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Production and actions of small RNAs

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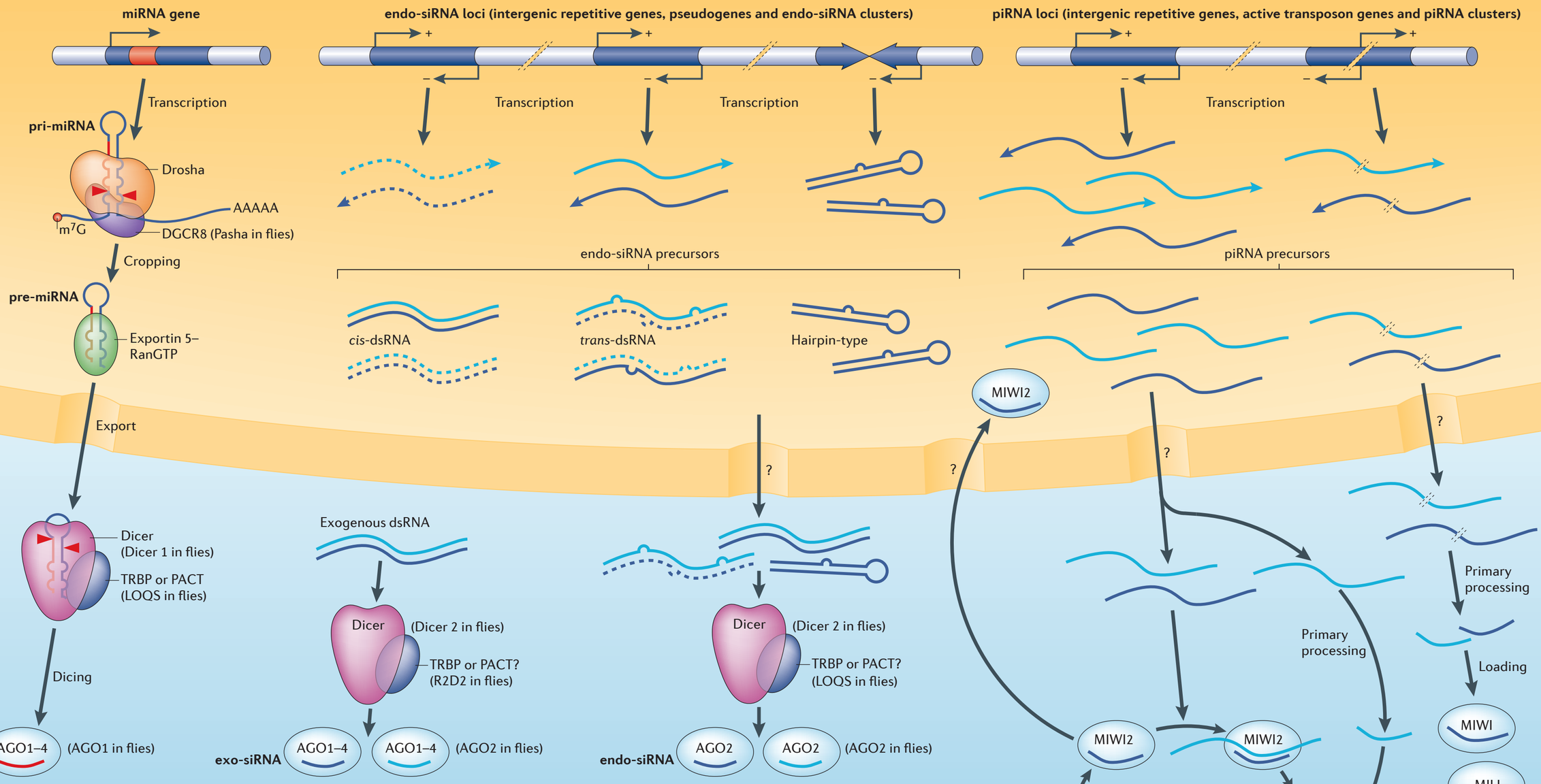
Small (20–30 nt) RNAs are associated with members of the Argonaute (Ago) family, which comprises two subfamilies: Ago and Piwi. Based on their biogenesis mechanism and the type of Argonaute proteins that they associate with, at least three classes of small RNAs can be distinguished in eukaryotes: microRNAs (miRNAs), endogenous small interfering RNAs (endo-siRNAs) and Piwi-interacting RNAs (piRNAs). miRNAs control mRNA stability and translation by targeting cognate mRNAs. endo-siRNAs suppress repetitive genes by cleaving their transcripts. Some piRNAs mediate RNA cleavage or heterochromatin formation of transposons, although the functions of most piRNAs are still unknown.



The figure shows the mammalian processing pathways with fly components in brackets. miRNAs are ssRNAs of ~22 nt that are generated from transcripts that can fold into hairpin structures. Two RNase III type proteins, Drosha and Dicer, mediate nuclear and cytoplasmic processing, respectively, thereby producing the miRNAs that subsequently associate with Ago-subfamily proteins (AGO1–4 in humans). In flies, most miRNAs are loaded onto AGO1, whereas miRNAs from highly base-paired precursors are sorted into AGO2. exo-siRNAs are derived from experimentally introduced dsRNAs or viral RNAs. Human exo-siRNAs are loaded onto AGO1–4, but only AGO2 has 'slicer' activity. endo-siRNAs are derived from *cis* or *trans* sense–antisense dsRNAs or long hairpins. They are produced by Dicer and interact with Ago-subfamily proteins. In humans, endo-siRNAs are thought to be loaded onto AGO2; it has not been determined whether they associate with other Ago proteins. piRNAs (24–31 nt) are processed from ssRNA precursors that are transcribed from transposons or large piRNA clusters. piRNAs associate with Piwi-subfamily proteins; they are not dependent on either Drosha or Dicer, although the requirement of Drosha has not been formally tested. They may instead use the nuclease activity of the Piwi proteins themselves for their processing. piRNA biogenesis involves primary and secondary processing mechanisms. The strand bias of piRNAs in secondary processing differs between mice and flies (for this reason, fly components are not shown). Primary processing and loading might occur in the cytoplasm because Piwi proteins MIWI and MILI are localized in the cytoplasm. Factors that are needed for primary processing are unknown. Not shown here are less abundant groups of miRNA-like RNAs that are derived from various sources of short hairpins in a Drosha-independent manner.

Possible mechanisms of action

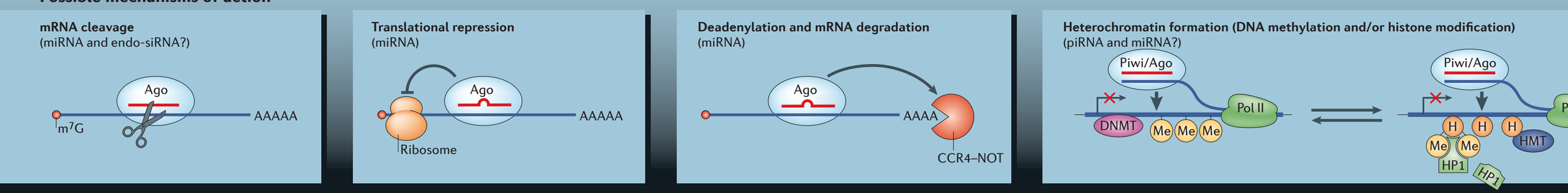
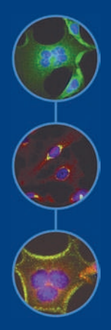


Table | **Eukaryotic small RNAs are associated with Argonaute-family proteins**

Subfamily	Ago-family protein	Class of small RNA*	Length of small RNA	Origin of small RNA‡	Mechanism of action
Mammals					
Ago	AGO1–4	miRNA	21–23 nt	miRNA genes	Translational repression, mRNA degradation, mRNA cleavage and heterochromatin formation?
		endo-siRNA§	21–22 nt	Intergenic repetitive genes, pseudogenes and endo-siRNA clusters	mRNA cleavage?
Piwi	MILI (PIWIL2 in humans)	Pre-pachytene piRNA and pachytene piRNA	24–28 nt	Transposons and piRNA clusters	Heterochromatin formation (DNA methylation)
	MIWI (PIWIL1 in humans)	Pachytene piRNA	29–31 nt	piRNA clusters	?
	MIWI2 (PIWIL4 in humans)	Pre-pachytene piRNA	27–29 nt	Transposons and piRNA clusters	Heterochromatin formation (DNA methylation)
	(PIWIL3 in humans)	?	?	?	?
Drosophila melanogaster					
Ago	AGO1	miRNA	21–23 nt	miRNA genes	Translational repression and mRNA degradation
	AGO2	endo-siRNA	~21 nt	Transposons, mRNAs and repeats	RNA cleavage
		exo-siRNA	~21 nt	Viral genome	Viral RNA cleavage
Piwi	AUB	piRNA	23–27 nt	Transposons, repeats, piRNA clusters and *Su(Ste)* locus	RNA cleavage
	AGO3	piRNA	24–27 nt	Transposons and repeats (unknown in testis)	RNA cleavage
	PIWI	piRNA	24–29 nt	Transposons, repeats and piRNA clusters	Heterochromatin formation?
Schizosaccharomyces pombe					
Ago	Ago1	endo-siRNA	~21 nt	Outer centromeric repeats, mating-type locus and subtelomeric regions	Heterochromatin formation
***Arabidopsis thaliana*‖**					
Ago	AGO1	miRNA	20–24 nt	miRNA genes	mRNA cleavage and translational repression
		endo-siRNA (tasiRNA including TAS3)	21 nt	TAS genes	mRNA cleavage
		exo-siRNA	20–22 nt	Viral genome	Viral RNA cleavage
	AGO4 and AGO6	rasiRNA	24 nt	Transposons and repetitive elements	Heterochromatin formation
	AGO7	miR-390	21 nt	miRNA gene	Cleavage of TAS3 RNA

*Small RNAs that are the main partners of a given Ago protein are listed. ‡miRNAs, as a class, are expressed in all cell types, whereas endo-siRNAs and piRNAs are expressed abundantly in germline cells and contribute to germline development. §So far, only AGO2 has been shown to be required for endo-siRNAs. ‖Plants have ten Ago proteins, but only those with known small RNA partners are shown.



Abbreviations

Ago, Argonaute; AUB, Aubergine; CCR4, C-C chemokine type 4; DGCR8, DiGeorge syndrome critical region gene 8; DNMT, DNA methyltransferase; dsRNA, double-stranded RNA; endo-siRNA; endogenous small interfering RNA; exo-siRNA, exogenous small interfering RNA; H, histone; HMT, histone methyltransferase; HP1, heterochromatin protein 1; LOQS, Loquacious; m7G, 7-methylguanosine; Me, methyl; miRNA, microRNA; nt, nucleotide; piRNA, Piwi-interacting RNA; Pol II, RNA polymerase II; PACT, PKR-activating protein; pre-miRNA, precursor miRNA; pri-miRNA, primary miRNA; rasiRNA, repeat-associated small interfering RNA; ssRNA, single-stranded RNA; *Su(Ste)*, *Suppressor of Stellate*; TAS, tasi gene; tasiRNA, *trans*-acting siRNA; TRBP, HIV-1 TAR RNA-binding protein.

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For further reading, see www.nature.com/nrm/posters/smallrnas

Linked review article

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