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**Suspended DNA structural characterization by TEM diffraction.**

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**Abstract**

In this work, micro-fabrication, super-hydrophobic properties and a physiologically compatible preparation step are combined and tailored to obtain background free biological samples to be investigated by Transmission Electron Microscopy (TEM) diffraction technique. The validation was performed evaluating a well-known parameter such as the DNA interbases value. The diffraction spacing measured is in good agreement with those obtained by HRTEM direct metrology and by traditional X-Ray diffraction. This approach addresses single molecule studies in a simplified and reproducible straightforward way with respect to more conventional and widely used techniques. In addition, it overcomes the need of long and elaborated samples preparations: the sample is in its physiological environment and the HRTEM data acquisition occurs without any background interference, coating, staining or additional manipulation. The congruence in the results reported in this paper makes the application of this approach extremely promising towards those molecules for which crystallization remains a hurdle, such as cell membrane proteins and fibrillar proteins.

**1. Introduction**

The diffraction pattern taken by Raymond Gosling under Rosalind Franklin [1] supervision, shown in the so-called “Photo 51”, has been a milestone for biology. The information contained in the image was crucial for the correct determination of the helical structure of DNA by Watson and Crick [2]. That moment changed the approach to biological sciences, highlighting the needed for new techniques capable to investigate biological matter down to the single molecule scale and below.

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