Determination Of Green Fluorescent Protein By Ultraviolet-Excited Gel Imaging

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ABSTRACT

The green fluorescent protein has gained interest in bioanalytical applications due to its visible fluorescence. As the usage of green fluorescent protein increases, more appropriate fluorescence instrumentation is required. Most fluorescence instrumentation uses ultraviolet light as the excitation source for the determination of green fluorescent protein. However, ultraviolet radiation may damage biological molecules and affect the quantitative analysis. In this study, the effects of the ultraviolet radiation period and the mass of green fluorescent protein on the fluorescence determination were characterized using gel imaging. The ultraviolet illumination period affected the green fluorescent protein fluorescence intensity. The fluorescence increased with the ultraviolet illumination time from 30–90 s. However, the fluorescence intensity decreased when the excitation period was longer, probably due to photobleaching. The photobleaching decreased when a higher concentration of enhanced green fluorescent protein was employed. This gel imaging study has provided a better understanding of the optimum conditions for the determination of green fluorescent protein.

KEYWORDS: Gel imaging, green fluorescent protein, photoactivation, photobleching, ultraviolet excitation

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