**Time-lapse movies linked to Geiser et al 2021, mBio**

***Salmonella enterica* Serovar Typhimurium Exploits Cycling through Epithelial Cells to Colonize Human and Murine Enteroids**

[**https://doi.org/10.1128/mBio.02684-20**](https://doi.org/10.1128/mBio.02684-20)

**Enteroid Time-lapse 1 - Geiser et al 2021:**

**Human enteroid microinjection procedure.**

A human enteroid was injected with *Salmonella* Typhimurium*wt* *rpsM*-mCherry and imaged by widefield DIC and fluorescence time-lapse microscopy at 1 second intervals during and after the microinjection procedure. The movie shows the middle plane of the enteroid at the z-location where the needle was inserted. The time stamper indicates the duration of the movie in seconds. Scale bar: 30µm.

**Enteroid Time-lapse 2 - Geiser et al 2021:**

***Salmonella* Typhimurium near-surface swimming atop the human enteroid epithelium.**

A human enteroid was injected with *Salmonella* Typhimurium*wt* *rpsM*-mCherry and imaged by fluorescence time-lapse microscopy at 500 millisecond intervals. The movie shows a bottom plane view on the surface of enteroid epithelium. The fluorescence time-lapse movie was overlaid with a DIC image acquired immediately before the fluorescent image series at the same location. The time stamper indicates the duration of the movie in seconds. Scale bar: 20µm.

**Enteroid Time-lapse 3 - Geiser et al 2021:**

**Expansion of the microinjected *Salmonella* Typhimurium population in the human enteroid lumen.**

A human enteroid was injected with *Salmonella* Typhimurium*wt* *rpsM*-mCherry and imaged by widefield DIC and fluorescence time-lapse microscopy at 2 minute intervals. The movie shows a middle plane view. The injected *S*.Tm population expands to fill up the entire enteroid lumen by ~5.5h p.i. Bacterial invasion events (examples indicated by arrows) and expulsion of infected intestinal epithelial cells (examples marked with asterisks) can be observed during the first hours of the time-lapse movie. The time stamper indicates the time p.i. in hours and minutes. Scale bar: 50µm.

**Enteroid Time-lapse 4 - Geiser et al 2021:**

***Salmonella* Typhimurium invasion of the murine enteroid epithelium.**

A murine enteroid was injected with *Salmonella* Typhimurium*wt* *rpsM*-mCherry and imaged by widefield DIC and fluorescence time-lapse microscopy at 3 minute intervals. The movie shows an epithelial close view. Individual bacterial invasion events (marked with arrows) can be followed over time. In addition, examples of expulsion of infected intestinal epithelial cells (marked with asterisks) can be observed. The time stamper indicates the time p.i. in hours and minutes. Scale bar: 20µm.

**Enteroid Time-lapse 5 - Geiser et al 2021:**

**Selective expulsion of *Salmonella* Typhimurium-infected intestinal epithelial cells from the murine enteroid epithelium.**

A murine enteroid was injected with *Salmonella* Typhimurium*wt* *rpsM*-mCherry and imaged by widefield DIC and fluorescence time-lapse microscopy at 3 minute intervals. The movie shows a bottom plane view. Two *S*.Tm invasion foci are marked with arrows. Please note that specifically the infected intestinal epithelial cells are expelled from the epithelial layer within the first ~75min p.i. Expelling/expelled intestinal epithelial cells are marked with asterisks. The time stamper indicates the time p.i. in hours and minutes. Scale bar: 20µm.