Elongation, Termination and Antitermination: The Final Stages of Transcription in Escherichia coli

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Termination signals in bacteria such as Escherichia coli trigger the end of transcript elongation by causing dissociation of RNA polymerase (RNAP) from both the RNA transcript and template DNA, thus preventing RNAP from carrying out further processive nucleotide addition to the 3’-end of the transcript. Two principal classes of transcription terminators are known to regulate gene expression in E. coli: intrinsic (Rho-independent) terminators – whereby termination is induced by the structure and nucleotide composition of the transcript at a specific template sequence, without the need for auxiliary factors; and Rho-dependent terminators – whereby termination is induced by the actions of Rho factor (a homohexameric ring-shaped RNA-dependent ATPase with translocase and RNA:DNA helicase activities). Although some finer molecular-level details remain elusive, many of the general mechanisms and consequences of transcript elongation, termination and antitermination are by now well-characterised and supported by a substantial body of evidence. A notable caveat, however, regarding termination, is that much of our current understanding stems from investigations involving only a few model terminators – for example, λ tR2 (intrinsic), and λ tR1 (Rho-dependent) – and despite recent advances in bioinformatics, computational methods of terminator identification in the E. coli genome are typically constrained by our limited understanding of the exact sequences and structural elements involved.
Introduction

The Gram-negative, rod-shaped bacterium *Escherichia coli* is by now one of the most intensively studied and best understood organisms on Earth (Blount, 2015; Keseler *et al.*, 2013). A hardy, versatile, easily manipulable model organism, *E. coli* played a key role in elucidating the fundamental principles of life in the early days of molecular biology: including the nature of the genetic code (Crick *et al.*, 1961), as well as the processes of DNA replication (Lehman *et al.*, 1958), gene regulation (Jacob and Monod, 1961), translation (Nirenberg *et al.*, 1965) and transcription (Stevens, 1960). Fifty to sixty years later, the study of transcription in *E. coli* continues to have a significant impact on the fields of prokaryotic and eukaryotic genetics, due, in part, to the fact that the principal structural determinants of multisubunit RNAPs have been highly conserved throughout evolution (Werner and Grohmann, 2011). In addition, our enhanced understanding of the structure and function of both the bacterial RNAP and Rho factor has contributed towards the development of clinically-relevant antibiotics used in the treatment of diseases such as tuberculosis and leprosy – namely, rifampicin and bicyclomycin, which inhibit RNAP and Rho respectively (Campbell *et al.*, 2001; Vincent *et al.*, 2000).

Transcription in *E. coli* can be divided into five major stages: (i) promoter engagement, (ii) initiation, (iii) promoter clearance, (iv) transcript elongation, and (v) termination (Mooney *et al.*, 1998). Intrinsic and Rho-dependent terminators situated at the ends of operons; as well as between, within and upstream from genes, are involved in the regulation of gene expression (Peters *et al.*, 2011), and can be overridden or inhibited by the actions of various opposing ‘antitermination’ mechanisms (Santangelo and Artsimovitch, 2011). The interconnected processes of transcript elongation, termination and antitermination will be discussed, with an emphasis on the two principal types of signal which give rise to termination in *E. coli* – intrinsic terminators, and Rho-dependent terminators. While there is currently little doubt about the broad, fundamental mechanisms and consequences of both intrinsic and Rho-dependent termination, many molecular-level ambiguities remain, and this has led to the development of three major competing models of termination: (i) the ‘hybrid shearing’ model, (ii) the ‘hypertranslocation’ model, and (iii) the ‘invasion’ (allosteric) model. All three models are supported to differing extents by biochemical and single-molecule analyses (Komissarova *et al.*, 2002, Santangelo and Roberts, 2004, Epshtein *et al.*, 2007, Datta and von Hippel, 2008, Larson *et al.*, 2008, Epshtein *et al.*, 2010), and are repeatedly posited in the literature without any clear consensus. The basis for each model will be addressed, along with a number of unanswered questions and recent developments (e.g. regarding the mechanism of Rho translocation). However, transcript elongation should be considered first, as it is the breakdown of this process, through the pausing of the transcription elongation complex (TEC), which ultimately allows termination to occur.
Reducing TEC Stability

The process of transcript elongation (Figure 1) is carried out by a single, multisubunit, DNA-dependent RNA polymerase (RNAP) and involves the synthesis, via nucleotide addition, of a single-stranded RNA molecule with a nucleotide sequence complementary to that of an antisense, template strand of DNA (Mooney et al., 1998, Nudler, 1999). The *E. coli* TEC consists of a characteristic and typically dynamic arrangement of RNAP, DNA and RNA, containing a short (~8–9 bp), stable RNA:DNA hybrid within a slightly longer (12–14 bp) transcription bubble of unwound (melted) DNA (Nudler et al., 1997, Korzheva et al., 2000). Inducing a reduction in *E. coli* TEC stability or in the rate of transcript elongation, in order to facilitate termination, is not easy. During a typical phase of uninterrupted elongation, the TEC is highly stable, continuing to successfully mediate nucleotide addition at temperatures as high as 70°C (Wilson and von Hippel, 1994) and against applied forces of up to 14 pN (Yin et al., 1995). Although the stability of the RNA:DNA hybrid, and its contacts with the RNAP, account for the larger part of overall TEC stability (Sidorenkov et al., 1998), RNAP also maintains contacts with ~7 nt of the single-stranded RNA transcript as it exits via the RNA exit channel; as well as with ~14 bp of unmelted, double-stranded DNA downstream of the transcription bubble (Vassylyev et al., 2007). These protein-nucleic acid interactions are now also thought to contribute to TEC stability: for example, the rudder loop of RNAP forms stabilizing bridging contacts with the RNA:DNA hybrid and the downstream DNA (Vassylyev et al., 2007). The rate of transcript elongation can be reduced by sequences in the template DNA (Bochkareva et al., 2012), as well as by the actions of many different accessory protein transcription factors (e.g. NusA and NusG) (Schmidt and Chamberlin, 1987, Sullivan and Gottesman, 1992). This can bring the TEC to a temporary halt (pausing) or a complete halt (arrest); or induce backwards movement of the TEC towards the promoter by 1 bp, known as ‘backstepping’, or by >1 bp, known as ‘backtracking’ (Washburn and Gottesman, 2015, Belogurov and Artsimovitch, 2015, Landick, 2006, Nudler, 2012). Low elongation rates can induce or enhance termination mechanisms via paused RNAP intermediates (Schmidt and Chamberlin, 1987, Sullivan and Gottesman, 1992). Hence, antiterminators often employ strategies to prevent pausing and/or increase the elongation rate (Santangelo and Artsimovitch, 2011).
Figure 1. Summary of transcript elongation, and the relationship between elongation, pausing and termination: (a) the most-recently-added nucleoside triphosphate (NTP) at the 3’-end of the RNA transcript (GTP in this example) is blocking the 3’-most part of the RNAP active site, and hence, binding of another NTP is not possible; (b) the incoming NTP (UTP in this example) is thought to enter via the RNAP secondary channel; (c) RNAP translocation allows addition of the incoming NTP to the 3’-end of the RNA transcript; (d) displacement of 1 nt of RNA from the upstream edge of the RNA:DNA hybrid re-establishes the 8 bp RNA:DNA hybrid (Santangelo and Artsimovitch, 2011). Adapted from Nudler et al., 1997; Korzheva et al., 2000; Santangelo and Artsimovitch, 2011.
Overview of Intrinsic Termination

Intrinsic (Rho-independent) terminators in *E. coli* typically involve a relatively short (~20 nt), guanine-and-cytosine-rich (G+C-rich) region of template DNA with dyad symmetry (i.e. consisting of inverted repeats joined by a short, unrepeated intervening sequence) followed by a sequence typically consisting entirely of deoxyadenosine (dA) residues – i.e. an oligo(dA) region (Wilson and von Hippel, 1995, Lesnik *et al.*, 2001, Gusarov and Nudler, 1999). Transcription of this template DNA gives rise to a correspondingly G+C-rich region of RNA with dyad symmetry, followed by a sequence, at the RNA 3’-end, typically consisting entirely of uridine (rU) residues – i.e. an oligo(rU) region. Base-pairing of the inverted repeats in the RNA induces the formation of a stable G+C-rich RNA hairpin (stem-loop structure), which, when combined with the destabilizing dA:rU duplex of the RNA:DNA hybrid, leads to transcription termination via breakdown of the TEC and dissociation of RNAP from both DNA and RNA (Tomizawa and Masukata, 1987, Wilson and von Hippel, 1995). The process of intrinsic termination can be divided into four major stages: (i) pausing, (ii) hairpin nucleation, (iii) hairpin completion (which causes disruption of the TEC), and (iv) TEC dissociation (Peters *et al.*, 2011). Approximately 80% of the known protein-coding (mRNA) transcripts in *E. coli* are terminated by intrinsic terminators (Peters *et al.*, 2009). One example of a model intrinsic terminator is the phage λ tR2 terminator (Wilson and von Hippel, 1995).

Overview of Rho-dependent Termination

Unlike intrinsic termination, which relies principally on the structure and nucleotide composition of the nascent RNA transcript at a specific template sequence, Rho-dependent termination relies on both cis-acting RNA elements and trans-acting factors, such as Rho, NusA and NusG (Richardson, 2002, Banerjee *et al.*, 2006, Ciampi, 2006). Rho-dependent terminators in *E. coli* are bipartite elements, consisting of a Rho-utilization (rut) site and termination sites called transcription stop points (tsp) (Ciampi, 2006). The rut site – an 83–87 nt Rho-binding site on an untranslated RNA transcript (Koslover *et al.*, 2012) – has a high affinity for Rho due to the fact that it has a high proportion of cytosine residues relative to guanine, and little secondary structure (Hart and Roberts, 1991, Platt, 1994, Richardson and Richardson, 1996). Rho-dependent termination requires an untranslated (i.e. ribosome-free) RNA transcript with a minimum length of 85–90 nt (Hart and Roberts, 1994). The tsp release sites, are the RNA sites situated at the TEC, at which Rho-dependent termination occurs – these can be separated from the rut site by hundreds of nucleotides (Richardson and Richardson, 1996). The protein which mediates this termination mechanism – Rho factor – is a homohexameric ring-shaped (Skordalakes and Berger, 2003) RNA-dependent ATPase (Lowery-Goldhammer and Richardson, 1974) with translocase and RNA:DNA helicase
activities (Brennan et al., 1987). Prior to termination, Rho binds to the rut site and begins to actively thread the RNA transcript through its central cavity using energy from ATP hydrolysis (Figure 2) (Gocheva et al., 2015). In this way, Rho translocates along the transcript in a 5’->3’ direction, while maintaining contacts with the rut site—a mechanism called ‘tethered tracking’ (Gocheva et al., 2015)–before inducing termination at a transcript release site by an unknown mechanism (Ciampi, 2006). Approximately 20% of the known protein-coding (mRNA) transcripts in *E. coli* are terminated by Rho-dependent terminators (Peters et al., 2009). One example of a model Rho-dependent terminator is the phage λ tr1 terminator, which terminates the rightward phage λ operon (Lau et al., 1982).

![Diagram of Rho-rut binding and Rho 5’->3’ translocation via tethered tracking](image)

**Figure 2.** *Mechanism of Rho-rut binding and Rho 5’->3’ translocation via tethered tracking:* (a) unbound Rho in the open (lock-washer) conformation; (b) primary binding of Rho to rut (Rho is in contact with 55–59 nt of RNA); (c) during secondary binding the transcript is passed through the central cavity of Rho, and Rho undergoes a conformation change to the closed (ring) conformation (Rho is in contact with 83–87 nt of RNA); (d) Rho 5’->3’ translocation occurs via tethered tracking, the RNA is actively threaded through the central cavity of Rho using energy from ATP hydrolysis; (e) translocation continues until Rho reaches a release site at the TEC, where it terminates transcription by an unknown mechanism. Adapted from Koslover et al. (2012).
Overview of Antitermination

A wide variety of host-generated (Table 1) and phage-generated (Table 2) mechanisms are known to prevent termination in *E. coli*. The modes and sites of action of these bacterial and phage antiterminators are highly varied (Santangelo and Artsimovitch, 2011): some require specific DNA and/or RNA sequences or auxiliary co-factors in order to carry out their function; others do not; some act as general antiterminators, preventing termination at many different sites; others may be specific to only one or a small number of sites, or may be specific to preventing either intrinsic or Rho-dependent termination. Antiterminators in *E. coli* typically act to prevent termination by one or more of the following strategies: (i) direct disruption of the termination signal – that is, in the case of intrinsic termination, by destabilizing RNA secondary structure and preventing formation of the terminator hairpin (e.g. *E. coli* cold-shock proteins or BglG) (Phadtare and Severinov, 2010, Nussbaum-Shochat and Amster-Choder, 1999); or in the case of Rho-dependent termination, by disrupting the actions of Rho either by preventing binding of Rho to the RNA rut site (e.g. *E. coli* protein YaeO) (Gutierrez et al., 2007), or preventing Rho translocation along the transcript (e.g. *E. coli* protein Hfq or phage P4 protein Psu) (Ranjan et al., 2013, Rabhi et al., 2011), (ii) converting RNAP into a pause-resistant or termination-resistant form (e.g. *E. coli* proteins RfaH and S4; phage λ proteins N and Q; HK022 polymerase utilization (put) RNAs; put/put-like RNAs of phage HK639, HK75 and prophages) (Artsimovitch and Landick, 2002, Torres et al., 2001, Mason et al., 1992, Shankar et al., 2007, King et al., 2011, Komissarova et al., 2008), or (iii) inhibiting RNAP translocation (e.g. phage HK022 protein Nun) (Vitiello et al., 2014).

Antiterminators which employ the first strategy are known as passive antiterminators, and typically enable RNAP to bypass a single terminator. Those which employ the second strategy are known as active (or processive) antiterminators, and typically allow RNAP to read through multiple, consecutive terminators. Although most known antiterminators in *E. coli* fall into one of the two aforementioned categories, the Nun protein of phage HK022 is a notable exception, as it instead acts to inhibit RNAP translocation entirely (Vitiello et al., 2014), as well as to strongly prevent both RNAP and transcript dissociation (Chung Hung and Gottesman, 1995). Phage HK022 protein Nun therefore represents a third class of antiterminator. Passive antitermination mechanisms not involving host or phage proteins are also known. These typically involve stalled or translating ribosomes bound to the transcript, which can either prevent terminator hairpin formation through attenuation (Henkin and Yanofsky, 2002), or obstruct and prevent Rho binding to the rut site (Konan and Yanofsky, 2000). Active antitermination can also occur in the regions of the *E. coli* genome which encode non-protein-coding RNA transcripts (e.g. the rRNA (rrn) operons). Rho-dependent termination of rRNA transcripts is rare, as they typically have considerable secondary structure and interact heavily with ribosomal proteins during transcription (Kaczanowska and Ryden-Aulin,
Both features contribute to preventing Rho binding to rut sites, and in addition to this, an rrn antitermination complex can convert RNAP to a Rho-resistant form (Condon et al., 1995).

Table 1. Simplified summary of bacterial antiterminators in E. coli (Santangelo and Artsimovitch, 2011, with additional data derived from Artsimovitch and Landick, 2002; Condon et al., 1995; Gutierrez et al., 2007; Henkin and Yanofsky, 2002; Konan and Yanofsky, 2000; Nussbaum-Shochat and Amster-Choder, 1999; Phadtare and Severinov, 2010; Rabhi et al., 2011; Torres et al., 2001). YaeO, antitermination protein (Rho-specific inhibitor); BglG, β-glucoside (bgl) operon antiterminator protein; Csp, cold-shock proteins; Hfq, RNA-binding protein (Rho-specific inhibitor); RfaH, antiterminator protein; S4, ribosomal protein (Rho-dependent antiterminator); rrn complex, rRNA (rrn) operon antiterminator protein complex.

<table>
<thead>
<tr>
<th>Name</th>
<th>Passive/Active</th>
<th>Mechanism</th>
<th>Reference</th>
</tr>
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<tbody>
<tr>
<td>YaeO</td>
<td>Passive</td>
<td>YaeO-Rho binding inhibits Rho-RNA binding</td>
<td>(Gutierrez et al., 2007)</td>
</tr>
<tr>
<td>BglG</td>
<td>Passive</td>
<td>BglG-RNA binding prevents hairpin formation</td>
<td>(Nussbaum-Shochat and Amster-Choder, 1999)</td>
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<tr>
<td>Csp</td>
<td>Passive</td>
<td>Csp-RNA binding destabilizes RNA secondary structures and prevents hairpin formation</td>
<td>(Phadtare and Severinov, 2010)</td>
</tr>
<tr>
<td>Hfq</td>
<td>Passive</td>
<td>Hfq-Rho binding prevents Rho translocation</td>
<td>(Rabhi et al., 2011)</td>
</tr>
<tr>
<td>RfaH</td>
<td>Active</td>
<td>RfaH-RNAP binding converts RNAP to termination-resistant form</td>
<td>(Artsimovitch and Landick, 2002)</td>
</tr>
<tr>
<td>S4</td>
<td>Active</td>
<td>S4-RNAP binding converts RNAP to Rho-resistant form</td>
<td>(Torres et al., 2001)</td>
</tr>
<tr>
<td>Ribosome (i)</td>
<td>Passive</td>
<td>Ribosome-RNA interaction prevents hairpin formation</td>
<td>(Henkin and Yanofsky, 2002)</td>
</tr>
<tr>
<td>(ii)</td>
<td>Passive</td>
<td>Ribosome-RNA interaction prevents Rho-RNA binding</td>
<td>(Konan and Yanofsky, 2000)</td>
</tr>
<tr>
<td>rrn complex</td>
<td>Active</td>
<td>Converts RNAP into a Rho-resistant form</td>
<td>(Condon et al., 1995)</td>
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Table 2. Simplified summary of phage antiterminators in E. coli (Santangelo and Artsimovitch, 2011, with additional data derived from King et al., 2011; Komissarova et al., 2008; Mason et al., 1992; Ranjan et al., 2013; Shankar et al., 2007 and Vitiello et al., 2014). Psu, polarity suppression protein (Rho-specific inhibitor); N, phage λ protein N; Q, phage λ protein Q; Nun, phage HK022 protein Nun (E. coli RNAP translocation inhibitor); put, polymerase utilization.

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<tr>
<th>Name</th>
<th>Phage</th>
<th>Passive/Active</th>
<th>Mechanism</th>
<th>Reference</th>
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</thead>
<tbody>
<tr>
<td>Psu</td>
<td>P4</td>
<td>Passive</td>
<td>Psu-Rho binding prevents Rho translocation</td>
<td>(Ranjan et al., 2013)</td>
</tr>
<tr>
<td>N</td>
<td>λ</td>
<td>Active</td>
<td>Converts RNAP to termination-resistant form (aided by several host Nus proteins)</td>
<td>(Mason et al., 1992)</td>
</tr>
<tr>
<td>Q</td>
<td>λ</td>
<td>Active</td>
<td>Converts RNAP to termination-resistant form (aided by NusA)</td>
<td>(Shankar et al., 2007)</td>
</tr>
<tr>
<td>Nun</td>
<td>HK022</td>
<td>–</td>
<td>Prevents RNAP translocation</td>
<td>(Vitiello et al., 2014)</td>
</tr>
<tr>
<td>put RNAs</td>
<td>HK022</td>
<td>Active</td>
<td>Convert RNAP to termination-resistant form, via put RNA-RNAP interaction</td>
<td>(Komissarova et al., 2008)</td>
</tr>
<tr>
<td>put/put-like RNAs</td>
<td>HK639, HK75 and prophages</td>
<td>Active</td>
<td>Convert RNAP to termination-resistant form, via put/put-like RNA-RNAP interaction</td>
<td>(King et al., 2011)</td>
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Comparing and Contrasting Intrinsic Termination and Rho-dependent Termination

Intrinsic Termination

Figure 3. Current competing models of intrinsic termination: Hybrid Shearing: the 3’-end of the transcript is pulled from the RNAP active site by the extending, upstream RNA hairpin, causing RNA:DNA hybrid shearing (i.e. disruption of the complementary base-pairing between template and transcript). Hypertranslocation: the extending, upstream RNA hairpin promotes forward translocation of RNAP (without nucleotide addition) while the RNA:DNA hybrid is maintained by translocation of RNA and DNA in the opposite direction – resulting in removal of the 3’-end of the transcript from the RNAP active site, as in the hybrid shearing model. Hairpin Invasion: the 3’-end of the transcript is not pulled from the active site, rather remains in place until movement of the hairpin into the RNAP main channel promotes hybrid melting via steric interactions (Peters et al., 2011).

Rho-dependent Termination

Figure 4. Current competing models of Rho-dependent termination: Hybrid Shearing: the 3’-end of the transcript is pulled from the RNAP active site by the pulling force of the increasingly taut RNA transcript being threaded through the central cavity of Rho. This causes RNA:DNA hybrid shearing (i.e. disruption of the complementary base-pairing between template and transcript). Hypertranslocation: Rho exerts a pushing force, promoting forward translocation of RNAP (without nucleotide addition) while the RNA:DNA hybrid is maintained by translocation of RNA and DNA in the opposite direction – resulting in removal of the 3’-end of the transcript from the RNAP active site as in the hybrid shearing model. Invasion: the 3’-end of the transcript is not pulled from the active site, rather remains in place until Rho directly unwinds the RNA:DNA hybrid using its RNA:DNA helicase activity (Peters et al., 2011).
Rho Translocation

An example of a recent major advance in our understanding of Rho-dependent termination, involves the mechanism of Rho translocation (Figure 2), and the nature of the Rho-RNAP interaction. Recent chromatin immunoprecipitation and DNA microarray (ChIP-chip) analysis revealed that Rho associates directly with RNAP throughout the entire process of transcript elongation (Mooney et al., 2009). This was confirmed by an in vitro study (Epshtein et al., 2010), which appeared to show direct Rho-RNAP binding independent of proteins, DNA and RNA. It was also suggested that Rho might play a role in altering the properties of RNAP during elongation (Epshtein et al., 2010). If these findings were to be confirmed, rut site binding (or Rho-RNA interactions of any kind), would be unnecessary for Rho-RNAP association; and, assuming a Rho-RNAP association during elongation, Rho binding to rut would result in an RNA loop which would become shorter and more taut as the transcript was actively threaded through the central cavity of Rho. In fact, the opposite is true, and hence these radical findings have been challenged by other investigations, which maintain that Rho does not bind to RNAP in the absence of a rut site, nor does Rho directly associate with RNAP except during termination (Kalyani et al., 2011).

Single-molecule studies also show no evidence of direct Rho-RNAP binding (Koslover et al., 2012). As such, alternative models of Rho translocation (e.g. pure/simple translocation and rut-free translocation) have become increasingly unlikely and the previously described ‘tethered tracking’ model has gradually emerged as the most probable (Koslover et al., 2012). Support for the tethered tracking model has come, in part, from the fact that Rho can generate >200 pN of force (Schwartz et al., 2007): more than enough to overcome the hindrance of maintaining rut contacts during translocation, and surely sufficient to provide the >30 pN of force required to displace RNA from RNAP at one of several tsp release sites (Dalal et al., 2006). Recent single-molecule manipulations and fluorescence methods seem to have confirmed tethered tracking as the principal mechanism of Rho translocation in bacteria such as E. coli (Gocheva et al., 2015), and further investigation has also led to the observation that Rho can translocate against a relatively large applied force (7 pN), and can translocate approximately 2-5 times faster than RNAP (Gocheva et al., 2015).

Discussion and Conclusions

Unanswered Questions and Future Prospects

Our knowledge of transcription termination has greatly accelerated in recent years, thanks to structural and biochemical advances in our understanding of the multisubunit bacterial RNAP and TEC, as well as genetic analyses of terminators and single-molecule analyses of their associated protein factors (e.g. Rho, NusA,
NusG). However, certain fundamental elements of the termination mechanism remain elusive: for example, how Rho binding to the RNA rut site activates its ATPase activity, and if the intrinsic-terminator-associated pause involves backtracking. Further study of the interconnected mechanisms of elongation, termination and antitermination, as well as the continual and rapid development of more sophisticated investigative techniques – such as monitoring methods which allow the real-time observation of transcription (Greive et al., 2008) and backtracking (Lass-Napiorkowska and Heyduk, 2016) by E. coli RNAP using surface plasmon resonance (SPR) and various biochemical approaches – will likely lead to answers for these questions and others sometime in the near future. In addition, a greater understanding of elongation, termination and antitermination could have many practical and economical implications, due to the widespread use of E. coli in both the biotechnological and pharmaceutical industries (Blount, 2015). Further elucidation of these processes could potentially act to improve the efficiencies or yields of many commercial processes involving E. coli, including the production of biofuels (Liu and Khosla, 2010) and the production of recombinant therapeutic proteins such as insulin (Goeddel et al., 1979).

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References


STEVENS, A. (1960). Incorporation of the adenine ribonucleotide into RNA by cell fractions from *E. coli*. *Biochemical and Biophysical Research Communications*, 3, 92-96.


