

PHOTOTOXICITY OF FLUORESCENT PROTEINS

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Fluorescent protein tags are generally regarded as fairly nonphototoxic. This appears true in the case of standard live cell imaging of fusion proteins, which employs low intensities (μW) of exciting light. However, the advent of new methods in optical microscopy, like MP, SHG, FRAP, STED brings about a need of using laser beams of considerably higher intensities.

During our studies of DNA repair mechanisms we detected DNA damage, which was apparently inflicted by eGFP tag attached to a core histone (H2B), a base excision repair protein (XRCC1) [1] or an epigenetic regulator, heterochromatin protein 1 (HP1) [1,2]. These observations constitute a warning that eGFP can cause DNA damage even when illuminated with moderate light intensities required for standard imaging. The adverse effects exerted on live cells expressing FP-tagged proteins may become even more pronounced in samples studied by new techniques, which require higher intensities of light. I will discuss the existing knowledge pertaining to adverse effects, which can be exerted on live cells by fluorescent proteins excited by light of various intensities, and consequences of these phenomena for interpretation of imaging data.

[1]. M.Kuzak et al. Exciting eGFP fusion proteins during live cell imaging can introduce DNA damage. Abstract, this book.

[2]. D. Żurek et al. Using local photodamage to study cellular repair mechanisms - potential pitfalls. Abstract, this book.