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Enzymatic Site Specific Labeling of RNA with Unnatural Nucleobases

Tech ID: 25472 / UC Case 2015-196-0

BACKGROUND

The detection and manipulation of RNA is greatly aided by chemical modification. Therefore, there is tremendous interest in novel methods to site-specifically associate RNA with small molecules such as imaging probes and affinity labels. Conventional methodologies for detecting RNA include the use of antisense probes, aptamers, and fusion proteins that recognize specific RNA secondary structures. Relatively less explored are the use of enzymatic reactions for site-specific RNA labeling. Notably, these past approaches have not demonstrated the ability to append large functional molecules directly onto the RNA of interest. Instead they typically rely upon small bio-orthogonal handles, which after undergoing a second chemical reaction, can be modified by functional probes such as fluorophores or affinity ligands.

An ideal enzymatic reaction for labeling RNA would involve recognition of a minimal RNA structural motif, result in irreversible covalent modification, and would be capable of directly incorporating a diverse array of functional molecules such as fluorophores, affinity ligands, *etc.* in a single step.

TECHNOLOGY DESCRIPTION

Chemists from UC San Diego have addressed this need by developing an enzymatic method (RNA-TAG; transglycosylation at guanosine), to directly append large functional molecules site-specifically to RNA.

This method utilizes a bacterial enzyme (tRNA guanine transglycosylase; TGT) to exchange specific guanine nucleobases with functional derivatives of the bacterial nucleobase PreQ1. Any RNA with this particular hairpin structure, even relatively large transcripts of genes, can be tagged in this way, which should prove useful in a wide variety of biomedical and biochemical applications.

APPLICATIONS

This patent-pending RNA-TAG methodology has applications for imaging, and may also play a key role in discovering new RNA-protein interactions, drug screening applications, and exploring RNA lifetime dynamics.

RELATED MATERIALS

News Release (23-Sept-2015): Chemists Devise a New, Versatile Way to Tag RNA (Susan Brown) - 09/23/2015

Alexander SC, Busby KN, Cole CM, Zhou CY, Devaraj NK. Site-specific covalent labeling of RNA by enzymatic transglycosylation. J Am Chem Soc. 2015 Sep 22. - 09/22/2015

PATENT STATUS

Country	Туре	Number	Dated	Case
United States Of America	Published Application	20180148764	05/31/2018	2015-196
Patent Cooperation Treaty	Published Application	2016141243	09/09/2016	2015-196

Additional Patent Pending

CONTACT

University of California, San Diego Office of Innovation and Commercialization innovation@ucsd.edu tel: 858.534.5815.



OTHER INFORMATION

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La Jolla,CA 92093-0910

https://innovation.ucsd.edu

Fax: 858.534.7345

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