The timsTOF with PASEF relies on a unique dual Trapped Ion Mobility Spectrometry (TIMS) front end to provide nearly 100% duty cycle with no loss of ions. When combined with the Parallel Accumulation Serial Fragmentation (PASEF) method, the timsTOF Pro with PASEF enables MS/MS acquisition speeds in excess of 100 Hz, with improved sensitivity while maintaining all of the well-known, desirable features of the Bruker QTOF such as ultra-high resolution, ppm mass accuracy and TIP (True Isotopic Patterns). The principles of operation of PASEF on a timsTOF Pro and applications showing how this technology enables the proteomics researcher to dig deeper into the proteome will be shown.