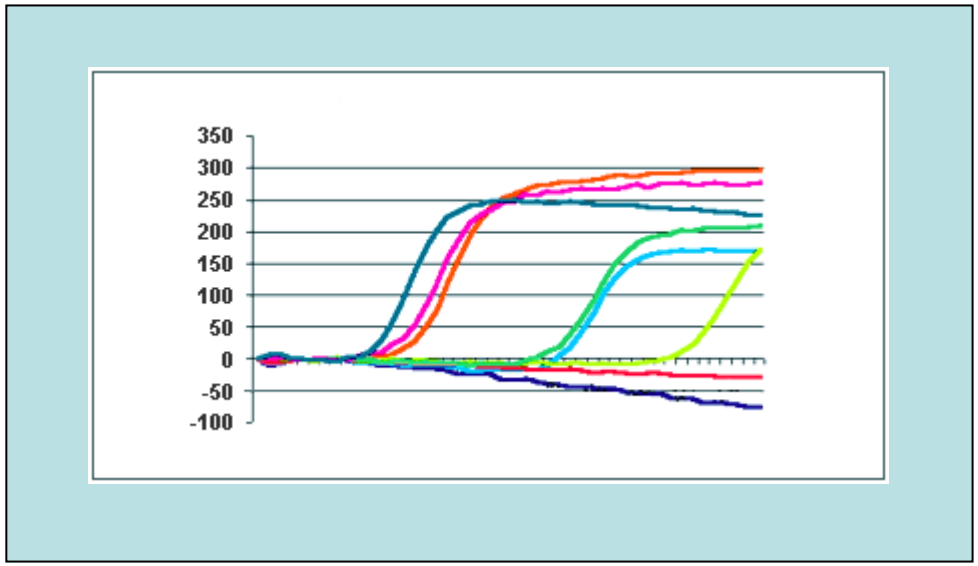
biocompare® Surveys

Real-Time PCR:

Quantifying the qPCR Marketplace



Executive Summary

Introduction

The conventional polymerase chain reaction (PCR) is a standard and essential tool in the research laboratory. With the development of real-time quantitative PCR (qPCR), the applications and acceptance of PCR have grown even further.[1] In conventional PCR, the amount of amplified product produced can only be quantified by using multiple samples or taking aliquots from a single sample at specific intervals. The samples are then analyzed with methods such as gel electrophoresis and Southern blot analysis. In real-time qPCR, the amount of amplified product is measured as the reaction proceeds (i.e. in real time) thus eliminating the need for post-PCR sample handling. This reduced handling minimizes carry-over contamination, improves reproducibility and enables the more rapid throughput of qPCR assays.[2]

Real-time detection of the qPCR amplicon is achieved through the use of a fluorescent reporter: either a general DNA-binding dye (e.g. SYBR Green) or a labeled probe which utilizes fluorescence resonance energy transfer (FRET) for quantitation. While DNA-binding dyes are the simpler and less expensive option, they are not specific to the amplicon. These dyes will bind to any double-stranded DNA (e.g. primer-dimers) and, as a result, can cause errors in quantitation. Alternatively, hybridization probes are specific to the DNA being amplified. There are two popular types of labeled probes: those which utilize the 5′ exonuclease activity of the polymerase in the real-time qPCR reaction (e.g. TaqMan® probes) and probes which self-hybridize into an hairpin stucture when free in solution (e.g. Molecular Beacons). According to the literature, the 5′ exonuclease probes are the most widely used probe format. However, the rate of publications describing other probe systems is increasing rapidly.1 The popularity of 5′ exonuclease probes is probably largely due to the early emergence of the TaqMan® probes on the commercial market. As alternate tools for the detection of qPCR products continue to be developed, we can anticipate an increasingly competitive real-time qPCR market.

The increased product choice which is generating a more competitive market has undoubtedly come about in response to the growth of the real-time qPCR market in recent years. The availability of sequence information created by numerous genome projects (e.g. human, mouse, dog, yeast) has enabled researchers to use oligonucleotide sequences to study previously unknown genes.3 This has led to the increased use of real-time qPCR in the study of gene expression and function. Its use in the field of diagnostics has also increased and real-time qPCR is now accepted as the standard method in a number of diagnostic assays.1 Undoubtedly, the number of applications for which real-time qPCR is used will also continue to increase. With all it has going for it, it's not surprising that individuals from a number of different companies agree that real-time qPCR is one of the fastest growing markets.4,5 It is the purpose of this study to help suppliers better understand the current real-time pPCR market and to identify where future opportunities are likely to exist.

Methodology

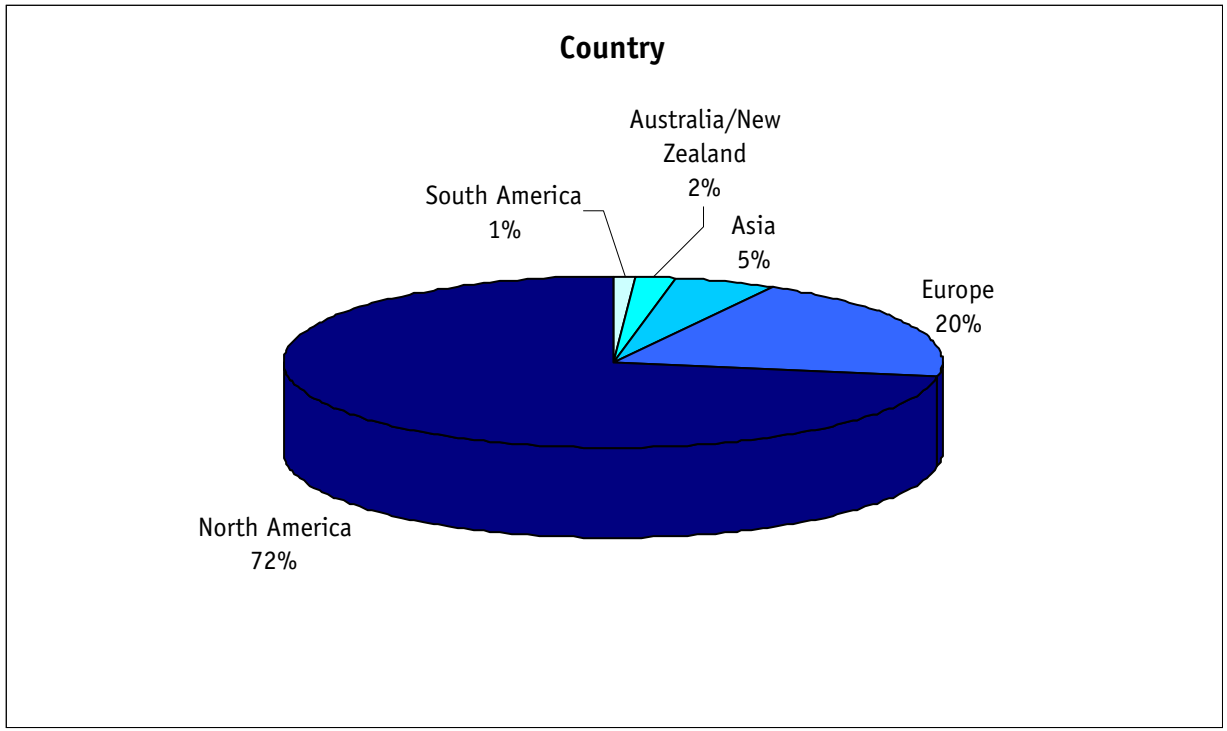
The *Real-Time PCR: Quantifying the qPCR Marketplace* survey was administered on-line between August 23 and 27, 2004. Invitations to complete the survey were emailed to life scientists who were interested in PCR technology. 746 researchers responded to the invitation. 567 of these indicated that they were using or planning to use Real-Time PCR technology and completed the survey.

The survey consisted of 21 closed-ended questions. Eleven questions included "other" as a response option, with space provided for the respondents to add the answer specific to their research.

Demographic information was evaluated from answers to 4 questions within the survey and from addresses provided by the respondents.

Results: Demographics

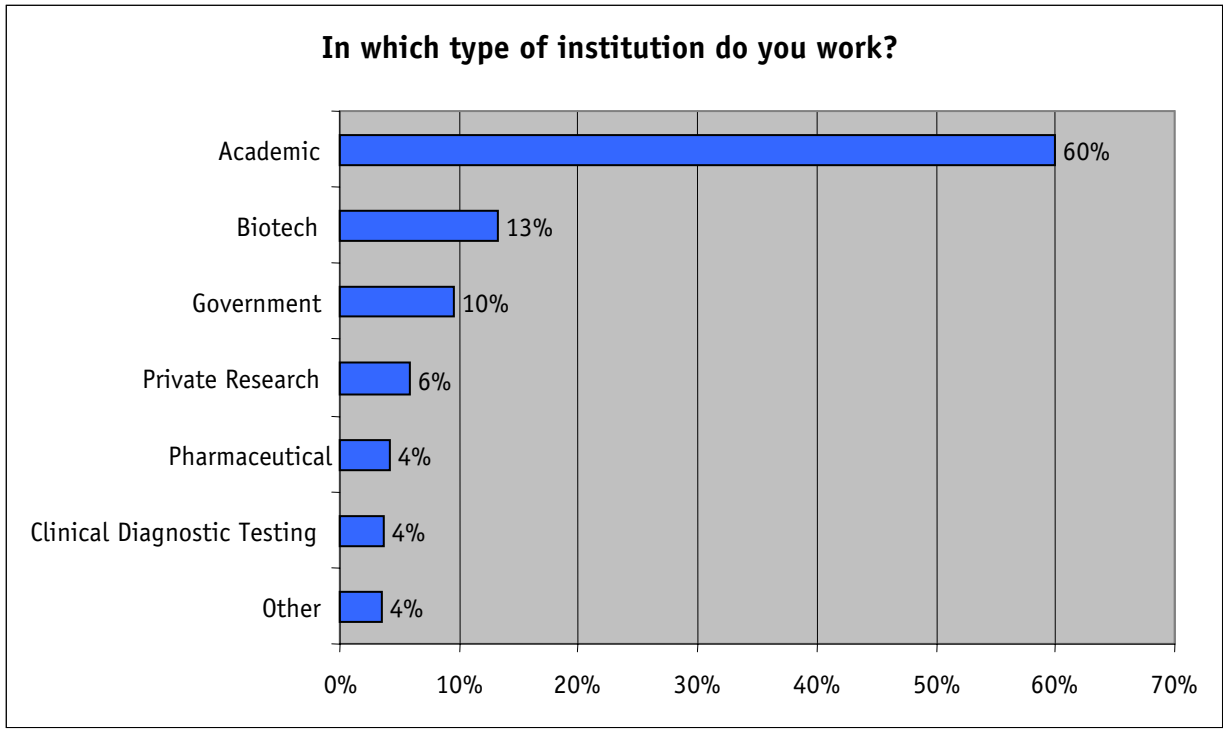
92% of respondents were in North America or Europe.



n=567

*Only those respondents who are either currently using or planning to use real-time PCR are included in this chart.

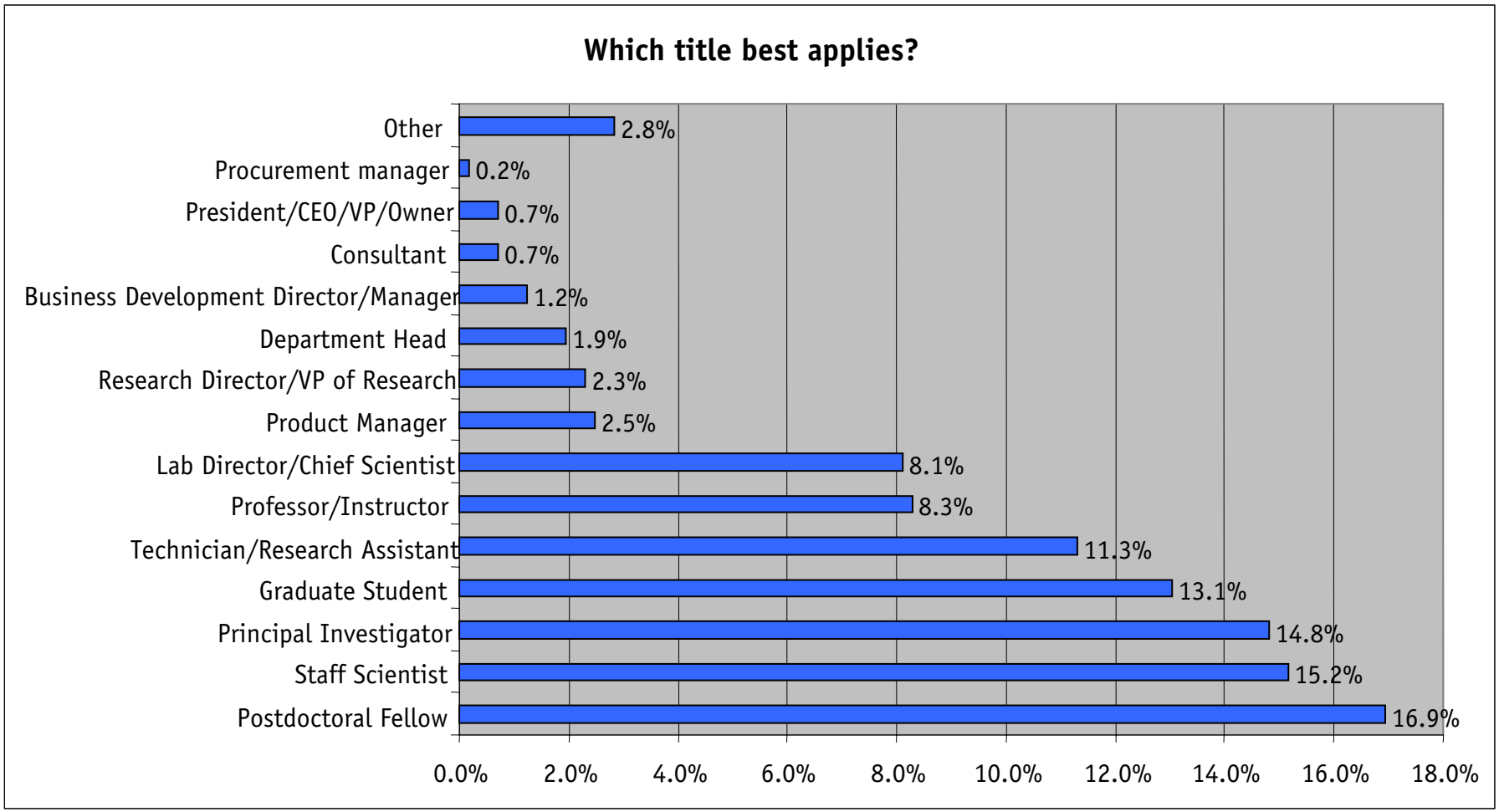
The majority of respondents worked in academic institutions while 17% were in either Biotech or Pharmaceutical companies and 10% worked in government labs.



n=567

*Only those respondents who are either currently using or planning to use real-time PCR are included in this chart.

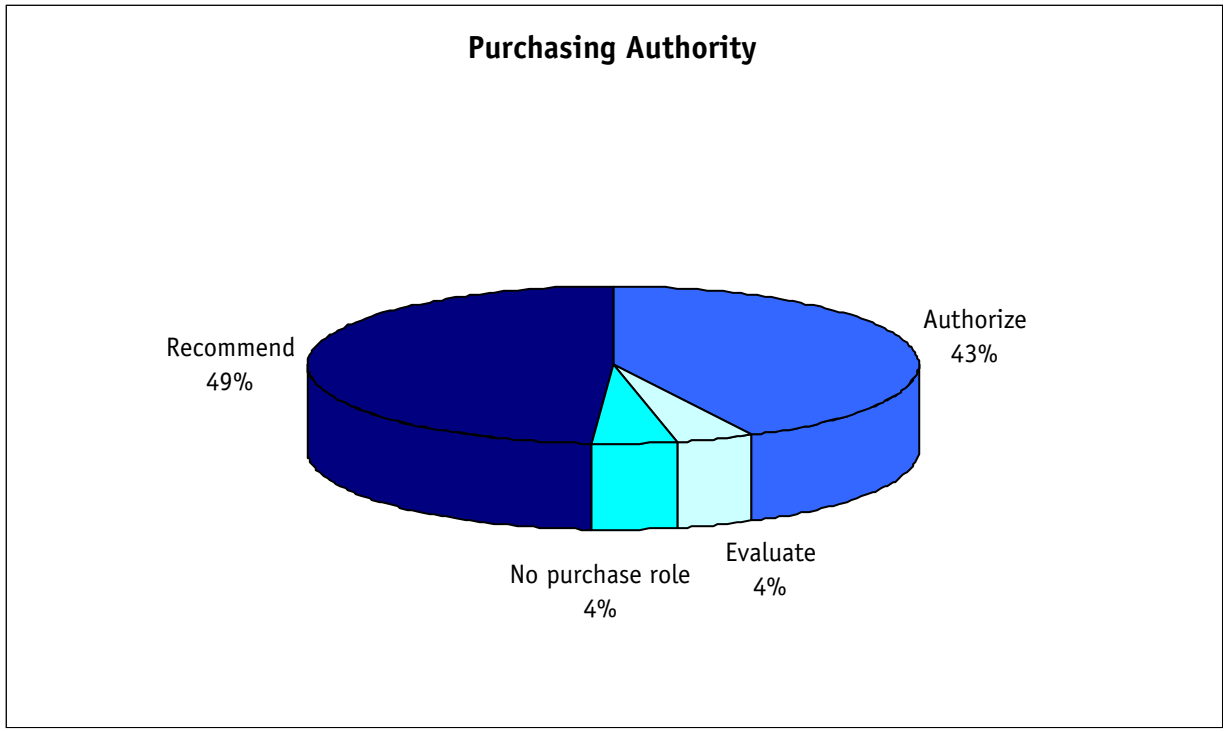
71.3% of respondents worked at the bench.



*Only those respondents who are either currently using or planning to use real-time PCR are included in this chart.

n=567

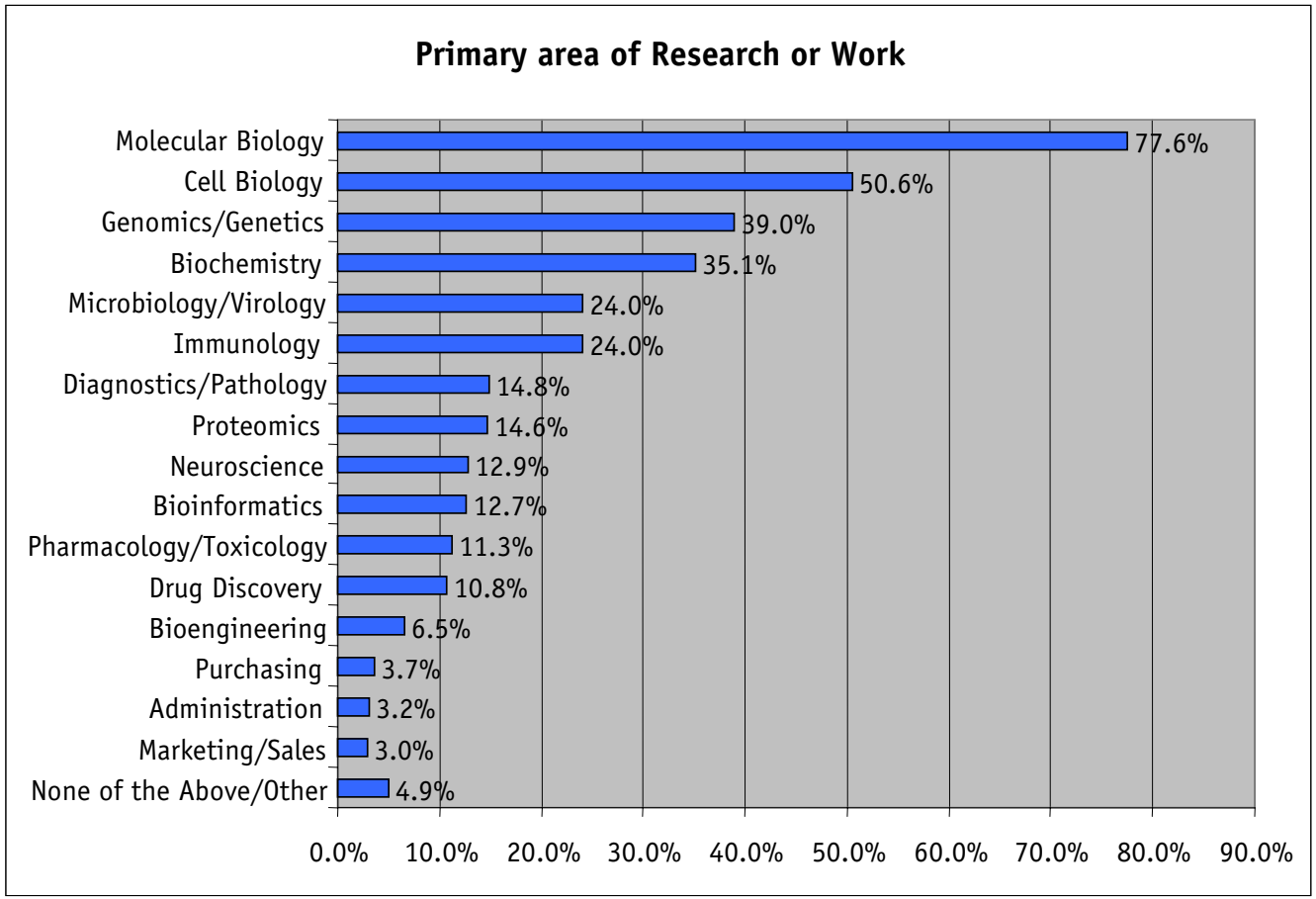
92% of respondents either authorized or recommended purchases.



n=567

*Only those respondents who are either currently using or planning to use real-time PCR are included in this chart.

Many respondents identified multiple areas of research. Molecular Biology, Cell Biology, Genomics/Genetics and Biochemistry were each identified as a primary area of work by greater than one-third of survey respondents.



*Only those respondents who are either currently using or planning to use real-time PCR are included in this chart. n=567

Do you currently perform or plan to perform real-time PCR?
Currently perform
Plan to perform
Neither currently perform nor plan to perform

What types of real-time assays do you perform?
5′ Exonuclease assay (ie. TaqMan probes)
DNA Dye Binding Assay (ie. SYBER Green)
Hybridization Assay (ie. Molecular Beacons, FRET probes/primers)
Other

Which of the following brands of real-time PCR kits/reagents do you use?
Invitrogen UltraSense™
BD Biosciences Clontech BD QZyme™
MJ Research DyNAmo™
Invitrogen ThermoScript™
Roche LightCycler
Epicentre MasterAmp™
Epicentre FailSafe™
QIAGEN® QuantiTect™
Applied Biosystems TaqMan®
Exiquon ProbeLibrary™
Stratagene Brilliant™
Other (please specify)

What format are you looking for when purchasing a PCR master mix?
Master mix for use with hybridization probes
Master mix with SYBR Green
I make my own PCR master mix
Other (please specify)

Do you own a real-time PCR thermocycler?
Yes
No

Which real-time PCR thermocycler do you own?
ABI Prism® 7000
MJ Research Chromo 4™
ABI Prism® 7900HT
ABI 7500
Stratagene Mx3000P™
Roche LightCycler® 2.0
Bio-Rad MyiQ
Roche LightCycler®
Cepheid Smart Cycler® TD
Idaho Technology R.A.P.I.D™
Bio-Rad iCycler iQ
Pyrosequencing Rotor-Gene 3000
ABI 7300
Stratagene Mx4000™
Cepheid Smart Cycler® II
MJ Research DNA Engine Opticon®
MJ Research DNA Engine Opticon® 2
Other (please specify)

Do you use the real-time PCR reagents sold by your instrument's manufacturer or those sold by another company?
I use kits and reagents sold by my instrument's manufacturer
I use kits and reagents sold by a company OTHER than my instrument's manufacturer
I use kits and reagents sold by BOTH my instrument's manufacturer and a company other than my instrument's manufacturer

What does the company you purchase real-time PCR reagents from offer that your instrument's manufacturer does not offer?
Lower prices
My instrument's manufacturer does not offer kits and reagents
Wider selection of novel kits
Faster delivery
Better technical support/customer service
Better quality reagents
Other (please specify)

Do you plan to purchase a real-time PCR thermocycler in the next 3-6 months?
Yes
No

Which real-time PCR thermocycler would you consider purchasing?
ABI Prism® 7000
MJ Research Chromo 4™
ABI Prism® 7900HT
ABI 7500
Stratagene Mx3000P™
Roche LightCycler® 2.0
Bio-Rad MyiQ
Roche LightCycler®
Cepheid Smart Cycler® TD
Idaho Technology R.A.P.I.D™
Bio-Rad iCycler iQ
Pyrosequencing Rotor-Gene 3000
ABI 7300
Stratagene Mx4000™
Cepheid Smart Cycler® II
MJ Research DNA Engine Opticon®
MJ Research DNA Engine Opticon® 2
Other (please specify)

Which features are important to you when choosing a thermocycler for real-time PCR?
Multiplexing
System Speed
Broad-range of detection
High throughput ability
Uniform temperature across the block/blocks
Online analysis capability
Random Access ability
Intuitive controls
Other (please specify)

What applications are you currently using or planning to use real-time PCR for?
Primary validation of gene expression
Pathogen detection
Cancer diagnostics
Microarray validation
Viral load titer
SNP analysis
Other

What type of template do you use?
Genomic DNA
cDNA
Plasmid DNA

Are you performing or planning to perform HIGH THROUGHPUT real-time PCR?
Yes, currently performing
Yes, planning to perform
No

Do you own a real-time PCR workstation?
Yes
No

Are you planning to purchase a real-time PCR workstation in the next 3-6 months?
Yes
No

References

1. Mackay IM. Real-time PCR in the microbiology laboratory. *Clinical Microbiology and Infection* 2004; 10:190 – 212.
2. Heid CA, Stevens J, Livak KJ, Williams PM. Real time quantitative PCR. *Genome Res* 1996; 6: 986-94
3. Gwynne P and Heebner G. Drug discovery and biotechnology trends – Biochips 1: Microarrays on the move. www.science-benchtop.org February 27, 2004.
4. Gwynne P and Heebner G. PCR and cloning: A technology for the 21st century. www.science-benchtop.org February 9, 2001.
5. Invitrogen launches new tool for functional genomics. www.bioexchange.com July 31, 2002.