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Nota di bibliografia	Includes bibliographical references and index.
Nota di contenuto	RNA Interference in Practice; Preface; Contents; 1 Introduction: RNA interference, the ""Breakthrough of the Year 2002""; 1.1 RNAi as a Tool for Functional Genomics; 1.2 Mechanism of RNAi; 1.3 Dicer - the Initiator to "Dice" the dsRNA?; 1.4 miRNAs versus siRNAs: Two Classes of Small RNAs Using the RNAi Pathway?; 1.5 RISC - the Effector to "Slice" the mRNA?; 1.6 Are RNA-Dependent RNA Polymerases (RdRps) Responsible for the Catalytic Nature of RNAi?; 1.7 Is RNAi Involved in the Regulation of Gene Expression?; 1.8 RNAi in Mammals; 1.9 Practical Approaches; 1.10 References 2 RNAi in Caenorhabditis elegans2.1 Introduction; 2.2 Application of RNAi in C. elegans; 2.3 Target Sequence Evaluation; 2.4 dsRNA Synthesis; 2.4.2 Generation of the DNA Template; 2.4.2.1 Plasmid Templates; 2.4.2.2 DNA Templates Derived by PCR/RT-PCR; 2.4.2 In-vitro dsRNA Transcription; 2.5 Delivery of dsRNA; 2.5.1 General Information on the C. elegans Anatomy; 2.5.2 C. elegans Strains for Silencing; 2.5.3 Culturing the Worms; 2.5.4 Microinjection Protocol; 2.5.5 Soaking Protocol; 2.5.6 RNAi Feeding Protocol; 2.5.7 DNA Templates for dsRNA Expression in Feeding E. coli 2.5.8 DNA Templates for Hairpin RNA Expression2.5.8.1 C. elegans Promoters; 2.5.8.2 Inverted Repeat Constructs; 2.6 Mounting Animals for Microscopy; 2.7 Genome Wide Screens; 2.7.1 C. elegans RNAi Library; 2.8 Selected Literature on C. elegans Research; 2.9 Useful C. elegans webpages; 2.10 References; 3 RNAi in Drosophila; 3.1 Introduction; 3.2 Application of RNAi in Drosophila; 3.2.1 dsRNA from Linear DNA Templates; 3.2.2 dsRNA from Inverted Repeat DNA; 3.2.3 Inducible Expression in Drosophila Cell Lines; 3.2.4 Limitations; 3.3 dsRNA Synthesis; 3.3.1 In-vitro dsRNA Transcription 3.3.2 Inverted Repeat DNA3.4 Injections; 3.4.1 Injection Services; 3.4.2 Injection Method; 3.4.3 DsRNA or Inverted Repeat DNA Preparation; 3.4.4 Embryo Collection; 3.5 Cell Lines; 3.6 Protocols; 3.6.1 Thawing and Maintenance of S2 Cells; 3.6.2 Freezing Protocol; 3.7 RNAi in S2 Cells; 3.7.1 dsRNA Transfection Using the Calcium Phosphate Method; 3.7.2 dsRNA Soaking of S2 Cells; 3.8 High-Throughput Screens; 3.8.1 Drosophila RNAi Library; 3.9 Useful Webpages for Drosophila Research; 3.10 Books and Literature on Drosophila; 3.11 References; 4 RNAi in Mammals; 4.1 Introduction 4.2 Transient RNAi in Cell Culture4.2.1 Chemical Synthesis and Modifications of siRNAs; 4.2.1.1 Advantages; 4.2.1.2 Limitations; 4.2.2 Custom Synthesis of siRNA Oligos; 4.2.3 siRNA Design Rules; 4.2.3.1 siRNA Strand Bias and Off-Gene Targeting; 4.2.3.2 Improvements in siRNA Stability; 4.2.3.3 siRNA Design: Novel Modifications of the "Tuschl Rules"; 4.2.3.4 Homology Search by BLAST, FASTA, or Smith-Waterman Algorithm; 4.2.3.5 Troubleshooting; 4.2.3.6 siRNA Design Programs and Algorithms; 4.2.3.7 Preparation of siRNA Duplexes; 4.2.4 Enzymatic Synthesis of siRNAs 4.2.4.1 Designing DNA Oligonucleotides
Sommario/riassunto	This hands-on guide to RNA interference brings the power of targeted gene silencing to any laboratory with the basic equipment for handling nucleic acids.In easy-to-follow, step-by-step protocols you will learn* how RNAi works in worms, flies and mammals,* how to design the most efficient RNAi constructs,* how to achieve transient, stable and conditional RNAi in cell cultures,* how to determine the efficiency of an RNAi experiment,* and how to use RNAi for gene therapy.All the protocols have been thoroughly tested in the author's own laboratory, and she provides exa