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Introduction

One of the difficulties in the development of the photodynamic therapy (PDT) is inherent to the multidisciplinary feature of this treatment gathering mainly clinicians, physicists, biologists, and chemists. Another issue is the great number of biophysical and biochemical parameters involved in the design of new photosensitizers as well as in the *in vivo* application of this treatment. We present a global development approach based on the methodology and tools of experimental design. Three study cases are developed to assess to potential relevance of such an empirical model-based approach for the development of PDT.

Methods & Results

In a first study, an *in vitro* screening experimental design was carried out. The addressed question dealt with the determination of influent factors on the phototoxicity of a new photosensitizer based on quantum dots. Five factors were examined: the nature of quantum dots, the excitation light wavelength, the incubation time with cells, the photoactivable compound concentration and the fluence level. Relevance of each factor was finally estimated and compared to identify the significant parameters. In comparison with a typical factorial design, the total number of experiments (42 trials) was divided by 5.

In a second study, an *in vivo* factorial experimental design was applied to detect potential synergic effects between four therapeutic factors: the phenotype of the cancer cell line, the food type, the nature of photosensitizer and the post-injection time, on the *in vivo* selectivity (cancer/normal tissue) of the tested photosensitizers. Results particularly pointed out the presence of a statistically significant synergic effect between these four factors and provided the optimal modalities to maximize the response in term of tumor-to-normal tissue ratio.

In a third study, a Doehlert experimental design associated with a response surface model was used to determine the *in vivo* PDT modalities (photosensitizer concentration, irradiance and fluence) to both minimize the post-treatment growth rate of the tumor and maximize its growth delay. Only 13 experimental conditions were tested and the relevance of the optimized condition was corroborated by *in vivo* validation experiments.

Conclusion

These studies have confirmed the applicability and attractive contributions of experimental design techniques in the development and determination of optimal modalities of new photosensitizers in PDT. Their main advantages are to *a priori* organize experiments according to specific questions while minimizing the experimental cost and controlling as much as possible the experimental uncertainty.