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Popular Article

RNA Interference and their application in Veterinary Parasitology

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History

The term 'RNA world' was first coined some four billion years ago when RNA was genetic material and catalyst for emerging life on Earth. The process that reflects an RNA world -RNA silencing or RNA interference (RNAi). When exposed to foreign genetic material (RNA or DNA), many organisms mount highly specific counter attacks to silence the invading nucleic-acid sequences before these sequences can integrate into the host genome or subvert cellular processes. At the heart of these sequence-directed immunity mechanisms is double-stranded RNA (dsRNA). dsRNA also guides endogenous developmental gene regulation, and can even control the modification of cellular DNA and associated chromatin. (Mello *et al*.,2004)

By Fire *et al*,1999 dsRNA is a potent trigger for RNAi in the nematode *Caenorhabditis elegans* accelerated the discovery of a unifying mechanism that underlies a host of cellular and developmental pathways. (Hammond *et al*.,2000)

dsRNA was thought to be a nonspecific silencing agent that triggers a general destruction of messenger RNAs and the complete suppression of protein translation in mammalian cells. Second, dsRNA is energetically stable and inherently incapable of further specific Watson–Crick base pairing.

Components of RNA Interference

✓ **Dicer**

RNase III family members are among the few nucleases that show specificity for dsRNAs (Nicholson *et al.*,1999) and cleave them with 3′ overhangs of 2 to 3 nucleotides and 5′-

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phosphate and 3′-hydroxyl termini (Elbashir *et al*.,2001).

✓ **Guide RNAs and RNA-Induced Silencing Complex**

The sequence-specific nuclease activity observed in the cellular extracts responsible for ablating target mRNAs are RNA-induced silencing complex (RISC) (Hammond *et al*.,2000).

- ✓ RNA and DNA Helicases
- ✓ Translation Initiation Factor
- ✓ RNA-Dependent RNA Polymerase
- ✓ Transmembrane Protein (Channel or Receptor)

Mechanism of RNA Interference

The first step involves degradation of dsRNA into small interfering RNAs (siRNAs), 21 to 25 nucleotides long, by an RNase III-like activity. In the second step, the siRNAs join an RNase complex, RISC (RNA-induced silencing complex), which acts on the cognate mRNA and degrades it. Duplex unwinder by helicases, and RNA get activated. Such complexes promote RNA degradation and inhibit translation. The siRNA binds to Argonaute protein and one of the strands from the double stranded is removed. The remaining strand binds to the mRNA target sequences. The Argonaute protein either cleaves the mRNA or recruits other factors to regulate the target sequence. Some of these components also control the development of many organisms by processing many noncoding RNAs, called micro-RNAs.

Terms for RNAi

Three phenotypically different but mechanistically similar forms of RNAi, co-suppression or PTGS in plants, quelling in fungi, and RNAi in the animal kingdom, have been described. More recently, micro-RNA formation, heterochromatinization, etc., have been revealed as other facets of naturally occurring RNAi processes of eukaryotic cells.

Current understanding of RNA interference

- ✓ Directs silencing of gene expression in a sequence specific manner
- ✓ Principal systems for achieving RNA interference are short synthetic double stranded RNA molecules and gene expression vectors that direct their production in the cell Libraries

RNA interference in mammals

Introduction of double stranded RNA into mammalian cells induces a powerful set of quite different antiviral responses characterized by production of interferons, resulting in inhibition of all gene expression and rapid cell death, limiting the ability of a virus to replicate

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and spread throughout the organism.

RNA interference as a functional genomics tool in Therapeutics

DNA microarray technology has now enabled the level of expression of every gene in the genome to be determined under any condition. This has led to a vast accumulation of information about genes whose expression is significantly altered in various disease states,for example, death of a cultured cancer cell but not a normal cell.(Agrawal *et al*.,2003)

RNA interference in Veterinary Parasitology

Gene knockdown has been reported for *Schistosoma mansoni* by soaking or electroporating different life-stages in dsRNA. In case of strongylid parasites where only a small number of genes were susceptible to RNAi-mediated gene knockdown.

The **tick histamine release factor (tHRF)** from I. scapularis was characterized by Dai et al. (2010). tHRF is secreted in tick saliva, upregulated in B. burgdorferi-infected ticks and it appears to have a role in tick engorgement and efficient B. burgdorferi transmission (Dai *et al*., 2010). Silencing tHRF by RNAi significantly impaired tick feeding and decreased B. burgdorferi infection levels in mice.

Ferritins are iron-storage proteins that play a pivotal role in the homeostasis of iron during tick feeding.Ferritin 2 (RmFER2) knockdown by RNAi and vaccination with the recombinant protein resulted in reduction of feeding, oviposition and fertility in I. ricinus, R. microplus and R. annulatus (Hajdusek *et al*., 2009, 2010) .

Blocking TROSPA with TROSPA antisera or via RNA interference (RNAi) reduces B. burgdorferi adherence to the gut of I. scapularis, and as a result reduces bacterial colonization of the vector and, potentially, pathogen transmission to the host (Pal *et al*., 2004).

RNA interference (RNAi) on parasitic nematodes has been described as successful and useful for the identification of novel drug and vaccine candidates. Cattle parasite *Ostertagia ostertagi*. Down-regulation of target transcript levels was evaluated by semi-quantitative reverse transcriptase (RT) PCR. (Visser *et al*.,2006)

Approaches which eliminate mRNA expression directly are ideally suited for reverse genetics applications by RNAi tests in *Leishmania*, that facilitated RNAi tests at the α-tubulin locus, whose inhibition gives a strong lethal phenotype in trypanosomatids,no effect on parasite morphology, growth or tubulin expression in *Leishmania major* or *L. donovani*. (Robinson *et al*.,2003)

RNA-interference (RNAi) to achieve targeted gene knockdown in larval stages of the human blood fluke, *Schistosoma mansoni*,that SGTP1 transcript knockdown results in the

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functional phenotype of reduced glucose transport activity. (Stefanic *et al*.,2010)

References

Agrawal N, Dasaradhi PV, Mohmmed A, Malhotra P, Bhatnagar RK, Mukherjee SK. RNA interference: biology, mechanism, and applications. Microbiol Mol Biol Rev. 2003 Dec;67(4):657-85. doi: 10.1128/MMBR.67.4.657-685.2003. PMID: 14665679; PMCID: PMC309050.

Dai, J., Narasimhan, S., Zhang, L., Liu, L., Wang, P. and Fikrig, E., 2010. Tick histamine release factor is critical for Ixodes scapularis engorgement and transmission of the lyme disease agent. *PLoS pathogens*, *6*(11), p.e1001205.

Elbashir, S. M., W. Lendeckel, and T. Tuschl. 2001. RNA interference is mediated by 21- and 22-nucleotide RNAs. Genes Dev. 15:188-200.

Mello, C., Conte, D. Revealing the world of RNA interference. *Nature* **431**, 338–342 (2004).

Pal, U., Li, X., Wang, T., Montgomery, R.R., Ramamoorthi, N., DeSilva, A.M., Bao, F., Yang, X., Pypaert, M., Pradhan, D. and Kantor, F.S., 2004. TROSPA, an Ixodes scapularis receptor for Borrelia burgdorferi. *Cell*, *119*(4), pp.457-468.

Robinson, K.A. and Beverley, S.M., 2003. Improvements in transfection efficiency and tests of RNA interference (RNAi) approaches in the protozoan parasite Leishmania. *Molecular and biochemical parasitology*, *128*(2), pp.217-228.

Visser, A., Geldhof, P., De Maere, V., Knox, D.P., Vercruysse, J. and Claerebout, E., 2006. Efficacy and specificity of RNA interference in larval life-stages of Ostertagia ostertagi. *Parasitology*, *133*(6), pp.777-783.

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