ISSN 0006-2979, Biochemistry (Moscow), 2010, Vol. 75, No. 11, pp. 1316-1323. © Pleiades Publishing, Ltd., 2010.
Original Russian Text © A. A. Zamyatnin, Jr., K. G. Lyamzaev, R. A. Zinovkin, 2010, published in Biokhimiya, 2010, Vol. 75, No. 11, pp. 1489-1498.

REVIEW

A-to-I RNA Editing: A Contribution to Diversity of the Transcriptome and an Organism's Development

A. A. Zamyatnin, Jr.*, K. G. Lyamzaev, and R. A. Zinovkin

Institute of Mitoengineering and Belozersky Institute of Physico-Chemical Biology, Lomonosov Moscow State University, 119991 Moscow, Russia; fax: (495) 939-5945; E-mail: zamyat@belozersky.msu.ru

Received June 30, 2010

Abstract—The complexity of multicellular organisms requires both an increase in genetic information and fine tuning in regulation of gene expression. One of the mechanisms responsible for these functions is RNA editing. RNA editing is a complex process affecting the mechanism of changes in transcriptome sequences. The best studied example of this process is A-to-I RNA editing. On the organism's level, RNA editing plays a key role during ontogenesis and in the defense against pathogens. Disorders in A-to-I RNA editing lead to serious abnormalities. The importance of RNA editing increases with an increase in the organism's complexity. Correct RNA editing is an indispensable factor of an organism's development and probably determines the lifespan of higher eukaryotes.

DOI: 10.1134/S0006297910110027

Key words: ADAR1, ADAR2, microRNA, RNA interference, alternative splicing, translational regulation

Many authors characterize the current state of molecular biology as a "postgenomic era" and thus emphasize that the enormous amount of information obtained as a result of the genome sequencing is insufficient for understanding the activities of living systems. Up to now, the central dogma of molecular biology (DNA → RNA → protein) is a subject of more precise corrections and gradually forms an extremely elaborated picture of gene expression. The discovery of post-transcriptional modification was an impressive example of "correcting" the basic postulate. Main processes of post-transcriptional modification in eukaryotes are alternative splicing, capping, polyadenylation, and also RNA editing [1, 2].

The best studied process of specific RNA editing is adenosine (A) deamination leading to its conversion into inosine (I), which is recognized by the cell as guanosine (G) (Fig. 1). A-to-I RNA editing is catalyzed by proteins of the adenosine deaminase family acting on RNA (ADAR family), and it occurs only on double-stranded regions of RNA. Thus, adenosine deaminases can cause point replacements in RNA converting A to an analog of G. In contrast to point mutations in DNA, which are irreversible for the cell genome, RNA editing mediated nucleotide replacements are really reversible [3, 4].

Functional and genetic studies have revealed that mutations in the ADAR genes or disturbances in the regulation of enzymatic activities of the ADAR proteins lead to some human diseases and also to various disorders during viral infections [5]. The correlation between the presence of single-nucleotide polymorphisms in human *ADAR1* and *ADAR2* genes and lifespan suggests a functional role of A-to-I RNA editing in aging process [6].

FAMILY OF dsRNA ADENOSINE DEAMINASES

The ADAR family proteins are thought to arise at the stage of appearance of multicellular animals. Adenosine deaminases modifying tRNA (ADAT), which are widely distributed in *Protozoa*, seem to be ancestors of these enzymes. The gene *ADAR1* duplication gave rise to *ADAR2* gene. Then, at the stage of appearance of chordates two more genes of this family evolved: *ADAR3* and *TENR* (testes-specific nuclear RNA-binding protein) [7].

In mammals there are two forms of the ADAR1 protein: p150 and p110. They are produced due to the use of

Abbreviations: ADAR, dsRNA adenosine deaminase; ADAT, tRNA adenosine deaminase; dsRNA, double-stranded RNA; HDV, hepatitis delta virus; HIV, human immunodeficiency virus; DSH, dyschromatosis symmetrica hereditaria; IP6, inositol hexaphosphate; TENR, testes-specific nuclear RNA-binding protein; UTR, untranslated region.

* To whom correspondence should be addressed.

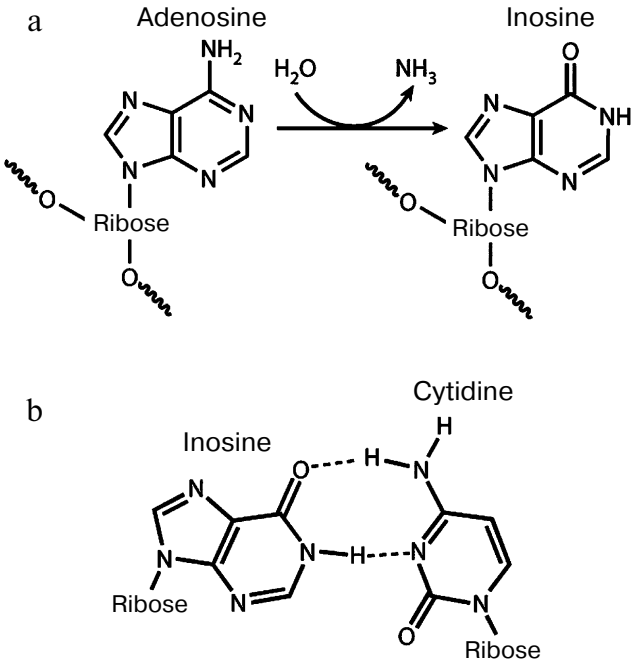

Fig. 1. Deamination reaction performed by ADAR family proteins. a) Deamination with adenosine converting to inosine; b) complementary interaction of inosine with cytidine.

two different promoters and alternative initiator codons. The protein p110 lacks the *N*-terminal 296 amino acid residues of protein p150. The promoter for p150 is induced only by interferon, whereas the promoter for p110 is constitutive. ADAR2 is also a constitutively expressed protein [3]. ADAR1 and ADAR2 are synthesized in all tissues of the organism. The expression of both ADAR3 and TENR is highly tissue-specific (in neurons and testes, respectively) [7].

All proteins of the ADAR family have similar structure (Fig. 2). The adenosine deaminase domain (Deaminase) is located close to the *C*-terminal region and is preceded by one, two, or three dsRNA binding motifs (dsRBD). In addition, ADAR1 and ADAR2 have an *N*-terminal z-DNA-binding domain, whereas ADAR3 has ssRNA-binding domain enriched with arginine on its *N*-terminus (R). A-to-I RNA editing activity was shown only for ADAR1 p150, ADAR1 p110, and ADAR2 [4]. *In vitro* experiments have shown that enzymatic activity of ADAR1 and ADAR2 requires protein homodimerization [8]. Unexpectedly, it was found that inositol hexaphosphate (IP6) is required for the enzymatic activity of ADAR2 [9]. It remains unclear whether the protein ADAR3 can function as adenosine deaminase, whereas the main function of TENR is only the binding with double-stranded regions of RNA [4, 10, 11].

The interferon-induced form ADAR1 p150 is located in both the nucleus and cytoplasm, whereas ADAR1 p110, ADAR2, ADAR3, and TENR are nuclear proteins [4, 11, 12].

ACTION OF A-TO-I DEAMINASES IN BIOGENESIS OF mRNA and miRNA

It has been theoretically predicted that >85% of mRNA precursors (pre-mRNA) can be A-to-I edited. The main targets of the editing are located in sequences of introns and also within untranslatable regions (UTR) [13].

It has been shown that A-to-I pre-mRNA editing can occur in all regions: in the region of exons, introns,

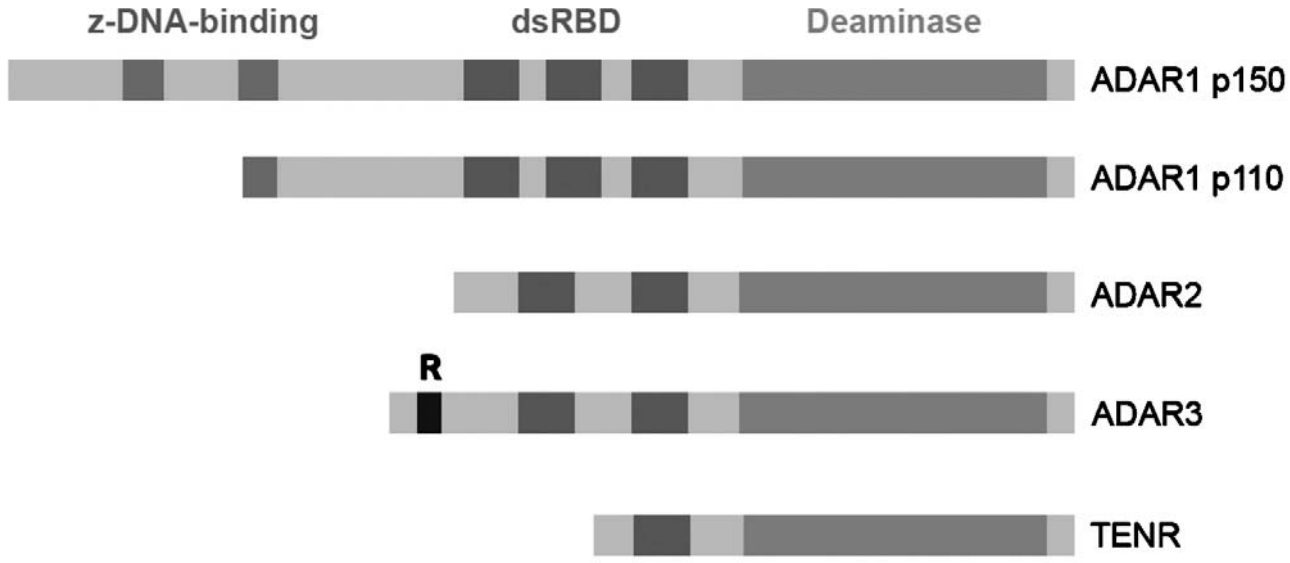

Fig. 2. Domain organization of proteins of the human dsRNA adenosine deaminase family.

Table 1. Influence of A-to-I editing in biogenesis of mRNA

Stages of mRNA biogenesis	Possible results of A-to-I editing
Splicing	Production of alternative sites of splicing [14]
Export from nucleus	Repression of mRNA export into cytoplasm [15, 16]
Regulation of translation through miRNA	mRNA–miRNA interactions [30]
Translation	Replacements within codon triplets [4]
Degradation	Specific degradation of I-containing RNAs [17]

and also within the 5′- and 3′-UTRs. Such editing can change the subsequent maturation of mRNA and/or functions of a protein product encoded by mRNA (Table 1).

Upon transcription and editing, pre-mRNA is subjected to splicing. The AA dinucleotide sequence can be edited into AI, which is equivalent to AG for the splicing machinery. AG in the 3′-region of the intron is a canonical splicing site. It was shown recently that AI site can function as a splicing site. This results in an additional exon in the mRNA sequence [14].

The next stage of mRNA biogenesis that can be corrected by A-to-I editing is the process of mRNA export from the nucleus. Recently it was shown that RNA-binding protein $p54^{nrb}$ interacts with inosine-enriched areas of 3′-UTR in mRNA and inhibits mRNA export from the nucleus [15, 16]. On entrance into the cytoplasm mRNA subjected to excessive A-to-I editing can be degraded by the cytoplasmic endonuclease specifically recognizing and hydrolyzing hyper-edited regions of RNA [17].

A-to-I editing is most strikingly exemplified by point replacements in the coding region of mRNA that are responsible for changes in functional properties of the final protein product [4, 18]. The available data present knowledge on functional consequences of the editing of mRNA coding regions that cause significant replacements in proteins of the glutamate receptor (GluR-B), serotonin receptor (5-HT_{2C}R), and ion channels: K^+-channel of mammals (Kv1.1), squid (Kv1.1A), and Na^+-channel of *Drosophila melanogaster* [4]. In all these cases the editing results in the synthesis of new protein isoforms with different functional properties. Thus, in the case of GluR-B subunit editing, the glutamine codon CAG is converted to the arginine codon CIG (Q/R-site). Incorporation of the GluR-B subunit containing the Q/R-replacement into the ion channel tetramer causes changes in the GluR permeability: the channel becomes impermeable for Ca^{2+} [19, 20]. Another example is the serotonin receptor 5-HT_{2C}R mRNA that contains a number of coding regions subjected to editing: the codon of isoleucine AUA, of asparagine AAU, and of isoleucine AUU. Twenty four isoforms of the final protein product are known possessing different affinities to serotonin [21, 22]. All these examples concern functions of neurons, which are the most highly specialized cells.

Besides mRNA precursors, ~16% of noncoding RNA precursors (pri-miRNA) undergo A-to-I editing [23]. This can influence the subsequent biogenesis of pri-miRNA (Table 2). Upon the transcription and a possible editing, pri-miRNAs are processed by endonuclease Drosha. The editing effect can be revealed even at this stage: the mouse pri-miRNA-142 containing I cannot be processed by Drosha but it is degraded under the influence of endonuclease Tudor-sn [24], which is highly specific to I-containing dsRNA substrates [25]. The opposite effect is also possible when the editing stimulates the processing of pri-miRNA [23]. In some cases deamination activity is not required to inhibit the pri-miRNA processing, only the RNA-binding properties of ADAR2 being sufficient [26].

Upon pri-miRNA processing, the produced precursor of microRNA (pre-miRNA) is exported from the nucleus. Up to now there is no experimental data on the influence of A-to-I editing of pre-miRNA on its export from the nucleus. Upon the entrance into the cytoplasm, pre-miRNA undergoes processing by endonuclease

Table 2. Influence of A-to-I editing in miRNA biogenesis

Stages of miRNA biogenesis	Possible results of A-to-I editing
Processing of pri-miRNA by endonuclease Drosha, production of pre-miRNA	Repression of processing by endonuclease Drosha [24], or, by contrast, stimulation of processing by endonuclease Drosha [23]
Export of pre-miRNA from nucleus	?
Processing of pre-miRNA by endonuclease Dicer, production of mature miRNA	Repression of processing by endonuclease Dicer [27]
Translation regulation due to production of mRNA–miRNA complex	miRNA–mRNA interactions [30]
Degradation	Specific degradation of I-containing pri-miRNAs [24]

Dicer. In the case of human pre-miRNA-151 the editing was shown to inhibit the pre-miRNA-151 processing by endonuclease Dicer [27].

Thus, A-to-I RNA editing regulates maturation of both mRNA and miRNA. These molecules are involved in RNA interference, which is based on formation of Watson–Crick pairs between miRNA and 3′-UTR of the target mRNA. Such interactions are regulated by rather strict rules: close to 100% complementarity leads to degradation of the target mRNA through mechanisms of RNA interference, whereas formation of complementary pairs between 3′-UTR of mRNA and nucleotides 2-8 of the 5′-terminal region of miRNA (seed-matched site) leads to repression of translation of the target mRNA [28, 29]. RNA editing can change sequences of both miRNA and 3′-UTR of mRNA, which influences the possibility of the miRNA–mRNA pair formation. The edited miRNA miR-376 interacts with mRNA of pyrophosphate synthase repressing translation of this enzyme and as a result influences the amount of synthesized uric acid [30].

It was predicted more than 3000 known sites of A-to-I editing in 3′-UTR of mRNA complementary to position 2-8 of 5′-terminal regions of miRNA [31], but their functional significance still needs experimental verification.

ANIMALS WITH *ADAR* GENE KNOCKOUT

During recent years many reports have been published describing phenotypes of animals with deletions in genes encoding proteins of the ADAR family.

Deletion mutants of *Caenorhabditis elegans* in the *ADAR1* and *ADAR2* genes and also the double mutant *ADAR1/ADAR2* are viable but unable to display chemotaxis. This suggests a dysfunction of the nervous system of mutants [32]. Moreover, the lifespan of the double mutant *ADAR1/ADAR2* was about twofold shorter compared to the wild type [33]. Chemotaxis function and the lifespan were recovered by a deletion of another gene (*RDE1*) encoding a protein product, which is involved in RNA interference. *RDE1* deletion mutant is lacking RNA interference. The recovery of the lifespan and chemotaxis in the triple mutant *RDE1/ADAR1/ADAR2* as compared to the mutant *ADAR1/ADAR2* was explained by the authors as proof of intersection between the functional pathways of RNA deamination and RNA interference [33, 34].

Drosophila melanogaster mutants in the *dADAR* gene displayed some behavioral defects notwithstanding the development of morphologically normal adults [35]. The authors emphasize a strong degeneration of the retina and age-related progress in brain damage, which becomes noticeable by the 30th day of the life of $dADAR^{-/-}$ flies.

In contrast to mutant worms and flies, mammalian mutants in genes *ADAR1* or *ADAR2* usually died before birth. Mutant mice $ADAR1^{-/-}$ or mutant mice $ADAR1^{+/-}$ in which the mRNA expression for ADAR1 was decreased up to 50-60%, displayed lethal phenotype during embryogenesis. The embryos died on the 12th or 14th day of the embryogenesis, respectively. The mutant embryos $ADAR1^{+/-}$ died due to abnormal proliferation and differentiation of blood cells during hemopoiesis [36]. In the mutant $ADAR1^{-/-}$ the authors observed a pronounced apoptosis in virtually all tissues [37]. As discriminated from the mutant $ADAR1^{+/-}$, the phenotype of adult heterozygous mice $ADAR2^{+/-}$ did not vary from that of the wild type, whereas homozygous $ADAR2^{-/-}$ mice died on the 15-20th day after birth. It is interesting that the lethal phenotype $ADAR2^{-/-}$ of mice disappeared on replacement of both *GluR-B* alleles of the wild type (encoding α-amino-3-hydroxy-5-methyl-4-isoxazol propionate (AMPA) receptor) by *GluR-B* with the edited replacement of the arginine codon [38]. Thus, it is concluded that the main function of ADAR2 is the editing of pre-mRNA of GluR-B. The absence of editing of other ADAR2 substrates is not so dramatic and probably is compensated by the activity of ADAR1. Homozygous $ADAR3^{-/-}$ mice did not display lethal phenotype; therefore, the function of ADAR3 was supposed to be compensated by other members of the ADAR family [4]. Mice with the genotype $TENR^{-/-}$ also survived, but the number of spermatozoa was lower than in wild type mice. Multiple morphological disorders were also found in the structure of their spermatozoa. Spermatozoa of the $TENR^{-/-}$ mice were unable to fertilize the oocyte [39].

RNA EDITING AND HUMAN DISEASES

Disorders in A-to-I RNA editing can cause some human diseases [5]. Independent genetic studies on patients with dyschromatosis symmetrica hereditaria (DSH) revealed genetic loci associated with this disease. Now >60 mutations in the gene *ADAR1* are known to be associated with DSH [40, 41]. Disorders associated with DSH begin early in childhood as pigment spots on the backs of hands and feet. This disease is supposed to be caused by disorders in RNA editing resulting in differentiation of melanoblasts into hyper- or hypoactive melanocytes, which are unequally distributed and produce pigment inclusions [41]. But this hypothesis has not been confirmed experimentally.

In some patients with amyotrophic lateral sclerosis the level of Q/R-site editing in GluR-B mRNA was decreased in motor neurons of spinal marrow leading to the death of the cells [5].

In mice with editing deficiency of the Q/R-site in mRNA of GluR-B, a phenotype similar to human temporal epilepsy was observed. Nevertheless there are no data on the relation of the deficient editing of GluR-B mRNA with this human disease [5, 42]. An affected editing of mRNA of the serotonin receptor 5-HT_{2C}R resulting in imbalance of its isoforms can cause various forms of

schizophrenia and depression [43]. Thus, RNA editing is necessary for the most important functions of the highest nervous activity.

Changes in A-to-I editing were also found in various tumors [44]. Thus, editing was shown of pre-mRNA of a tumor-suppressing factor tyrosine phosphatase. The editing results in formation of new sites of splicing and, as a consequence, of alternative forms of mRNA. The authors suppose that the final protein product loses its functional activity thus promoting the development of neoplasms [45]. A decreased level of Q/R-site editing in GluR-B mRNA was found in patients with multiform glioblastoma or with astrocytoma, which are very aggressive tumors. In this case the activity of ADAR2 editing the Q/R-site leads to suppression of proliferation and migration of tumor cells and finally to arresting the tumor growth [46, 47].

ADAR1 p150 is a protein with expression induced by type I interferons [48]. These interferons are synthesized by the cell in response to viral infection and significantly change the expression profile of the cell's genes. Thus, expression of protein kinase PKR, which phosphorylates the translation factor eIF-2a, is induced leading to translational inhibition; expression of various RNases cleaving viral and cellular mRNAs is also promoted. It is rather difficult to experimentally show the functional role of ADAR1 p150 because of abundance of the factors induced [49]. It is not always obvious whether the functional properties of ADAR1 p150 can contribute to antiviral protection of the cell, or, by contrast, the virus uses the enzymatic activity of ADAR1 p150 for its own purpose. The combination of both ways is also possible. RNA editing of genomes of hepatitis delta virus (HDV), human herpes virus 8 (HHV8), and of human immunodeficiency virus (HIV) is very important for development of the viral infection. The large delta antigen is produced in HDV in the case of editing the Amber-stop-codon gene of the small delta antigen. This results in formation of UIG-triplet recognized by the ribosome as an UGG-codon encoding tryptophan [50, 51]. In the case of HHV8, ADAR1 p150 can edit the caposine transcript leading to virus multiplication by the lytic pathway [52]. A specific editing by ADAR1 p150 of the gene *ENV* site is shown to increase the production of HIV1 [53].

By contrast, some viruses use inhibitors of the deaminase activity of ADAR p150 because the hyper-editing the viral RNA leads to multiple mutations and appearance of unviable viral progeny. Such inhibitors include, in particular, adenoviral noncoding RNA VAI and the protein E3L of the vaccinia virus [54, 55].

It is now clear that adenosine deaminases are also actively involved in inflammatory processes. Acute inflammation is accompanied by a excessive deamination in many tissues of the organism of a wide spectrum of cellular RNAs, and the amount of inosine in the total RNA raises up to 5% [56]. The importance of ADAR1 in innate immunity and inflammatory response was confirmed by further studies. The necessity of ADAR1 for viability of hemopoietic stem cells was shown [57]. In the absence of this enzyme the expression of genes induced by type I interferons is greatly induced leading to rapid apoptosis. Thus, ADAR1 was shown to be necessary for defense of the organism against destructive effects associated with interferon pathway activation accompanied by some pathologic processes, such as chronic inflammation, autoimmune diseases, and tumorigenesis. Moreover, RNA editing is an important participant of processes related to damage of the spinal marrow and its subsequent recovery [58].

Studies on markers of exceptional longevity in humans revealed a correlation between lifespan and some genes. Products of these genes are mainly involved in lipid metabolism and are connected with proteins of the FOXO family and also with signaling pathways of insulin and IGF1 [59]. It is quite possible that the genetic predisposition for longevity is determined by a much larger number of genes. Recent studies in this field revealed a positive correlation between single nucleotide polymorphism in the *ADAR1* and *ADAR2* genes and the lifespan in humans [33]. This confirms a significant role of A-to-I RNA editing during aging.

ROLE OF DEAMINATION DURING ONTOGENESIS

For a long time it was thought that the complexity of multicellular organisms can be reached first of all due to increase in the number of genes. However, results of genome sequencing revealed just another picture: *C. elegans* has ~20,000 genes and *D. melanogaster* has ~14,000 genes. The genome of mammals also contains ~20,000 genes [60]. Nevertheless, it is evident that mammals are much more complicated living organisms than flies and worms. Thus, with the same number of genes the nervous system of nematodes consists of few hundred cells while human nervous system includes several billion cells. How can such an obvious discrepancy be explained? It can be explained by the increase of the genome capacity due to specific mechanisms utilizing alternative promoters, transcriptome editing, splicing, alternative start- and stop-codons, post-translational modifications, and proteolytic processing.

The more complicated is an organism the more complicated are these mechanisms. The simplest way to increase genome capacity is using alternative promoters and alternative initiatory codons leading to expression of alternative transcripts and protein products from a single gene. These mechanisms are widely distributed even in prokaryotes.

Splicing is now the best studied mechanism of increase of genome capacity in eukaryotes [60]. The extent of this phenomenon can be exemplified by *D.*

melanogaster: during splicing of the *Dscam* transcript up to 38,016 different isoforms of mRNA can be produced [61]. This is about three times greater than the number of all genes of the fruit fly! However, the splicing and other known mechanisms of genome capacity increase, such as post-translational modifications and processing, is not the subject of this review.

Genome diversity can also be increased by correcting RNA sequences on the post-translational level by editing. The RNA editing can lead to deletions and insertions of single or multiple nucleotides as well as nucleotide substitutions [62]. Examples of A-to-I RNA editing by ADAR proteins and some known functional consequences of these processes have been considered above. We believe these examples are only the tip of the iceberg of various processes of editing of genetic information on the transcriptome level. It seems that the RNA editing mechanism can partially answer the question how the human genome consisting of 20,000 genes can express hundreds of thousands of different proteins.

A-to-I editing concerns another basic process that is necessary for functioning of complex multicellular organisms: the miRNA-mediated regulation of gene expression. The post-transcriptional regulation of mRNA biogenesis through miRNA is responsible for the well-timed "setting on" or "setting off" translation of a certain mRNA or for triggering its degradation. The regulation of at least 30% of expressed human genes is thought to be mediated through miRNA [63]. This regulation is based on production of canonical Watson–Crick interactions between miRNA and target mRNA. A-to-I editing of both miRNA and the site of interaction with miRNA in 3′-UTR of the target mRNA can change the character of interactions of miRNA with mRNA and thus fully change the expression of an individual gene or a group of genes [64, 65]. At present the only example of changing mRNA target of miRNA due to RNA editing is known. The editing of miR-376 miRNA results in changing of target mRNAs that in turn results in a decrease in the expression of pyrophosphate synthase-1 and finally influences the production of uric acid [30]. The potential of gene expression regulation via editing regions responsible for miRNA interactions with mRNA is supposed to be very high. It may occur due to editing of both 3′-terminal mRNA sequences and miRNA [65].

The origin of multicellular organisms required the cells to be specialized for performing various functions. The more complex the organism, the more specialized cell types it contains raising need for the coordinated regulation of differentiation processes and normal functioning. Both miRNA and programmed cell death mechanisms were recently found to be involved in all basic processes of regulation of organogenesis and differentiation in higher organisms. Disorders in the regulation of miRNA lead to severe diseases [66]. In turn, the caspase activity specific for apoptosis is detected during differentiation of many types of mammalian cells and is necessary for the process [67, 68]. Expression of virtually all members of the apoptotic cascades is regulated through miRNA [69]. The inverse regulation also exists: endonuclease Dicer, an inevitable member of miRNA biogenesis, is a substrate for caspases, and moreover, upon proteolysis becomes a deoxyribonuclease that continues apoptotic degradation [70]. Furthermore, at least two participants of A-to-I editing-connected cascades, ADAR1 p150 and endonuclease Tudor-sn, are cleaved by caspases *in vivo* [71, 72]. We believe that A-to-I editing is involved in control of gene expression for apoptotic cascade members that contribute to the cell differentiation.

We have mentioned above that A-to-I editing is necessary during ontogenesis of higher organisms for development of normal animals. Mutants with disturbed deamination of RNA can develop with disorders in the nervous system in worms and fruit flies, whereas similar mutants in mice die before birth [4]. Note that in invertebrates these disorders were found in the nervous system, i.e. the most differentiated cells, while mice died at the stage of active cell differentiation. The hypothesis about the increasing role of A-to-I editing along with development the organisms' complexity is also supported by an extremely high level of A-to-I editing in the human brain as compared to other primates [73, 74].

During ontogenesis of mammals many tissues need constant renewal. The involvement of programmed cell death apparatus in cell differentiation requires a very fine regulation of this process. The smallest disorders can switch differentiation into apoptosis. A-to-I editing may be a mechanism of such a fine tuning.

Disorders in A-to-I editing cause some human diseases [5, 44]. Possibly, correct RNA editing is necessary not only for differentiation and functioning of specialized cells but also for the whole ontogenesis process, including the organism's aging as the final stage of ontogenesis [75]. This hypothesis is supported by the correlation between the human lifespan and the polymorphism of the *ADAR* genes [33].

The authors are grateful to V. P. Skulachev for his attention and useful advice and remarks.

This work was supported by the Mitotech company.

REFERENCES

1. Licatalosi, D. D., and Darnell, R. B. (2010) *Nat. Rev. Genet.*, **11**, 75-87.
2. Gott, J. M., and Emeson, R. B. (2000) *Annu. Rev. Genet.*, **34**, 499-531.
3. Bass, B. L. (2002) *Annu. Rev. Biochem.*, **71**, 817-846.
4. Nishikura, K. (2010) *Annu. Rev. Biochem.*, **79**, 321-349.
5. Maas, S., Kawahara, Y., Tamburr, K. M., and Nishikura, K. (2006) *RNA Biol.*, **3**, 1-9.

6. Montano, M., and Long, K. (2010) *Ageing Res. Rev.*, doi: 10.1016/j.arr.2010.02.002.
7. Jin, Y., Zhang, W., and Li, Q. (2009) *IUBMB Life*, **61**, 572-578.
8. Cho, D. S., Yang, W., Lee, J. T., Shiekhattar, R., Murray, J. M., and Nishikura, K. (2003) *J. Biol. Chem.*, **278**, 17093-17102.
9. Macbeth, M. R., Schubert, H. L., Vandemark, A. P., Lingam, A. T., Hill, C. P., and Bass, B. L. (2005) *Science*, **309**, 1534-1539.
10. Saunders, L. R., and Barber, G. N. (2003) *FASEB J.*, **17**, 961-983.
11. Maas, S., and Gommans, W. M. (2009) *Nucleic Acids Res.*, **37**, 5822-5829.
12. Schumacher, J. M., Lee, K., Edelhoff, S., and Braun, R. E. (1995) *Biol. Reprod.*, **52**, 1274-1283.
13. Athanasiadis, A., Rich, A., and Maas, S. (2004) *PLoS Biol.*, **2**, e391.
14. Rueter, S. M., Dawson, T. R., and Emeson, R. B. (1999) *Nature*, **399**, 75-80.
15. Zhang, Z., and Carmichael, G. G. (2001) *Cell*, **106**, 465-475.
16. Prasanth, K. V., Prasanth, S. G., Xuan, Z., Hearn, S., Freier, S. M., Bennett, C. F., Zhang, M. Q., and Spector, D. L. (2005) *Cell*, **123**, 249-263.
17. Scadden, A. D., and Smith, C. W. (2001) *EMBO J.*, **20**, 4243-4252.
18. Pankratova, E. V., and Stepchenko, A. G. (2010) *Genetika*, **46**, 5-13.
19. Higuchi, M., Single, F. N., Kohler, M., Sommer, B., Sprengel, R., and Seeburg, P. H. (1993) *Cell*, **75**, 1361-1370.
20. Seeburg, P. H., and Hartner, J. (2003) *Curr. Opin. Neurobiol.*, **13**, 279-283.
21. Burns, C. M., Chu, H., Rueter, S. M., Hutchinson, L. K., Canton, H., Sanders-Bush, E., and Emeson, R. B. (1997) *Nature*, **387**, 303-308.
22. Wang, Q., O'Brien, P. J., Chen, C. X., Cho, D. S., Murray, J. M., and Nishikura, K. (2000) *J. Neurochem.*, **74**, 1290-1300.
23. Kawahara, Y., Megraw, M., Kreider, E., Iizasa, H., Valente, L., Hatzigeorgiou, A. G., and Nishikura, K. (2008) *Nucleic Acids Res.*, **36**, 5270-5280.
24. Yang, W., Chendrimada, T. P., Wang, Q., Higuchi, M., Seeburg, P. H., Shiekhattar, R., and Nishikura, K. (2006) *Nat. Struct. Mol. Biol.*, **13**, 13-21.
25. Scadden, A. D. (2005) *Nat. Struct. Mol. Biol.*, **12**, 489-496.
26. Heale, B. S., Keegan, L. P., McGurk, L., Michlewski, G., Brindle, J., Stanton, C. M., Caceres, J. F., and O'Connell, M. A. (2009) *EMBO J.*, **28**, 3145-3156.
27. Kawahara, Y., Zinshteyn, B., Chendrimada, T. P., Shiekhattar, R., and Nishikura, K. (2007) *EMBO Rep.*, **8**, 763-769.
28. Pillai, R. S., Bhattacharyya, S. N., and Filipowicz, W. (2007) *Trends Cell Biol.*, **17**, 118-126.
29. Baek, D., Villen, J., Shin, C., Camargo, F. D., Gygi, S. P., and Bartel, D. P. (2008) *Nature*, **455**, 64-71.
30. Kawahara, Y., Zinshteyn, B., Sethupathy, P., Iizasa, H., Hatzigeorgiou, A. G., and Nishikura, K. (2007) *Science*, **315**, 1137-1140.
31. Borchert, G. M., Gilmore, B. L., Spengler, R. M., Xing, Y., Lanier, W., Bhattacharya, D., and Davidson, B. L. (2009) *Hum. Mol. Genet.*, **18**, 4801-4807.
32. Tonkin, L. A., Saccomanno, L., Morse, D. P., Brodigan, T., Krause, M., and Bass, B. L. (2002) *EMBO J.*, **21**, 6025-6035.
33. Sebastiani, P., Montano, M., Puca, A., Solovieff, N., Kojima, T., Wang, M. C., Melista, E., Meltzer, M., Fischer, S. E., Andersen, S., Hartley, S. H., Sedgewick, A., Arai, Y., Bergman, A., Barzilai, N., Terry, D. F., Riva, A., Anselmi, C. V., Malovini, A., Kitamoto, A., Sawabe, M., Arai, T., Gondo, Y., Steinberg, M. H., Hirose, N., Atzmon, G., Ruvkun, G., Baldwin, C. T., and Perls, T. T. (2009) *PLoS One*, **4**, e8210.
34. Tonkin, L. A., and Bass, B. L. (2003) *Science*, **302**, 1725.
35. Palladino, M. J., Keegan, L. P., O'Connell, M. A., and Reenan, R. A. (2000) *Cell*, **102**, 437-449.
36. Wang, Q., Khillan, J., Gadue, P., and Nishikura, K. (2000) *Science*, **290**, 1765-1768.
37. Wang, Q., Miyakoda, M., Yang, W., Khillan, J., Stachura, D. L., Weiss, M. J., and Nishikura, K. (2004) *J. Biol. Chem.*, **279**, 4952-4961.
38. Higuchi, M., Maas, S., Single, F. N., Hartner, J., Rozov, A., Burnashev, N., Feldmeyer, D., Sprengel, R., and Seeburg, P. H. (2000) *Nature*, **406**, 78-81.
39. Connolly, C. M., Dearth, A. T., and Braun, R. E. (2005) *Dev. Biol.*, **278**, 13-21.
40. Miyamura, Y., Suzuki, T., Kono, M., Inagaki, K., Ito, S., Suzuki, N., and Tomita, Y. (2003) *Am. J. Hum. Genet.*, **73**, 693-699.
41. Li, M., Yang, L., Li, C., Jin, C., Lai, M., Zhang, G., Hu, Y., Ji, J., and Yao, Z. (2010) *Arch. Dermatol. Res.*, **302**, 469-476.
42. Krestel, H. E., Shimshek, D. R., Jensen, V., Nevian, T., Kim, J., Geng, Y., Bast, T., Depaulis, A., Schonig, K., Schwenk, F., Bujard, H., Hvalby, O., Sprengel, R., and Seeburg, P. H. (2004) *J. Neurosci.*, **24**, 10568-10578.
43. Iwamoto, K., Bundo, M., and Kato, T. (2009) *RNA Biol.*, **6**, 248-253.
44. Gallo, A., and Galardi, S. (2008) *RNA Biol.*, **5**, 135-139.
45. Beghini, A., Ripamonti, C. B., Peterlongo, P., Roversi, G., Cairoli, R., Morra, E., and Larizza, L. (2000) *Hum. Mol. Genet.*, **9**, 2297-2304.
46. Maas, S., Patt, S., Schrey, M., and Rich, A. (2001) *Proc. Natl. Acad. Sci. USA*, **98**, 14687-14692.
47. Cenci, C., Barzotti, R., Galeano, F., Corbelli, S., Rota, R., Massimi, L., Di Rocco, C., O'Connell, M. A., and Gallo, A. (2008) *J. Biol. Chem.*, **283**, 7251-7260.
48. George, C. X., and Samuel, C. E. (1999) *Proc. Natl. Acad. Sci. USA*, **96**, 4621-4626.
49. George, C. X., Li, Z., Okonski, K. M., Toth, A. M., Wang, Y., and Samuel, C. E. (2009) *J. Interferon Cytokine Res.*, **29**, 477-487.
50. Polson, A. G., Bass, B. L., and Casey, J. L. (1996) *Nature*, **380**, 454-456.
51. Casey, J. L. (2006) *Curr. Top. Microbiol. Immunol.*, **307**, 67-89.
52. Gandy, S. Z., Linnstaedt, S. D., Muralidhar, S., Cashman, K. A., Rosenthal, L. J., and Casey, J. L. (2007) *J. Virol.*, **81**, 13544-13551.
53. Phuphuakrat, A., Kraiwong, R., Boonarkart, C., Lauhakirti, D., Lee, T. H., and Auewarakul, P. (2008) *J. Virol.*, **82**, 10864-10872.
54. Lei, M., Liu, Y., and Samuel, C. E. (1998) *Virology*, **245**, 188-196.

55. Liu, Y., Wolff, K. C., Jacobs, B. L., and Samuel, C. E. (2001) *Virology*, **289**, 378-387.
56. Yang, J. H., Luo, X., Nie, Y., Su, Y., Zhao, Q., Kabir, K., Zhang, D., and Rabinovici, R. (2003) *Immunology*, **109**, 15-23.
57. Hartner, J. C., Walkley, C. R., Lu, J., and Orkin, S. H. (2009) *Nat. Immunol.*, **10**, 109-115.
58. Barbon, A., Fumagalli, F., Caracciolo, L., Madaschi, L., Lesma, E., Mora, C., Carelli, S., Slotkin, T. A., Racagni, G., Di Giulio, A. M., Gorio, A., and Barlati, S. (2010) *J. Neurochem.*, **114**, 397-407.
59. Fontana, L., Partridge, L., and Longo, V. D. (2010) *Science*, **328**, 321-326.
60. Nilsen, T. W., and Graveley, B. R. (2010) *Nature*, **463**, 457-463.
61. Schmucker, D., Clemens, J. C., Shu, H., Worby, C. A., Xiao, J., Muda, M., Dixon, J. E., and Zipursky, S. L. (2000) *Cell*, **101**, 671-684.
62. Gott, J. M. (2003) *Comptes Rendus Biol.*, **326**, 901-908.
63. Lewis, B. P., Burge, C. B., and Bartel, D. P. (2005) *Cell*, **120**, 15-20.
64. Winter, J., Jung, S., Keller, S., Gregory, R. I., and Diederichs, S. (2009) *Nat. Cell Biol.*, **11**, 228-234.
65. Hundley, H. A., and Bass, B. L. (2010) *Trends Biochem. Sci.*, **35**, 377-383.
66. Alvarez-Garcia, I., and Miska, E. A. (2005) *Development*, **132**, 4653-4662.
67. Fernando, P., and Megeney, L. A. (2007) *FASEB J.*, **21**, 8-17.
68. Yi, C. H., and Yuan, J. (2009) *Dev. Cell.*, **16**, 21-34.
69. Subramanian, S., and Steer, C. J. (2010) *J. Cell Physiol.*, **223**, 289-298.
70. Nakagawa, A., Shi, Y., Kage-Nakadai, E., Mitani, S., and Xue, D. (2010) *Science*, **328**, 327-334.
71. Toth, A. M., Li, Z., Cattaneo, R., and Samuel, C. E. (2009) *J. Biol. Chem.*, **284**, 29350-29356.
72. Sundstrom, J. F., Vaculova, A., Smertenko, A. P., Savenkov, E. I., Golovko, A., Minina, E., Tiwari, B. S., Rodriguez-Nieto, S., Zamyatnin, A. A., Jr., Valineva, T., Saarikettu, J., Frilander, M. J., Suarez, M. F., Zavialov, A., Stahl, U., Hussey, P. J., Silvennoinen, O., Sundberg, E., Zhivotovsky, B., and Bozhkov, P. V. (2009) *Nat. Cell Biol.*, **11**, 1347-1354.
73. Gommans, W. M., Mullen, S. P., and Maas, S. (2009) *BioEssays*, **31**, 1137-1145.
74. Paz-Yaacov, N., Levanon, E. Y., Nevo, E., Kinar, Y., Harmelin, A., Jacob-Hirsch, J., Amariglio, N., Eisenberg, E., and Rechavi, G. (2010) *Proc. Natl. Acad. Sci. USA*, **107**, 12174-12179.
75. Skulachev, V. P., and Longo, V. D. (2005) *Ann. N. Y. Acad. Sci.*, **1057**, 145-164.