Unraveling the kinetics of aggregation of single peptide-DNA complexes using force spectroscopy

J. Camunas-Soler[1,2*],S. Frutos[1,2], C.V. Bizarro[1,2], S. de Lorenzo[1,2],M.E. Fuentes-Perez[3], R. Ramsch[4,2], S. Vilchez[4,2], C. Solans[4,2], F. Moreno-Herrero[3], F. Albericio[5,2], R. Eritja[4,5,2], E. Giralt[5,2], S.B. Dev[5], F. Ritort[1,2]

1) Small Biosystems Lab, Departament de Física FonamentalFacultat de Física Universitat de Barcelona, Barcelona, Spain

2) CIBER de Bioingeniería, Biomateriales y Nanomedicina, Instituto de Salud Carlos III, Madrid, Spain

3) Centro Nacional de Biotecnologia, CSIC, Cantoblanco, Madrid, Spain

4) Institut de Química Avançada de Catalunya Consejo Superior de Investigaciones Científicas (IQAC-CSIC), Barcelona, Spain

5) Institute for Research in Biomedicine(IRB Barcelona), Barcelona Science Park Baldiri Reixac 10-12,08028, Barcelona, Spain

The knowledge of the mechanisms of interaction between hydrophobic molecules and essential cellular components is key to the understanding of many aggregation processes underlying several human diseases. Kahalalide F (KF) is an hydrophobic marine-derived peptide with a strong anticancer activity which contains a positively charged residue (L-Orn). KF is an ideal model to elucidate the mechanisms by which self-aggregation competes with binding to a strongly charged polyelectrolite such as DNA. Here we carry out mechanical stretching and unzipping experiments of single DNA molecules (in double and single stranded form) complexed with KF using optical tweezers. We show that KF and DNA interact forming large aggregate complexes promoted by the recruitment and binding of DNA to the aggregate surface that are further stabilized by hydrophobic interactions. These experiments reveal unique features of the aggregation process, and the proposed methodology might be useful to quantitatively characterize other compounds or proteins in which the formation of aggregates is of relevance.

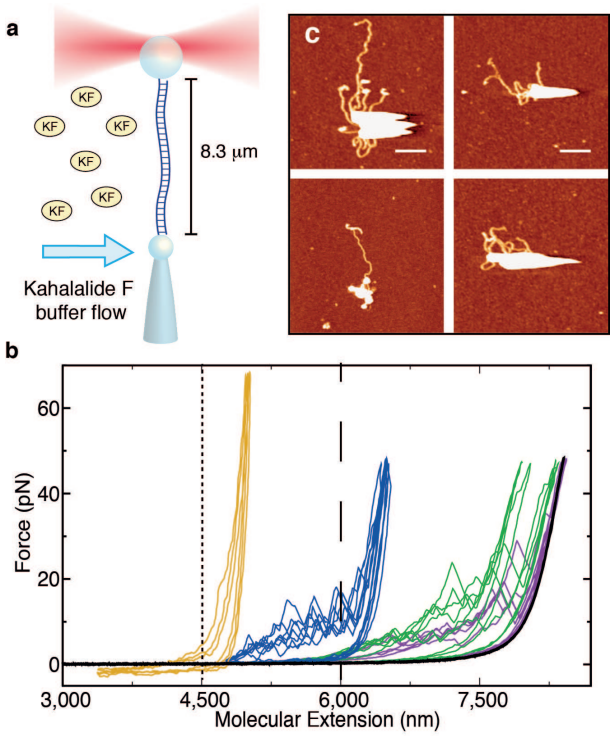

FIG. 1. **(a)** Optical tweezers experimental setup. **(b)** DNA pulling curves before (black) and after flowing KF at different waiting times: 5 min (purple), 15 min (green) and 30 min (blue). The sawtooth pattern observed indicates that KF induces the compaction of DNA. Pulling cycles reaching end-to-end distances lower than 4 μm are shown in yellow. v=500 nm/s. **(c)** AFM images of reactions of 1.65 ng linearized pGEM plasmid (2743-bp) and 100 μM KF obtained at 30 min incubation times at room temperature. The number of free DNA molecules decrease with the incubation time and large compaction blobs are observed.

* joan.camunas@gmail.com