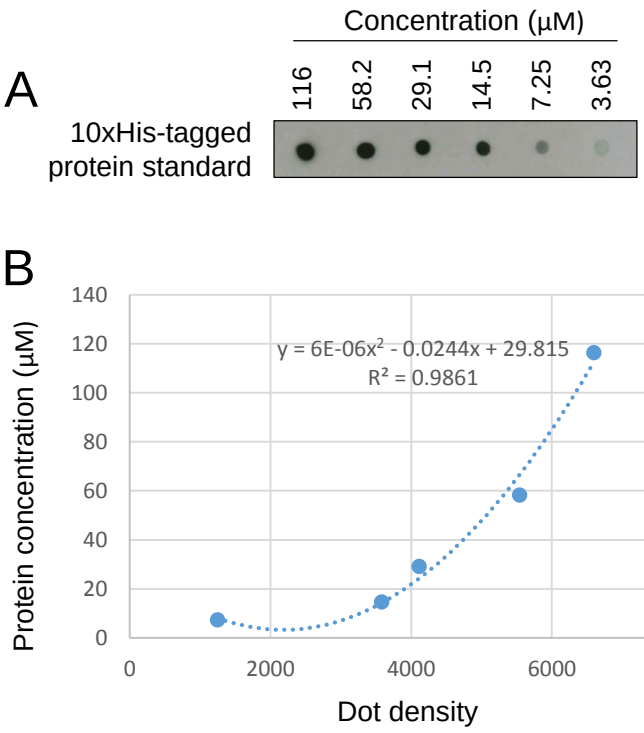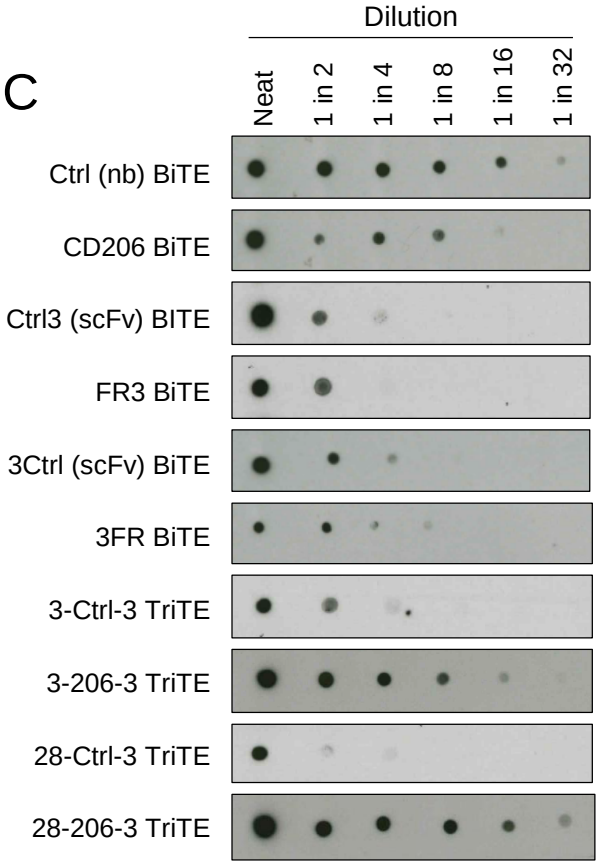Additional File 1