

RT-PCR Protocols, 2nd edition
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The first record I have of it, is when I made a computer file which I usually did whenever I had an idea, that would have been on the Monday when I got back, and I called it Chain Reaction.POL, meaning polymerase. That was the identifier for it and later I called the thing the Polymerase Chain Reaction, which a lot of people thought was a dumb name for it, but it stuck, and it became PCR.

With these words the Nobel Prize winner, Kary Mullis, explains how he named the PCR: one of the most important techniques ever invented and currently used in molecular biology. This book *RT-PCR Protocols* covers a wide range of aspects important for the setting of a PCR experiment for both beginners and advanced users. In my opinion the book is very well structured in three different sections. The first one describes the different technologies now available, like competitive RT-PCR, nested RT-PCR or RT-PCR for cloning. An important part regards the usage of PCR in single cell mouse embryos, stressing how important is to find a quantitative method to analyze gene expression in single mouse blastomeres, since different cells of the same embryo showed to have different genomic reprogramming. Single cells analyses have also been carried out using a Poly(A) cDNA real-time PCR as indicator for gene measurement in cancer, enabling global mRNA amplification of small amount of RNA from fresh to paraffin embedded tissue. In general, all the protocols described in this first

section have a detailed description of the methods and of the notes enriched with troubleshooting details and advices to improve the success of each experiment. The second section of the book describes the importance of PCR normalization and quantification, detailing which are the best protocols and the best housekeeping genes to be amplified together with target genes. An entire chapter is dedicated to the comparative qRT-PCR, an important method that uses conventional RT-PCR followed by electrophoresis and scanning densitometry. This is an important technique that can be used, for example, to investigate the effects of any compound on any gene in different cell population. The protocols described in each chapter stress the importance of having a good normaliser gene and, as expected, GAPDH seem to be the most suitable housekeeping gene, regardless of the type of the assay used. The third section of the book is more focused to the *initial steps*, fundamental for the success of the experiment. The name of the section is very intriguing and invites the reader to act like a chef mixing the right ingredients in order to make the PCR reaction, a successful one! This part gives a detailed description on how to design suitable primers, how to isolate, extract and quantify a good RNA and how to set up a working reaction for a working assay, starting from the first reverse transcription to the last amplification steps. Considering the versatility of the PCR and the importance of this technique in a wide range of studies, this book is very useful for those who face the magic world of the PCR for the first time and/or for those who are looking for advices or tricks able to improve their own home-made protocols.

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