Molecular mechanism of antitermination

ANASTASIA SEVOSTYANOVA
AND IRINA ARTSIMOVITCH*

Department of Microbiology, The Ohio State University, Columbus, OH 43210, USA.
(*correspondence: artsimovitch.1@osu.edu)

Bacterial RNA polymerase (RNAP, $\alpha_2\beta'\beta\omega$ complex) mediates expression of all types of genes and responds to diverse regulatory inputs that fine-tune gene expression to the environmental cues. RNAP is an obligatory processive enzyme that must complete synthesis of the entire nascent RNA without releasing the transcript. While transcribing polycistronic messages, RNAP encounters numerous roadblocks; as a result, the distal genes are expressed at lower levels than the promoter-proximal ones. Diverse antiterminators act to increase RNAP processivity and ensure the coordinated expression of RNA messages.

We have previously proposed that RNAP exists in two states. In a processive, rapidly elongating state, the β' clamp and the β lobe domains tightly grip on the nucleic acid chains, forming a clamp around the downstream DNA. When RNAP encounters a roadblock, the clamp is thought to open, weakening RNAP/nucleic acids interactions and favoring isomerization into pause, arrest, and termination states. Indeed, different states of the clamp were reported in X-ray structures of bacteria and yeast transcription complexes.

Here, we use bacterial antiterminator RfaH to elucidate the molecular mechanism of antitermination modification of RNAP. Once RfaH is recruited to RNAP in the beginning of long virulence operons, it remains bound to the enzyme throughout transcription. RfaH switches RNAP into a processive state using four mechanisms. *First*, RfaH competes with σ and prevents σ-dependent arrest. Second, RfaH competes with NusG and thus indirectly reduces Rho-dependent termination. *Third*, RfaH induces forward translocation of RNAP, thereby inhibiting isomerization into an off-pathway state that occurs from a pre-translocated conformation. *Fourth*, RfaH establishes simultaneous contacts with the β' clamp and the β lobe thereby preventing the clamp opening. While the first three mechanisms may be RfaH-specific, the last mechanism is likely broadly conserved. We propose that structurally diverse antiterminators solidify the transcription elongation complex to inhibit conformational transitions that underlie pausing and termination.

Arsenic localization, speciation, and co-occurrence with Fe on rice (*Oryza sativa* L.) roots with variable Fe plaque coatings

ANGELIA L. SEYFFERTH[1]*, SAMUEL M. WEBB[2], JOY C. ANDREWS[2] AND SCOTT FENDORF[1]

[1]Stanford University, Department of Environmental Earth System Science, Stanford, CA, USA, 94305 (*correspondence: angelias@stanford.edu)
[2]Stanford Synchrotron Radiation Lightsource, SLAC National Accelerator Laboratory, Menlo Park, CA, USA 94025

Arsenic contamination of rice is widespread, but questions remain regarding the predominant root-uptake and possible attenuation mechanisms. The formation of Fe plaque around rice roots is thought to be an important barrier to As uptake, but the relative importance of this mechanism is not well characterized. The aim of this investigation was to elucidate the co-localization of As species and Fe on rice roots; we used a combination of techniques—X-ray fluorescence imaging, μXANES, transmission X-ray microscopy, and tomography—for this purpose. Two dominant As species were observed in fine roots–inorganic As (V) and As (III)–with a minor amount of dimethylarsinic acid (DMA). Our investigation shows that in roots with Fe plaque, As and Fe were strongly correlated; however, As and Fe were not correlated in roots with minimal Fe plaque. Moreover, arsenic was not exclusively associated with Fe plaque in the rice root system; in fact, Fe plaque does not coat many of the young roots or the younger portion of mature roots. Young, fine roots, important for solute uptake, have little to no iron plaque. Thus, Fe plaque does not directly intercept (and hence restrict) As supply to and uptake by rice roots but rather serves as a bulk scavenger of As predominantly near the root base.