## Fluctuation Relations and Entropy production in a Dual Trap Optical Tweezers Setup

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Recent theoretical developments in nonequilibrium statistical physics have shown how it is possible to recover free energy differences (FED) and free energy landscapes (FEL) from irreversible work measurements in thermodynamic transformations of small systems.

These developments go under the name of Fluctuation Relation (FR) and have found several applications in the field of single molecule manipulation [1], [2], [3]. A crucial concern in using FRs is the identification of the right observables which satisfy the relation: the use of the wrong observable can lead to a large error in the estimated FED or FEL [4]. In this communication we will discuss the applicability of FRs to non-equilibrium measurements obtained in Dual Trap Optical Tweezers setups (DTOTs), the standard high resolution tool in single molecule biophysics. We will show that in general three different observables satisfy a FR. In particular our study proves that the differential work measurement, based on the differential coordinate [5] (which provides the highest resolution in DTOTs) satisfies a FR and can be used in FED or FEL estimates, independently of how the pulling is carried on. The theoretical results will be confronted with experimental results obtained on several DNA tethers in a novel counter-propagating DTOT setup which directly measures forces by linear momentum conservation.

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