iSpinach: an optimized fluorogenic RNA aptamer

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Status des brevets

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Exclusive or non-exclusive license

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CONTEXT

The recent development of new fluorogenic dyes and the concomitant isolation of specific RNA aptamers offer the possibility to monitor in real-time the synthesis of a specific RNA both in vitro and in vivo. Among this new generation of dyes, the 3,5-difluoro-4-hydroxybenzylidene imidazolinone (DFHBI), a commercially available dye mimicking the natural fluorophore of the green fluorescent protein, proved to be particularly well suited for live-cell imaging as it is nontoxic, cell membrane-permeable, does not interact with cell components and has a low fluorescence in it is free state. These attractive properties led to the isolation of a first DFHBI-binding aptamer termed Spinach. While Spinach was able to enhance DFHBI fluorescence more than ~2000 times upon binding, it however suffered from several limitations such as limited folding efficiency and thermal instability. These limitations were partly overcome in a second version of the aptamer (Spinach2) obtained by rational design.

TECHNICAL DESCRIPTION

This invention provides an improved version of the RNA aptamer Spinach = iSpinach, a nucleic acid molecule capable of binding to a fluorophore molecule, in particular DFHBI. iSpinach is not only able to interact with the fluorophore DFHBI in the presence of potassium to form a complex more fluorescent but it can also do it in the absence of any monovalent cation and at physiological temperature.

BENEFITS

iSpinach has been developed to circumvent important limitations of the existing DFHBI-binding aptamer:
- **Efficient folding in potassium-free environment**
- **High folding efficiency**
- **High thermal stability**
- **Improved brightness**
- **1.3 times shorter than Spinach2**

INDUSTRIAL APPLICATIONS

This innovation could be used for:
- High throughput screening
- Live-cell imaging / in vitro / ex vivo imaging of small molecules, RNA and proteins

References:

Autour et al., 2016, Nucleic Acid Res. 44(6):2491-500

For further information, please contact us (Ref 09016-01)