July 16-21, 2017 at UC San Diego, CA, USA

Simple non-coded peptides enhance RNA polymerase ribozyme function

S. Tagami[1, 2], J. Attwater[1], and P. Holliger[1*]
[1]MRC Laboratory of Molecular Biology, Francis Crick Avenue, Cambridge Biomedical Campus, Cambridge CB2 0QH, UK, [2]Present address: RIKEN Center for Life Science Technologies, 1-7-22 Suehiro-cho, Tsurumi-ku, Yokohama 230-0045, Japan
*ph1@mrc-lmb.cam.ac.uk

Introduction: Protein synthesis in modern biology depends on multiple elaborate machineries (eg. ribosomes, mRNA, tRNA, etc.). However, at the early stage of the evolution of life the translation system must have emerged as a much simpler system (or just one small ribozyme). Therefore, the peptides synthesized at that time were probably simple non-coded peptides. It is still elusive how such simple peptides could be functional without specific sequences encoded by the genetic code. To answer this question we tested if simple non-coded peptides have any benefit on function and survival of an RNA polymerase ribozyme (RPR) [1].

Experiments: RPRs have been developed and engineered in several laboratories as modern-day models of RNA replicase in the RNA world [2–5]. However, They require high concentration of Mg^{2+} (200 mM) to perform RNA synthesis efficiently [6,7]. This is problematic because the ribozyme itself is unstable in high Mg^{2+} condition [8]. Here, we describe oligo-lysine and its analogues stimulate RPR function in low Mg^{2+} condition by promoting docking between RPR and substrate RNAs (primer and template) [1]. Furthermore, oligo-lysine could accelerate in vitro evolution of RPR in low Mg^{2+} conditions. The newly engineered RPR could perform RNA synthesis at near physiological Mg^{2+} concentration in the presence of oligo-lysine (Fig 1).

Discussion: Considering RNA has strong negative charges, positively charged peptides like oligo-lysine probably enhanced interaction between RNA molecules in the RNA world. Therefore, such simple peptides could have enhanced functional networks among ribozymes and helped the life in the RNA world to evolve into more complicated cellular systems.

[1] Tagami S et al. (2017) Nature Chemistry 9:325–332. [2] Johnston WK et al. (2001) Science 292:1319–1325. [3] Wochner A et al. (2011) Science 332:209–212. [4] Attwater J et al. (2013) Nature Chemistry 5:1011–1018. [5] Horning DP & Joyce GF (2016) PNAS 113:9786–9791. [6] Attwater J et al. (2013) Chemical Science 4:2804–2814. [7] Muller UF & Bartel DP (2008) RNA 14:552-562. [8] Attwater J et al. (2010) Nature communications 1:1-8.

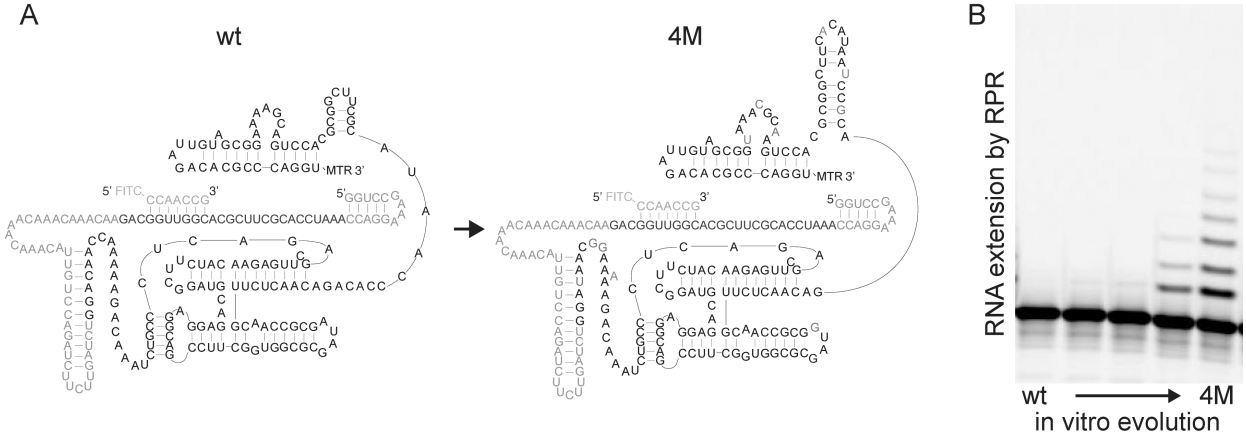

Figure 1. A) Secondary structures of RPRs before (wt) and after (4M) in vitro evolution in low Mg^{2+} conditions. B) RNA primer extension by RPR variants at 50 mM Tris-HCl (pH 8.3), 2 mM free [Mg^{2+}] and 6 μM oligo-lysine.