OS RNAs E O CONTROLE DA EXPRESSÃO GÊNICA

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Departamento de Genética
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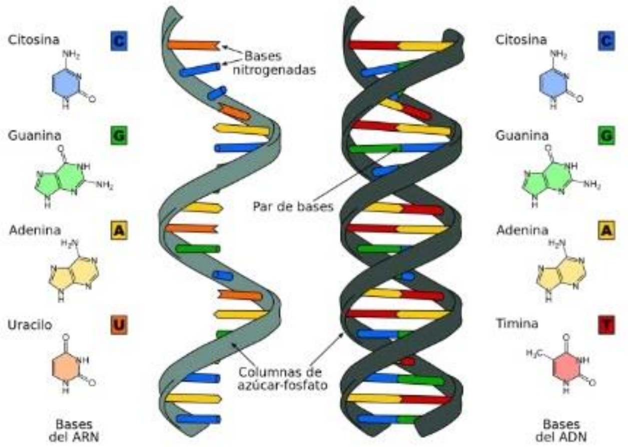
SUMÁRIO

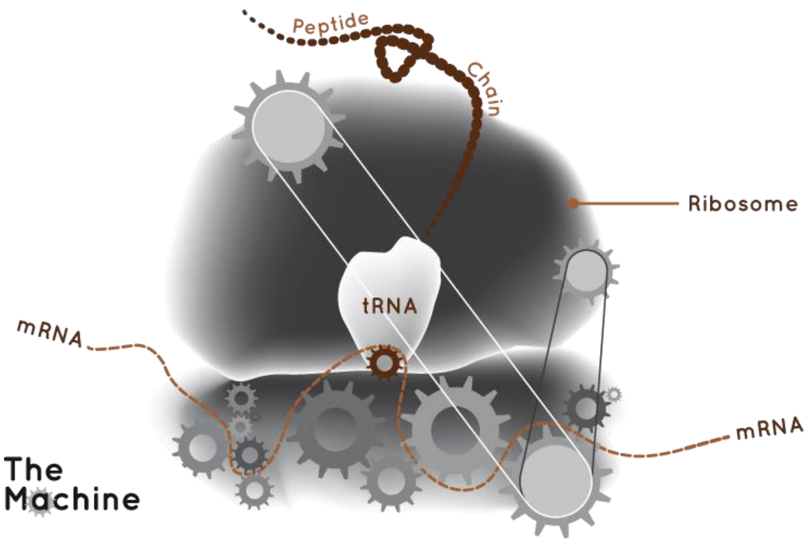
- Histórico do RNA
- RNA interferencia – conceito e histórico
- siRNA
- microRNA
- RNAi em plantas
- Próxima aula

1890 e 1950 – ENTENDO A ESTRTURURA DO RNA

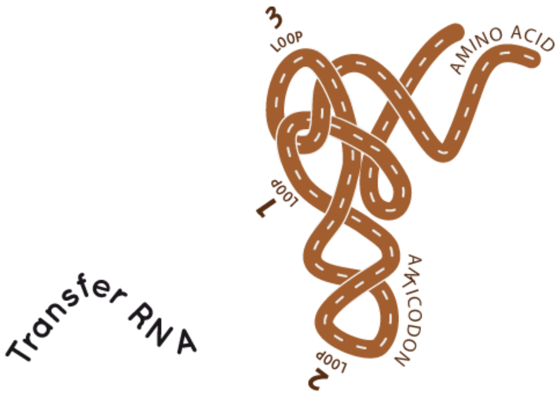


1951 e 1965 – RNA FAZ PROTEÍNA

Severo Ochoa
Nobel 1959

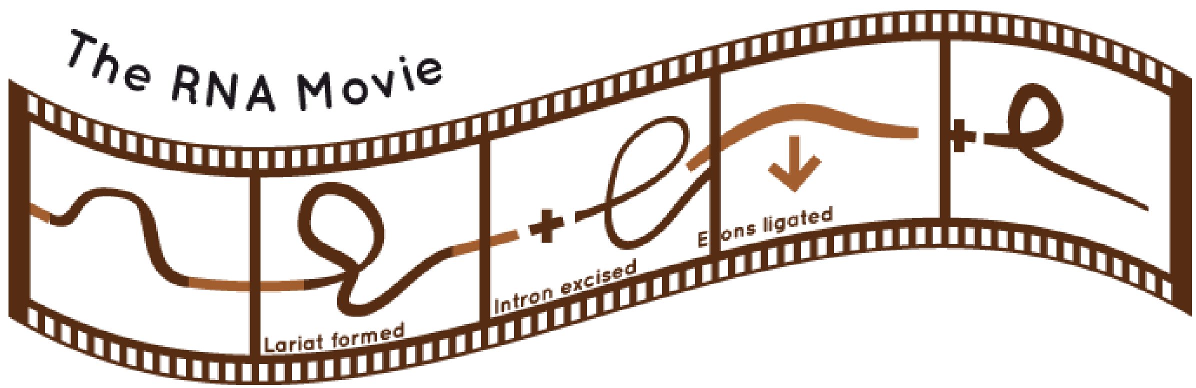


1966 e 1975 – “RNA Seq”

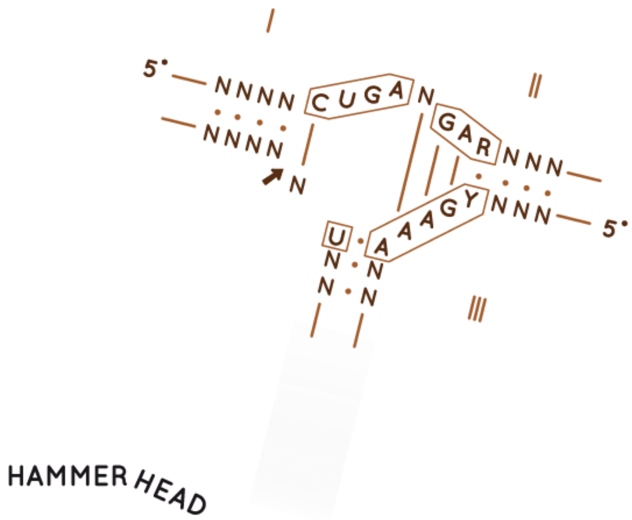


Robert W. Holley
Nobel 1968

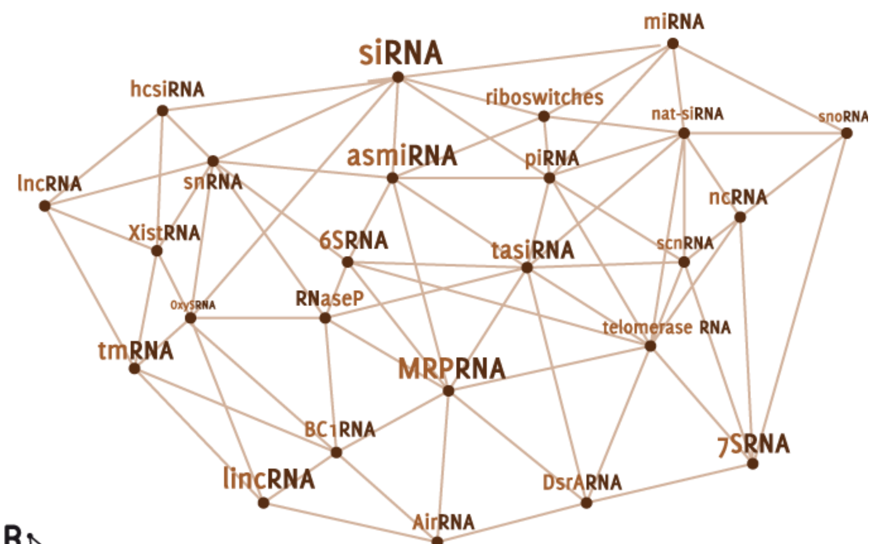
1976 e 1985 – RNA – “Top Hit” da biologia



1986 a 2000 – RNA e atividade catalítica



2000 ao presente – Controle da expressão gênica



The RNA Syndicate

NON-CODING RNA

Unique features of long non-coding RNA biogenesis and function

Jeffrey J. Quinn[1,2] and Howard Y. Chang[1]

The biogenesis and emerging roles of circular RNAs

Ling-Ling Chen

Nonsense-mediated mRNA decay: an intricate machinery that shapes transcriptomes

Søren Lykke-Andersen and Torben Heick Jensen

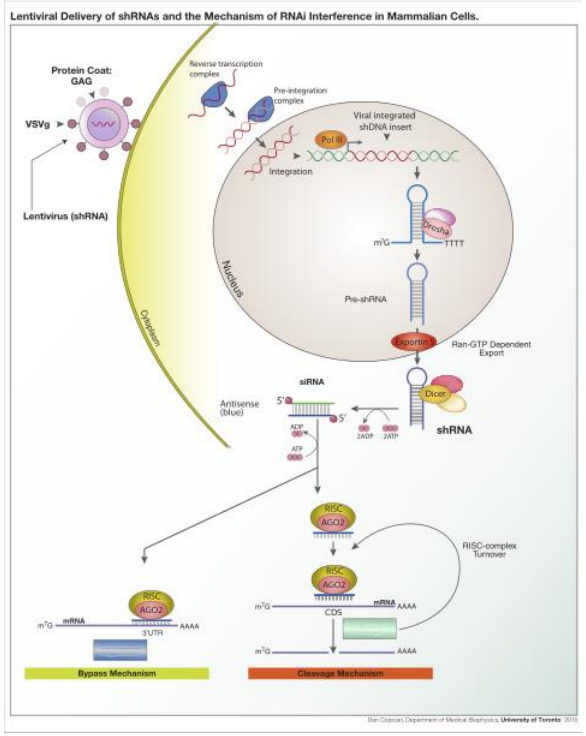
RNAi – fenômeno biológico!

Controle de expressão mediado por RNA!!!

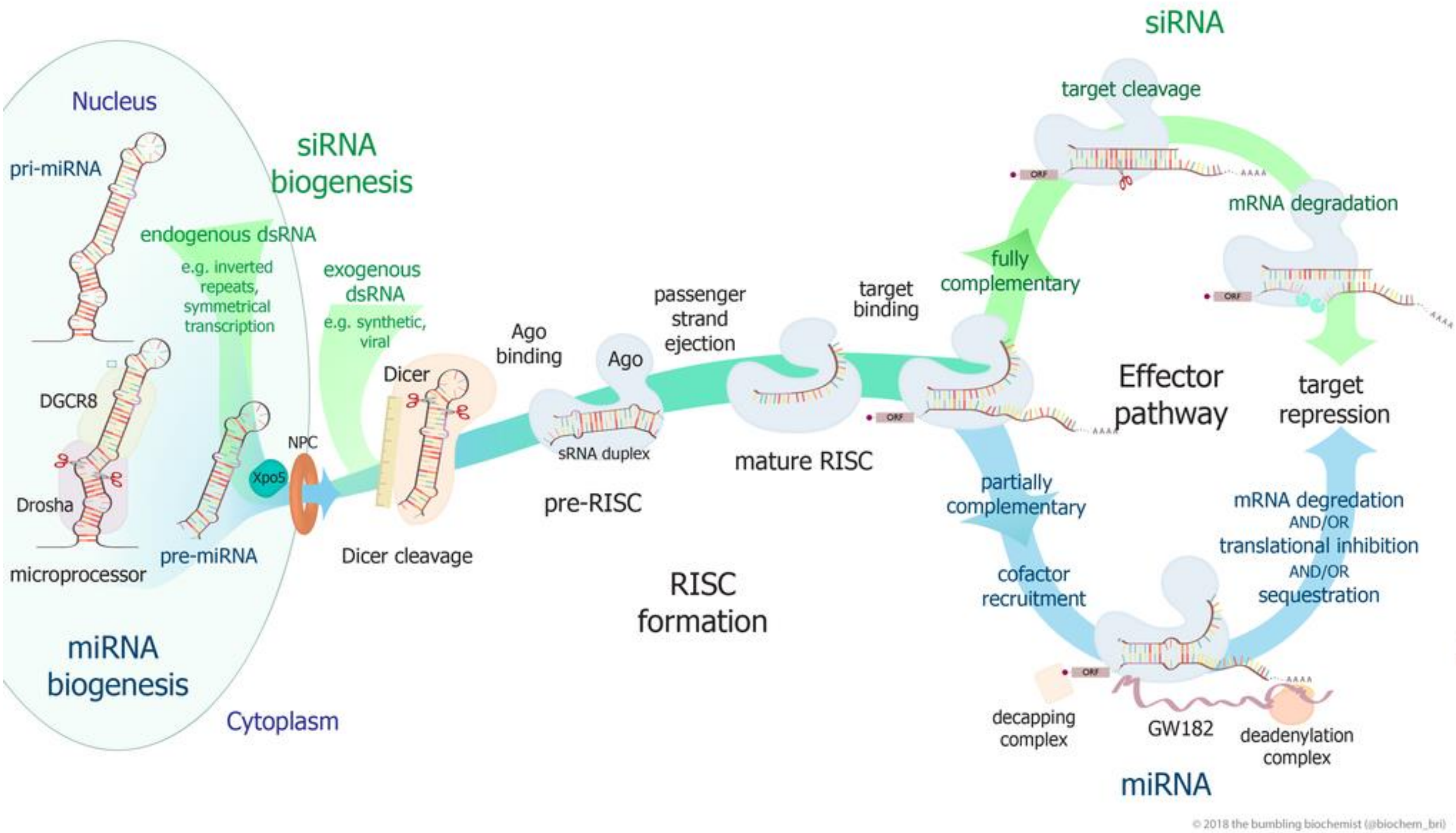
- microRNAs (miRNAs) - imperfeita complementariedade com RNA alvos causando repressão pós-transcricional e traducional;
- small interfering RNAs (siRNAs) –perfeita complementariedade com o alvo e causa degradação de transcritos

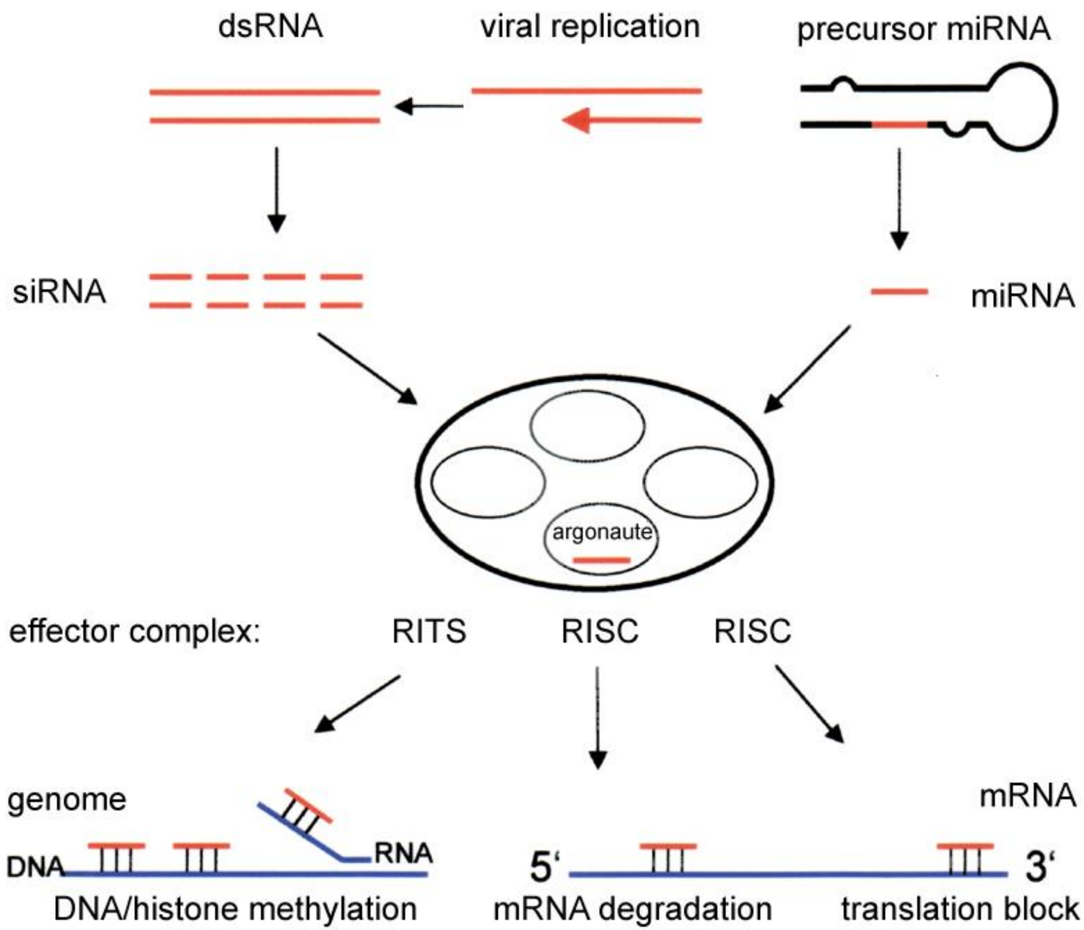
Local de ação:
Citoplasma
Núcleo

Função:
Proteção contra vírus
Silenciamento de transposons
Desenvolvimento



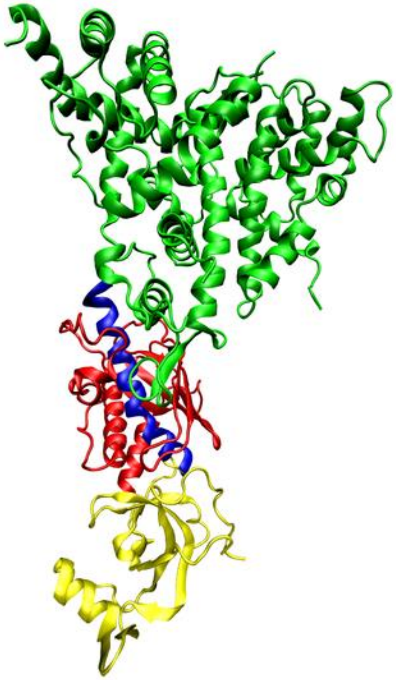
O FENÔMENO RNAi





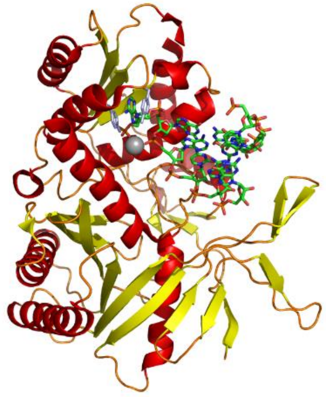
A enzima Dicer reconhece dsRNA para formar siRNA ou miRNA. Esses RNAs são incorporados no RNA-induced silencing complex (RISC), que se liga ao mRNA.

ALGUMAS PROTEÍNAS CHAVES…



Dicer

Argonatas



RNA-dependent RNA polymerase (RdRP)

Swiss army knives: non-canonical functions of nuclear Drosha and Dicer

Kaspar Burger and Monika Gullerova

Argonaute proteins: key players in RNA silencing

Gyorgy Hutvagner and Martin J. Simard‡*

Abstract | During the past decade, small non-coding RNAs have rapidly emerged as important contributors to gene regulation. To carry out their biological functions, these small RNAs require a unique class of proteins called Argonautes. The discovery and our comprehension of this highly conserved protein family is closely linked to the study of RNA-based gene silencing mechanisms. With their functional domains, Argonaute proteins can bind small non-coding RNAs and control protein synthesis, affect messenger RNA stability and even participate in the production of a new class of small RNAs, Piwi-interacting RNAs.

Table 1 | **Functions of Argonaute proteins in different species**

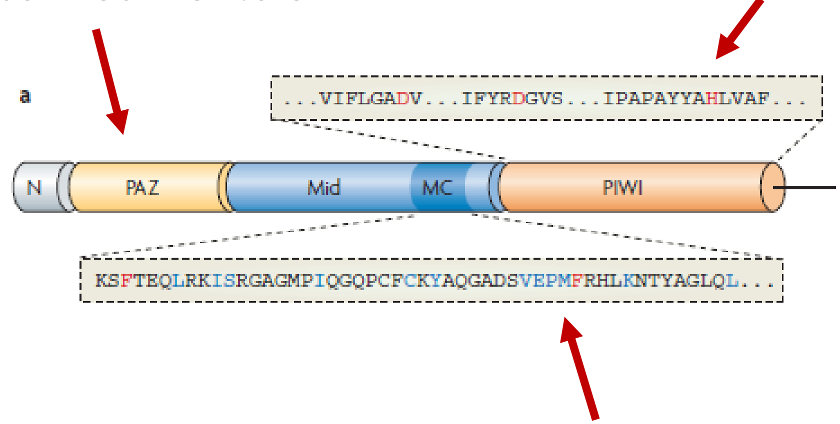
Argonaute protein	Molecular function	References
Neurospora crassa		
QDE2	Quelling*	120, 121
SMS-2	Meiotic silencing of unpaired DNA	122
Schizosaccharomyces pombe		
Ago1	Heterochromatin silencing, TGS, PTGS	11, 12, 103, 123
Tetrahymena		
Twi1	DNA elimination	124
Arabidopsis thaliana		
AGO1	miRNA-mediated gene silencing, tasiRNA	125, 126
AGO4	rasiRNA, heterochromatin silencing	127
AGO6	rasiRNA, heterochromatin silencing	128
AGO7	tasiRNA, heteroblasty, leaf development	129
Caenorhabditis elegans		
RDE-1	Exogenous RNAi	7, 59
ALG-1	miRNA-mediated gene silencing, TGS	60, 130
ALG-2	miRNA-mediated gene silencing	60
ERGO-1	Endogenous RNAi	7
CSR-1	Chromosome segregation and RNAi	7
SAGO-1	Endogenous and exogenous RNAi	7
SAGO-2	Endogenous and exogenous RNAi	7
PPW-1	Endogenous and exogenous RNAi	7, 131
PPW-2	Endogenous and exogenous RNAi	7
F58G1.1	Endogenous and exogenous RNAi	7
C16C10.3	Endogenous and exogenous RNAi	7
PRG-1	Germline maintenance	90
Drosophila melanogaster		
AGO1	miRNA-mediated gene silencing	58
AGO2	RNAi	58, 132
AGO3	piRNA, transposon silencing	38, 39
PIWI	piRNA, transposon silencing, germline stem-cell maintenance, RNAi	38, 39, 98
Aubergine	piRNA, transposon silencing, stellate silencing, RNAi	38, 39, 97
Zebrafish		
Ziwi	piRNA, germ-cell maintenance, transposon silencing	99
Murine/Human		
AGO1	Heterochromatin silencing	133, 134
AGO2	RNAi, miRNA-mediated gene silencing, heterochromatin silencing	40, 41, 133
MIWI (mouse)	piRNA, spermatogenesis	88, 92, 95
MILI (mouse)	piRNA, spermatogenesis	87, 96
RIWI (rat)	piRNA	94

The table contains Argonaute-like and Piwi-like proteins that have been associated with either a small RNA or cellular functions. Mammals encode two further Argonaute-like and Piwi-like proteins, *A. thaliana* has six further Argonaute-like proteins and *C. elegans* has 15 further Argonaute proteins with no described function(s). *Quelling is a term that describes post-transcriptional gene silencing in *Neurospora crassa*. miRNA, microRNA; piRNA, Piwi-interacting RNA; PTGS, post-transcriptional gene silencing; rasiRNA, repeat-associated small interfering RNA; RNAi, RNA interference; tasiRNA, trans-acting small interfering RNA; TGS, transcriptional gene silencing.

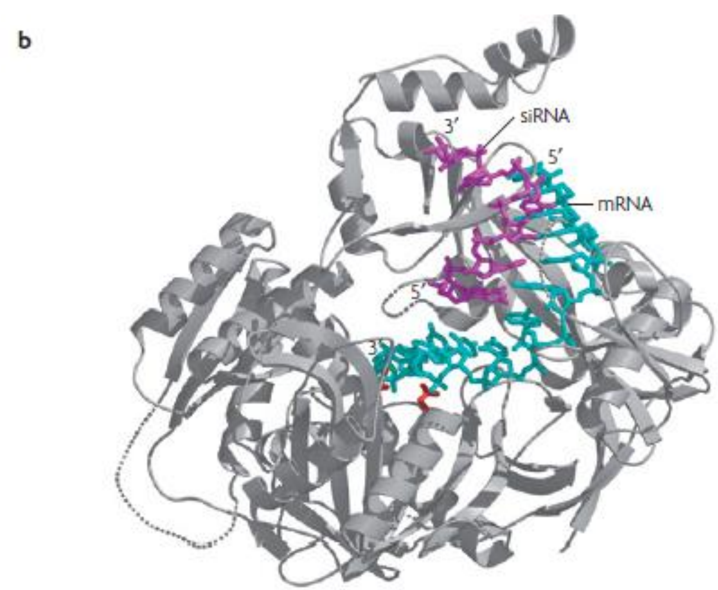
doi:10.1038/nrm2321

Sítio de clivagem

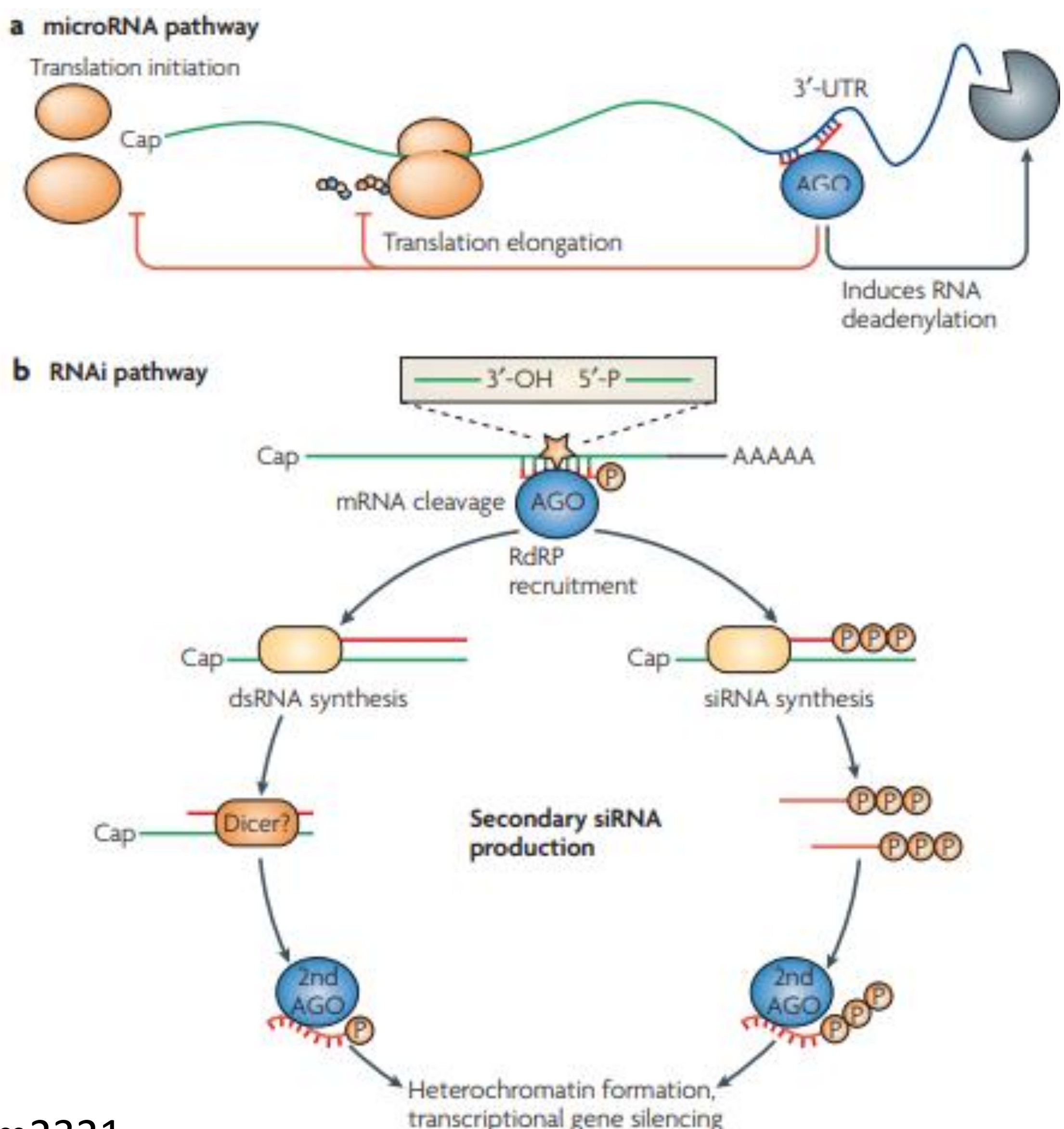
Reconhecimento 3’



Similaridade com eIF4E



Função das Argonautas



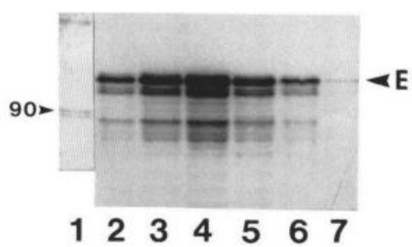
doi:10.1038/nrm2321

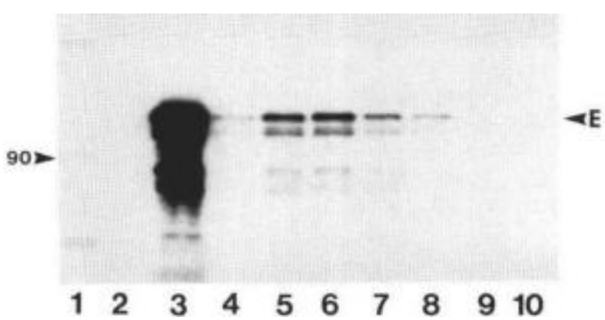
O início de tudo...

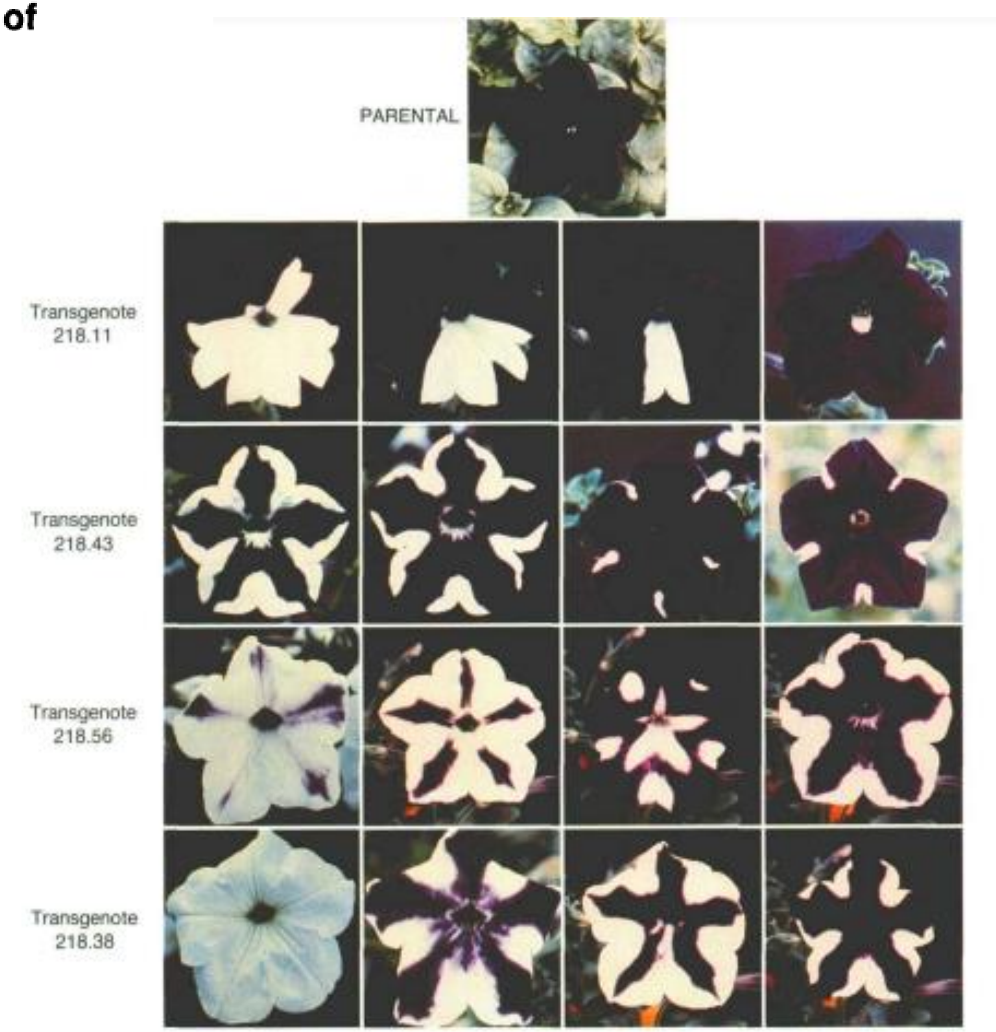
The Plant Cell, Vol. 2, 279–289, April 1990 © 1990 American Society of Plant Physiologists

Introduction of a Chimeric Chalcone Synthase Gene into Petunia Results in Reversible Co-Suppression of Homologous Genes *in trans*

Carolyn Napoli,[1] Christine Lemieux, and Richard Jorgensen[2]
DNA Plant Technology Corporation, 6701 San Pablo Avenue, Oakland, California 94608



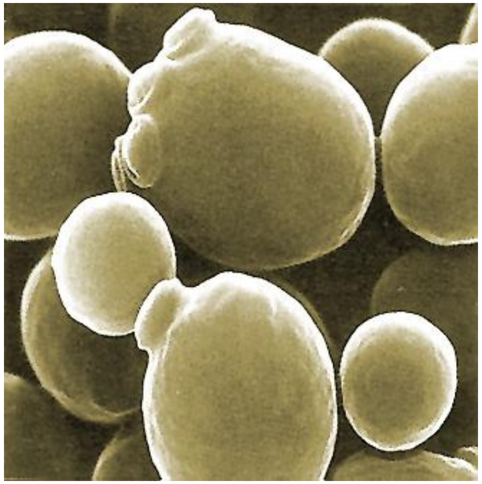


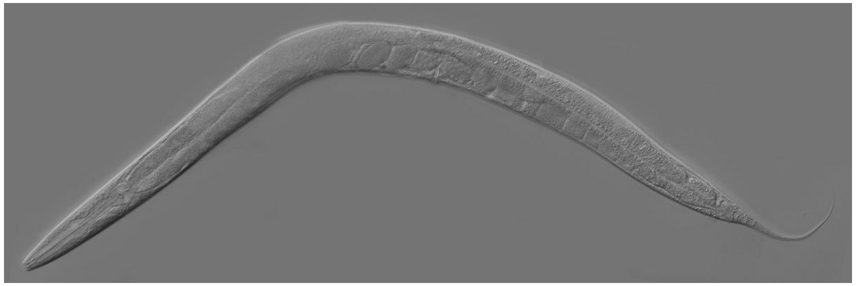


doi: 10.1105/tpc.2.4.279

Sistema presente em dierentes organismos modelo…





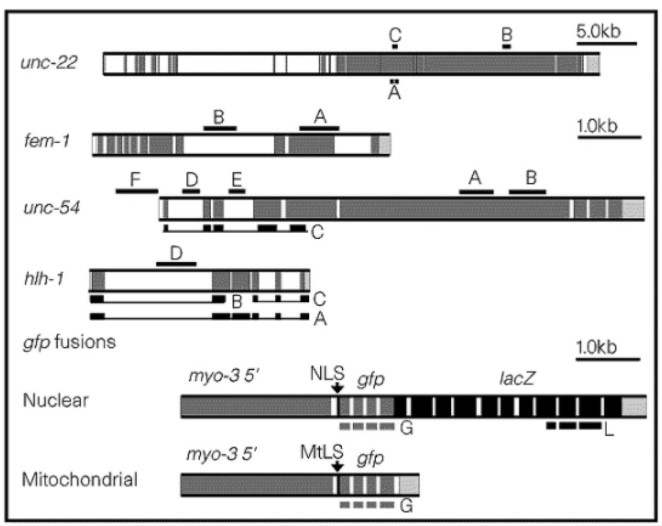


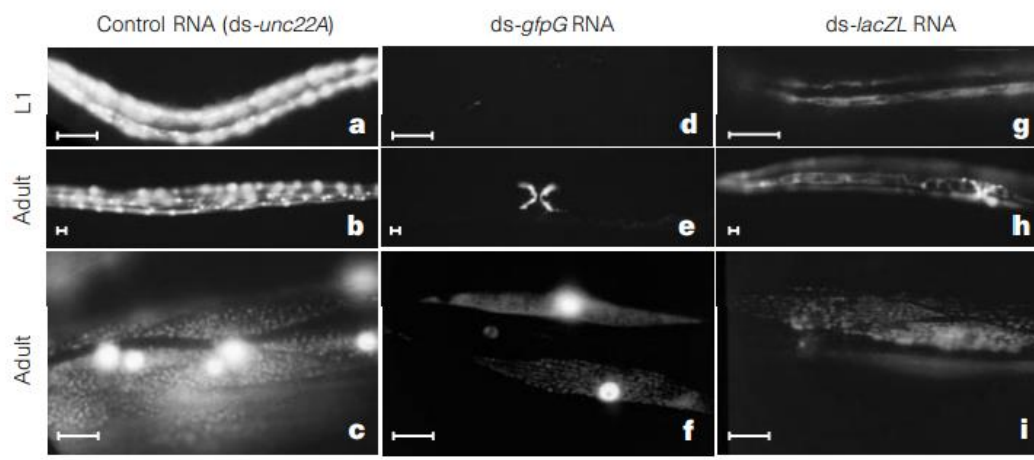
Potent and specific genetic interference by double-stranded RNA in *Caenorhabditis elegans*

Andrew Fire*, SiQun Xu*, Mary K. Montgomery*, Steven A. Kostas*†, Samuel E. Driver‡ & Craig C. Mello‡

* *Carnegie Institution of Washington, Department of Embryology, 115 West University Parkway, Baltimore, Maryland 21210, USA*
† *Biology Graduate Program, Johns Hopkins University, 3400 North Charles Street, Baltimore, Maryland 21218, USA*
‡ *Program in Molecular Medicine, Department of Cell Biology, University of Massachusetts Cancer Center, Two Biotech Suite 213, 373 Plantation Street, Worcester, Massachusetts 01605, USA*

Figure 1: Genes used to study RNA-mediated genetic interference in *C.elegans*.





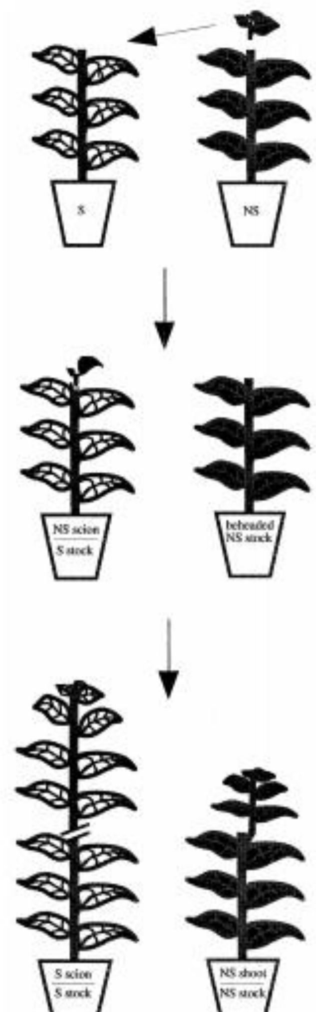
The EMBO Journal Vol.16 No.15 pp.4738–4745, 1997

Systemic acquired silencing: transgene-specific post-transcriptional silencing is transmitted by grafting from silenced stocks to non-silenced scions

Jean-Christophe Palauqui, Taline Elmayan, Jean-Marie Pollien and Hervé Vaucheret[1]

Laboratoire de Biologie Cellulaire, INRA, 78026 Versailles Cedex, France

[1]Corresponding author



articles

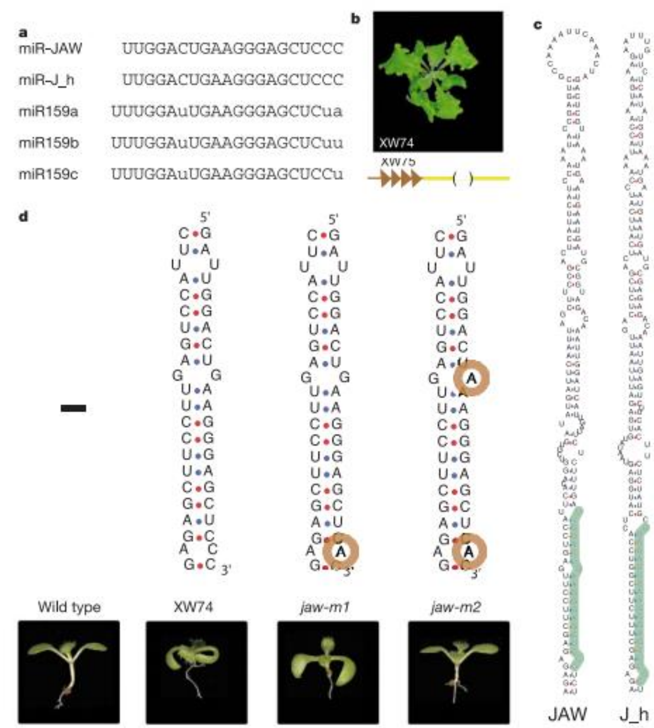
Control of leaf morphogenesis by microRNAs

Javier F. Palatnik[1,2], Edwards Allen[3], Xuelin Wu[2*], Carla Schommer[1*], Rebecca Schwab[1*], James C. Carrington[3] & Detlef Weigel[1,2]

[1]*Department of Molecular Biology, Max Planck Institute for Developmental Biology, D-72076 Tübingen, Germany*
[2]*Plant Biology Laboratory, The Salk Institute for Biological Studies, La Jolla, California 92037, USA*
[3]*Center for Gene Research and Biotechnology and Department of Botany and Plant Pathology, Oregon State University, Corvallis, Oregon 97331, USA*

* These authors contributed equally to this work

JAW locus – produz um microRNA que degrada genes TCP controladores de desenvolviemnto foliar. microRNA do TCP4 mRNA é necessário para previnir atividade aberrante TCP4. TCP genes com microRNA correspondentes é encontrado em várias espécies vegetais.



TRENDS in Genetics Vol.17 No.8 August 2001

RNA silencing as a plant immune system against viruses

Olivier Voinnet

Table 1. Virus-induced gene silencing as a widespread phenomenon in plants

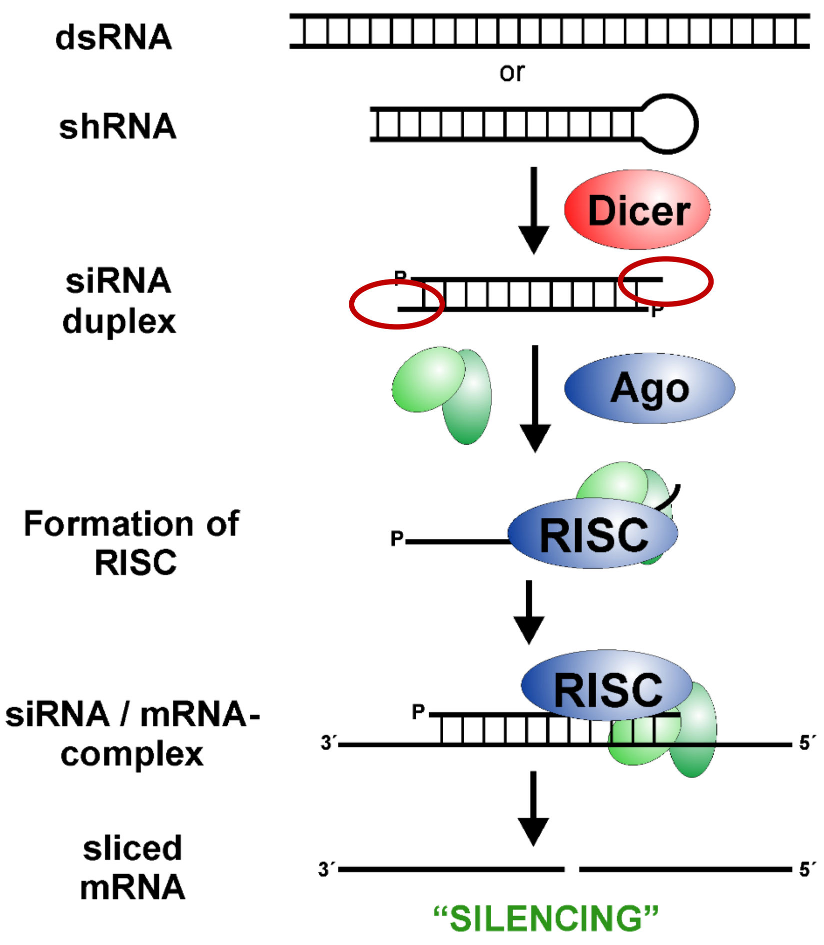
Virus[a]	Group	Genome	Plant species	Target nuclear gene	Refs
TMV	Tobamovirus	RNA	*Nicotinia benthamiana*	Phytoene desaturase	61
PVX	Potexvirus	RNA	*Nicotinia benthamiana*	Phytoene desaturase, Rubisco small subunit, cellulose synthase, GFP transgene	62,63
TRV	Tobravirus	RNA	*Nicotinia benthamiana*	Phytoene desaturase, Rubisco small subunit, LEAFY homologue[b], GFP transgene	47,64
			Arabidopsis	Phytoene desaturase, GFP transgene	
TEV	Potyvirus	RNA	*Nicotinia benthamiana*	TEV coat protein transgene	65
PSbMV	Potyvirus	RNA	*Pisum satvium*	PSbMV replicase transgene	66
PPV	Potyvirus	RNA	*Nicotinia benthamiana*	PPV replicase transgene	67
TYDV	Geminivirus	DNA	*Petunia hybrida*	Chalcone synthase transgene	68
TGMV	Geminivirus	DNA	*Nicotinia benthamiana*	Magnesium chelatase, luciferase transgene	69

[a]Abbreviations: TMV, tobacco mosaic virus; PVX, potato virus X; TRV, tobacco rattle virus; TEV, tobacco etch virus; PSbMV, pea seed-borne mosaic virus; PPV plum pox virus; TYDV, tobacco yellow dwarf virus; TGMV, tomato golden mosaic virus.
[b]LEAFY is an *Arabidopsis* gene involved in flower development.

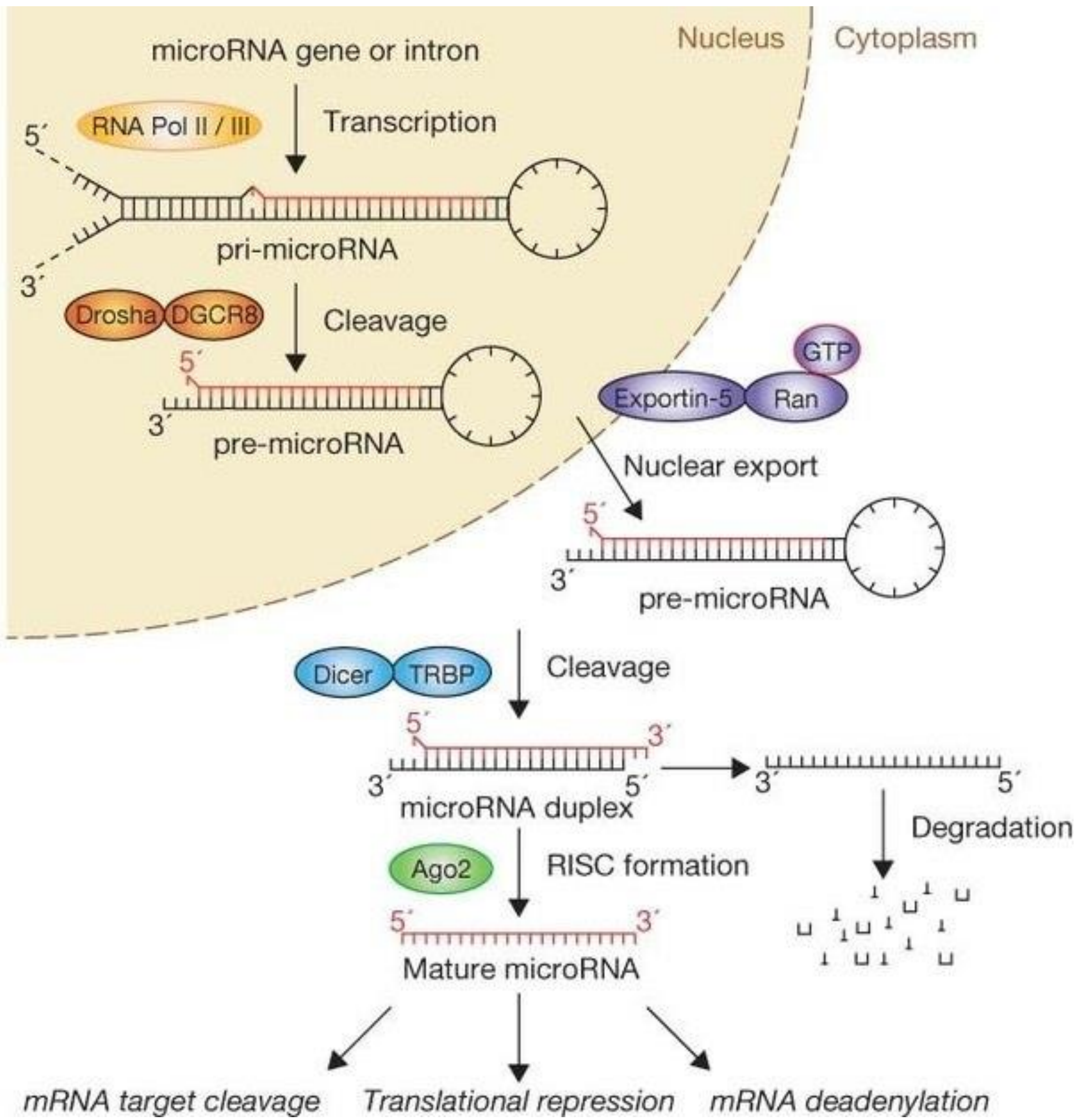
DE ONDE VEM Os siRNAs?

- transcrição convergente,
- complementariedade de transcritos,
- *harpin* em transcritos,
- RNA-directed RNA polymerase (RdRP).

EXÓGENO!!!!



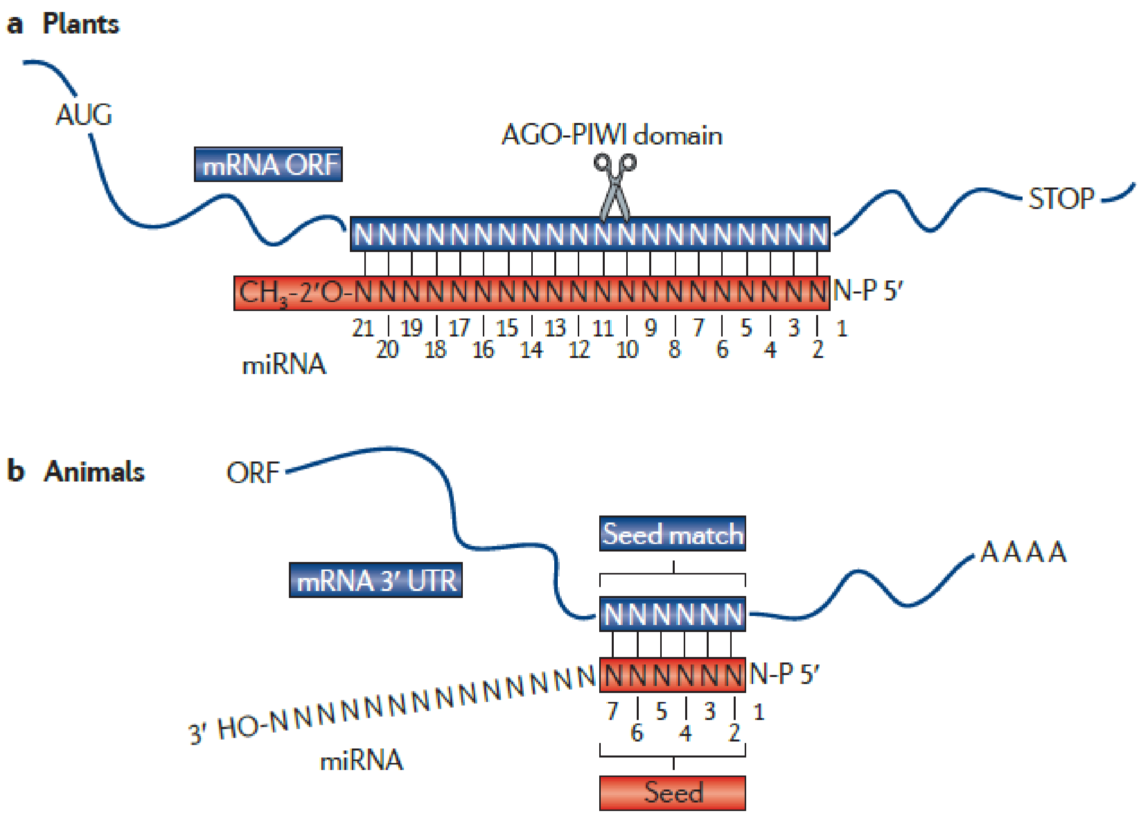
E OS miRNA?



Nucleus
Cytoplasm
microRNA gene or intron
RNA Pol II / III
Transcription
5´
3´
pri-microRNA
Drosha
DGCR8
Cleavage
5´
3´
pre-microRNA
GTP
Exportin-5
Ran
Nuclear export
5´
3´
pre-microRNA
Dicer
TRBP
Cleavage
5´
3´
3´
5´
microRNA duplex
3´
5´
Degradation
Ago2
RISC formation
5´
3´
Mature microRNA
mRNA target cleavage
Translational repression
mRNA deadenylation

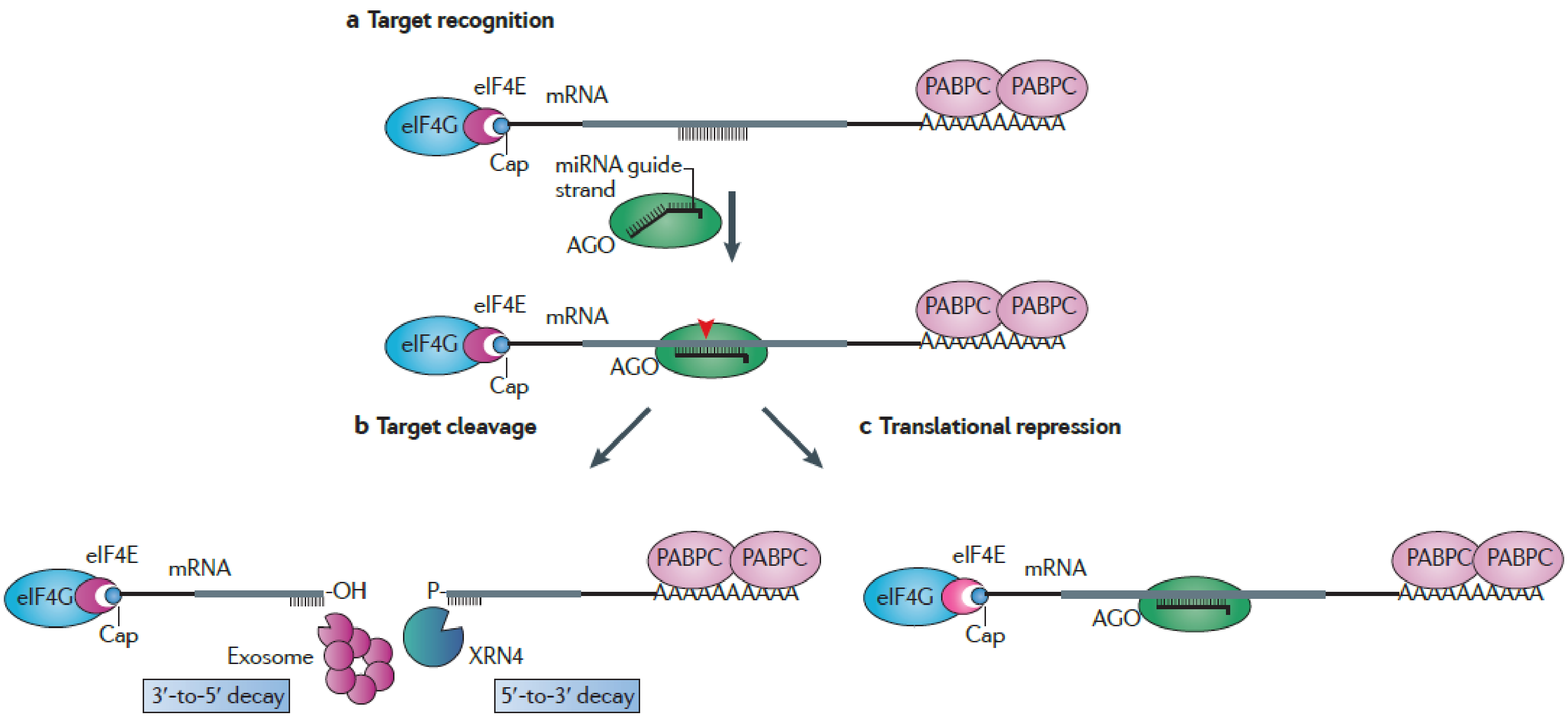
Gene silencing by microRNAs: contributions of translational repression and mRNA decay

Eric Huntzinger and Elisa Izaurralde



doi:10.1038/nrg2936

miRNA EM PLANTAS



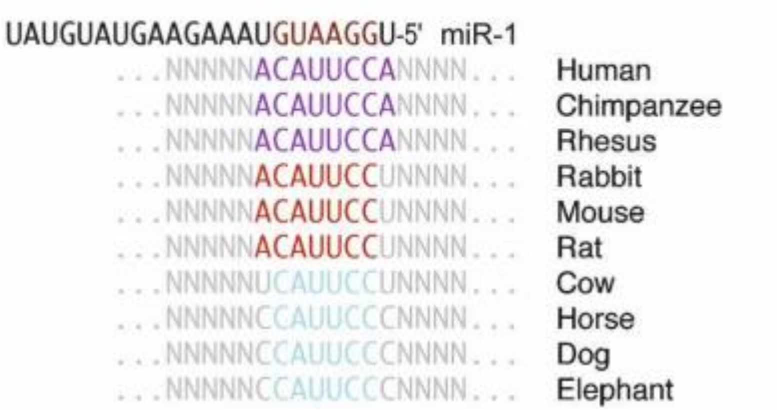
doi:10.1038/nrg2936

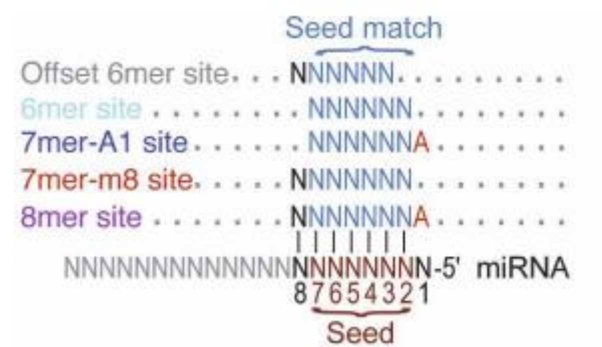
Methods

Most mammalian mRNAs are conserved targets of microRNAs

Robin C. Friedman,[1,2,3] Kyle Kai-How Farh,[1,2,4] Christopher B. Burge,[1,5] and David P. Bartel[1,2,5]

[1]Department of Biology, Massachusetts Institute of Technology, Cambridge, Massachusetts 02139, USA; [2]Whitehead Institute for Biomedical Research and Howard Hughes Medical Institute, Cambridge, Massachusetts 02142, USA; [3]Computational and Systems Biology Program, Massachusetts Institute of Technology, Cambridge, Massachusetts 02139, USA; [4]Division of Health Sciences and Technology, Massachusetts Institute of Technology, Cambridge, Massachusetts 02139, USA





É estimado que >45,000 miRNA sítios alvos em humanos mRNA 3'UTRs, >60% de genes codificadores de proteínas tem pressão seletiva para manter MREs

Biological Psychiatry

MicroRNA and Posttranscriptional Dysregulation in Psychiatry

Michael Geaghan and Murray J. Cairns

Table 1. Psychiatric Disease–Associated miRNAs Identified by Multiple Studies

miRNA	Authors	miRNA	Authors
Schizophrenia		Major Depressive Disorder	
miR-106b[a]	Perkins et al., 2007 (14); Moreau et al., 2011 (41); Smalheiser et al., 2014 (43)	miR-107[a]	Smalheiser et al., 2011 (53); Belzeaux et al., 2012 (85)
miR-107[a]	Beveridge et al., 2010 (15); Santarelli et al., 2011 (21)	miR-125a	Smalheiser et al., 2011 (53); Cao et al., 2013 (86)
miR-132[a]	Kim et al., 2010 (22); Miller et al., 2012 (42)	miR-132[a]	Smalheiser et al., 2011 (53); Li et al., 2013 (56)
miR-132*[a]	Kim et al., 2010 (22); Miller et al., 2012 (42)	miR-142-3p	Smalheiser et al., 2011 (53); Smalheiser et al., 2012 (52)
miR-134	Santarelli et al., 2011 (21); Gardiner et al., 2012 (25)	miR-145[a]	Smalheiser et al., 2011 (53); Belzeaux et al., 2012 (85)
miR-137	Ripke et al., 2011 (30); Whalley et al., 2012 (45); Green et al., 2013 (79)	miR-182	Smalheiser et al., 2011 (53); Li et al., 2013 (56); Cao et al., 2013 (86)
miR-150	Santarelli et al., 2011 (21); Miller et al., 2012 (42)	miR-200c	Smalheiser et al., 2011 (53); Belzeaux et al., 2012 (85)
miR-15a[a]	Beveridge et al., 2010 (15); Moreau et al., 2011 (41)	miR-298	Smalheiser et al., 2011 (53); Cao et al., 2013 (86)
miR-16	Beveridge et al., 2010 (15); Smalheiser et al., 2014 (43)	miR-376a*[a]	Smalheiser et al., 2011 (53); Smalheiser et al., 2012 (52); Belzeaux et al., 2012 (85)
miR-17[a]	Santarelli et al., 2011 (21); Wong et al., 2013 (80); Smalheiser et al., 2014 (43)	miR-381	Smalheiser et al., 2011 (53); Belzeaux et al., 2012 (85)
miR-181b	Beveridge et al., 2008 (16); Beveridge et al., 2010 (15); Shi et al., 2012 (29)	miR-494	Smalheiser et al., 2012 (52); Belzeaux et al., 2012 (85)
miR-195[a]	Perkins et al., 2007 (14); Beveridge et al., 2010 (15); Shi et al., 2012 (29)	miR-497	Smalheiser et al., 2011 (53); Smalheiser et al., 2012 (52)
miR-212	Perkins et al., 2007 (14); Kim et al., 2010 (22)	Autism Spectrum Disorders	
miR-219-3p	Shi et al., 2012 (29); Smalheiser et al., 2014 (43)	miR-106b[a]	Abu-Elneel et al., 2008 (59); Sarachana et al., 2010 (62)
miR-24	Perkins et al., 2007 (14); Xu et al., 2010 (81)	miR-1288	Vaishnavi et al., 2013 (67); Mamale et al., 2014 (88)
miR-26b	Perkins et al., 2007 (14); Beveridge et al., 2010 (15)	miR-1308	Vaishnavi et al., 2013 (67); Mamale et al., 2014 (88)
miR-29c[a]	Perkins et al., 2007 (14); Beveridge et al., 2010 (15)	miR-132[a]	Abu-Elneel et al., 2008 (59); Talebizadeh et al., 2008 (61); Sarachana et al., 2010 (62)
miR-30b	Perkins et al., 2007 (14); Mellios et al., 2012 (82)	miR-146b	Abu-Elneel et al., 2008 (59); Talebizadeh et al., 2008 (61)
miR-30e	Perkins et al., 2007 (14); Xu et al., 2010 (81)	miR-148b*[a]	Abu-Elneel et al., 2008 (59); Sarachana et al., 2010 (62)
miR-320	Tabares-Seisdedos et al., 2009 (83); Miller et al., 2012 (42)	miR-149	Vaishnavi et al., 2013 (67); Mamale et al., 2014 (88)
miR-34a	Kim et al., 2010 (22); Lai et al., 2011 (28)	miR-17*[a]	Kannu et al., 2013 (65); Hemmat et al., 2014 (64)
miR-432	Lai et al., 2011 (28); Gardiner et al., 2012 (25)	miR-185	Sarachana et al., 2010 (62); Vaishnavi et al., 2013 (67); Mamale et al., 2014 (88)
miR-544	Kim et al., 2010 (22); Gardiner et al., 2012 (25)	miR-18a	Kannu et al., 2013 (65); Hemmat et al., 2014 (64)
miR-652	Lai et al., 2011 (28); Santarelli et al., 2011 (21)	miR-195[a]	Sarachana et al., 2010 (62); Vaishnavi et al., 2013 (67)
miR-7	Perkins et al., 2007 (14); Beveridge et al., 2010 (15); Kim et al., 2010 (22)	miR-199b-5p	Sarachana et al., 2010 (62); Ghahramani Seno et al., 2011 (63)
Bipolar Disorder		miR-19a	Kannu et al., 2013 (65); Hemmat et al., 2014 (64)
let-7b	Miller et al., 2012 (42); Shih et al., 2012 (84)	miR-19b-1	Kannu et al., 2013 (65); Hemmat et al., 2014 (64)
miR-106b[a]	Moreau et al., 2011 (41); Shih et al., 2012 (84); Smalheiser et al., 2014 (43)	miR-200a	Vaishnavi et al., 2013 (67); Mamale et al., 2014 (88)
miR-132[a]	Miller et al., 2012 (42); Whalley et al., 2012 (45)	miR-200b	Vaishnavi et al., 2013 (67); Mamale et al., 2014 (88)
miR-133b	Kim et al., 2010 (22); Shih et al., 2012 (84)	miR-20a	Kannu et al., 2013 (65); Hemmat et al., 2014 (64)
miR-145[a]	Kim et al., 2010 (22); Smalheiser et al., 2014 (43)	miR-211	Miller et al., 2009 (66); Sarachana et al., 2010 (62); Vaishnavi et al., 2013 (67)
miR-148b*[a]	Moreau et al., 2011 (41); Shih et al., 2012 (84)	miR-23a	Abu-Elneel et al., 2008 (59); Talebizadeh et al., 2008 (61); Sarachana et al., 2010 (62)
miR-15a[a]	Moreau et al., 2011 (41); Shih et al., 2012 (84)	miR-320a	Abu-Elneel et al., 2008 (59); Talebizadeh et al., 2008 (61)
miR-17[a]	Shih et al., 2012 (84); Smalheiser et al., 2014 (43)	miR-429	Vaishnavi et al., 2013 (67); Mamale et al., 2014 (88)
miR-27b	Moreau et al., 2011 (41); Shih et al., 2012 (84)	miR-484	Abu-Elneel et al., 2008 (59); Vaishnavi et al., 2013 (67)
miR-29a[a]	Kim et al., 2010 (22); Shih et al., 2012 (84)	miR-598	Abu-Elneel et al., 2008 (59); Vaishnavi et al., 2013 (67)
miR-29c	Shih et al., 2012 (84); Banigan et al., 2013 (44); Smalheiser et al., 2014 (43)	miR-649	Vaishnavi et al., 2013 (67); Mamale et al., 2014 (88)
miR-874	Kim et al., 2010 (22); Miller et al., 2012 (42)	miR-650	Ghahramani Seno et al., 2011 (63); Mamale et al., 2014 (88)
		miR-92a-1	Kannu et al., 2013 (65); Hemmat et al., 2014 (64)
		miR-93	Abu-Elneel et al., 2008 (59); Sarachana et al., 2010 (62)

These miRNAs have been associated with schizophrenia, bipolar disorder, major depressive disorder, or autism spectrum disorders by more than one study.

miRNA, microRNA.

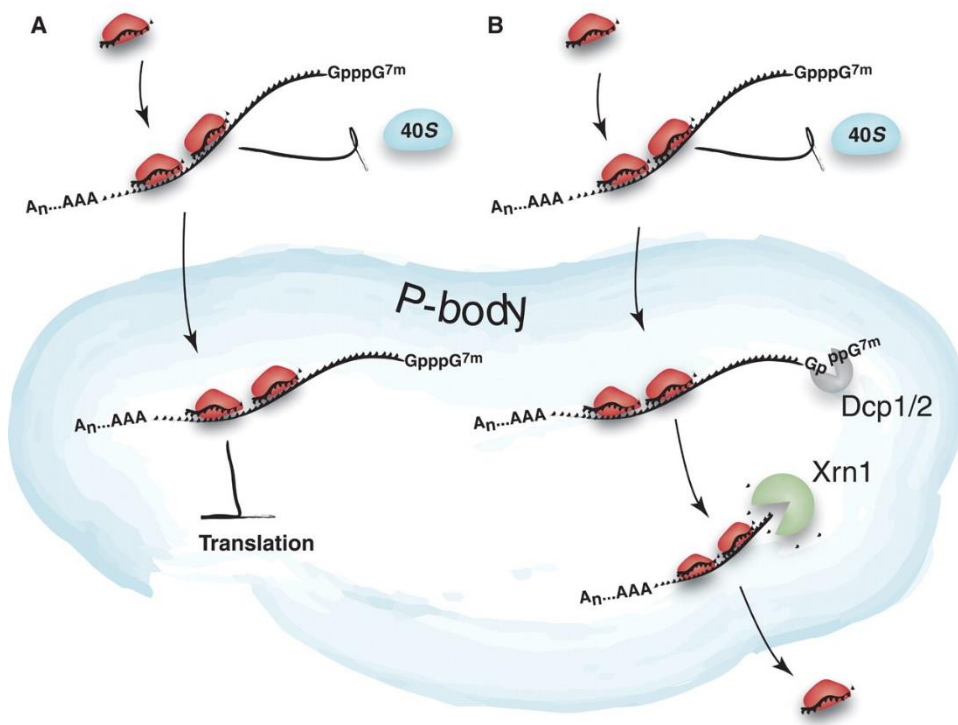
[a]MiRNA has been identified multiple times in more than one of these conditions.

MicroRNA-dependent localization of targeted mRNAs to mammalian P-bodies

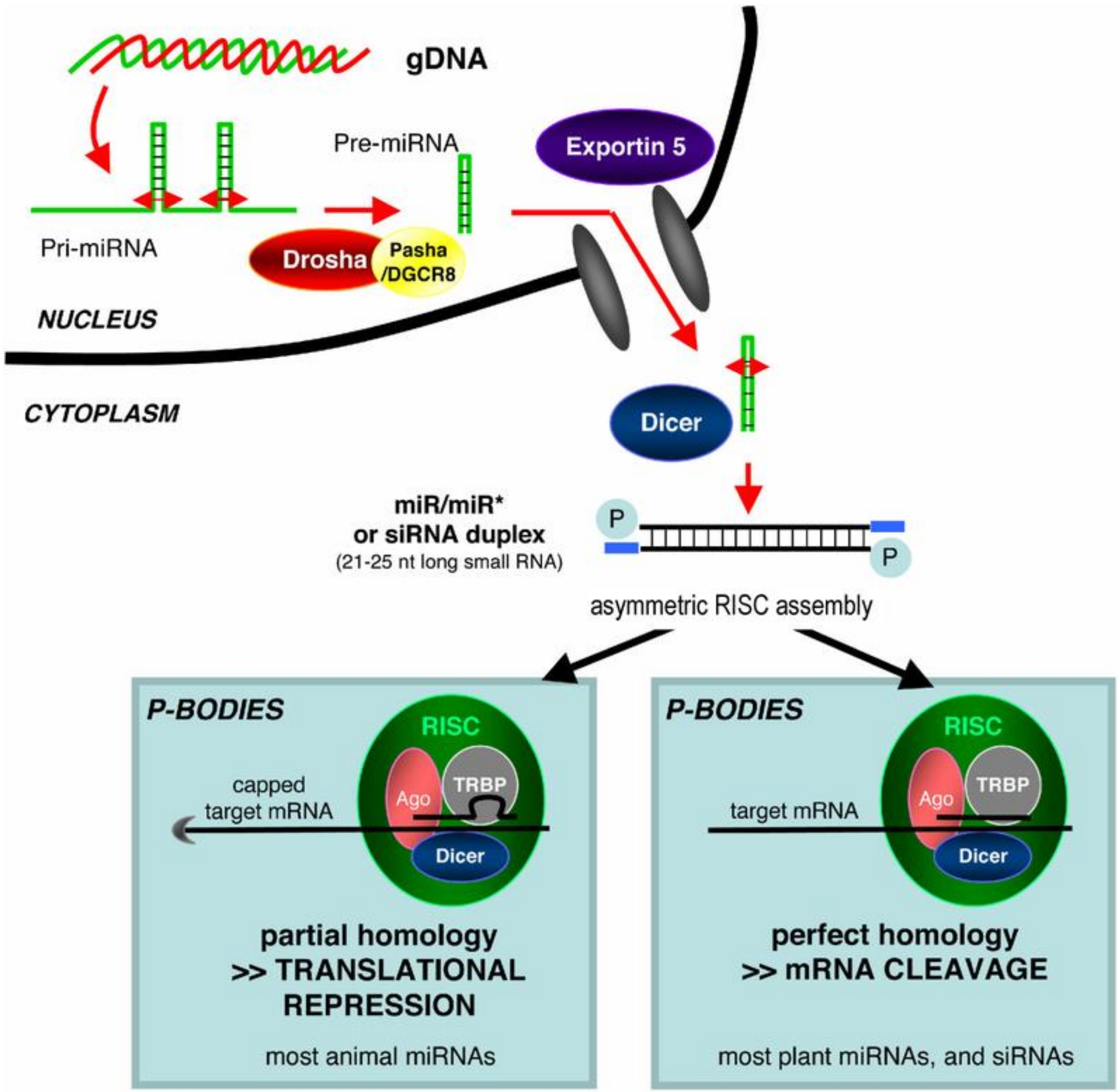
Jidong Liu[1], Marco Antonio Valencia-Sanchez[2], Gregory J. Hannon[1,3], and Roy Parker[2,3]

1 *Cold Spring Harbor Laboratory, Watson School of Biological Sciences, 1 Bungtown Road, Cold Spring Harbor, NY 11724, USA*

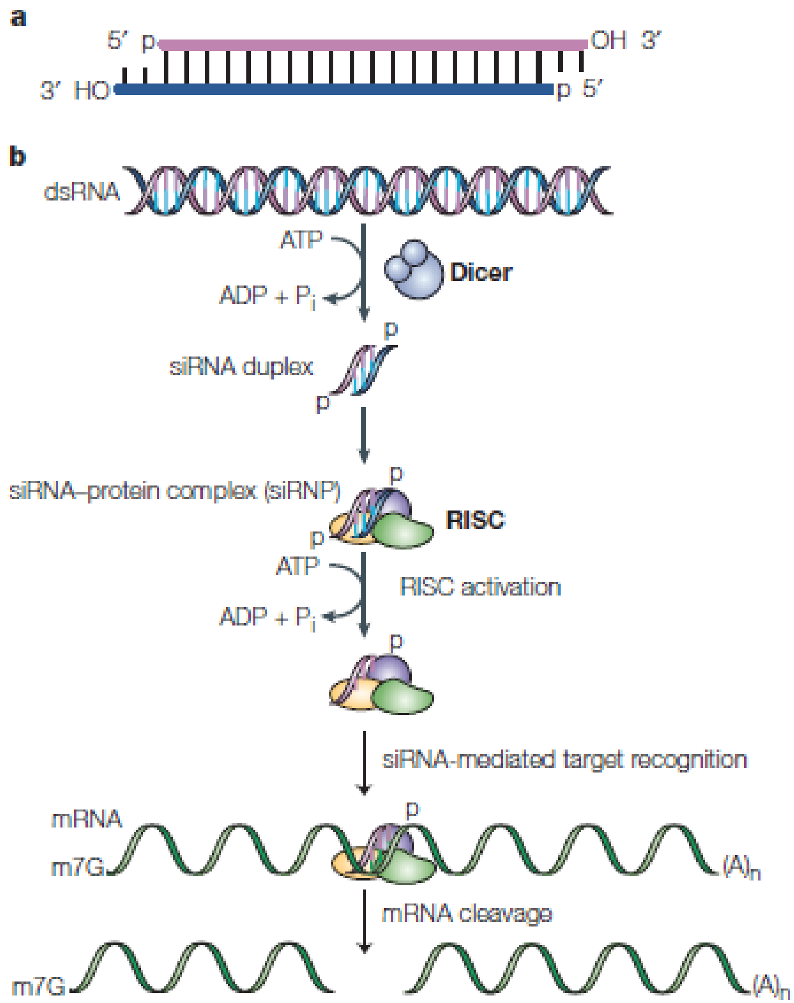
2 *Department of Molecular and Cellular Biology & Howard Hughes Medical Institute, University of Arizona, Tucson, AZ 85721, USA*



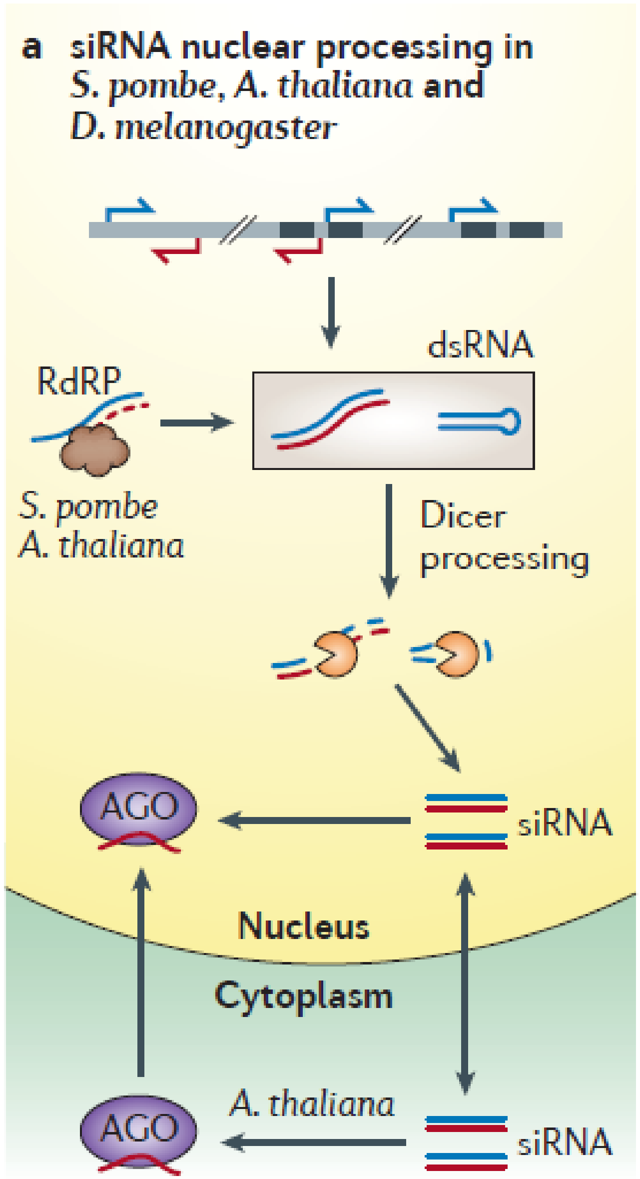
PLANTAS X ANIMAIS: SIMILARIDADES E DIVERGÊNCIAS



EM PLANTAS É COMUM OCORRER siRNA e miRNA



doi: 10.1038/nrm1129



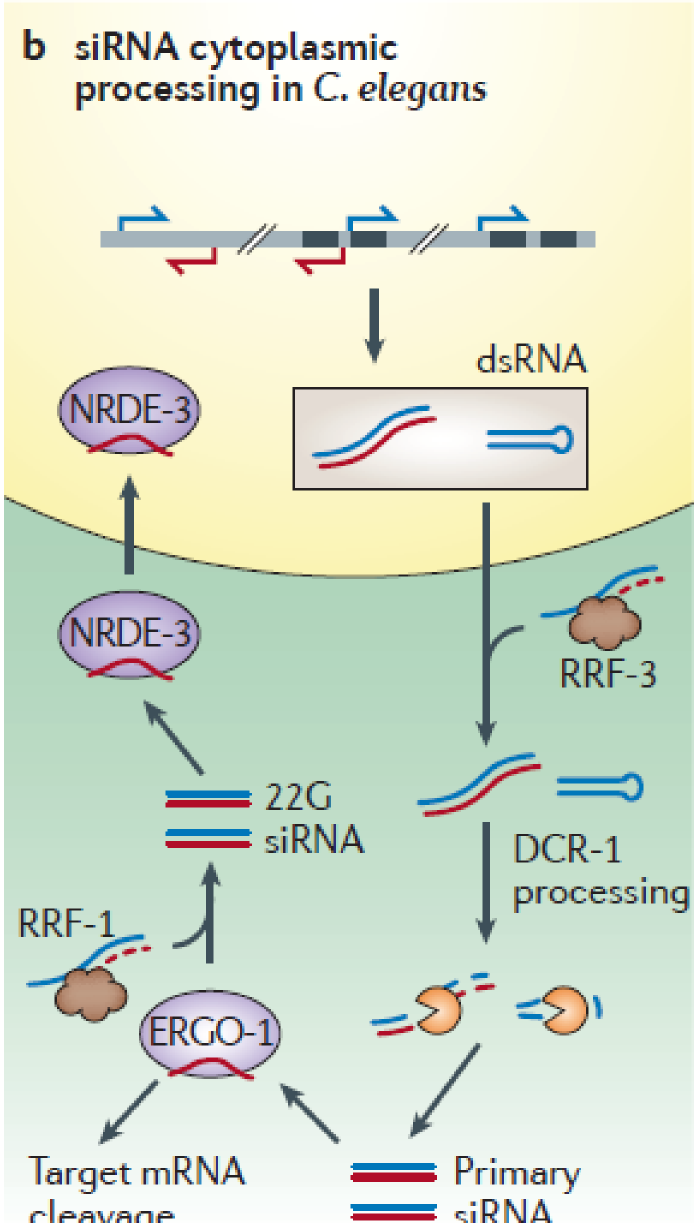
NON-CODING RNA

RNA interference in the nucleus: roles for small RNAs in transcription, epigenetics and beyond

Stephane E. Castel[1] and Robert A. Martienssen[1,2]

Small interfering RNA (siRNA) – o processamento ocorre no núcleo de *Schizosaccharomyces pombe* e *Drosophila melanogaster* e no nucléolo de *Arabidopsis thaliana*. Dupla-fita RNA (dsRNA) pode ser produzida por transcrição convergente, complementariedade de transcritos ou RNA-directed RNA polymerase (RdRP). As proteínas Dicer geram siRNAs que são acopladas em proteínas Argonaute (AGO). Em *A. thaliana*, siRNAs são transportados para o citoplasma acopladas à Argonatas e então importada para o núcleo.

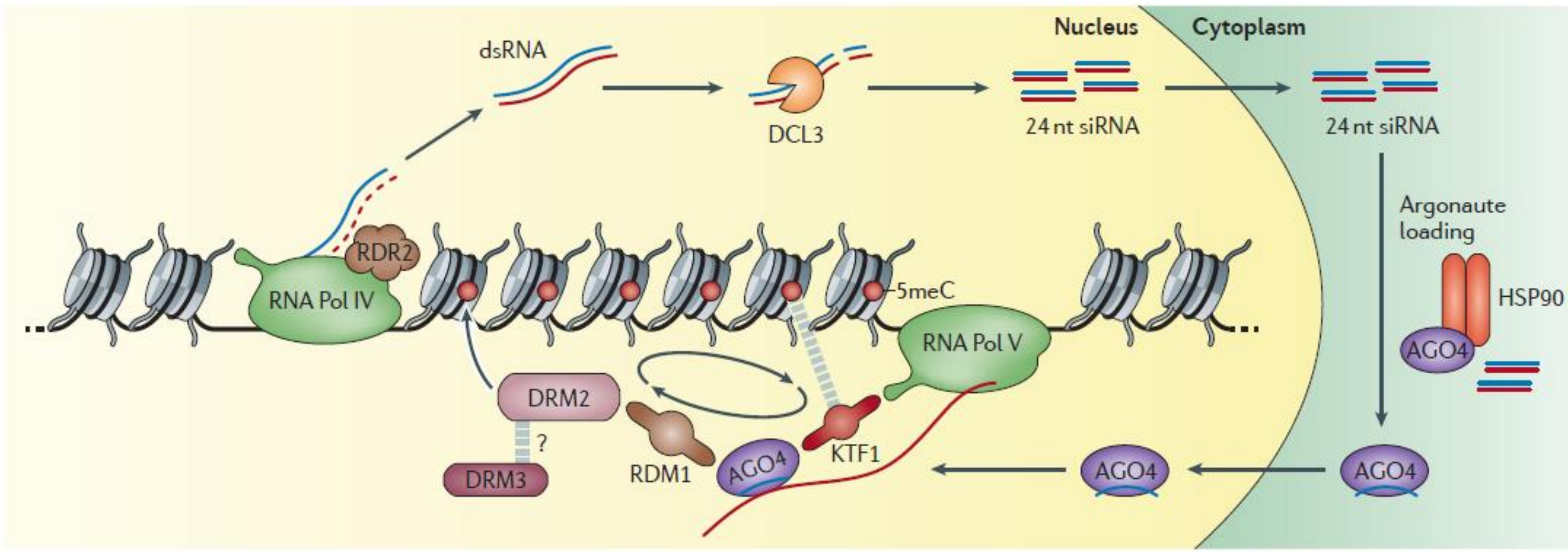
doi:10.1038/nrg3355



Caenorhabditis elegans, processo de siRNA ocorre no citoplasmaem duas etapas. dsRNA vem do núcleo ou RdRP de RRF-3, que age em transcritos do citoplasma. O DCR-1 produz primário 26-nucleotide siRNAs, que se liga a Argonaute ERGO-1 que facilita PTGS no citoplasma. Com RRF-1 pode gerar 22G siRNAs. No citoplasma 22G siRNAs são acopalhados no Argonaute NRDE-3 que são transportadas para o núcleo

doi:10.1038/nrg3355

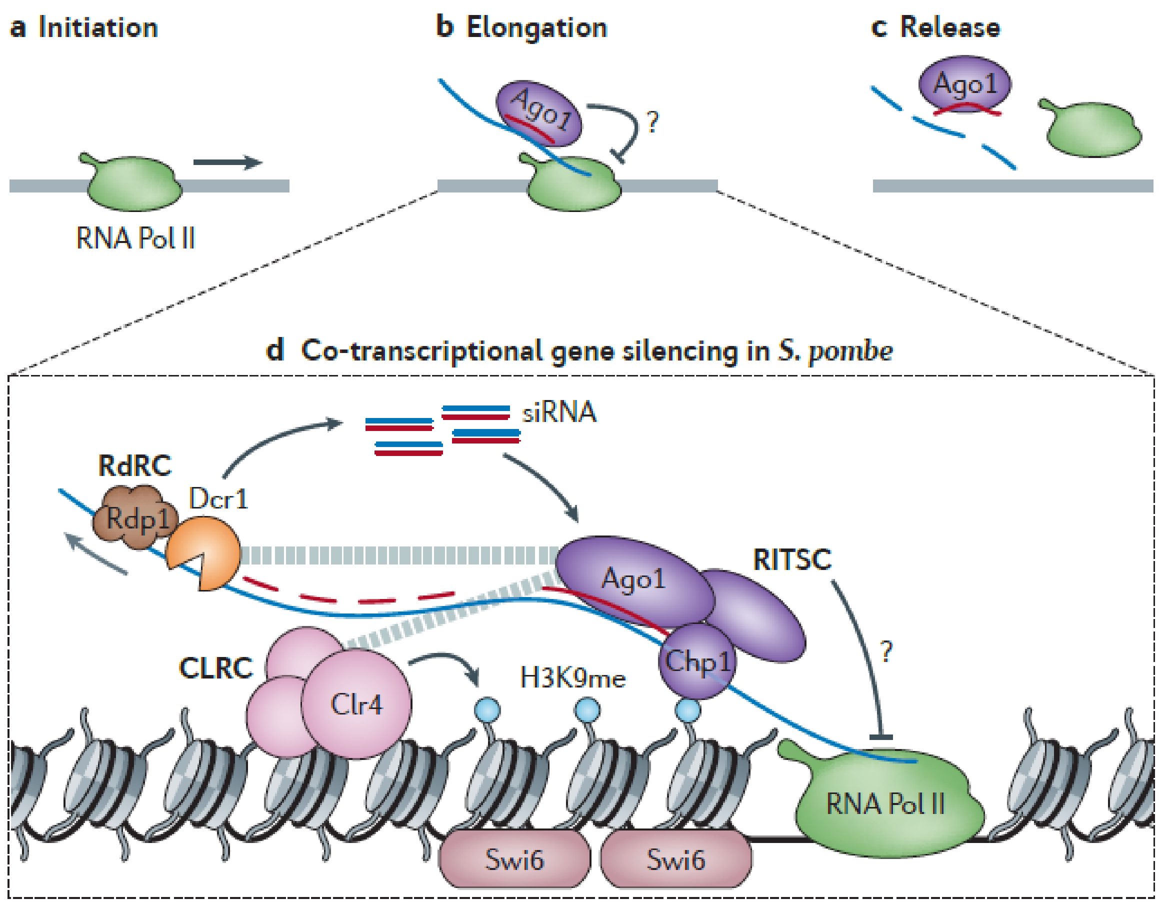
RNA-DEPENDENT DNA METHYLATION EM *Arabidopsis thaliana*



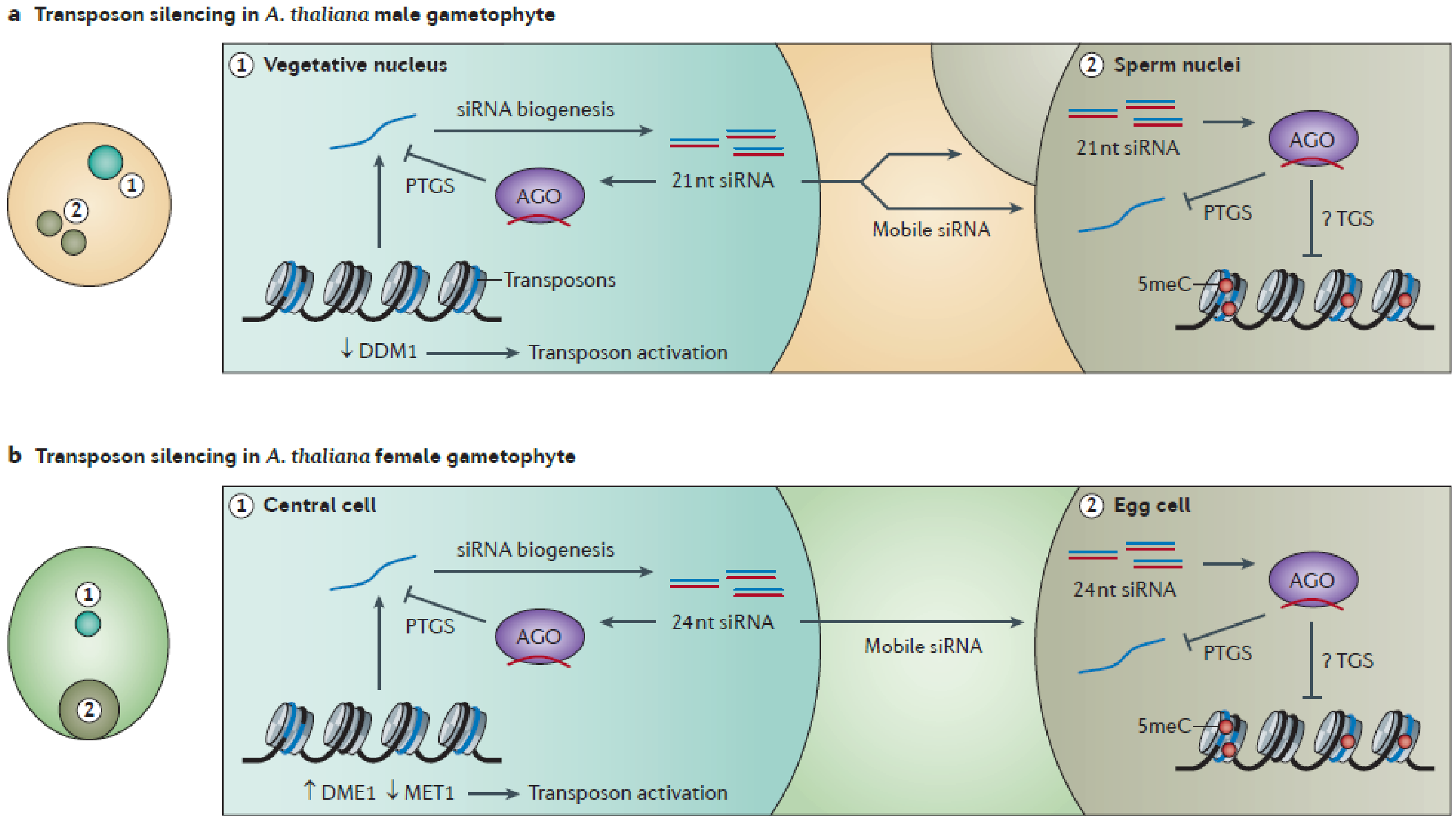
Transcritos provenientes da RNA Pol IV servem como substrato para a geração de siRNA, enquanto nascentes transcritos provenientes da RNA Pol V são alvos para RNAi

doi:10.1038/nrg3355

Silencimento co-trancricional em *S. pomboe*



doi:10.1038/nrg3355



DDM1 – remodelador de hetocromatina

DME1 - DNA glycosylase DEMETER

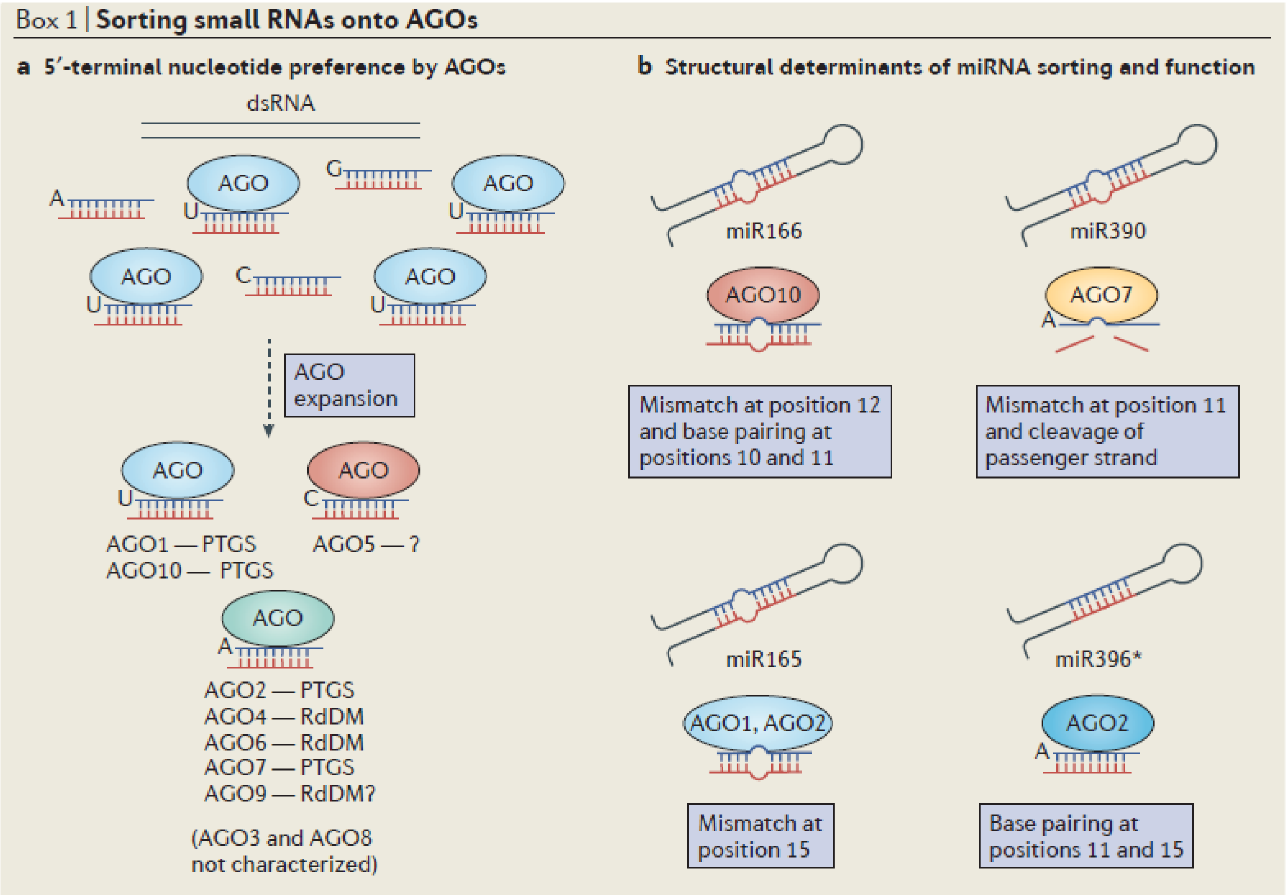
MET1- DNA metilase

doi:10.1038/nrg3355

The expanding world of small RNAs in plants

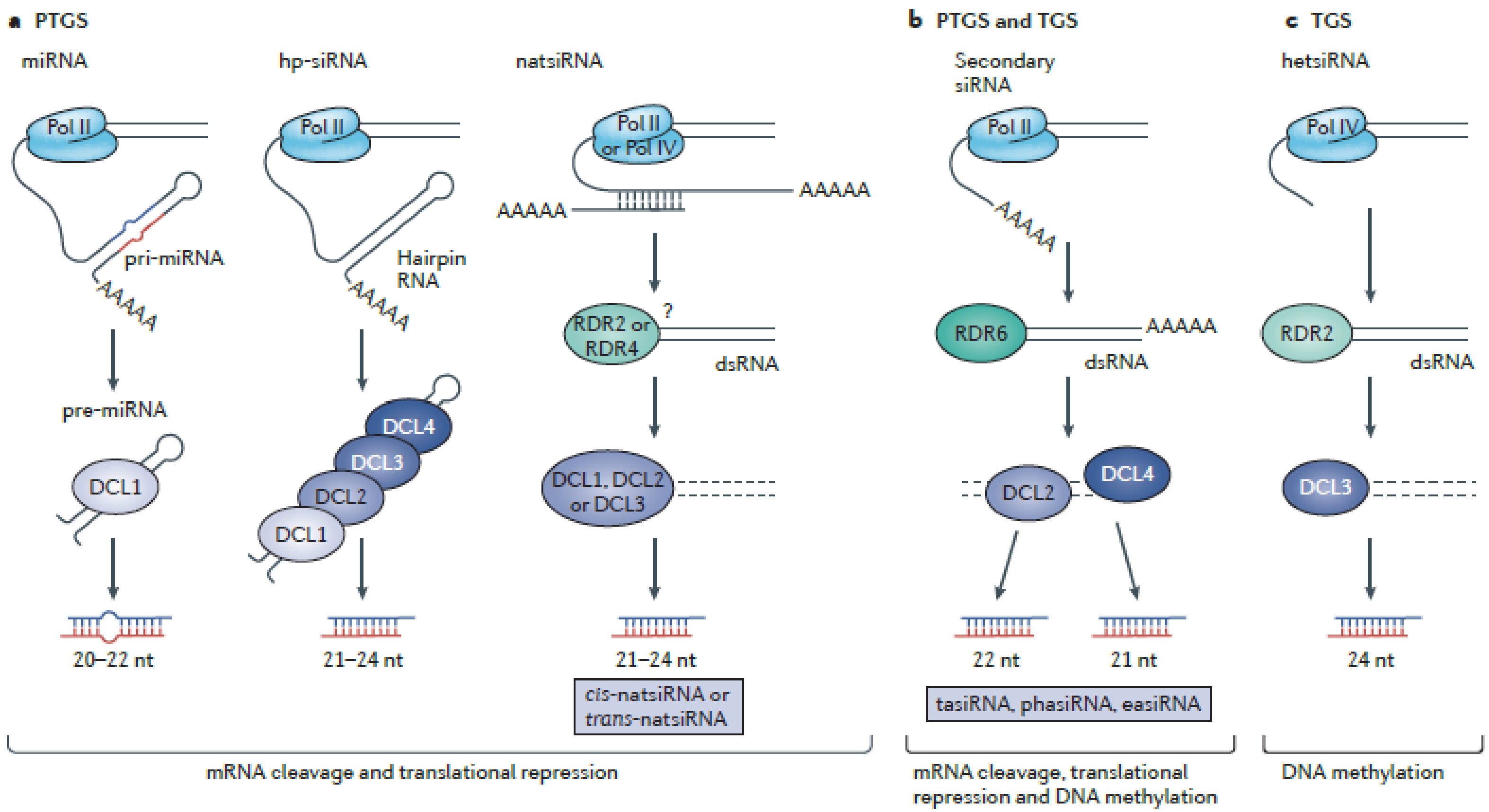
Filipe Borges and Robert A. Martienssen

doi:10.1038/nrm4085



Enorme diversidade de AGO em plantas!!

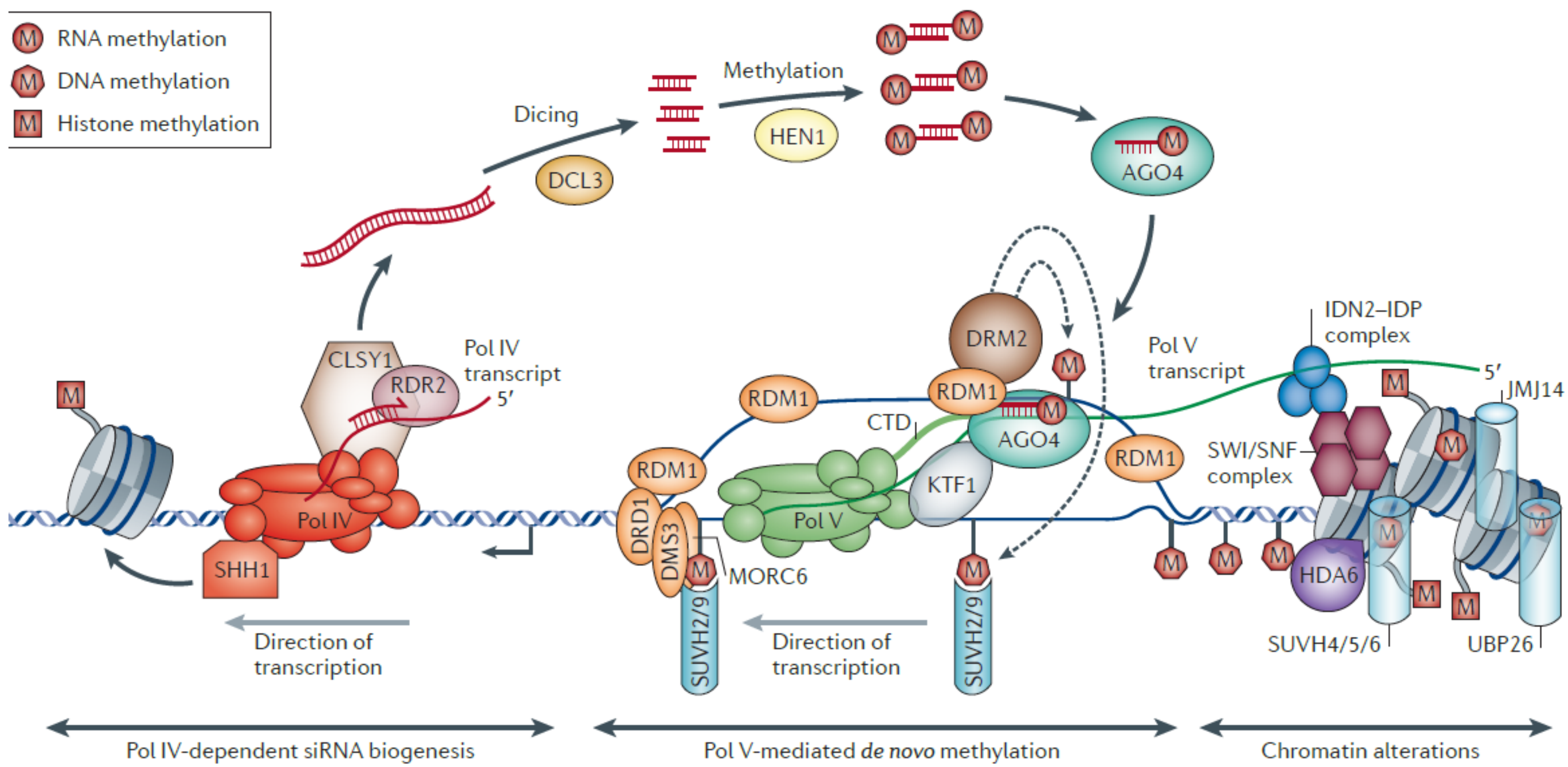
DIFERENTE VIAS PARA A BIOGENESE DE SMALL RNAs EM PLANTAS



doi:10.1038/nrm4085

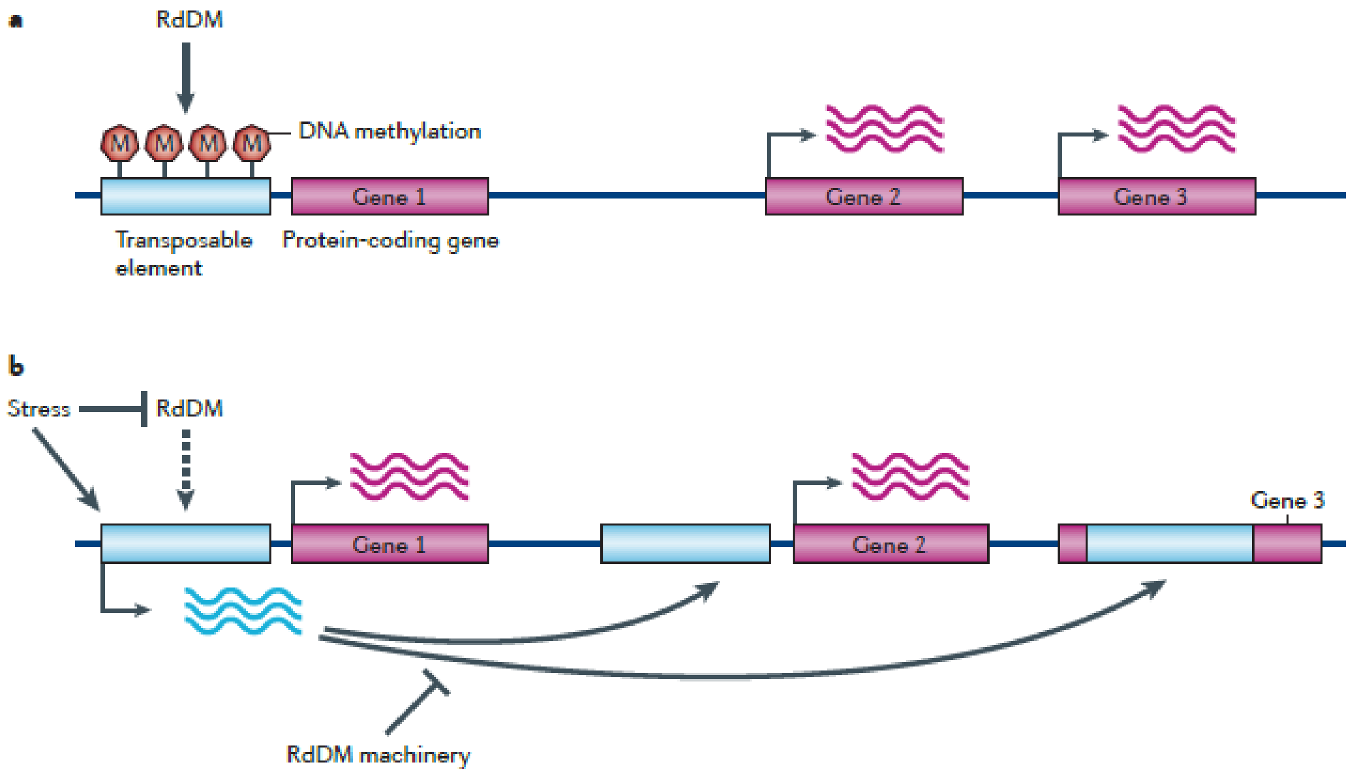
RNA-directed DNA methylation: an epigenetic pathway of increasing complexity

Marjori A. Matzke[1] and Rebecca A. Mosher[2]



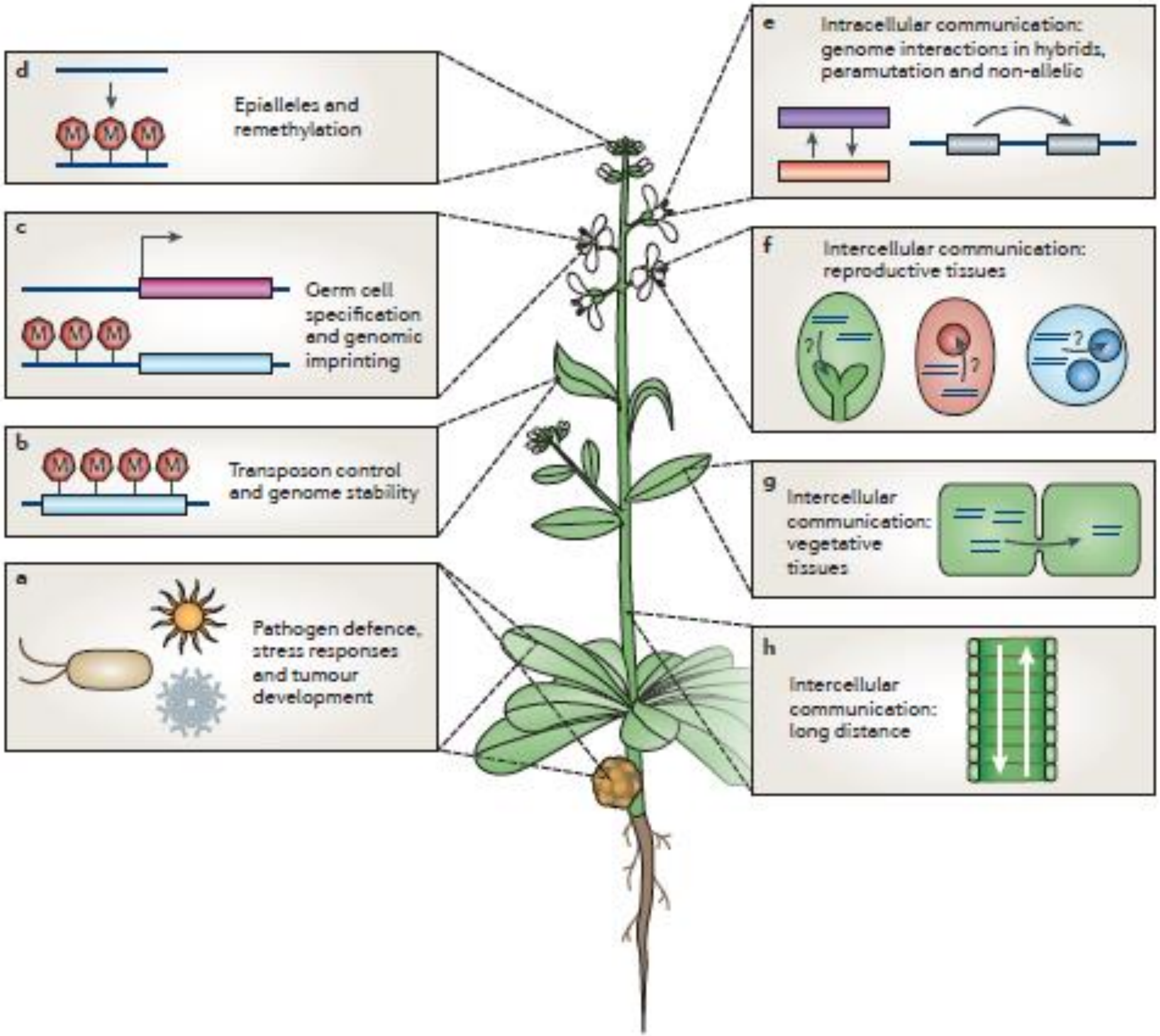
doi:10.1038/nrg3683

LIBERAÇÃO DE RDDM DURANTE STRESS



doi:10.1038/nrg3683

PROCESSOS BIOLÓGICOS ENVOLVENDO siRNAs E RdDM



Uma vasta literatura usando a técnica

Box 2 | **Potential determinants of efficient siRNA-directed gene silencing**

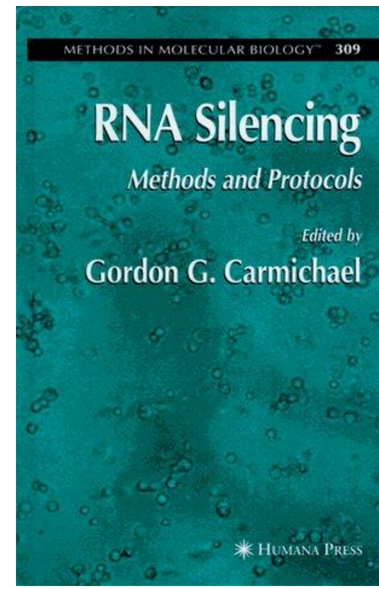
Sequence determinants intrinsic to the short interfering (si)RNA, the messenger RNA or both might affect the efficiency of each step of the siRNA-directed mRNA cleavage that results in efficient gene silencing.

siRNA

- Incorporation into the RNA-inducing silencing complex (RISC) and stability in RISC.
- Basepairing with mRNA.
- Cleavage of mRNA.
- Turnover of mRNA after cleavage.

mRNA

- The position of the siRNA-binding target region.
- Secondary and tertiary structures in mRNA.
- Binding of mRNA-associated proteins.
- Basepairing with siRNA.
- The rate of mRNA translation.
- The number of polysomes that are associated with translating mRNA.
- The abundance and half-life of mRNA.
- The subcellular location of mRNA.



http://profissaobiotec.com.br/primeira-planta-transgenica-contendo-tecnologia-rnai-com-atividade-inseticida-e-aprovada-nos-eua/

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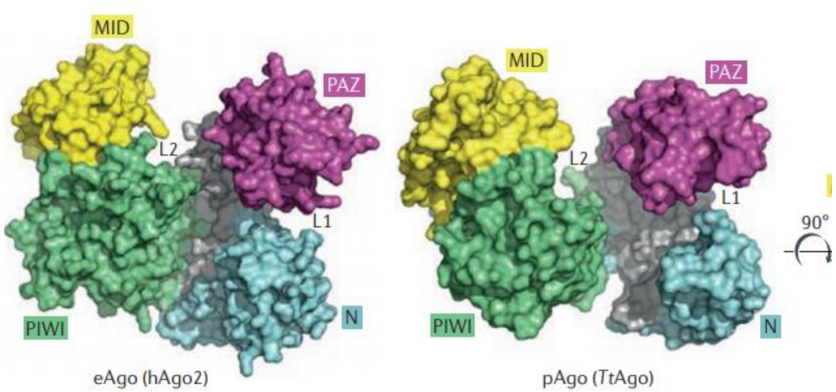
Table 1 | **Guide and target preferences of characterized eAgo and long pAgo proteins**

eAgo and long pAgo proteins		Activity				Refs
Host	Argonaute name	RNA-guided RNA interference	RNA-guided DNA interference	DNA-guided RNA interference	DNA-guided DNA interference	
Eukaryotes	eAgo	+	–	–	–	3
*Marinitoga piezophila**	*MpAgo*	+	+	–	–	14
*Thermotoga profunda**	*TpAgo*	–	+	–	–	14
Rhodobacter sphaeroides	*RsAgo*	–	+	–	–	11,20
Aquifex aeolicus	*AaAgo*	–	–	+	–	8
Natronobacterium gregoryi[‡]	*NgAgo*	–	–	unknown	unknown	35,40
Thermus thermophilus	*TtAgo*	–	–	+	+	12
Methanocaldococcus jannaschii	*MjAgo*	–	–	–	+	15,17
Pyrococcus furiosus	*PfAgo*	–	–	–	+	13

Plus signs indicate activity that has been demonstrated. Minus signs indicate either inactivity under tested conditions or untested. eAgo, eukaryotic Argonaute protein; pAgo, prokaryotic Argonaute protein. *Determined using 5′-hydroxylated guides. ‡Activity under investigation.

Jorrit W. Hegge, Daan C. Swarts and John van der Oost



PRÓXIMA AULA – 23 de junho

CRISPR-Cas9 – início 9:00 horas