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REVIEW

mRNA Degradation Machinery in Plants

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Abstract Control of gene expression is exerted by multiple steps such as transcription, mRNA processing, mRNA export, mRNA degradation, translation, and posttranslational events. Recent discovery of small RNAs has enhanced the impact of posttranscriptional regulation, in particular, alterations in mRNA stability in the regulation of gene expression. Therefore, mRNA turnover is an important process not only for setting the basal level of gene expression but also as a regulatory step. Compared to the mechanism of transcription, much less information is available regarding mRNA degradation machineries. However, in the past several years, various components involved in the mRNA degradation process have been identified in eukaryotes. In particular, progress in the plant field has revealed the involvement of mRNA turnover in a wide variety of developmental processes and hormonal responses. Here, we provide an overview of machineries involved in general mRNA degradation and mRNA surveillance systems in plants.

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Introduction

The control of mRNA stability is important for regulation of gene expression. Accumulating findings regarding general mRNA degradation pathways in yeast indicated that there are two general pathways, a deadenylation-dependent decapping pathway and a 3′ to 5′ decay pathway (Fig. 1). In each case, the removal of poly(A) tails from the 3′ end of transcripts, called deadenylation, is the initial and rate-limiting step of the mRNA degradation. The complex of Ccr4p and Pop2p has been identified as a predominant deadenylase (Tucker et al. 2001, 2002; Chen et al. 2002). This event triggers the removal of the cap at the 5′ end of the mRNA by the decapping enzyme, a complex of Dcp2p and Dcp1p (Beelman et al. 1996; LaGrandeur and Parker 1998; Dunckley and Parker 1999). Following decapping, the message is degraded by Xrn1p, a processive exoribonuclease that hydrolyzes RNA in a 5′ to 3′ direction to completion (Muhlrad and Parker 1994). Alternatively, transcripts can be degraded in a 3′ to 5′ direction by the exosome following deadenylation. These two pathways comprise the bulk of mRNA decay in yeast (Anderson and Parker 1998). As specialized mRNA turnover pathways, nonsense-mediated mRNA decay (NMD), nonstop mRNA decay (NSD), and no-go decay (NGD) have been reported (Fig. 2). The NMD pathway is responsible for the degradation of mRNAs bearing premature nonsense codons. Such transcripts are degraded either through the deadenylation-independent decapping pathway or through accelerated deadenylation probably followed by 3′ to 5′ decay (Elliott et al. 1989; Muhlrad and Parker 1994; Cao and Parker 2003; Lejeune et al. 2003; Mitchell and Tollervey 2003; Takahashi et al. 2003). In the NSD pathway, transcripts lacking translation termination codons are degraded rapidly from 3′ to 5′ direction by exosome (Frischmeyer et al. 2002; van Hoof et al. 2002). Translation elongation arrest

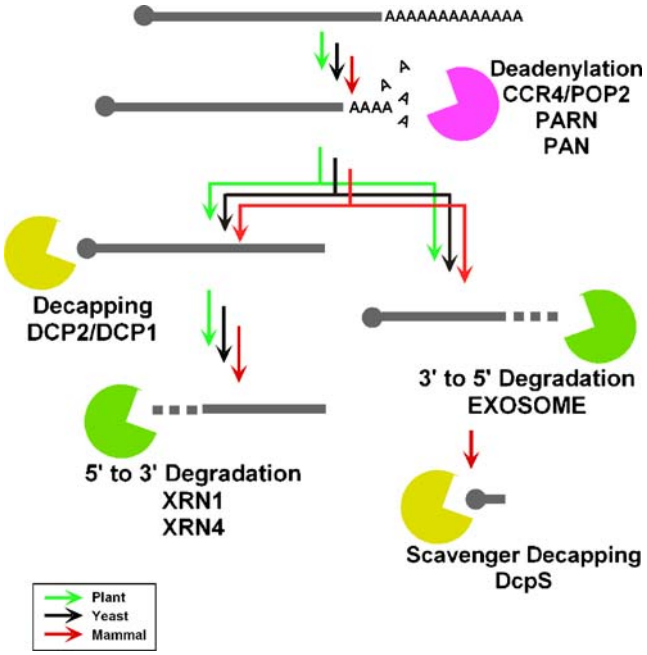

Fig. 1 General mRNA degradation pathways in eukaryotes. The mRNA body is protected by a cap structure and poly(A) tail at the 5′ and 3′ ends, respectively. mRNA degradation pathways in yeast (*black*), mammal (*red*), and plant (*green*) systems are indicated by arrows. Initially, the poly(A) tail is removed by deadenylation. Deadenylated mRNA is degraded by 5′ to 3′ degradation followed by decapping, or alternatively by 3′ to 5′ degradation. In mammals, the scavenger decapping step is reported to follow. The responsible enzyme(s) for each step is represented by a Pac-Man. Also, endonuclease activity of the exosome, not shown in the figure, is likely involved

induced by strong secondary structure triggers the NGD, in which an endonucleolytic cleavage occurs in the vicinity of the stalled ribosome leading to exonucleolytic degradation of the mRNA by Xrn1p and exosome. In addition to an active translation, Dom34p and Hbs1p, homologs of translation termination factors eRF1 and eRF3, are required for this cleavage (Doma and Parker 2006). Interestingly, the complex of Dom34p and Hbs1p itself shows endoribonuclease activity *in vitro*, with Dom34p as an active unit (Lee et al. 2007).

Information about mRNA degradation pathways in multicellular eukaryotes, including plants, is limited by contrast to yeast. Considering the fact that most components for the general mRNA degradation machinery are conserved in eukaryotes, multicellular eukaryotes and yeast are likely to use similar basic mechanisms for mRNA degradation. However, some different aspects have also been reported. For example, several *in vitro* studies showed that 3′ to 5′ degradation pathway by exosome is the main degradation pathway for at least some mRNAs in mammals, although it is not clear yet what pathway is dominant *in vivo* (Chen et al. 2001; Wang and Kiledjian 2001; Mukherjee et al. 2002). In this 3′ to 5′ degradation pathway, the resulting oligonucleotide cap structure is removed by a scavenger decapping enzyme, DcpS (Liu et al. 2002). Moreover, analyses of enzymes responsible for mRNA degradation in plants revealed the link between mRNA turnover and gene silencing as well as the involvement of mRNA turnover in the developmental processes and hormonal responses. Therefore, figuring out the mRNA degradation mechanisms in plants is important in that it will increase understanding of the general significance of mRNA turnover control. In this review, we will summarize information regarding mRNA degradation pathways and responsible components identified in plants.

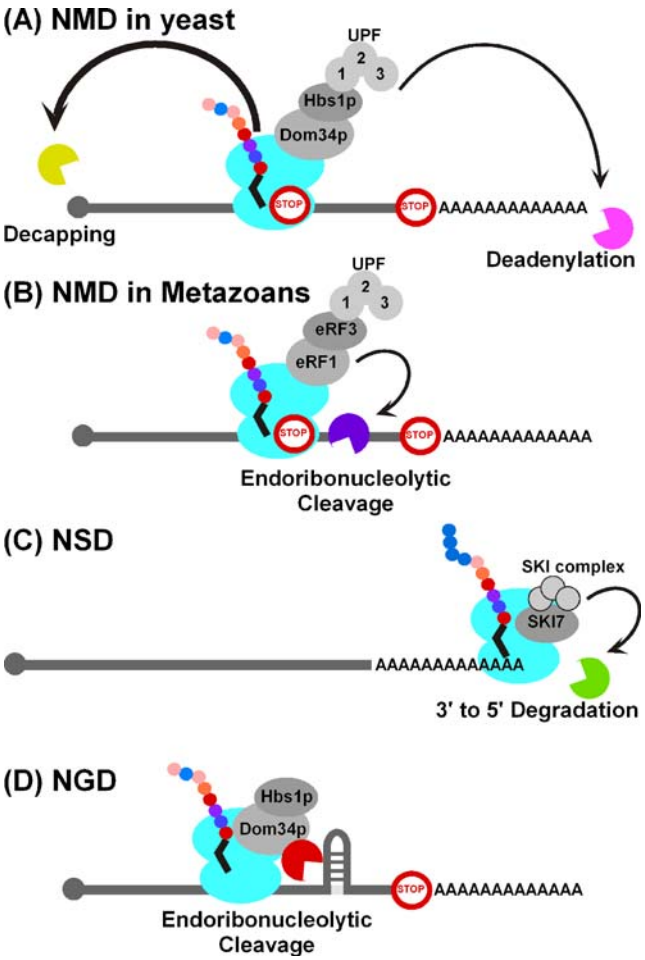

Fig. 2 mRNA surveillance system. Schematic view of nonsense-mediated mRNA decay (NMD), nonstop mRNA decay (NSD), and no-go decay (NGD) is indicated. *A* Premature termination codon is recognized by a surveillance complex, UPF1, UPF2, and UPF3. NMD in yeast triggers deadenylation-independent decapping or accelerates deadenylation. *B* NMD in metazoans induces endoribonucleolytic cleavage in the vicinity of premature codon. *C* In NSD, a stalled ribosome at the 3′ end of the transcript recruits Ski7p, Ski complex, and exosome to degrade mRNA from 3′ to 5′ direction. *D* In NGD, translation elongation arrest triggers endoribonucleolytic cleavage in the vicinity of the stalled ribosome. eRF3 and eRF1 (Dom34p and Hbs1p are their respective homologs in yeast) are translation termination factors

General mRNA Degradation Pathways

Deadenylases

Two deadenylase complexes responsible for the shorting of the poly(A) tail have been studied in yeast. As a major cytoplasmic deadenylase, the complex of Ccr4p/Pop2p (also called as Caf1p) and several associated factors have been identified (Tucker et al. 2001; Denis and Chen 2003). Based on the mutant analysis, Ccr4p seems to be the catalytic subdomain of this complex (Tucker et al. 2002), and mutations in the catalytic domains of Ccr4p abolish its functions *in vivo* (Chen et al. 2002). It has been suggested that Pop2p might also have nuclease activity (Daugeron et al. 2001), though it has a role in enhancing the activity of Ccr4p. The second enzyme complex is a poly(A) binding-protein (PABP)-dependent poly(A) nuclease, referred to as PAN (Lowell et al. 1992; Sachs and Deardorff 1992). This enzyme is composed of at least two subunits, Pan2p and Pan3p, of which Pan2p seems to be the catalytic subunit. Studies carried out using mutants deficient in Pan2p and/or Pan3p have shown that PAN is responsible for the control of poly(A) length of newly synthesized mRNA. It shortens the poly(A) tail from about 150–200 nucleotides to the 70–90 nucleotide length, which is characteristic of mature yeast mRNAs (Brown and Sachs 1998). For cytoplasmic deadenylation, PAN has a function for residual deadenylation process in the yeast strain lacking the predominant deadenylase, Ccr4p (Tucker et al. 2001).

Homologous proteins of Ccr4p/Pop2p complex and PAN have been identified in multicellular eukaryotes, including plants (Dupressoir et al. 2001; Chen et al. 2002; Uchida et al. 2004). Functional analysis of mammalian CCR4/POP2 and PAN revealed the biphasic deadenylation pathway by the consecutive action of these two distinct deadenylases. During the first phase of deadenylation, PAN shortens the poly(A) tail to about 110 nucleotides with a relatively slow process. PAN is then replaced by CCR4/POP2 complex for the second phase. In this phase, not only is the remaining poly(A) tail removed by CCR4/POP2-mediated deadenylation, but DCP2-directed decapping is also active, which renders the transcripts susceptible to the following rapid 5′ to 3′ exonucleolytic degradation (Yamashita et al. 2005a). Interestingly, the deadenylation mediated by PAN and CCR4/POP2 is coupled with the translation termination through the competitive interaction of translation termination complex eRF1/eRF3 and the two deadenylase complexes to PABP. The translation-dependent exchange of eRF1/eRF3 for deadenylase complexes that occurs on PABP is a trigger of deadenylation (Funakoshi et al. 2007). This deadenylation coupled with the translation termination is conserved in yeast, and the involvement of two deadenylase complexes, Pan2p/Pan3p and Ccr4p/Pop2p, is suggested (Hosoda et al. 2003; Funakoshi et al. 2007; Siddiqui et al. 2007).

Arabidopsis has multiple potential homologs of the CCR4/POP2 complex, therefore functional analysis is hampered by the possible redundancy and regulatory complexity of gene families encoding the components. However, gain- and loss-of-function analyses of the POP2 homolog (CaCAF1) of chili pepper, *Capsicum annuum*, indicate interesting phenotypes. Overexpression of *CaCAF1* in tomato shows significant growth enhancement and increase of pathogen resistance, while silencing of *CaCAF1* in the pepper plants shows growth retardation and enhancement of susceptibility to various types of pathogens. The level of several transcripts that might contribute to these phenotypes, such as those related to cell wall formation and salicylic acid- and jasmonic acid-mediated signaling pathways, is increased in the *CaCAF1*-overexpressing plants (Sarowar et al. 2007). Considering the fact that CaCAF1 shares high homology with POP2 of many other eukaryotes, CaCAF1 might be a part of a poly(A)-specific exoribonuclease, as in yeast and mammals, and the role of CaCAF1 in plant growth and pathogen resistance would then implicate the biological significance of deadenylation in plants.

Multicellular eukaryotes have an additional poly(A) ribonuclease, designated PARN, that is absent in yeast. *In vitro* analysis indicated that PARN is a Mg^{2+}-dependent deadenylase and its activity is inhibited by PABP under the physiological salt concentration (Korner and Wahle 1997). PARN has cap-binding activity and preferentially deadenylates capped mRNAs (Dehlin et al. 2000b; Gao et al. 2000). Immunodepletion experiments in HeLa cell extracts suggested that PARN is a major cytoplasmic deadenylase (Korner et al. 1998; Dehlin et al. 2000a; Gao et al. 2000). Moreover, PARN is also involved in default poly(A) tail removal during *Xenopus* meiotic maturation (Korner et al. 1998; Copeland and Wormington 2001). SiRNA-mediated downregulation studies revealed the involvement of PARN in the NMD pathway (Lejeune et al. 2003). PARN also functions in the rapid deadenylation of mRNA containing AU-rich element (ARE) promoted by the ARE-binding protein, tristetraprolin (Lai et al. 2003).

Homologous proteins of PARN are found in many eukaryotes, including plants. *Arabidopsis thaliana* PARN-like protein, AtPARN, showed poly(A)-specific degradation activity *in vitro*. GFP fusion experiments suggested that, similar to human PARN, AtPARN is localized both in cytoplasm and the nucleus. Based on the mutant analysis, it appears that AtPARN is essential for embryo development (Chiba et al. 2004). Some embryo-specific transcripts have longer poly(A) tails in the mutant, suggesting that deadenylation of several specific mRNAs by AtPARN is critical for proper embryo development (Reverdatto et al. 2004). Interestingly, a weak and viable AtPARN mutant was

isolated during the ABA-hypersensitive mutant screening. Characterization of this leaky allele revealed the involvement of AtPARN in ABA, salicylic-acid, and stress responses in *Arabidopsis* (Nishimura et al. 2005). However, direct substrates for AtPARN have not been elucidated yet.

Decapping Enzymes

In yeast and multicellular eukaryotes, decapping is catalyzed by a complex consisting of two subunits, Dcp1 and Dcp2, with Dcp2 as the catalytic subunit. Several different regulatory factors are required for its activity. Edc1p-Edc3p, Dhh1p, Pat1p, and Lsm1p–Lsm7p are reported as association factors in yeast (Coller and Parker 2004). hEDC3 and Rck/p54, homologs of Edc3p and Dhh1p, respectively, and an additional factor, Hedls/Ge1, are identified as associating proteins in humans. Hedls/Ge1 is required for the interaction of hDCP1 and hDCP2 (Fenger-Gr et al. 2005).

The critical role of decapping in cell growth and development is suggested by the pleiotropic phenotype displayed by decapping component mutants in several species, such as *Drosophila*, *Saccharomyces cerevisiae*, and *Schizosaccharomyces pombe* (Beelman et al. 1996; Hatfield et al. 1996; Dunckley and Parker 1999; Sakuno et al. 2004; Lin et al. 2006). Additionally, the important role of decapping in postembryonic development has been indicated in *Arabidopsis* (Xu et al. 2006; Iwasaki et al. 2007). AtDCP2, AtDCP1, and VARICOSE (VCS) are isolated as proteins homologous to hDCP2, hDCP1, and Hedls/Ge1, respectively. AtDCP2 is an active subunit of a decapping complex, and the mutation analysis indicated that the Nudix domain is important for the activity. AtDCP1 and VCS can enhance the AtDCP2 activity *in vitro*. Analyses of interacting factors both *in vivo* and *in vitro* suggested that these three components interact with one another within a big complex. AtDCP1, AtDCP2, and VCS are localized in cytoplasmic foci called the processing bodies (P-bodies), where most mRNA degradation machinery is accumulated. Interestingly, T-DNA insertion lines of these three components showed a similar seedling lethal phenotype having growth that is severely perturbed at the cotyledon stage (Xu et al. 2006).

Global analysis of mRNA profiles in the mutant of AtDCP2 using microarrays indicated that the mRNA level of 142 transcripts was elevated more than fivefold compared to wild type. These putative targets of AtDCP2 include mRNAs of many different functional categories, such as signaling molecules, transcription factors, transporters, and metabolism-related proteins. In the mutants of AtDCP2 and VCS, some of these putative targets showed higher levels of expression and the capped state, suggesting that these two proteins are components of decapping machinery for several specific targets (Goeres et al. 2007). Although AtDCP2 and XRN4, 5′ to 3′ exoribonuclease (see below), are likely to work in the sequential steps of the same degradation pathway, none of the transcripts elevated in the *xrn4* mutant showed greater expression in the mutant of AtDCP2. This result suggests that other unidentified exoribonucleases might exist (Souret et al. 2004; Olmedo et al. 2006; Goeres et al. 2007).

Isolation and characterization of *Arabidopsis* mutants defective in miRNA-guided silencing revealed that translational repression is a mode of plant miRNA action that is more widespread than originally thought (Brodersen et al. 2008). Decapping component VCS is required for this miRNA-mediated translational repression, as in animals (Eulalio et al. 2007; Brodersen et al. 2008), providing evidence of the link between mRNA decapping and action of at least some of the plant miRNAs.

5′ to 3′ Exoribonucleases

Yeast has two 5′ to 3′ exoribonucleases. Xrn1p, localized in the cytoplasm, is a main enzyme catalyzing mRNA degradation following decapping, whereas Xrn2p (also called Rat1p), localized primarily in the nucleus, is an essential enzyme involved in the processing of rRNA and small nucleolar RNAs (snoRNAs; Hsu and Stevens 1993; Johnson 1997). Three members of the *Arabidopsis* XRN family were isolated as homologs of Xrn2p/Rat1p, and no Xrn1p-like gene was identified. All of them showed 5′ to 3′ exoribonuclease activity in the yeast heterologous expression system. Among them, AtXRN4 is localized in the cytoplasm; therefore, it is the best candidate for a functional homolog of the yeast Xrn1p (Kastenmayer and Green 2000). Although several substrates of AtXRN4 were identified using reverse genetics and microarray analysis, AtXRN4 is only required for the degradation of a few of the transcripts and not critical for the general degradation of unstable transcripts tested. Interestingly, the substrates of AtXRN4 include the 3′ end products produced by miRNA-mediated cleavage (Souret et al. 2004; German et al. 2008).

During the search of *Arabidopsis* mutants that promote RNA-dependent RNA polymerase (RdRp)-dependent cosuppression, a novel biological role of AtXRN4 in transgene-dependent gene silencing was revealed. AtXRN4 functions to negatively regulate gene silencing by degrading decapped transgene mRNA, which is required to initiate or maintain silencing by serving as a template for RdRp (Gazzani et al. 2004). Additionally, deep sequencing analysis of small RNAs in the *xrn4* mutant indicated that AtXRN4 negatively regulates the level of 21-nucleotide small RNAs processed from the endogenous transcripts as well. Endogenous uncapped transcripts, which likely act as substrates of small RNA biogenesis, accumulate more in the *xrn4* mutant compared to the wild type. Simultaneous

loss of ABH1, a subunit of the mRNA cap binding complex, enhances the level of these small RNAs, suggesting that an integral function of XRN4 and ABH1 is to prevent the production of small RNAs from the endogenous transcripts (Gregory et al. 2008). AtXRN4 is also involved in the degradation of target mRNAs of long siRNAs (lsiRNAs), a new class of endogenous small RNAs with size range of 30 to 40 nucleotides in *Arabidopsis*. *AtlsiRNA-1*, induced by *Pseudomonas syringae* infection, is processed from the natural antisense transcript. This lsiRNA down-regulates the expression of the antisense transcript, *AtRAP*, by AtXRN4-mediated 5′ to 3′ exoribonucleolytic degradation following decapping. Down-regulation of *AtRAP* results in the increased resistance to both virulent and avirulent strains of *P. syringae* (Katiyar-Agarwal et al. 2007).

Involvement of AtXRN4 in the ethylene response pathway is uncovered by identification of *ETHYLENE-INSENSITIVE 5* (*EIN5*) as *AtXRN4*. Based on genetic analysis, EIN5 appeared to be acting in the ethylene signaling pathway (Olmedo et al. 2006; Potuschak et al. 2006). EIN3, a main transcription factor, is rapidly induced in response to ethylene, whereas in the absence of ethylene, the EIN3 protein is quickly degraded through a ubiquitin/proteosome pathway mediated by F-box proteins, EBF1 and EBF2. Interestingly, EBF1 and EBF2 mRNAs are ethylene inducible, suggesting the existence of a negative feedback regulation for fine-tuning the EIN3 level (Binder et al. 2007; Kendrick and Chang 2008). The function of AtXRN4 in the ethylene signaling pathway is likely to antagonize the negative feedback regulation of EIN3 by down-regulating EBF1 and EBF2 mRNA levels (Olmedo et al. 2006). However, EBF1 and EBF2 mRNAs are not likely to be direct targets for AtXRN4 because the half-life of both transcripts in the *ein5/atxrn4* mutant is the same as in the wild-type (Souret et al. 2004; Potuschak et al. 2006), suggesting that more complex action must be involved.

Similar to AtXRN4, AtXRN2 and AtXRN3 also function as endogenous suppressors of transgene-induced silencing, probably by degrading aberrant transgene RNAs in the nucleus. In addition, AtXRN2 and 3 are involved in the maturation process of miRNA, such as degradation of looped end products derived from miRNA precursors (Gy et al. 2007). Interestingly, levels of these three AtXRNs are likely to be controlled by another repressor, FIERY1 (FRY1), which is a bifunctional nucleotidase/phosphatase converting 3′-phosphoadenosine 5′-phosphate (PAP), a toxic by-product of sulfate assimilation, to 5′AMP and Pi (Saito 2004; Gy et al. 2007). Increased PAP in the *fry1* mutant could be an inhibitor of AtXRNs as observed in yeast (Dichtl et al. 1997). Recent genetic analyses of the *fry1* mutants indicated the role of FRY1 as a negative regulator of drought stress response (Wilson et al. 2009), in addition to its involvement of FRY1 in photomorphogenesis (Kim and von Arnim 2008). The phenotypic similarity of the *xrn2/xrn3* double mutant suggested that the photomorphogenic phenotypes of *fry1* mutant are likely attributed to modified XRN activity (Kim and von Arnim 2008).

3′ to 5′ Exoribonucleases

The exosome is a multisubunit complex responsible for the 3′ to 5′ exoribonucleolytic degradation of RNAs, both in the nucleus and the cytoplasm. The yeast exosome, identified first and best analyzed, consists of a core complex containing ten different subunits and nucleus- or cytoplasmic-specific-associating proteins that are required for its function. Six subunits (Rrp41p, Rrp42p, Rrp43p, Rrp45p, Rrp46p, and Mtr3p) show homology to the catalytic domain of phosphorolytic bacterial RNase PH and archaeal exosome. Three other subunits (Rrp40p, Rrp4p, and Csl4p) indicate homology to S1 and KH RNA-binding proteins. These nine subunits form a ring-type structure. The tenth subunit, Dis3p (also known as Rrp44), is homologous to a bacterial RNase II that is a hydrolytic 3′ to 5′ exoribonuclease (Mitchell et al. 1997; Allmang et al. 1999). Structural and functional analysis revealed that Dis3p is the only subunit responsible for the hydrolytic 3′ to 5′ exonucleolytic activity of the exosome core (Liu et al. 2006; Dziembowski et al. 2007). Moreover, this subunit also has recently been shown to contain endonuclease activity that is of functional consequence *in vivo* (Lebreton et al. 2008; Schaeffer et al. 2009). Other subunits do not have enzymatic activity, but are required for the interaction of associated proteins with Dis3p (Liu et al. 2006; Dziembowski et al. 2007). In the nucleus, the exosome is involved in the processing of rRNA, snoRNA, and snRNA. It is also involved in the quality control of many types of RNA, such as mRNAs, tRNAs, and rRNAs, together with the nucleus association factors, an RNase D-like protein (Rrp6p) and a putative RNA-binding protein (Lrp1p). In the cytoplasm, the exosome functions in minor degradation pathways for normal transcripts. It also participates in NMD, NSD, and NGD. The exosome in the cytoplasm is activated by Ski7p, a putative GTPase and the Ski2p/Ski3p/Ski8p complex (Houseley et al. 2006). The exosome is also required for ARE-mediated mRNA degradation in human cells and for the degradation of the 5′ fragment cleaved by RNA interference (RNAi) in *Drosophila* (Mukherjee et al. 2002; Orban and Izaurralde 2005).

Arabidopsis also conserved several proteins homologous to the exosome components. Two of them, AtRRP41 and AtRRP4, have been characterized in detail. AtRRP41 is a phosphorolytic exoribonuclease that complements the defects in the 5.8S rRNA processing and 3′ to 5′ degradation observed in the yeast temperature-sensitive mutant of Rrp41p, as well as the lethal phenotype of its null mutant

(Chekanova et al. 2000). AtRRP4 is a hydrolytic 3′ to 5′ exoribonuclease that contains S1 and KH RNA-binding domain. Both are physically interacting and identified in a high molecular mass complex in *Arabidopsis*, strongly suggesting that AtRRP41 and AtRRP4 are likely to be members of the plant exosome (Chekanova et al. 2002). Other components of the complex, as well as AtRRP41 and AtRRP4, are identified by tandem affinity purification. Nine subunits of the exosome core, except for the Dis3p homolog, are identified. There are two genes that may encode the Dis3p homolog in the *Arabidopsis* genome, suggesting that the exosome composition may differ between yeast and multicellular eukaryotes (Chekanova et al. 2007). In fact, the Dis3p homologue in humans does not seem to be associated stably with the exosome complex, although the rest of the components appear to be quite similar to yeast (Allmang et al. 1999; Raijmakers et al. 2004; Liu et al. 2006). Additionally, AtRRP41 is unique in that it retains the full catalytic activity (Chekanova et al. 2000). Interestingly, mutant analyses of AtCSL4, AtRRP41, and AtRRP4 in *Arabidopsis* implicated the distinct functions of each subunit of the exosome. AtRRP41 and AtRRP4 are essential for the developmental of female gametophytes and embryogenesis, respectively, whereas AtCSL4 is dispensable for growth and development. Moreover, the exosome targets are identified by applying tilling microarray analysis to the mutants of AtRRP41 and AtRRP4, in which these genes are knocked down by the inducible RNAi system. In addition to a subset of snRNAs, snoRNAs, tRNAs, and mRNAs, a wide variety of RNAs are identified as the exosome substrates, including mRNAs that did not properly complete 3′ end processing, primary microRNA processing intermediates, tandem-repeat-associated siRNA precursors, and noncoding RNAs (Chekanova et al. 2007). As an additional physiological impact of exosome function, involvement of the exosome in the cuticle wax biosynthesis in *Arabidopsis* is indicated by the analysis of a wax-deficient *eceriferum7* (*cer7*) mutant. The CER7, a functional homolog of yeast Rrp45p, plays a role in keeping the cuticular wax levels high in stems and siliques by controlling transcription of a key gene for wax production in an indirect way (Hooker et al. 2007).

Specialized mRNA Degradation Pathways

mRNA Surveillance System

The NMD pathway, which is responsible for the degradation of selective mRNAs containing a premature termination codon (PTC), is well conserved among eukaryotes. The mechanism of NMD is studied extensively in yeast and mammals, leading to the identification of three core components: UPF1, UPF2, and UPF3. It has been postulated that mammalian transcripts are targeted for NMD only if a stop codon is located more than 50 nucleotides upstream of an exon–exon junction (Nagy and Maquat 1998). During the splicing, an exon junction complex (EJC) interacting with UPF2 and UPF3 binds to 20 to 25 nucleotides upstream of the exon–exon junction (Le Hir et al. 2000). In the absence of PTC, the ribosomes can displace EJC from mRNAs, but when termination is premature, the remaining EJC downstream of the aberrant stop codon triggers NMD (Chang et al. 2007). UPF1 is recruited to prematurely terminating ribosomes through interactions with the release factors, where it can complex with UPF3 via UPF2. Then, the phosphorylation of UPF1 is promoted by SMG1 (Kashima et al. 2006; Ivanov et al. 2008). The phosphorylation/dephosphorylation of UPF1 controlled by SMG proteins is critical for NMD (Yamashita et al. 2005b). The phosphorylated UPF1 inhibits a new round of translation initiation and triggers degradation of mRNA (Isken et al. 2008). Yeast, which contains fewer introns, has a different strategy to distinguish an aberrant stop codon from a normal one. The main determinant to trigger NMD is an exceptionally long 3′ UTR, which prevents normal translation termination by association of eRF1 and eRF3 with cytoplasmic PABP (Amrani et al. 2004). Although this "*faux* 3′-UTR" model is incompatible with some observations in yeast—such as NMD still being active in mRNAs without poly(A) tails or in the PABP-deleted strains (Meaux et al. 2008)—an abnormal 3′UTR can activate NMD in a similar way in *Drosophila* as well (Behm-Ansmant et al. 2007). Recent studies in mammalian NMD indicate that mammalian NMD is also generally triggered by a longer 3′ UTR, as observed in yeast and *Drosophila* and the EJC located downstream of the PTC functions as an enhancer (Buhler et al. 2006).

Degradation of PTC-containing transcripts derived both from intron-containing and intronless genes has been observed in plants (Jofuku et al. 1989; Voelker et al. 1990; Dickey et al. 1994; van Hoof and Green 1996; Isshiki et al. 2001; Arciga-Reyes et al. 2006; Wu et al. 2007). Unusually long 3′ UTRs in the intronless NMD targets are indicated as a key feature to distinguish a PTC from the authentic termination codon in plants. mRNAs with longer 3′ UTRs are more effectively processed by NMD pathways (Kertesz et al. 2006; Hori and Watanabe 2007). Moreover, introns in the 3′ UTR act as *cis*-elements for NMD as well. As in mammals, the distance between PTC and its downstream intron is important to trigger NMD (Kertesz et al. 2006; Hori and Watanabe 2007; Wu et al. 2007). However, there are some exceptions suggesting that the effect of introns on NMD might be more complex in plants. In the case of the rice *WAXY* gene, proper splicing of an intron upstream of the PTC is required for NMD (Isshiki et al. 2001). Even in the last

exon, a PTC presented in a magnesium chelatase mutant of barley also induced NMD (Gadjieva et al. 2004).

The core components UPF1, UPF2, and UPF3 have been identified in plants. Additionally, homologues of mammalian SMG7 and components of EJC are found in *Arabidopsis* (Pendle et al. 2005; Kerenyi et al. 2008). Mutant analyses of *upf1* and *upf3* in *Arabidopsis* (Hori and Watanabe 2005; Arciga-Reyes et al. 2006) as well as down-regulation of *UPF2* expression by virus-induced gene silencing (VIGS) in *Nicotiana attenuata* indicated that all three components are required for NMD function in plants (Wu et al. 2007). Detailed analysis of these three factors by the transient NMD assay in *Nicotiana benthamiana* combined with VIGS revealed that UPF1, UPF2 and SMG7 are involved both in long 3′ UTR-based and intron-based NMD, whereas EJC components are required only for intron-based NMD, as expected (Kerenyi et al. 2008). The *low-beta-amylase1* (*lba1*), a missense mutant of *upf1*, and an allelic series of *ufp1* mutants in *Arabidopsis* indicate pleiotropic phenotypes, such as floral abnormalities, altered seed development, and seedling lethality, suggesting that NMD regulates a subset of genes important for plant development and survival. Indeed, gene expression profile analysis in the *lba1* mutant identified several putative targets of UPF1-dependent NMD, including many transcription factors and metabolic enzymes involved in a wide variety of functions (Yoine et al. 2006). Moreover, an analysis of a hypomorphic *smg7* mutant in *Arabidopsis* revealed that an essential function of SMG7 in NMD is required for exit from meiosis (Riehs et al. 2008). Interestingly, SMG7 in flowering plants conserved the exceptionally long 3′ UTR containing two introns, suggesting that SMG7 itself is also a direct target for NMD. In fact, SMG7 expression in *N. benthamiana* is increased in leaves where UPF1 and UPF2 are silenced, but not in leaves where EJC components are silenced, indicating that SMG7 mRNAs are negatively regulated by long 3′ UTR-based NMD. Since SMG7 is functional for both types of NMD in plants, intensity of NMD is likely feedback-controlled through the autoregulation of SMG7 (Kerenyi et al. 2008).

Once PTC-containing transcripts are recognized as target mRNAs for NMD, they are degraded by mechanisms similar to those of general mRNA decay. However, the selection of degradation strategies after the recognition of a PTC differs among species. In yeast, PTC-containing transcripts are degraded exonucleolyticly from both RNA ends (Muhlrad and Parker 1994; Hagan et al. 1995; Cao and Parker 2003), whereas in flies, endonucleolytic cleavage in the vicinity of the nonsense codon leads to elimination of PTC-containing transcripts (Gatfield and Izaurralde 2004). Mammalian NMD has been suggested to use similar degradation mechanisms as in yeast NMD (Chen and Shyu 2003; Lejeune et al. 2003; Couttet and Grange 2004). A revised mechanistic model was provided recently, in which the degradation induced by NMD is initiated by endonucleolytic cleavage near PTC, indicating that the degradation mechanism of PTC-containing mRNAs is conserved in metazoans (Eberle et al. 2009). Furthermore, SMG6 is reported to be a responsible endonuclease both in *Drosophila* and human cells (Huntzinger et al. 2008; Eberle et al. 2009). Together with the newly discovered endonuclease activity in the exosome, it is clear that endonuclease activity is more important for eukaryotic decay processes than originally thought (Wilusz 2009). Although plants have similar mRNA degradation machinery, further investigation is needed to determine which pathway contributes to NMD and the contribution of endonuclease activity. Two other mRNA surveillance systems, NSD and NGD, have been demonstrated in yeast; however, similar degradation pathways have not been identified in plants.

mRNA Degradation Coupled with Translation

In addition to NMD mentioned above, NSD and NGD are mRNA degradation systems coupled with translation. The link between mRNA degradation and translation other than the rapid decay of nonsense mRNAs is evident in plants, such as for the *ferredoxin-1* (*Fed1*) mRNA in pea and the *cystathionine gamma-synthase-1 (CGS1*) mRNA in *Arabidopsis*.

Fed1 mRNA, encoding the major chloroplast isoform of ferredoxin, is regulated in response to light/dark conditions at the level of transcription in etiolated seedlings and at the posttranscriptional level in green leaves (Elliott et al. 1989; Gallo-Meagher et al. 1992; Petracek et al. 1998). In green leaves, *Fed1* mRNA is rapidly degraded in the dark and is stabilized in the light (Petracek et al. 1998). The *cis*-element responsible for this posttranscriptional light regulation is the internal light regulatory element (iLRE) that spans the 5′-UTR and the first one-third of the *Fed1* coding region (Dickey et al. 1992). Several lines of evidence indicated that the change in the translation efficiency/mode of mRNA association with polyribosomes is a factor for determining mRNA stability of *Fed1* in response to light conditions (Dickey et al. 1994, 1998; Petracek et al. 1997, 1998; Hansen et al. 2001).

Cystathionine gamma-synthase (CGS) catalyzes the first committed step of the methionine biosynthesis pathway in higher plants (Matthews 1999). The *CGS1* gene is negatively feedback regulated at the mRNA stability level in response to *S*-adenosyl-L-methionine (AdoMet), a direct metabolite of methionine (Chiba et al. 1999, 2003). A nascent peptide designated as the MTO1 region, encoded by the first exon of *CGS1* itself, is sufficient and essential for this process (Ominato et al. 2002). Moreover, ongoing translation is critical for the posttranscriptional regulation of

CGS1 mRNA (Lambein et al. 2003). Consistent with this result, detailed analyses using a cell-free system have revealed that temporal translation elongation arrest immediately downstream of the MTO1 region occurs in response to AdoMet prior to *CGS1* mRNA degradation (Onouchi et al. 2005). In NMD, NSD, and NGD pathways, the responsible role players are recruited to an empty A-site of the ribosome to trigger mRNA degradation. In contrast, when the translation elongation of CGS1 mRNA is temporarily arrested, the A-site is occupied by the peptidyl-tRNA, suggesting that this nascent peptide-mediated posttranscriptional regulation is conducted by a novel mechanism (Onouchi et al. 2005).

Conclusions

We are still far from a complete understanding of the machinery for general or specific mRNA degradation in plants. However, unique features of mRNA degradation mechanisms have been identified. For example, several components responsible for mRNA decay are involved in hormonal responses, such as ABA and ethylene. In addition, mutant analyses of these components suggested that mRNA turnover is also involved in developmental processes. Studies using *Arabidopsis* and other model systems, such as forward and reverse genetics and functional genomics, will help to address how mRNA stability is regulated to conduct multiple physiological activities in plants. Finally, the recent demonstration that Parallel Analysis of RNA Ends (PARE) (German et al. 2008) and other next-generation deep sequencing strategies (Addo-Quaye et al. 2008; Gregory et al. 2008) can be used to detect mRNA decay intermediates with high sensitivity, should contribute to elucidating mRNA decay pathways and players in eukaryotic systems in general.

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References

Addo-Quaye C, Eshoo TW, Bartel DP, Axtell MJ (2008) Endogenous siRNA and miRNA targets identified by sequencing of the *Arabidopsis* degradome. Current Biology 18(10):758–762

Allmang C, Petfalski E, Podtelejnikov A, Mann M, Tollervey D, Mitchell P (1999) The yeast exosome and human PM-Scl are related complexes of 3′ –> 5′ exonucleases. Genes Dev 13 (16):2148–2158

Amrani N, Ganesan R, Kervestin S, Mangus DA, Ghosh S, Jacobson A (2004) A faux 3′-UTR promotes aberrant termination and triggers nonsense-mediated mRNA decay. Nature 432(7013):112–118

Anderson JS, Parker RP (1998) The 3′ to 5′ degradation of yeast mRNAs is a general mechanism for mRNA turnover that requires the SKI2 DEVH box protein and 3′ to 5′ exonucleases of the exosome complex. EMBO J 17(5):1497–1506

Arciga-Reyes L, Wootton L, Kieffer M, Davies B (2006) UPF1 is required for nonsense-mediated mRNA decay (NMD) and RNAi in *Arabidopsis*. Plant J 47(3):480–489

Beelman CA, Stevens A, Caponigro G, LaGrandeur TE, Hatfield L, Fortner DM, Parker R (1996) An essential component of the decapping enzyme required for normal rates of mRNA turnover. Nature 382(6592):642–646

Behm-Ansmant I, Gatfield D, Rehwinkel J, Hilgers V, Izaurralde E (2007) A conserved role for cytoplasmic poly(A)-binding protein 1 (PABPC1) in nonsense-mediated mRNA decay. Embo J 26 (6):1591–1601

Binder BM, Walker JM, Gagne JM, Emborg TJ, Hemmann G, Bleecker AB, Vierstra RD (2007) The *Arabidopsis* EIN3 binding F-Box proteins EBF1 and EBF2 have distinct but overlapping roles in ethylene signaling. Plant Cell 19(2):509–523

Brodersen P, Sakvarelidze-Achard L, Bruun-Rasmussen M, Dunoyer P, Yamamoto YY, Sieburth L, Voinnet O (2008) Widespread translational inhibition by plant miRNAs and siRNAs. Science 320(5880):1185–1190

Brown CE, Sachs AB (1998) Poly(A) tail length control in *Saccharomyces cerevisiae* occurs by message-specific deadenylation. Mol Cell Biol 18(11):6548–6559

Buhler M, Steiner S, Mohn F, Paillusson A, Muhlemann O (2006) EJC-independent degradation of nonsense immunoglobulin-mu mRNA depends on 3′ UTR length. Nat Struct Mol Biol 13 (5):462–464

Cao D, Parker R (2003) Computational modeling and experimental analysis of nonsense-mediated decay in yeast. Cell 113(4):533–545

Chang YF, Imam JS, Wilkinson MF (2007) The nonsense-mediated decay RNA surveillance pathway. Annu Rev Biochem 76:51–74

Chekanova JA, Shaw RJ, Wills MA, Belostotsky DA (2000) Poly(A) tail-dependent exonuclease AtRrp41p from *Arabidopsis thaliana* rescues 5.8 S rRNA processing and mRNA decay defects of the yeast ski6 mutant and is found in an exosome-sized complex in plant and yeast cells. J Biol Chem 275(42):33158–33166

Chekanova JA, Dutko JA, Mian IS, Belostotsky DA (2002) *Arabidopsis thaliana* exosome subunit AtRrp4p is a hydrolytic 3′–>5′ exonuclease containing S1 and KH RNA-binding domains. Nucleic Acids Res 30(3):695–700

Chekanova JA, Gregory BD, Reverdatto SV, Chen H, Kumar R, Hooker T, Yazaki J, Li P, Skiba N, Peng Q, Alonso J, Brukhin V, Grossniklaus U, Ecker JR, Belostotsky DA (2007) Genome-wide high-resolution mapping of exosome substrates reveals hidden features in the *Arabidopsis* transcriptome. Cell 131(7):1340–1353

Chen CY, Shyu AB (2003) Rapid deadenylation triggered by a nonsense codon precedes decay of the RNA body in a mammalian cytoplasmic nonsense-mediated decay pathway. Mol Cell Biol 23 (14):4805–4813

Chen CY, Gherzi R, Ong SE, Chan EL, Raijmakers R, Pruijn GJ, Stoecklin G, Moroni C, Mann M, Karin M (2001) AU binding proteins recruit the exosome to degrade ARE-containing mRNAs. Cell 107(4):451–464

Chen J, Chiang YC, Denis CL (2002) CCR4, a 3′–5′ poly(A) RNA and ssDNA exonuclease, is the catalytic component of the cytoplasmic deadenylase. Embo J 21(6):1414–1426

Chiba Y, Ishikawa M, Kijima F, Tyson RH, Kim J, Yamamoto A, Nambara E, Leustek T, Wallsgrove RM, Naito S (1999) Evidence for autoregulation of cystathionine gamma-synthase mRNA stability in *Arabidopsis*. Science 286(5443):1371–1374

Chiba Y, Sakurai R, Yoshino M, Ominato K, Ishikawa M, Onouchi H, Naito S (2003) *S*-adenosyl-L-methionine is an effector in the posttranscriptional autoregulation of the cystathionine gamma-

synthase gene in *Arabidopsis*. Proc Natl Acad Sci USA 100 (18):10225–10230
Chiba Y, Johnson MA, Lidder P, Vogel JT, van Erp H, Green PJ (2004) AtPARN is an essential poly(A) ribonuclease in *Arabidopsis*. Gene 328:95–102
Coller J, Parker R (2004) Eukaryotic mRNA decapping. Annu Rev Biochem 73:861–890
Copeland PR, Wormington M (2001) The mechanism and regulation of deadenylation: identification and characterization of *Xenopus* PARN. Rna 7(6):875–886
Couttet P, Grange T (2004) Premature termination codons enhance mRNA decapping in human cells. Nucleic Acids Res 32(2):488–494
Daugeron MC, Mauxion F, Seraphin B (2001) The yeast POP2 gene encodes a nuclease involved in mRNA deadenylation. Nucleic Acids Res 29(12):2448–2455
Dehlin E, Wormington M, Korner CG, Wahle E (2000a) Cap-dependent deadenylation of mRNA. EMBO J 19(5):1079–1086
Dehlin E, Wormington M, Korner CG, Wahle E (2000b) Cap-dependent deadenylation of mRNA. Embo J 19(5):1079–1086
Denis CL, Chen J (2003) The CCR4-NOT complex plays diverse roles in mRNA metabolism. Prog Nucleic Acid Res Mol Biol 73:221–250
Dichtl B, Stevens A, Tollervey D (1997) Lithium toxicity in yeast is due to the inhibition of RNA processing enzymes. EMBO J 16 (23):7184–7195
Dickey LF, Gallo-Meagher M, Thompson WF (1992) Light regulatory sequences are located within the 5′ portion of the Fed-1 message sequence. Embo J 11(6):2311–2317
Dickey LF, Nguyen TT, Allen GC, Thompson WF (1994) Light modulation of ferredoxin mRNA abundance requires an open reading frame. Plant Cell 6(8):1171–1176
Dickey LF, Petracek ME, Nguyen TT, Hansen ER, Thompson WF (1998) Light regulation of Fed-1 mRNA requires an element in the 5′ untranslated region and correlates with differential polyribosome association. Plant Cell 10(3):475–484
Doma MK, Parker R (2006) Endonucleolytic cleavage of eukaryotic mRNAs with stalls in translation elongation. Nature 440 (7083):561–564
Dunckley T, Parker R (1999) The DCP2 protein is required for mRNA decapping in Saccharomyces cerevisiae and contains a functional MutT motif. EMBO J 18(19):5411–5422
Dupressoir A, Morel AP, Barbot W, Loireau MP, Corbo L, Heidmann T (2001) Identification of four families of yCCR4- and Mg^{2+}-dependent endonuclease-related proteins in higher eukaryotes, and characterization of orthologs of yCCR4 with a conserved leucine-rich repeat essential for hCAF1/hPOP2 binding. BMC Genomics 2(1):9
Dziembowski A, Lorentzen E, Conti E, Seraphin B (2007) A single subunit, Dis3, is essentially responsible for yeast exosome core activity. Nat Struct Mol Biol 14(1):15–22
Eberle AB, Lykke-Andersen S, Muhlemann O, Jensen TH (2009) SMG6 promotes endonucleolytic cleavage of nonsense mRNA in human cells. Nat Struct Mol Biol 16(1):49–55
Elliott RC, Dickey LF, White MJ, Thompson WF (1989) *cis*-Acting elements for light regulation of pea ferredoxin I gene expression are located within transcribed sequences. Plant Cell 1(7):691–698
Eulalio A, Rehwinkel J, Stricker M, Huntzinger E, Yang SF, Doerks T, Dorner S, Bork P, Boutros M, Izaurralde E (2007) Target-specific requirements for enhancers of decapping in miRNA-mediated gene silencing. Genes Dev 21(20):2558–2570
Fenger-Gr M, Fillman C, Norrild B, Lykke-Andersen J (2005) Multiple processing body factors and the are binding protein TTP activate mRNA decapping. Molecular Cell 20(6):905–915
Frischmeyer PA, van Hoof A, O'Donnell K, Guerrerio AL, Parker R, Dietz HC (2002) An mRNA surveillance mechanism that eliminates transcripts lacking termination codons. Science 295 (5563):2258–2261
Funakoshi Y, Doi Y, Hosoda N, Uchida N, Osawa M, Shimada I, Tsujimoto M, Suzuki T, Katada T, Hoshino S (2007) Mechanism of mRNA deadenylation: evidence for a molecular interplay between translation termination factor eRF3 and mRNA deadenylases. Genes Dev 21(23):3135–3148
Gadjieva R, Axelsson E, Olsson U, Vallon-Christersson J, Hansson M (2004) Nonsense-mediated mRNA decay in barley mutants allows the cloning of mutated genes by a microarray approach. Plant Physiol Biochem 42(7–8):681–685
Gallo-Meagher M, Sowinski DA, Elliott RC, Thompson WF (1992) Both internal and external regulatory elements control expression of the pea Fed-1 gene in transgenic tobacco seedlings. Plant Cell 4(4):389–395
Gao M, Fritz DT, Ford LP, Wilusz J (2000) Interaction between a poly (A)-specific ribonuclease and the 5′ cap influences mRNA deadenylation rates in vitro. Molecular Cell 5(3):479–488
Gatfield D, Izaurralde E (2004) Nonsense-mediated messenger RNA decay is initiated by endonucleolytic cleavage in *Drosophila*. Nature 429(6991):575–578
Gazzani S, Lawrenson T, Woodward C, Headon D, Sablowski R (2004) A link between mRNA turnover and RNA interference in *Arabidopsis*. Science 306(5698):1046–1048
German MA, Pillay M, Jeong DH, Hetawal A, Luo S, Janardhanan P, Kannan V, Rymarquis LA, Nobuta K, German R, De Paoli E, Lu C, Schroth G, Meyers BC, Green PJ (2008) Global identification of microRNA-target RNA pairs by parallel analysis of RNA ends. Nat Biotechnol 26(8):941–946
Goeres DC, Van Norman JM, Zhang W, Fauver NA, Spencer ML, Sieburth LE (2007) Components of the *Arabidopsis* mRNA decapping complex are required for early seedling development. Plant Cell 19(5):1549–1564
Gregory BD, O'Malley RC, Lister R, Urich MA, Tonti-Filippini J, Chen H, Millar AH, Ecker JR (2008) A link between RNA metabolism and silencing affecting *Arabidopsis* development. Dev Cell 14(6):854–866
Gy I, Gasciolli V, Lauressergues D, Morel JB, Gombert J, Proux F, Proux C, Vaucheret H, Mallory AC (2007) *Arabidopsis* FIERY1, XRN2, and XRN3 are endogenous RNA silencing suppressors. Plant Cell 19(11):3451–3461
Hagan KW, Ruiz-Echevarria MJ, Quan Y, Peltz SW (1995) Characterization of cis-acting sequences and decay intermediates involved in nonsense-mediated mRNA turnover. Mol Cell Biol 15(2):809–823
Hansen ER, Petracek ME, Dickey LF, Thompson WF (2001) The 5′ end of the pea ferredoxin-1 mRNA mediates rapid and reversible light-directed changes in translation in tobacco. Plant Physiol 125 (2):770–778
Hatfield L, Beelman CA, Stevens A, Parker R (1996) Mutations in trans-acting factors affecting mRNA decapping in *Saccharomyces cerevisiae*. Mol Cell Biol 16(10):5830–5838
Hooker TS, Lam P, Zheng H, Kunst L (2007) A core subunit of the RNA-processing/degrading exosome specifically influences cuticular wax biosynthesis in *Arabidopsis*. Plant Cell 19(3):904–913
Hori K, Watanabe Y (2005) UPF3 suppresses aberrant spliced mRNA in *Arabidopsis*. Plant J 43(4):530–540
Hori K, Watanabe Y (2007) Context analysis of termination codons in mRNA that are recognized by plant NMD. Plant Cell Physiol 48 (7):1072–1078
Hosoda N, Kobayashi T, Uchida N, Funakoshi Y, Kikuchi Y, Hoshino S, Katada T (2003) Translation termination factor eRF3 mediates mRNA decay through the regulation of deadenylation. J Biol Chem 278(40):38287–38291
Houseley J, LaCava J, Tollervey D (2006) RNA-quality control by the exosome. Nat Rev Mol Cell Biol 7(7):529–539

Hsu CL, Stevens A (1993) Yeast cells lacking 5′–>3′ exoribonuclease 1 contain mRNA species that are poly(A) deficient and partially lack the 5′ cap structure. Mol Cell Biol 13(8):4826–4835

Huntzinger E, Kashima I, Fauser M, Sauliere J, Izaurralde E (2008) SMG6 is the catalytic endonuclease that cleaves mRNAs containing nonsense codons in metazoan. Rna 14(12):2609–2617

Isken O, Kim YK, Hosoda N, Mayeur GL, Hershey JW, Maquat LE (2008) Upf1 phosphorylation triggers translational repression during nonsense-mediated mRNA decay. Cell 133(2):314–327

Isshiki M, Yamamoto Y, Satoh H, Shimamoto K (2001) Nonsense-mediated decay of mutant waxy mRNA in rice. Plant Physiol 125 (3):1388–1395

Ivanov PV, Gehring NH, Kunz JB, Hentze MW, Kulozik AE (2008) Interactions between UPF1, eRFs, PABP and the exon junction complex suggest an integrated model for mammalian NMD pathways. Embo J 27(5):736–747

Iwasaki S, Takeda A, Motose H, Watanabe Y (2007) Characterization of *Arabidopsis* decapping proteins AtDCP1 and AtDCP2, which are essential for post-embryonic development. FEBS Lett 581 (13):2455–2459

Jofuku KD, Schipper RD, Goldberg RB (1989) A frameshift mutation prevents Kunitz trypsin inhibitor mRNA accumulation in soybean embryos. Plant Cell 1(5):567

Johnson AW (1997) Rat1p and Xrn1p are functionally interchangeable exoribonucleases that are restricted to and required in the nucleus and cytoplasm, respectively. Mol Cell Biol 17(10):6122–6130

Kashima I, Yamashita A, Izumi N, Kataoka N, Morishita R, Hoshino S, Ohno M, Dreyfuss G, Ohno S (2006) Binding of a novel SMG-1-Upf1-eRF1-eRF3 complex (SURF) to the exon junction complex triggers Upf1 phosphorylation and nonsense-mediated mRNA decay. Genes Dev 20(3):355–367

Kastenmayer JP, Green PJ (2000) Novel features of the XRN-family in *Arabidopsis*: evidence that AtXRN4, one of several orthologs of nuclear Xrn2p/Rat1p, functions in the cytoplasm. Proc Natl Acad Sci USA 97(25):13985–13990

Katiyar-Agarwal S, Gao S, Vivian-Smith A, Jin H (2007) A novel class of bacteria-induced small RNAs in *Arabidopsis*. Genes Dev 21(23):3123–3134

Kendrick MD, Chang C (2008) Ethylene signaling: new levels of complexity and regulation. Curr Opin Plant Biol 11(5):479–485

Kerenyi Z, Merai Z, Hiripi L, Benkovics A, Gyula P, Lacomme C, Barta E, Nagy F, Silhavy D (2008) Inter-kingdom conservation of mechanism of nonsense-mediated mRNA decay. Embo J 27 (11):1585–1595

Kertesz S, Kerenyi Z, Merai Z, Bartos I, Palfy T, Barta E, Silhavy D (2006) Both introns and long 3′-UTRs operate as *cis*-acting elements to trigger nonsense-mediated decay in plants. Nucleic Acids Res 34(21):6147–6157

Kim B-H, von Arnim AG (2008) *FIERY1* regulates light-mediated repression of cell elongation and flowering time via its 3′(2′), 5′-bisphosphate nucleotidase activity. The Plant Journal (in press). doi.10.1111/j.1365-313X.2008.03770.x

Korner CG, Wahle E (1997) Poly(A) tail shortening by a mammalian poly(A)-specific 3′-exoribonuclease. J Biol Chem 272(16):10448–10456

Korner CG, Wormington M, Muckenthaler M, Schneider S, Dehlin E, Wahle E (1998) The deadenylating nuclease (DAN) is involved in poly(A) tail removal during the meiotic maturation of *Xenopus* oocytes. EMBO J 17(18):5427–5437

LaGrandeur TE, Parker R (1998) Isolation and characterization of Dcp1p, the yeast mRNA decapping enzyme. EMBO J 17 (5):1487–1496

Lai WS, Kennington EA, Blackshear PJ (2003) Tristetraprolin and its family members can promote the cell-free deadenylation of AU-rich element-containing mRNAs by poly(A) ribonuclease. Mol Cell Biol 23(11):3798–3812

Lambein I, Chiba Y, Onouchi H, Naito S (2003) Decay kinetics of autogenously regulated CGS1 mRNA that codes for cystathionine gamma-synthase in *Arabidopsis thaliana*. Plant Cell Physiol 44(9):893–900

Le Hir H, Moore MJ, Maquat LE (2000) Pre-mRNA splicing alters mRNP composition: evidence for stable association of proteins at exon–exon junctions. Genes Dev 14(9):1098–1108

Lebreton A, Tomecki R, Dziembowski A, Seraphin B (2008) Endonucleolytic RNA cleavage by a eukaryotic exosome. Nature 456(7224):993–996

Lee HH, Kim YS, Kim KH, Heo I, Kim SK, Kim O, Kim HK, Yoon JY, Kim HS, Kim do J, Lee SJ, Yoon HJ, Kim SJ, Lee BG, Song HK, Kim VN, Park CM, Suh SW (2007) Structural and functional insights into Dom34, a key component of no-go mRNA decay. Mol Cell 27(6):938–950

Lejeune F, Li X, Maquat LE (2003) Nonsense-mediated mRNA decay in mammalian cells involves decapping, deadenylating, and exonucleolytic activities. Mol Cell 12(3):675–687

Lin MD, Fan SJ, Hsu WS, Chou TB (2006) *Drosophila* decapping protein 1, dDcp1, is a component of the oskar mRNP complex and directs its posterior localization in the oocyte. Dev Cell 10 (5):601–613

Liu H, Rodgers ND, Jiao X, Kiledjian M (2002) The scavenger mRNA decapping enzyme DcpS is a member of the HIT family of pyrophosphatases. Embo J 21(17):4699–4708

Liu Q, Greimann JC, Lima CD (2006) Reconstitution, activities, and structure of the eukaryotic RNA exosome. Cell 127(6):1223–1237

Lowell JE, Rudner DZ, Sachs AB (1992) 3′-UTR-dependent deadenylation by the yeast poly(A) nuclease. Genes Dev 6(11):2088–2099

Matthews BF (1999) Lysine, thereonine, and methionine biosynthesis. In: Singh BK (ed) Plant amino acids: biochemistry and biotechnology, vol. Marcel Dekker, New York, pp 205–225

Meaux S, van Hoof A, Baker KE (2008) Nonsense-mediated mRNA decay in yeast does not require PAB1 or a poly(A) tail. Mol Cell 29(1):134–140

Mitchell P, Tollervey D (2003) An NMD pathway in yeast involving accelerated deadenylation and exosome-mediated 3′–>5′ degradation. Mol Cell 11(5):1405–1413

Mitchell P, Petfalski E, Shevchenko A, Mann M, Tollervey D (1997) The exosome: a conserved eukaryotic RNA processing complex containing multiple 3′–>5′ exoribonucleases. Cell 91(4):457–466

Muhlrad D, Parker R (1994) Premature translational termination triggers mRNA decapping. Nature 370(6490):578–581

Mukherjee D, Gao M, O'Connor JP, Raijmakers R, Pruijn G, Lutz CS, Wilusz J (2002) The mammalian exosome mediates the efficient degradation of mRNAs that contain AU-rich elements. EMBO J 21(1–2):165–174

Nagy E, Maquat LE (1998) A rule for termination-codon position within intron-containing genes: when nonsense affects RNA abundance. Trends Biochem Sci 23(6):198–199

Nishimura N, Kitahata N, Seki M, Narusaka Y, Narusaka M, Kuromori T, Asami T, Shinozaki K, Hirayama T (2005) Analysis of ABA hypersensitive germination2 revealed the pivotal functions of PARN in stress response in *Arabidopsis*. Plant J 44 (6):972–984

Olmedo G, Guo H, Gregory BD, Nourizadeh SD, Aguilar-Henonin L, Li H, An F, Guzman P, Ecker JR (2006) Ethylene-insensitive5 encodes a 5′–>3′ exoribonuclease required for regulation of the EIN3-targeting F-box proteins EBF1/2. Proc Natl Acad Sci USA 103(36):13286–13293

Ominato K, Akita H, Suzuki A, Kijima F, Yoshino T, Yoshino M, Chiba Y, Onouchi H, Naito S (2002) Identification of a short highly conserved amino acid sequence as the functional region required for posttranscriptional autoregulation of the cystathionine gamma-synthase gene in *Arabidopsis*. J Biol Chem 277(39):36380–36386

Onouchi H, Nagami Y, Haraguchi Y, Nakamoto M, Nishimura Y, Sakurai R, Nagao N, Kawasaki D, Kadokura Y, Naito S (2005) Nascent peptide-mediated translation elongation arrest coupled with mRNA degradation in the CGS1 gene of *Arabidopsis*. Genes Dev 19(15):1799–1810

Orban TI, Izaurralde E (2005) Decay of mRNAs targeted by RISC requires XRN1, the Ski complex, and the exosome. RNA 11(4):459–469

Pendle AF, Clark GP, Boon R, Lewandowska D, Lam YW, Andersen J, Mann M, Lamond AI, Brown JW, Shaw PJ (2005) Proteomic analysis of the *Arabidopsis* nucleolus suggests novel nucleolar functions. Mol Biol Cell 16(1):260–269

Petracek ME, Dickey LF, Huber SC, Thompson WF (1997) Light-regulated changes in abundance and polyribosome association of ferredoxin mRNA are dependent on photosynthesis. Plant Cell 9 (12):2291–2300

Petracek ME, Dickey LF, Nguyen TT, Gatz C, Sowinski DA, Allen GC, Thompson WF (1998) Ferredoxin-1 mRNA is destabilized by changes in photosynthetic electron transport. Proc Natl Acad Sci USA 95(15):9009–9013

Potuschak T, Vansiri A, Binder BM, Lechner E, Vierstra RD, Genschik P (2006) The exoribonuclease XRN4 is a component of the ethylene response pathway in *Arabidopsis*. Plant Cell 18(11):3047–3057

Raijmakers R, Schilders G, Pruijn GJ (2004) The exosome, a molecular machine for controlled RNA degradation in both nucleus and cytoplasm. Eur J Cell Biol 83(5):175–183

Reverdatto SV, Dutko JA, Chekanova JA, Hamilton DA, Belostotsky DA (2004) mRNA deadenylation by PARN is essential for embryogenesis in higher plants. Rna 10(8):1200–1214

Riehs N, Akimcheva S, Puizina J, Bulankova P, Idol RA, Siroky J, Schleiffer A, Schweizer D, Shippen DE, Riha K (2008) *Arabidopsis* SMG7 protein is required for exit from meiosis. J Cell Sci 121(Pt 13):2208–2216

Sachs AB, Deardorff JA (1992) Translation initiation requires the PAB-dependent poly(A) ribonuclease in yeast. Cell 70(6):961–973

Saito K (2004) Sulfur assimilatory metabolism. The long and smelling road. Plant Physiol 136(1):2443–2450

Sakuno T, Araki Y, Ohya Y, Kofuji S, Takahashi S, Hoshino S, Katada T (2004) Decapping reaction of mRNA requires Dcp1 in fission yeast: its characterization in different species from yeast to human. J Biochem 136(6):805–812

Sarowar S, Oh HW, Cho HS, Baek KH, Seong ES, Joung YH, Choi GJ, Lee S, Choi D (2007) Capsicum annuum CCR4-associated factor CaCAF1 is necessary for plant development and defence response. Plant J 51(5):792–802

Schaeffer D, Tsanova B, Barbas A, Reis FP, Dastidar EG, Sanchez-Rotunno M, Arraiano CM, van Hoof A (2009) The exosome contains domains with specific endoribonuclease, exoribonuclease and cytoplasmic mRNA decay activities. Nat Struct Mol Biol 16 (1):56–62

Siddiqui N, Mangus DA, Chang TC, Palermino JM, Shyu AB, Gehring K (2007) Poly(A) nuclease interacts with the C-terminal domain of polyadenylate-binding protein domain from poly(A)-binding protein. J Biol Chem 282(34):25067–25075

Souret FF, Kastenmayer JP, Green PJ (2004) AtXRN4 degrades mRNA in *Arabidopsis* and its substrates include selected miRNA targets. Mol Cell 15(2):173–183

Takahashi S, Araki Y, Sakuno T, Katada T (2003) Interaction between Ski7p and Upf1p is required for nonsense-mediated 3′-to-5′ mRNA decay in yeast. Embo J 22(15):3951–3959

Tucker M, Valencia-Sanchez MA, Staples RR, Chen J, Denis CL, Parker R (2001) The transcription factor associated Ccr4 and Caf1 proteins are components of the major cytoplasmic mRNA deadenylase in *Saccharomyces cerevisiae*. Cell 104(3):377–386

Tucker M, Staples RR, Valencia-Sanchez MA, Muhlrad D, Parker R (2002) Ccr4p is the catalytic subunit of a Ccr4p/Pop2p/Notp mRNA deadenylase complex in *Saccharomyces cerevisiae*. Embo J 21(6):1427–1436

Uchida N, Hoshino S, Katada T (2004) Identification of a human cytoplasmic poly(A) nuclease complex stimulated by poly(A)-binding protein. J Biol Chem 279(2):1383–1391

van Hoof A, Green PJ (1996) Premature nonsense codons decrease the stability of phytohemagglutinin mRNA in a position-dependent manner. Plant J 10(3):415–424

van Hoof A, Frischmeyer PA, Dietz HC, Parker R (2002) Exosome-mediated recognition and degradation of mRNAs lacking a termination codon. Science 295(5563):2262–2264

Voelker TA, Moreno J, Chrispeels MJ (1990) Expression analysis of a pseudogene in transgenic tobacco: a frameshift mutation prevents mRNA accumulation. Plant Cell 2(3):255–261

Wang Z, Kiledjian M (2001) Functional link between the mammalian exosome and mRNA decapping. Cell 107(6):751–762

Wilson PB, Estavillo GM, Field KJ, Pornsiriwong W, Carroll AJ, Howell KA, Woo NS, Lake JA, Smith SM, Millar AH, von Caemmere S, Pogson BJ (2009) The nucleotidase/phosphatase SAL1 is a negative regulator of drought tolerance in *Arabidopsis*. The Plant Journal (in press). doi:10.1111/j.1365–313X.2008.03780.x

Wilusz J (2009) RNA stability: is it the endo' the world as we know it? Nat Struct Mol Biol 16(1):9–10

Wu J, Kang JH, Hettenhausen C, Baldwin IT (2007) Nonsense-mediated mRNA decay (NMD) silences the accumulation of aberrant trypsin proteinase inhibitor mRNA in *Nicotiana attenuata*. Plant J 51 (4):693–706

Xu J, Yang JY, Niu QW, Chua NH (2006) *Arabidopsis* DCP2, DCP1, and VARICOSE form a decapping complex required for postembryonic development. Plant Cell 18(12):3386–3398

Yamashita A, Chang TC, Yamashita Y, Zhu W, Zhong Z, Chen CY, Shyu AB (2005a) Concerted action of poly(A) nucleases and decapping enzyme in mammalian mRNA turnover. Nat Struct Mol Biol 12(12):1054–1063

Yamashita A, Kashima I, Ohno S (2005b) The role of SMG-1 in nonsense-mediated mRNA decay. Biochim Biophys Acta 1754 (1–2):305–315

Yoine M, Ohto MA, Onai K, Mita S, Nakamura K (2006) The lba1 mutation of UPF1 RNA helicase involved in nonsense-mediated mRNA decay causes pleiotropic phenotypic changes and altered sugar signalling in *Arabidopsis*. Plant J 47(1):49–62