REVIEW

ACS chemical biology

Alternative Polyadenylation: A Twist on mRNA 3′ End Formation

Carol S. Lutz*

Department of Biochemistry and Molecular Biology, University of Medicine and Dentistry of New Jersey—New Jersey Medical School, MSB E671, 185 South Orange Avenue, Newark, New Jersey 07101

ABSTRACT Regulation of gene expression by RNA processing mechanisms is now understood to be an important level of control in mammalian cells. Regulation at the level of RNA transcription, splicing, polyadenylation, nucleo-cytoplasmic transport, and translation into polypeptides has been well-studied. Alternative RNA processing events, such as alternative splicing, also have been recognized as key contributors to the complexity of mammalian gene expression. Pre-messenger RNAs (pre-mRNAs) may be polyadenylated in several different ways due to more than one polyadenylation signal, allowing a single gene to encode multiple mRNA transcripts. However, alternative polyadenylation has only recently taken the field as a major player in gene regulation. This review summarizes what is currently known about alternative polyadenylation. It covers results from bioinformatics, as well as those from investigations of viral and tissue-specific studies and, importantly, will set the stage for what is yet to come.

RNA is the intermediary in the information transfer process, standing between the genetic material, DNA, and the end product, the translated protein. Messenger RNA transcripts (mRNAs) in eukaryotic cells have, with few exceptions, similar architecture and features. Most mRNAs have 5′ and 3′ untranslated regions (UTRs), exons, and introns, usually thought of as "coding" and "noncoding" regions, respectively, a special modified base at the 5′ end, called a "cap", and a number of signals present within the mRNA that signal translational "start" and "stop" (Figure 1).

Almost all eukaryotic mRNAs acquire an uncoded poly(A) tail at their 3′ ends in a process called polyadenylation (reviewed in refs 1—5). This process involves two tightly coupled steps, in which the mRNA is first cleaved at a specific site, and then adenosine (A) residues are added in a nontemplated fashion. The process of 3′ end formation is interconnected to other transcriptional and post-transcriptional processes, such as splicing and transcriptional termination (reviewed in refs 6—8). Defects in 3′ end formation can drastically affect the development, growth, and viability of a cell.

The Mechanism of Polyadenylation. Virtually all mammalian polyadenylation signals contain the consensus sequence AAUAAA (or a variant) between 10 and 35 nucleotides upstream of the actual cleavage and polyadenylation site (Figure 2; ref 9 and references therein). In addition, sequences 14—70 nucleotides downstream of the polyadenylation signal (the U/GU rich binding site for the multisubunit cleavage stimulation factor, or CstF) are known to be involved in directing polyadenylation (*10–22*). These two elements are often termed the "core" polyadenylation elements. Auxiliary elements both upstream and downstream of the core elements have also been characterized that can enhance polyadenylation efficiency and have been identified in both

*Corresponding author, lutzcs@umdnj.edu.

Received for review June 10, 2008 and accepted August 21, 2008.

Published online September 26, 2008

10.1021/cb800138w CCC: $40.75

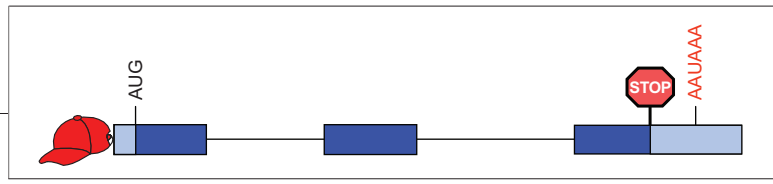

Figure 1. A schematic of a mammalian pre-mRNA. Blue boxes indicate exons, and black lines indicate introns. AUG indicates the translational start codon for the open reading frame, and the stop sign indicates the translational stop codon. AAUAAA indicates a polyadenylation signal, and the red baseball cap is representative of the monomethylated guanosine cap added at the 5′ end of the mRNA. Lighter blue regions represent the 5′ and 3′ UTRs.

viral and cellular systems (*23–41*). Spacing of these elements relative to the core elements is significant in their influence on polyadenylation (*24, 26, 27, 30, 41*). Often these auxiliary elements are polyadenylation efficiency elements; however, they may also provide additional functions in proper processing.

In addition to the RNA sequence elements, five multisubunit protein factors make up the core mammalian polyadenylation and cleavage machinery: cleavage and polyadenylation specificity factor (CPSF), CstF, cleavage factors I_m and II_m (CF I_m and CF II_m), and poly(A) polymerase (Figure 2; refs 1−5).The polyadenylation machinery assembles on the pre-mRNA before any reactions take place. Cleavage occurs 10−30 residues after the AAUAAA; the nuclease responsible for this specific cleavage has been reported to be CPSF 73 (*42*).

Polyadenylation and 3′ end formation play a critical role in gene expression because improperly processed mRNAs are not transported out of the nucleus into the cytoplasm to be translated. Likewise, inefficiently processed mRNAs may be transported out of the nucleus with lower frequency than those mRNAs that are efficiently processed. Poly(A) tails have also been shown to influence mRNA stability, and translation (reviewed in refs 43−46). Polyadenylation, therefore, is a fundamental aspect of gene expression.

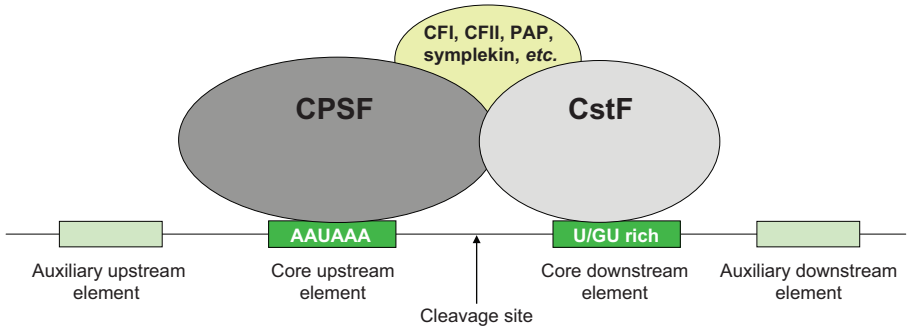

Figure 2. A schematic of RNA sequences and protein factors involved in polyadenylation. Green boxes are core polyadenylation elements; light green boxes are potential auxiliary elements that may or may not be present in a polyadenylation signal. Ovals are basal polyadenylation machinery as indicated.

A single series of processing events may take place on a pre-mRNA in the simplest of scenarios. However, it is becoming increasingly clear that this simplest scenario is often not the rule. Alternative processing, that is, choice of one or more distinct processing events, of mRNAs is now recognized as an important regulatory level in gene expression control (reviewed in refs *9* and 47−49). In alternative splicing events, different mRNA transcripts could be generated from alternative use of splicing signals. Alternative polyadenylation is defined as use of more than one polyadenylation signal. Taken together, alternative processing events could result in RNA transcripts that could differ in subtle ways, such as having different length 3′ UTRs, or in more dramatic ways, such as encoding different proteins with different domains. These alternative processing events can then, in turn, influence transcript localization, stability, and transport. Although alternative splicing is an incredibly important mechanism of generating transcript diversity and in regulation of gene expression (reviewed in refs 47 and 48), this review will focus on the lesser-known and previously under-appreciated mechanism of alternative polyadenylation and, in particular, alternative polyadenylation in mammals.

It has only been recently appreciated that more than half of the genes in the human genome are alternatively polyadenylated (*9*). Therefore, a closer understanding of alternative polyadenylation as a mechanism for transcript diversity and gene regulation is warranted. The current review is directed primarily at highlighting discoveries on mammalian alternative polyadenylation that have been made in the past several years. The many consequences of alternative polyadenylation will be discussed at the end of this review, as well as emerging questions in the field. Several other reviews (*4, 5, 47*) have discussed aspects of polyadenylation and alternative polyadenylation, including the implications of polyadenylation on health and disease (*50*). In addition, recent studies on alternative polyadenylation in plants and algae, specifically rice, *Arabidopsis*, and *Chlamydomonas*, is yielding fascinating results that may have important ramifications for our understanding of mammalian alternative polyadenylation (*51–53*).

Global Studies on Alternative Polyadenylation. With the completion of the human genome sequencing project, as well as other mammalian sequencing projects, the opportunities for exploring and understanding polyadenylation and alternative polyadenylation events expanded tremendously. Many of these studies have used the large amount of available data such as expressed sequence tags (ESTs), cDNAs, or microarray data in new and different ways, or developed new techniques such as tiling arrays to explore the complex transcriptional landscape. One of the most striking findings was that the number of transcripts encoded in the mouse and human genomes were much larger, up to 10 times or more, than the number of "genes" (refs 54 and 55 and references therein). This transcript variation represents, among other things, alternatively polyadenylated mRNAs.

Several recent reports have examined mammalian polyadenylation signals and patterns using available genomic information coupled with bioinformatics algorithms. These studies have determined that more than half of mammalian genes are alternatively polyadenylated and, furthermore, that many alternative polyadenylation signals are evolutionarily conserved (*9, 56, 57*). The numbers are much higher than had previously been expected, perhaps because less information was available five or six years ago. What this means in the most simplistic of terms is that the mRNA landscape is richer and more complex than previously anticipated.

The knowledge that alternative polyadenylation is more widespread and complex than previously anticipated also impacts other studies of gene expression regulation, for example, splicing. Alternative polyadenylation may or may not be affected by splicing; type I polyadenylation is characterized by only one polyadenylation signal in an mRNA and is not affected by splicing (Figure 3; *9*). Type II alternative polyadenylation as described by Tian et al. (*9*) has all alternative polyadenylation signals in the 3′ most exon and therefore is not affected by splicing either (Figure 3). However, type III alternative polyadenylation is characterized as having at least one alternative polyadenylation event coupled with an alternative splicing event (Figure 3; *9*). A classic example of type III polyadenylation is the immunoglobulin mu system in B cells, where use of one polyadenylation signal during development leads to the secreted form, while use of the other polyadenylation and the accompanying alternative splice leads to the membrane-bound form (*58*). Now that alternative polyadenylation is known to occur quite frequently, a number of studies have shown dynamic interplay between these two processes (splicing and polyadenylation), the results of many of which lead to variant protein products (*59, 60*). It should also be noted that in one of the most common techniques used to examine gene expression, the microarray, resulting data can also be affected by alternative splicing and polyadenylation (*61*), depending on the probe used for the microarray experiments. This could affect inaccurate interpretation of data from such experiments and underscores the need for further appreciation and study of alternative polyadenylation.

Tissue-Specific Studies on Alternative Polyadenylation. Alternative polyadenylation of specific mRNAs may be regulated differently in different tissues in response to spatial, developmental, or functional needs. The question of tissue-specific alternative polyadenylation has recently generated a number of studies, some of which are bioinformatic reports, while other studies used more conventional molecular biology approaches. The bioinformatic approaches, by nature, examined broad databases of information to search for patterns, as opposed to research on a specific gene or family of genes. An early bioinformatic approach using available EST data (Oct 2000 dbEST) and their program termed *ESTparser* first suggested a correlation between alternative polyadenylation and tissue- or disease-specific forms (*62*). Since then, EST databases have improved and grown such that a more comprehensive and refined picture is now being revealed. One recent study found that distinct alternative polyadenylation signal biases, that is, biased use of a particular alternative polyadenylation signal, are found in reproductive tissues, such as testis, uterus, and placenta, and eye tissues, including the retina (*63*). Also, many interesting patterns of usage

KEYWORDS

3′ UTR: The noncoding or untranslated region at the 3′ end of a mRNA. It immediately follows the stop codon and includes regulatory sequence elements, such as those that direct formation of the poly(A) tail, as well as sequence elements that regulate mRNA translation, mRNA stability, and binding sites for microRNAs (miRNAs).

mRNA: Messenger ribonucleic acid. mRNA is transcribed from DNA by an RNA polymerase. Most mRNAs serve as templates for protein products. Mature mRNAs contain untranslated regions at the 5′ and 3′ ends, as well as a modified base at the 5′ end called a "cap" and a poly(A) tail at the 3′ end.

Polyadenylation: The process by which an unencoded polyadenylyl (poly(A)) chain is added to the 3′ end of a maturing mRNA. This reaction is mediated by multiprotein complexes, which first cleave the pre-mRNA and subsequently add the poly(A) tail.

Alternative polyadenylation: Pre-mRNAs may be polyadenylated in several different ways due to more than one polyadenylation signal, allowing a single gene to encode multiple mRNA transcripts. Sometimes these transcripts differ only in their 3′ end, and sometimes they encode entirely different proteins.

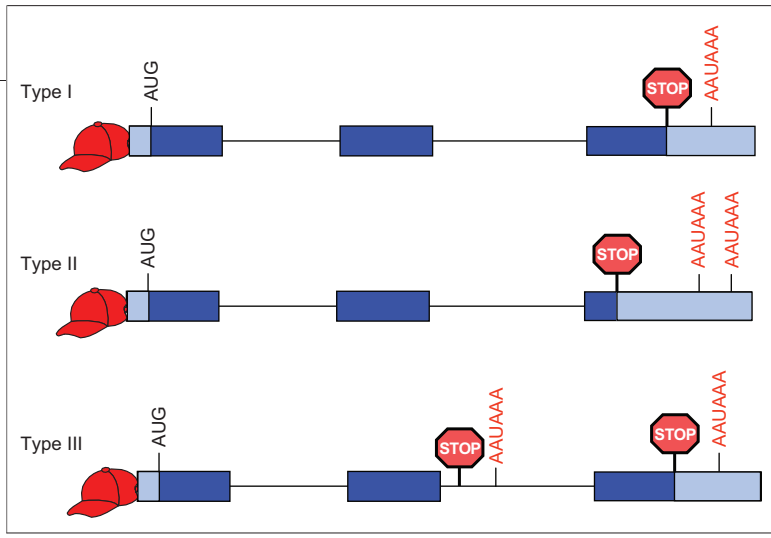

Figure 3. Types of alternative polyadenylation. As described in ref 9, there are three basic types of alternative polyadenylation. Type I polyadenylation can be characterized as constitutive polyadenylation, because only one polyadenylation signal is present in the 3′ UTR, and is actually not alternative polyadenylation but is included here for comparison. Type II alternative polyadenylation has more than one polyadenylation signal, but all polyadenylation signals are present in the 3′ most exon. Type III alternative polyadenylation involves alternative splicing coupled with alternative polyadenylation.

were found in the brain (*63*) and in spermatogenesis (*64*). Finally, alternative polyadenylation patterns in mammals seem to be conserved in evolution (*56*), suggesting selection pressure exists to keep these in place. These bioinformatic studies will provide an interesting basis for validation in molecular biology laboratories in the near future.

Tissue-specific polyadenylation of mRNA transcripts has also been explored for a number of specific genes. Although tissue-specific expression of some core polyadenylation factors themselves can and does occur (*65–67*), this has not yet been found to be a general driving force in tissue-specific alternative polyadenylation. Here, I will mention a few examples of tissue-specific alternative polyadenylation of mRNA transcripts from specific, noncore polyadenylation factor genes; there are certainly many more that could be discussed. Several examples are present in the recent literature demonstrating tissue-specific alternative splicing resulting in alternatively polyadenylated mRNAs (*68–72*). These are all excellent examples of type III polyadenylation. Because of the recent nature of these studies, the mechanism(s) of the alternative splicing (i.e., tissue-specific expression of a regulatory splicing factor) have not yet been elucidated.

A connection with transcription and alternative polyadenylation has also been explored. The β-adducin gene gives rise to two transcripts, an ~8–9 kb mRNA only present in brain tissues in human, rat, and mouse and an ~3–4 kb mRNA is found in other tissues (*73*). The alternative polyadenylation signal that is used in this unusually long 3′ UTR in the brain is highly conserved over ~180 bases in these three species and uses a suboptimal, conserved AGUAAA core upstream element. In an interesting twist, a brain-specific promoter was found in conjunction with the rat and mouse β-adducin brain-specific alternatively polyadenylated mRNAs, but this promoter is not found in humans (*73*). The connection between cellular promoter choice and polyadenylation signal choice has been further studied in some other genes. Interestingly, a recent study suggests that tissue-specific expression of the X-linked gene MID1, involved in a malformation syndrome of midline structures called Opitz syndrome, is regulated by concerted action of alternative promoters and alternative polyadenylation signals (*74*). In this study, it was shown that cellular choice of a particular promoter dictated which polyadenylation signal would define the 3′ end of the mRNA; however, a mechanism for how this works is still unclear (*74*), although links between transcription initiation and transcription termination have been discussed (*7, 75*).

Finally, two recent studies report a connection between tissue-specific alternative polyadenylation and translational efficiency (*76, 77*). A human apoptosis-inducing gene called hap was identified to have two different sized mRNA transcripts, ~1.8 and ~2.7 kb, generated by alternative polyadenylation (*76*). The larger of the two transcripts was found to be highest in brain and lowest in liver, while the ~1.8 kb transcript was highest in testis and pancreas (*76*). Chloramphenicol acetyltransferase (CAT) assays using reporter constructs containing the polyadenylation signals from each of the two transcripts showed that the ~2.7 kb transcript polyadenylation signal up-regulated translational activity ~3-fold, while the overall levels of mRNA remained the same (*76*). Translational regulation coupled with alternative polyadenylation can have a negative effect as well. BZW1 is a member of the bZIP superfamily of transcription factors, with a regular mRNA size of ~2.9 kb, but a novel ~1.8 kb transcript was recently identified in the mouse testis (*77*). An AGUAAA suboptimal polyadenylation signal is found 19 nucleotides downstream of the stop codon, and use of this polyadenylation signal gives rise to the testis-specific transcript (*77*). Most interestingly, reporter constructs using this suboptimal poly-

adenylation signal, as compared with the optimal AAUAAA associated with the ~2.9 kb transcript, had the lowest translational efficiency. This is likely due to different sequences in the 3′ UTR, directed by the chosen polyadenylation signal, suggesting that choice of a particular polyadenylation signal may be able to regulate translation of the mRNA. Although having a poly(A) tail has long been correlated with efficient translation of an mRNA, this particular connection has not been well explored and warrants further study.

Viral Studies on Alternative Polyadenylation. It has long been appreciated that viral genomes are often very compact and use genetic information to its fullest potential. Many viral genomes are highly alternatively spliced and polyadenylated. Recent studies describe transcriptional profiles of previously uncharacterized viral transcripts. Transcripts from two parvoviruses, Aleutian mink disease virus and Bocavirus bovine parvovirus, have been recently characterized by the Pintel laboratory and show multiple, extensively processed mRNAs that are both alternatively spliced and polyadenylated, but in each case, they are derived from a single transcriptional promoter (*78, 79*). Papillomavirus type 16 (HPV16) and Kaposi's sarcoma herpesvirus also have many bicistronic or polycistronic transcripts that are widely processed through both alternative splicing and polyadenylation mechanisms (*80–84*). HPV16 has been well-characterized; it has two polyadenylation signals, early and late, which are defined relative to their temporal use during infection. The early polyadenylation signal is a type III polyadenylation signal and is found in an intron (reviewed in refs 81−84). The late polyadenylation signal of HPV16 is tightly linked to keratinocyte differentiation and is "off" at early times of infection and is turned "on" at late times. The on/off switch is controlled by a complex of cellular proteins that interact with the HPV 3′ UTR, including SF2/ASF, the U1snRNP, and CUG-BP1 (*85–87*). Avian retroviruses have inefficient polyadenylation and are actually often polyadenylated at the polyadenylation signal of downstream host genes, yet another twist on the alternative polyadenylation story (reviewed in ref 88). Although the HPV16 story has been well-studied, it is not known or yet fully appreciated in the cases of Kaposi's sarcoma-associated and avian retroviruses the mechanisms by which alternative polyadenylation takes place in the virally infected cells, for example, if viral proteins promote or interfere with polyadenylation as is the case for influenza virus (*89–91*).

Alternative polyadenylation in HIV presents a different set of interesting challenges because HIV viral RNA sequences have repetitive regions at their 5′ and 3′ ends. Both ends contain polyadenylation signals, but the 3′ polyadenylation signal would direct cleavage and polyadenylation of the mRNA transcript. Many studies over the years have postulated and demonstrated various reasons for why the 5′ polyadenylation signal is not used, including promoter occlusion, lack of an upstream enhancer element, and presence of a major splice donor site for U1snRNP binding, which is inhibitory (28, 34−36, and 92 and references therein). A new twist was introduced in a recent study by the Shapiro laboratory (*93*), which used computational folding programs to examine multiple strains of HIV-1 to make RNA structural predictions. They used the massively parallel genetic algorithm (MPGA fold) to examine the folding propensities of the 5′ and 3′ poly(A) signals from nine different HIV-1 strains and found that the 5′ poly(A) signal is dominantly in a hairpin motif that inhibits premature polyadenylation (*93*). This structural inhibition has previously been shown experimentally. The 3′ polyadenylation signal was shown, on the other hand, to have a propensity to be associated with a linear structure, which would also be more advantageous to polyadenylation signal use (*93*). These studies raise questions for future exploration of the role of structure in polyadenylation signal choice, not only of viral mRNAs but also of cellular mRNAs.

Emerging Questions in the Field. While the observations described here make it clearer than ever that alternative polyadenylation is a powerful yet underexplored gene expression mechanism, there are many large questions in the field that remain unanswered. Is the mechanism of alternative polyadenylation different than constitutive polyadenylation, or does it use the same machinery? Are additional protein factors or sequence elements involved, as in the case of alternative polyadenylation of the COX-2 gene (*26, 94*) and prothrombin (*95, 96*)? Many of the protein factors involved in alternative polyadenylation of COX-2 and prothrombin are also splicing factors, suggesting further coordination of polyadenylation and splicing events (*94, 95*). Other as-yet unexplored genes may also have a similar set of RNA elements and RNA binding proteins that may regulate their expression in a similar fashion. Do these factors in-

teract with the basal polyadenylation machinery to accomplish alternative polyadenylation? If so, how? If not, is there an alternative machinery or parts of the machinery? And how did alternative polyadenylation evolve in the first place? Might alternative polyadenylation regulate gene expression in some very interesting genetic ways, such as the non-Mendelian inheritance mode termed genomic imprinting, as recently suggested for the mouse H13 gene (*97*)? Or might specific histone modification and chromatin architecture influence alternative polyadenylation (*98*)? And would these mechanisms be universal or gene-specific?

It should also be considered that there may be an as-yet undeciphered "code" that may regulate aspects of alternative polyadenylation. Candidates for such a code might include post-transcriptional modification of the core polyadenylation machinery, such as phosphorylation or dephosphorylation (*99, 100*). A complex, combinatorial code involving RNA elements, RNA-binding proteins, and specific progesterone-mediated phosphorylation/dephosphorylation of polyadenylation factors was recently described for the different yet related process of cytoplasmic polyadenylation in *Xenopus* oocytes (*101*). Might nuclear alternative polyadenylation use a similar complex but elegant code?

More unanswered questions arise when we consider the ever-expanding field of microRNA (miRNA) regulation of gene expression. miRNAs are small, noncoding RNAs that can regulate mRNA expression *via* up- or down-regulating transcription, splicing, translation, differentiation, and development (reviewed in refs 102−105 and references therein). Might miRNAs also be involved in regulating alternative polyadenylation? Because miRNA target binding sites are often found in 3′ UTRs, the connection is quite plausible and was recently suggested to play a part in regulation of such global cellular programs as cellular proliferation (*106*).

Ultimately, what are the consequences of alternative polyadenylation? One consequence is the generation of different mRNAs that encode different protein products, such as would be the result of type III alternative polyadenylation. Examples include related proteins, such as the immunoglobulin example given, but also very different proteins such as calcitonin and CGRP. Another straightforward consequence of alternative polyadenylation is the prospect of generating mRNA products with differential stability, which could ultimately lead to more or less protein product, depending on enhanced or diminished RNA half-life. Might alternative polyadenylation be a "rescue pathway" from nonsense-mediated decay surveillance, as suggested by Gilat and Shweiki (*107*)? It is appreciated that polyadenylation is directly linked to transcription termination (reviewed in refs 8, 73, and 90), but how does alternative polyadenylation interface with termination? Can alternative polyadenylation be post-transcriptionally modulated by nuclear mRNA retention and cell stress, as was suggested for the CTN−mCAT2 gene pair (*108*)? Perhaps there are even more under-explored consequences of alternative polyadenylation, including subcellular localization and translational regulation. Taken together, the emerging topic of alternative polyadenylation has many interesting questions to address in the years to come.

Acknowledgment: The author wishes to thank Jeff Wilusz, Bin Tian, Lisa Canto, Sarah Darmon, and Catherine Newnham for comments and critical reading of the manuscript. C.S.L. was supported by National Science Foundation Award MCB-0462195 and National Institutes of Health Award 5R03AR052038-03.

REFERENCES

1. Wahle, E., and Kuhn, U. (1997) The mechanism of 3′ cleavage and polyadenylation of eukaryotic pre-mRNA, *Prog. Nucl. Acids Res. Mol. Biol. 57*, 41–71.
2. Keller, W., and Minvielle-Sebastia, L. (1997) A comparison of mammalian and yeast pre-mRNA 3′ end processing, *Curr. Opin. Cell Biol. 9*, 329–336.
3. Colgan, D. F., and Manley, J. L. (1997) Mechanism and regulation of mRNA polyadenylation, *Genes Dev. 11*, 2755–2766.
4. Zhao, J., Hyman, L., and Moore, C. (1999) Formation of mRNA 3′ ends in eukaryotes: mechanism, regulation and interrelationships with other steps in mRNA synthesis, *Microbiol. Mol. Biol. Rev. 63*, 405–445.
5. Edmonds, M. (2002) A history of poly(A) sequences: from formation to factors to function, *Prog. Nucl. Acids Res. Mol. Biol. 71*, 285–389.
6. Shatkin, A. J., and Manley, J. L. (2000) The ends of the affair: capping and polyadenylation, *Nat. Struct. Biol. 7*, 838–842.
7. Maniatis, T., and Reed, R. (2002) An extensive network of coupling among gene expression machines, *Nature 416*, 499–506.
8. Proudfoot, N. J, Furger, A., and Dye, M. J. (2002) Integrating mRNA processing with transcription, *Cell 108*, 501–512.
9. Tian, B., Hu, H., Zhang, H., and Lutz, C. S. (2005) A large-scale analysis of mRNA polyadenylation of human and mouse genes, *Nucleic Acids Res. 33*, 201–212.
10. Gil, A., and Proudfoot, N. J. (1984) A sequence downstream of AAUAAA is required for rabbit beta-globin mRNA 3′ end formation, *Nature 312*, 473–474.
11. Gil, A., and Proudfoot, N. J. (1987) Position-dependent sequence elements downstream of AAUAAA are required for efficient rabbit beta-globin mRNA formation, *Cell 49*, 399–406.

12. McDevitt, M. A., Imperiale, M. J., Ali, H., and Nevins, J. R. (1984) Requirement of a downstream sequence for generation of a poly(A) addition site, *Cell 37*, 993–999.
13. McDevitt, M. A., Hart, R. P., Wong, W. W., and Nevins, J. R. (1986) Sequences capable of restoring poly(A) site function define two distinct downstream elements, *EMBO J. 5*, 2907–2913.
14. Cole, C. N., and Stacy, T. P. (1985) Identification of sequences in the herpes virus thymidine kinase gene required for efficient processing and polyadenylation efficiency, *Mol. Cell. Biol. 4*, 1460–1468.
15. Sadofsky, M., and Alwine, J. C. (1984) Sequences on the 3′ side of hexanucleotide AAUAAA affect efficiency of cleavage at the polyadenylation site, *Mol. Cell. Biol. 4*, 1460–1468.
16. Sadofsky, M., Connelly, S., Manley, J. L., and Alwine, J. C. (1985) Identification of a sequence element on the 3′ side of AAUAAA which is necessary for simian virus 40 late mRNA 3′ end processing, *Mol. Cell. Biol. 5*, 2713–2719.
17. Conway, L., and Wickens, M. (1985) A sequence downstream of AAUAAA is required for formation of simian virus 40 late mRNA in 3′ termini in frog oocytes, *Proc. Natl. Acad. Sci. U.S.A. 82*, 3949–3953.
18. Zhang, F., and Cole, C. N. (1987) Purification of a complex associated with processing and polyadenylation in vitro of herpes simplex type 1 thymidine kinase precursor mRNA, *Mol. Cell. Biol. 7*, 3277–3286.
19. Wilusz, J., Feig, D. I., and Shenk, T. (1988) The C proteins of heterogeneous nuclear ribonucleoprotein complexes interact with RNA sequences downstream of the polyadenylation cleavage sites, *Mol. Cell. Biol. 8*, 4477–4483.
20. Wilusz, J., and Shenk, T. (1990) A uridylate tract mediates efficient heterogeneous nuclear ribonucleoprotein C protein-RNA crosslinking and functionally substitutes for the downstream element of the polyadenylation signal, *Mol. Cell. Biol. 10*, 6397–6407.
21. Ryner, L. C., Takagaki, Y., and Manley, J. L. (1989) Sequences downstream of AAUAAA signals affect pre-mRNA cleavage and polyadenylation in vitro both directly and indirectly, *Mol. Cell. Biol. 9*, 1759–1771.
22. Chen, F., MacDonald, C. C., and Wilusz, J. (1995) Cleavage site determinants in the mammalian polyadenylation signal, *Nucleic Acids Res. 23*, 2614–2620.
23. Chen, F., and Wilusz, J. (1998) Auxiliary downstream elements are required for efficient polyadenylation of mammalian pre-mRNAs, *Nucleic Acids Res. 26*, 2891–2898.
24. Bagga, P. S., Ford, L. P., Chen, F., and Wilusz, J. (1995) The G-rich auxiliary downstream element has distinct sequence and position requirements and mediates efficient 3′ end and pre-mRNA processing through a trans-acting factor, *Nucleic Acids Res. 23*, 1625–1631.
25. Bagga, P. S., Arhin, G. K., and Wilusz, J. (1998) DSEF-1 is a member of the hnRNP H family of RNA-binding proteins and stimulates pre-mRNA cleavage and polyadenylation in vitro, *Nucleic Acids Res. 26*, 5343–5350.
26. Hall-Pogar, T., Zhang, H., Tian, B., and Lutz, C. S. (2005) Alternative polyadenylation of cyclooxygenase-2, *Nucleic Acids Res. 33*, 2565–2579.
27. Natalizio, B. J., Muniz, L. C., Arhin, G. K., Wilusz, J., and Lutz, C. S. (2002) Upstream elements present in the 3′ untranslated region of collagen genes influence the processing efficiency of overlapping polyadenylation signals, *J. Biol. Chem. 277*, 42733–42740.
28. Brown, P. H., Tiley, L. S., and Cullen, B. R. (1991) Efficient polyadenylation within the human immunodeficiency type 1 long terminal repeat inhibits polyadenylation of its own pre-mRNA, *J. Virol. 65*, 3340–3343.
29. Carswell, S., and Alwine, J. C. (1989) Efficiency of utilization of the simian virus 40 late polyadenylation site: effects of upstream sequences, *Mol. Cell. Biol. 9*, 4248–4258.
30. Schek, N., Cooke, C., and Alwine, J. C. (1992) Definition of the upstream efficiency element of the simian virus 40 late polyadenylation signal by using in vitro analyses, *Mol. Cell. Biol. 12*, 5386–5393.
31. Valsamakis, A., Zeichner, S., Carswell, S., and Alwine, J. C. (1991) The human immunodeficiency virus type 1 polyadenylation signal: a long terminal repeat element upstream of the AAUAAA necessary for efficient polyadenylation, *Proc. Natl. Acad. Sci. U.S.A. 88*, 2108–2112.
32. Valsamakis, A., Schek, N., and Alwine, J. C. (1992) Elements upstream of the AAUAAA within the human immunodeficiency virus polyadenylation signal are required for efficient polyadenylation in vitro, *Mol. Cell. Biol. 12*, 3699–3705.
33. DeZazzo, J. D., and Imperiale, M. J. (1989) Sequences upstream of the AAUAAA influence poly(A) selection in a complex transcriptional unit, *Mol. Cell. Biol. 9*, 4951–4961.
34. DeZazzo, J. D., Kilpatrick, J. E., and Imperiale, M. J. (1991) Involvement of long terminal repeat U3 sequences overlapping the transcriptional control region in human immunodeficiency virus type 1 mRNA 3′ end formation, *Mol. Cell. Biol. 11*, 1624–1630.
35. Gilmartin, G. M., Fleming, E. S., and Oetjen, J. (1992) Activation of HIV-1 pre-mRNA 3′ processing in vitro requires both an upstream element and TAR, *EMBO J. 11*, 4419–4428.
36. Gilmartin, G. M., Fleming, E. S., Oetjen, J., and Gravely, B. R. (1995) CPSF recognition of and HIV-1 mRNA 3′ processing enhancer: multiple sequence contacts involved in poly(A) site definition, *Genes Dev. 9*, 72–83.
37. Moreira, A., Wollerton, M., Monks, J., and Proudfoot, N. J. (1995) Upstream sequence elements enhance poly(A) site efficiency of the C2 complement gene and are phylogenetically conserved, *EMBO J. 14*, 3809–3819.
38. Moreira, A., Takagaki, Y., Brackenridge, S., Wollerton, M., Manley, J. L., and Proudfoot, N. J. (1998) The upstream sequence element of the C2 complement poly(A) signal activates mRNA 3′ end formation by two distinct mechanisms, *Genes Dev. 12*, 2522–2534.
39. Sanfacon, H., Brodmann, P., and Hohn, T. (1991) A dissection of the cauliflower mosaic virus polyadenylation signal, *Genes Dev. 5*, 141–149.
40. Russnak, R., and Ganem, D. (1990) Sequences 5′ to the polyadenylation signal mediate differential poly(A) site use in hepatitis B virus, *Genes Dev. 4*, 764–776.
41. Russnak, R. (1991) Regulation of polyadenylation in hepatitis B viruses: stimulation by the upstream activating signal PS1 is orientation-dependent, distance-dependent, and additive, *Nucleic Acids Res. 19*, 6449–6456.
42. Ryan, K., Calvo, O., and Manley, J. L. (2004) Evidence that polyadenylation factor CPSF-73 is the mRNA 3′ processing endonuclease, *RNA 10*, 565–573.
43. Lewis, J., Gunderson, S., and Mattaj, I. W. (1995) The influence of 5′ and 3′ end structures on pre-mRNA metabolism, *J. Cell Sci. (Suppl.) 19*, 13–19.
44. Jacobson, A., and Peltz, S. W. (1996) Interrelationships of the pathways of mRNA decay and translation in eukaryotic cells, *Annu. Rev. Biochem. 65*, 693–739.
45. Sachs, A. B., Sarnow, P., and Hentze, M. W. (1997) Starting at the beginning, middle and end: translation initiation in eukaryotes, *Cell 89*, 831–838.
46. Wickens, M., Anderson, P., and Jackson, R. J. (1997) Life and death in the cytoplasm: messages from the 3′ end, *Curr. Opin. Genet. Dev. 7*, 220–232.
47. Edwalds-Gilbert, G., Veraldi, K. L., and Milcarek, C. (1997) Alternative poly(A) site selection in complex transcription units: means to an end? *Nucleic Acids Res. 25*, 2547–2561.
48. Chabot, B. (1996) Directing alternative splicing: cast and scenarios, *Trends Genet. 12*, 472–478.

49. Lopez, A. J. (1998) Alternative splicing of pre-mRNA: developmental consequences and mechanisms of regulation, *Annu. Rev. Genet 32*, 279–305.
50. Danckwardt, S., Hentze, M. W., and Kulozik, A. E. (2008) 3′ End mRNA processing: molecular mechanisms and implications for health and disease, *EMBO J. 27*, 482–498.
51. Loke, J. C., Stahlberg, E. A., Strenski, D. G., Haas, B. J., Wood, P. C., and Li, Q. Q. (2005) Compilation of mRNA polyadenylation signals in *Arabidopsis* revealed a new signal element and potential secondary structures, *Plant Physiol. 138*, 1457–1468.
52. Shen, Y., Ji, G., Haas, B. J., Wu, X., Zheng, J., Reese, G. J., and Li, Q. Q. (2008) Genome level analysis of rice mRNA 3′ end processing signals and alternative polyadenylation, *Nucleic Acids Res. 36*, 3150–3161.
53. Shen, Y., Liu, Y., Liu, L., Liang, C., and Li, Q. Q. (2008) Unique features of nuclear mRNA poly(A) signals and alternative polyadenylation in *Chlamydomonas reinhardtii*, *Genetics 179*, 167–176.
54. The Fantom Consortium, Carninci, P., Kasukawa, T., Katayama, S., Gough, J., Frith, M. C., Maeda, N., Oyama, R., Ravasi, T., Lenhard, B., Wells, C., Kodzius, R., Shimokawa, K., Bajic, V. B., Brenner, S. E., Batalov, S., Forrest, A. R. R., Zavolan, M., Davis, M. J., Wilming, L. G., Aidinis, V., Allen, J. E., Ambesi-Impiombato, A., Apweiler, R., Aturaliya, R. N., Bailey, T. L., Bansal, M., Baxter, L., Beisel, K. W., Bersano, T., Bono, H., Chalk, A. M., Chiu, K. P., Choudhary, V., Christoffels, A., Clutterbuck, D. R., Crowe, M. L., Dalla, E., Dalrymple, B. P., de Bono, B., Della Gatta, G., di Bernardo, D., Down, T., Engstrom, P., Fagiolini, M., Faulkner, G., Fletcher, C. F., Fukushima, T., Furuno, M., Futaki, S., Gariboldi, M., Georgii-Hemming, P., Gingeras, T. R., Gojobori, T., Green, R. E., Gustincich, S., Harbers, M., Hayashi, Y., Hensch, T. K., Hirokawa, N., Hill, D., Huminiecki, L., Iacono, M., Ikeo, K., Iwama, A., Ishikawa, T., Jakt, M., Kanapin, A., Katoh, M., Kawasawa, Y., Kelso, J., Kitamura, H., Kitano, H., Kollias, G., Krishnan, S. P. T., Kruger, A., Kummerfeld, S. K., Kurochkin, I. V., Lareau, L. F., Lazarevic, D., Lipovich, L., Liu, J., Liuni, S., McWilliam, S., Madan Babu, M., Madera, M., Marchionni, L., Matsuda, H., Matsuzawa, S., Miki, H., Mignone, F., Miyake, S., Morris, K., Mottagui-Tabar, S., Mulder, N., Nakano, N., Nakauchi, H., Ng, P., Nilsson, R., Nishiguchi, S., Nishikawa, S., Nori, F., Ohara, O., Okazaki, Y., Orlando, V., Pang, K. C., Pavan, W. J., Pavesi, G., Pesole, G., Petrovsky, N., Piazza, S., Reed, J., Reid, J. F., Ring, B. Z., Ringwald, M., Rost, B., Ruan, Y., Salzberg, S. L., Sandelin, A., Schneider, C., Schönbach, C., Sekiguchi, K., Semple, C. A. M., Seno, S., Sessa, L., Sheng, Y., Shibata, Y., Shimada, H., Shimada, K., Silva, D., Sinclair, B., Sperling, S., Stupka, E., Sugiura, K., Sultana, R., Takenaka, Y., Taki, K., Tammoja, K., Tan, S. L., Tang, S., Taylor, M. S., Tegner, J., Teichmann, S. A., Ueda, H. R., van Nimwegen, E., Verardo, R., Wei, C. L., Yagi, K., Yamanishi, H., Zabarovsky, E., Zhu, S., Zimmer, A., Hide, W., Bult, C., Grimmond, S. M., Teasdale, R. D., Liu, E. T., Brusic, V., Quackenbush, J., Wahlestedt, C., Mattick, J. S., Hume, D. A., RIKEN Genome Exploration Research Group and Genome Science Group (Genome Network Project Core Group), Kai, C., Sasaki, D., Tomaru, Y., Fukuda, S., Kanamori-Katayama, M., Suzuki, M., Aoki, J., Arakawa, T., Iida, J., Imamura, K., Itoh, M., Kato, T., Kawaji, H., Kawagashira, N., Kawashima, T., Kojima, M., Kondo, S., Konno, H., Nakano, K., Ninomiya, N., Nishio, T., Okada, M., Plessy, C., Shibata, K., Shiraki, T., Suzuki, S., Tagami, M., Waki, K., Watahiki, A., Okamura-Oho, Y., Suzuki, H., Kawai, J., and Hayashizaki, Y. (2005) The transcriptional landscape of the mammalian genome, *Science* 309, 1559–1563.
55. Cheng, J., Kapranov, P., Drenkow, J., Dike, S., Brubaker, S., Paterl, S., Long, J., Stern, D., Tammana, H., Helt, G., Sementchenko, V., Piccolboni, A., Bekiranov, S., Bailey, D. K., Ganesh, M., Ghosh, S., Bell, I., Gerhard, D. S., and Gingeras, T. R. (2005) Transcriptional maps of 10 human chromosomes and 5-nucleotide resolution, *Science 308*, 1149–1154.
56. Ara, T., Lopez, F., Ritchie, W., Benech, P., and Gautheret, D. (2006) Conservation of alternative polyadenylation patterns in mammalian genes, *BMC Genomics 7*, 189–200.
57. Lee, J. Y., Yeh, I., Park, J. Y., and Tian, B. (2007) PolyA_DB2: mRNA polyadenylation sites in vertebrate genes, *Nucleic Acids Res. 35*, D165–D168.
58. Peterson, M. L., and Perry, R. P. (1989) The regulated production of mu m and mu s mRNA is dependent on the relative efficiencies of mu s poly(A) site usage and the c mu 4-to-M1 splice, *Mol. Cell. Biol. 9*, 726–738.
59. LeTexier, V., Riethoven, J. J., Kumanduri, V., Gopalakrishnan, C., Lopez, F., Gautheret, D., and Thanaraj, T. A. (2006) AltTrans: transcript pattern variants annotated for both alternative splicing and alternative polyadenylation, *BMC Bioinformatics 7*, 169–181.
60. Tian, B., Pan, Z., and Lee, J. Y. (2007) Widespread mRNA polyadenylation events in introns indicate dynamic interplay between polyadenylation and splicing, *Genome Res. 17*, 156–165.
61. D'mello, V., Lee, J. Y., MacDonald, C. C., and Tian, B. (2006) Alternative mRNA polyadenylation can potentially affect detection of gene expression by Affymetrix GeneChip® arrays, *Appl. Bioinformatics 5*, 249–253.
62. Beaudoing, E., and Gautheret, D. (2001) Identification of alternate polyadenylation sites and analysis of their tissue distribution using EST data, *Genome Res. 11*, 1520–1526.
63. Zhang, H., Lee, J. Y., and Tian, B. (2005) Biased alternative polyadenylation in human tissues, *Genome Biol. 6*, R100–R113.
64. Liu, D., Brockman, J. M., Dass, B., Hutchins, L. N., Singh, P., McCarrey, J. R., MacDonald, C. C., and Graber, J. H. (2007) Systematic variation in mRNA 3′-processing signals during mouse spermatogenesis, *Nucleic Acids Res. 35*, 234–246.
65. Dass, B., McMahon, K. W., Jenkins, N. A., Gilbert, D. J., Copeland, N. G., and MacDonald, C. C. (2001) The gene for a variant form of the polyadenylation protein CstF-64 is on chromosome 19 and is expressed in pachytene spermatocytes in mice, *J. Biol. Chem. 276*, 8044–8050.
66. Wallace, A. M, Denison, T. L., Attaya, E. N., and MacDonald, C. C. (2004) Developmental distribution of the polyadenylation protein CstF-64 and the variant tauCstF-64 in mouse and rat testis, *Biol. Reprod. 70*, 1080–1087.
67. Dass, B., Tardif, S., Park, J. Y., Tian, B., Weitaluf, H. M., Hess, R. A., Carnes, K., Griswold, M. D., Small, C. L., and MacDonald, C. C. (2007) Loss of polyadenylation protein tauCstF-64 causes spermatogenic defects and male infertility, *Proc. Natl. Acad. Sci. U.S.A. 104*, 20374–20379.
68. Rigault, C., Le Borgne, F., and Demarquoy, J. (2006) Genomic structure, alternative maturation and tissue expression of the human BBOX1 gene, *Biochem. Biophys. Acta 1761*, 1469–1481.
69. Farris, W., Leissring, M. A., Hemming, M. L., Chang, A. Y., and Selkoe, D. J. (2005) Alternative splicing of human insulin-degrading enzyme yields a novel isoform with a decreased ability to degrade insulin and amyloid β-protein, *Biochemistry 44*, 6513–6525.
70. Liu, H., and Johnson, E. M. (2002) Distinct proteins encoded by alternative transcripts of the PURG gene, located contrapodal to WRN on chromosome 8, determined by differential termination/polyadenylation, *Nucleic Acids Res. 30*, 2417–2426.
71. Lu, H., Huang, X., Zhang, L., Guo, Y., Cheng, H., and Zhou, R. (2007) Multiple alternative splicing of mouse Dmrt1 during gonadal differentiation, *Biochem. Biophys. Res. Commun. 352*, 630–634.
72. Yam, J. W. P., Chan, K. W., Ngan, E. S. W., and Hsiao, W. L. W. (2005) Genomic structure, alternative splicing, and tissue expression of rFrp/sFRP-4, the rat frizzled related protein gene, *Gene 357*, 55–62.
73. Costessi, L., Devescov, G., Baralle, F. E., and Muro, A. F. (2006) Brain-specific promoter and polyadenylation sites of the beta-adducin pre-mRNA generate an unusually long 3′ UTR, *Nucleic Acids Res. 34*, 243–253.

74. Winter, J., Kunath, M., Roepcke, S., Krause, S., Schneider, R., and Schweiger, S. (2007) Alternative polyadenylation signals and promoters act in concert to control tissue-specific expression of the Opitz Syndrome gene MID1, *BMC Mol. Biol. 8*, 105.
75. Proudfoot, N. (2004) New perspectives on connecting messenger 3′ end formation to transcription, *Curr. Opin. Cell Biol. 16*, 272–278.
76. Qu, X., Qi, Y., and Qi, B. (2002) Generation of multiple mRNA transcripts from the novel human apoptosis-inducing gene hap by alternative polyadenylation utilization and the translational activation function of 3′ untranslated region, *Arch. Biochem. Biophys. 400*, 233–244.
77. Yu, M., Sha, H., Gao, Y., Zeng, H., Zhu, M., and Gao, X. (2006) Alternative 3′ UTR polyadenylation of Bzw1 transcripts display differential translation efficiency and tissue-specific expression, *Biochem. Biophys. Res. Commun. 345*, 479–485.
78. Qiu, J., Cheng, F., Burger, L. R., and Pintel, D. (2006) The transcription profile of Aleutian mink disease virus in CRFK cells is generated by alternative processing of pre-mRNAs produced from a single promoter, *J. Virol. 80*, 654–662.
79. Qiu, J., Cheng, F., Johnson, F. B., and Pintel, D. (2007) The transcription profile of the Bocavirus bovine parvovirus is unlike those of previously described parvoviruses, *J. Virol. 81*, 12080–12085.
80. Majerciak, V., Yamanegi, K., and Zheng, Z. M. (2006) Gene structure and expression of Kaposi's sarcoma-associated herpesvirus ORF56, ORF 57, ORF 58, and ORF 59, *J. Virol. 80*, 11968–11981.
81. Milligan, S. G., Veerapraditsin, T., Ahamet, B., Mole, S., and Graham, S. V. (2007) Analysis of novel human papillomavirus type 16 late mRNAs in differentiated W12 cervical epithelial cells, *Virology 360*, 172–181.
82. Schwartz, S. (2008) HPV-16 RNA processing, *Front. Biosci. 13*, 5880–5891.
83. Graham, S. V. (2008) Papillomavirus 3′ UTR regulatory elements, *Front. Biosci. 13*, 5646–5663.
84. Zheng, Z. M., and Baker, C. C. (2006) Papillomavirus genome structure, expression, and post-transcriptional regulation, *Front. Biosci. 11*, 2286–2302.
85. McPhillips, M. G., Veerapraditsin, T., Cumming, S. A., Karali, D., Milligan, S. G., Boner, W., Morgan, I. M., and Graham, S. V. (2004) SF2/ ASF binds the human papillomavirus type 16 late RNA control element and is regulated during differentiation of virus-infected epithelial cells, *J. Virol. 78*, 10598–10605.
86. Goraczniak, R., and Gunderson, S. I. (2008) The regulatory element in the 3′ untranslated region of human papillomavirus 16 inhibits expression by binding CUG-binding protein 1, *J. Biol. Chem. 283*, 2286–2296.
87. Furth, P. A., Chow, W. T., Rex, J. H., Byrne, J. C., and Baker, C. C. (1994) Sequences homologues to 5′ splice sites are required for the inhibitory activity of papillomavirus late 3′ untranslated regions, *Mol. Cell. Biol. 14*, 5278–5289.
88. McNally, M. T. (2008) RNA processing control in avian retroviruses, *Front. Biosci. 13*, 3869–3883.
89. Nemeroff, M. E., Barabino, S. M., Li, Y., Keller, W., and Krug, R. M. (1998) Influenza virus NS1 protein interacts with the cellular 30 kDa subunit of CPSF and inhibits 3′ end formation of cellular pre-mRNAs, *Mol. Cell 1*, 991–1000.
90. Chen, Z., Li, Y., and Krug, R. M. (1999) Influenza A virus NS1 protein targets poly(A)-binding protein II of the cellular 3′-end processing machinery, *EMBO J. 18*, 2273–2283.
91. Noah, D. L., Twu, K. Y., and Krug, R. M. (2003) Cellular antiviral responses against influenza A virus are countered at the posttranscriptional level by the viral NS1A protein via its binding to a cellular protein required for the 3′ end processing of cellular pre-mRNAs, *Virology 307*, 386–395.
92. Ashe, M. P., Furger, A., and Proudfoot, N. J. (2000) Stem-loop 1 of the U1snRNP plays a critical role in the suppression of HIV-1 polyadenylation, *RNA 6*, 170–177.
93. Gee, A. H., Kasprzak, W., and Shapiro, B. A. (2006) Structural differentiation of the HIV-1 poly(A) signals, *J. Biomol. Struct. Dyn. 23*, 417–428.
94. Hall-Pogar, T., Liang, S., Hague, L. K., and Lutz, C. S. (2007) Specific trans-acting proteins interact with auxiliary RNA polyadenylation elements in the COX-2 3′ UTR, *RNA 13*, 1103–1115.
95. Danckwardt, S., Kaufman, I., Gentzel, M., Foerstner, K. U., Gantzert, A. S., Gehring, N. H., Neu-Yilik, G., Bork, P., Keller, W., Wilm, M., Hentze, M. W., and Kulozik, A. E. (2007) Splicing factors stimulate polyadenylation via USEs at non-canonical 3′ end formation signals, *EMBO J. 26*, 2658–2669.
96. Ceelie, H., Spaargaren-van Riel, C. C., Bertina, R. M., and Vos, H. L. (2004) G20210A is a functional mutation in the prothrombin gene; effect on protein levels and 3′ end formation, *J. Thromb. Haemostasis 2*, 119–127.
97. Wood, A. J., Schulz, R., Woodfine, K., Koltowska, K., Beechey, C. V., Peters, J., Bourc'his, D., and Oakey, R. J. (2008) Regulation of alternative polyadenylation by genomic imprinting, *Genes Dev. 22*, 1141–1146.
98. Lian, Z., Karpikov, A., Lian, J., Mahajan, M. C., Hartman, S., Gerstein, M., Snyder, M., and Weissman, S. M. (2008) A genomics analysis of RNA polymerase II modification and chromatin architecture related to 3′ end polyadenylation, *Genome Res. 18*, 1224–1237.
99. Ryan, K. (2006) Pre-mRNA 3′ cleavage is reversibly inhibited in vitro by cleavage factor dephosphorylation, *RNA Biol. 4*, 26–33.
100. Ryan, K., and Bauer, D. L. V. (2008) Finishing touches: post-translational modification of protein factors involved in mammalian pre-mRNA 3′ end formation, *Int. J. Biochem. Cell. Biol. 40*, 2384–2396.
101. Pique, M., Loez, J. M., Foissac, S., Guigo, R., and Mendez, R. (2008) A combinatorial code of CPE-mediated translational control, *Cell 132*, 434–448.
102. Makeyev, E. V., and Maniatis, T. (2008) Multilevel regulation of gene expression by microRNAs, *Science 319*, 1789–1790.
103. Hobert, O. (2008) Gene regulation by transcription factors and microRNAs, *Science 319*, 1785–1786.
104. Guarnieri, D. J., and DiLeone, R. J. (2008) MicroRNAs: a new class of gene regulators, *Ann. Med. 40*, 197–208.
105. Vasudevan, S., Tong, Y., and Steitz, J. A. (2007) Switching from repression to activation: microRNAs can up-regulate translation, *Science 318*, 1931–1934.
106. Sandberg, R., Neilson, J. R., Sarma, A., Sharp, P., and Burge, C. B. (2008) Proliferating cells express mRNAs with shortened 3′ untranslated regions and fewer microRNA target sites, *Science 320*, 1643–1646.
107. Gilat, R., and Shweiki, D. (2007) A novel function for alternative polyadenylation as a rescue pathway from NMD surveillance, *Biochem. Biophys. Res. Commun. 353*, 487–492.
108. Prasanth, K. V., Prasanth, S. G., Xuan, Z., Hearn, S., Frier, S. M., Bennett, C. F., Zhang, M. Q., and Spector, D. L. (2005) Regulating gene expression through RNA nuclear retention, *Cell 123*, 249–263.