after CD28, or CD11a triggering. Time-lapse video of lytic granule dynamics in a LY294002-treated YTS GFP-tubulin cell on anti-CD28- (left) or anti-CD11a- (right) coated imaging chambers (Figure 3). Green fluorescence represents GFP-tubulin and red, LysoTracker-loaded acidified lysosomes. The MTOC is defined as the focus of GFP intensity. Images were obtained at a rate of 6 frames per minute and at least one image per 60s is shown from T = 0min to 15min.

Video 4: Wortmannin does not block NK cell lytic granule convergence after CD28, or CD11a triggering. Time-lapse video of lytic granule dynamics in a Wortmannin-treated YTS GFP-tubulin cell on anti-CD28- (left) or anti-CD11a- (right) coated imaging chambers (Figure 3). Green fluorescence represents GFP-tubulin and red, LysoTracker-loaded acidified lysosomes. The MTOC is defined as the focus of GFP intensity. Images were obtained at a rate of 6 frames per minute and at least one image per 60s is shown from T = 0min to 15min.

Video 5: PD98059 does not block NK cell lytic granule convergence after CD28, or CD11a triggering. Time-lapse video of lytic granule dynamics in a PD98059-treated YTS GFP-tubulin cell on anti-CD28- (left) or anti-CD11a- (right) coated imaging chambers (Figure 3). Green fluorescence represents GFP-tubulin and red, LysoTracker-loaded acidified lysosomes. The MTOC is defined as the focus of GFP intensity. Images were obtained at a rate of 6 frames per minute and at least one image per 60s is shown from T = 0min to 15min.

Video 6: U73122 does not block NK cell lytic granule convergence after CD28, or CD11a triggering. Time-lapse video of lytic granule dynamics in a U73122-treated YTS GFP-tubulin cell on anti-CD28- (left) or anti-CD11a- (right) coated imaging chambers (Figure 3). Green fluorescence represents GFP-tubulin and red, LysoTracker-loaded acidified lysosomes. The MTOC is defined as the focus of GFP intensity. Images were obtained at a rate of 6 frames per minute and at least one image per 60s is shown from T = 0min to 15 or 13min.

Video 7: Lytic granules converge to a granule centroid in a YTS GFP-KIR2DL1 cell conjugated to a non-cognate KIR ligand expressing target cell. Time-lapse movie of lytic granule dynamics in a YTS GFP-KIR2DL1 cell (top) conjugated to a susceptible 721 HLA-Cw3 cell (bottom; Figure 5). Green fluorescence represents GFP-KIR2DL1 and red LysoTracker-loaded acidified lysosomes. Images were obtained at a rate of 6 frames per minute and at least one image per 60 s is shown from T = 0min to 17min.

Video 8: Lytic granules still converge to a granule centroid in a YTS GFP-KIR2DL1 cell conjugated to a cognate KIR ligand expressing target cell. Time-lapse movie of lytic granule dynamics in a YTS GFP-KIR2DL1 cell (top) conjugated to a non-susceptible 721 HLA-Cw4 cell (bottom; Figure 5). Green fluorescence represents GFP-KIR2DL1 and red, LysoTracker-loaded acidified lysosomes. Images were obtained at a rate of 6 frames per minute and at least one image per 60s is shown from T = 0min to 17min.

Video 9: IL-2 induces MTOC-directed lytic granule movement in an NK cell. Time-lapse video of lytic granule dynamics in a resting YTS GFP-tubulin cell on a plain imaging chamber (Figure 7) with IL-2 added at 4 min. Green fluorescence represents GFP-tubulin and red, LysoTracker-loaded acidified lysosomes. The MTOC is defined as the focus of GFP intensity. Images were obtained at a rate of 6 frames per minute and at least one image per 60s is shown from T = 0min to 15min.

Video 10: IL-10 does not induce NK cell lytic granule convergence to the MTOC. Time-lapse video of lytic granule dynamics in a resting YTS GFP-tubulin cell on a plain imaging chamber (Figure 7) with IL-10 added at 0min. Green fluorescence represents GFP-tubulin and red, LysoTracker-loaded acidified lysosomes. The MTOC is defined as the focus of GFP intensity. Images were obtained at a rate of 6 frames per minute and at least one image per 60s is shown from T = 0min to 17min.

Video 11: ZM449829 does not block IL-2 induced NK cell lytic granule convergence. Time-lapse video of lytic granule dynamics in a ZM449829-treated YTS GFP-tubulin cell on a plain imaging chamber (Figure 7) with IL-2 added at 2min. Green fluorescence represents GFP-tubulin and red LysoTracker-loaded acidified lysosomes. The MTOC is defined as the focus of GFP intensity. Images were obtained at a rate of 6 frames per minute and at least one image per 60s is shown from T = 0min to 10min.

Video 12: PP2 inhibits IL-2 induced NK cell lytic granule convergence. Time-lapse video of lytic granule dynamics in a PP2-treated YTS GFP-tubulin cell on a plain imaging chamber (Figure 7) with IL-2 added at 4min. Green fluorescence represents GFP-tubulin and red, LysoTracker-loaded acidified lysosomes. The MTOC is defined as the focus of GFP intensity. Images were obtained at a rate of 6 frames per minute and at least one image per 60s is shown from T = 0min to 20min.