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Nota di contenuto	Lab Session 1 Getting Oriented: Practicing with Micropipettes -- Lab Session 2 Purification and Digestion of Plasmid (Vector) DNA -- Lab Session 3 PCR Amplification of egfp and Completion of Vector Preparation -- Lab Session 4 Preparation of Insert DNA (egfp) PCR Product -- Lab Session 5 DNA Ligation and Transformation of Escherichia coli -- Lab Session 6 Colony Hybridization -- Lab Session 7 Characterization of Recombinant Clones: Part 1 -- Lab Session 8 Characterization of Recombinant Clones: Part 2 -- Lab Session 9 Characterization of Recombinant Clones: Part 3 -- Lab Session 10 Computational Analysis of DNA Sequence from a Positive Clone: Part 2 -- Lab Session 11 Expression of Fusion Protein from Positive Clones, SDS-PAGE and Western Blot: Part 1 -- Lab Session 12 Expression of Fusion Protein from Positive Clones, SDS-PAGE and Western Blot: Part 2 -- Lab Session 13 Extraction of Recombinant Protein from Escherichia coli Using a Glutathione Affinity Column -- Lab Session 14 Analysis of

Purification Fractions -- Lab Session 15 Total RNA Purification -- Lab Session 16 Analysis of *gst::egfp* mRNA Levels by RT-qPCR: Part 1 -- Lab Session 17 Analysis of *gst::egfp* mRNA Levels by RT-qPCR: Part 2 -- Lab Session 18 Analysis of *gst::egfp* mRNA Levels by Semi-Quantitative RT-PCR: Part 1 -- Lab Session 19 Analysis of *gst::egfp* mRNA Levels by Semi-Quantitative RT-PCR: Part 2 -- Appendix 1 Equipment -- Appendix 2 Prep List -- Appendix 3 Preparation of Competent *E. coli* Cells -- Appendix 4 Pre-Lab Questions.

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## Sommario/riassunto

This manual is an indispensable tool for introducing advanced undergraduates and beginning graduate students to the techniques of recombinant DNA technology, or gene cloning and expression. The techniques used in basic research and biotechnology laboratories are covered in detail. Students gain hands-on experience from start to finish in subcloning a gene into an expression vector, through purification of the recombinant protein. The second edition has been completely re-written, with new laboratory exercises and all new illustrations and text, designed for a typical 15-week semester,

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