PCRBIO **Rapid Extract PCR Kit**

- Column-free
- Convenient
- Fast

PCRBIO Rapid Extract PCR Kit combines rapid DNA extraction with fast, highly specific DNA amplification in a convenient, easy-to-use format. Eliminate the need for laborious and time-consuming DNA extraction methods with this simple, integrated extraction and amplification PCR kit powered by the latest advances in hot start polymerase technology.

Features

- Rapid, convenient, single-tube DNA extraction
- Produces high yield, PCR-ready DNA in 15 minutes
- Minimised contamination risks and sample loss
- Maximised PCR speed, yield and specificity
- Powered by PCRBIO HS Taq Mix Red for direct gel loading
- Ideal for complex templates
- Also available as a lysis-only kit for downstream PCR and qPCR reactions

Figure 1. Mouse tail DNA rapid extraction comparison

PCRBIO Rapid Extract PCR Kit was used to extract DNA from 3mg of mouse tail clipping following the standard 15 minute protocol. The extraction was repeated using equivalent extraction kits from alternative manufacturers. A serial three-fold dilution series was made from each supernatant. PCRBIO HS Taq Mix Red was used to amplify a 1kb fragment of mouse GAPDH gene from each dilution. Results were compared by agarose gel electrophoresis. Row 1 shows results from PCR Biosystems, row 2 from Kapa Biosystems, row 3 from Bioline, row 4 from Sigma and row 5 from Fermentas.

Applications

- Genotyping
- Transgene detection
- Knockout analysis
- Sequencing

Samples

- Mouse tail clip and ear punch
- Animal tissue
- Hair follicle
- Buccal swab
- Mammalian blood
- FFPE tissue

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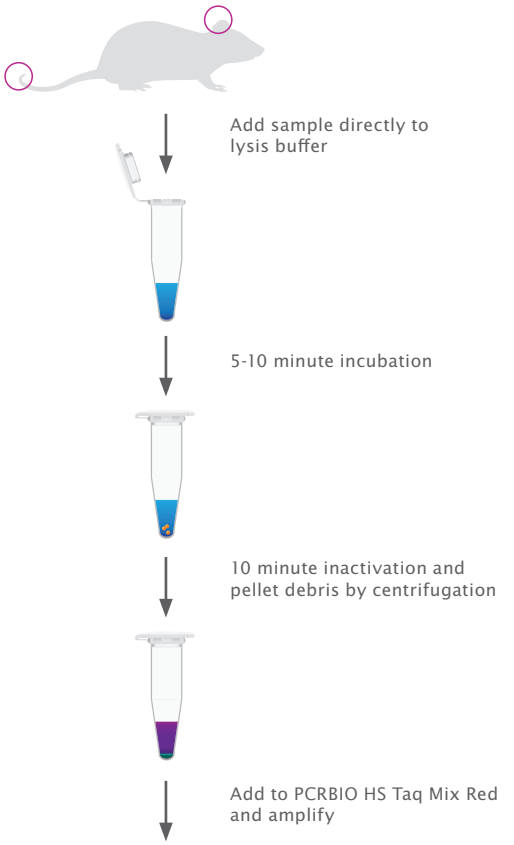

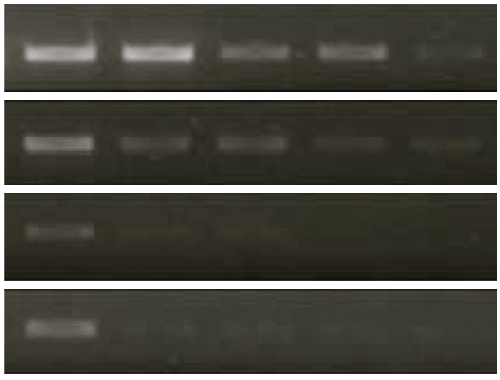

PCRBIO Rapid Extract PCR Kit has been developed for fast, efficient amplification of DNA from a variety of tissues and is particularly suited to solid tissue such as mouse tail or mouse ear. Sample processing is simplified and contamination risks minimised as DNA extraction is performed in a single tube, removing the need for multiple washing steps.

Extracted DNA is amplified in a proprietary buffer system using PCRBIO HS Taq Mix Red. Our antibody-mediated hot start polymerase uses the latest developments in polymerase technology and buffer chemistry to enhance PCR speed, yield and sensitivity. Primer dimer formation and non-specific amplification are prevented giving superior specificity ideal for complex templates such as mammalian genomic DNA. The mix contains a red dye suitable for direct gel loading without the need for additional loading buffer.

PCRBIO Rapid Extract Lysis Kit contains only the lysis and protease buffer system allowing the generation of PCR-ready DNA for use in downstream PCR or qPCR reactions.

Figure 2. Mouse ear DNA rapid extraction and 2.5kb amplification using supplied PCR reagent

PCRBIO Rapid Extract PCR Kit was used to extract DNA from 3mg of mouse ear clipping following the standard 15 minute protocol. The extraction was repeated using equivalent extraction kits from alternative manufacturers. A serial two fold dilution series was made from each supernatant. The supplied polymerase was used to amplify a 2.5kb fragment of the mouse Calnexin gene from each dilution. Results were compared by agarose gel electrophoresis. Row 1 shows results from PCR Biosystems, row 2 from Kapa Biosystems, row 3 from Bioline and row 4 from Sigma.

Catalogue Number	Product Name	Pack Size	Presentation
PB10.24-08	PCRBIO Rapid Extract PCR Kit	80 Reactions	PCRBIO HS Taq Mix Red 2 x 1ml, Buffer A 1 x 1.6ml, Buffer B 1 x 0.8ml
PB10.24-40	PCRBIO Rapid Extract PCR Kit	400 Reactions	PCRBIO HS Taq Mix Red 10 x 1ml, Buffer A 5 x 1.6ml, Buffer B 5 x 0.8ml
PB15.11-08	PCRBIO Rapid Extract Lysis Kit	80 Reactions	Buffer A 1 x 1.6ml, Buffer B 1 x 0.8ml
PB15.11-24	PCRBIO Rapid Extract Lysis Kit	240 Reactions	Buffer A 3 x 1.6ml, Buffer B 3 x 0.8ml

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qPCRBIO **Genotyping Mix**

- Allelic discrimination
- Reproducible
- Tight clustering

Features

- Accurate genotype calling
- Superior allele clustering
- Novel hot start for improved sensitivity
- Compatible on all real-time PCR platforms
- Standard and fast cycling conditions

Applications

- Genotyping single nucleotide polymorphisms (SNPs)
- TaqMan® allelic discrimination assays
- High throughput genotyping studies
- TaqMan® pre-designed SNP genotyping assays

qPCRBIO Genotyping Mix is a kit designed for use in TaqMan® and other dual-labelled probe based genotyping assays. PCR Biosystems provide a fast, accurate, reliable 2x mix for reproducible allelic discrimination.

qPCRBIO Genotyping Mix is able to accurately call class I to class IV mutations. The mix is compatible with LNA and PNA probes, which offer more stringent allele calling.

Proprietary small molecular inhibitor technology prevents formation of primer dimer and non-specific products leading to improved reaction sensitivity and specificity. Combining the latest advancements in polymerase technology and advanced buffer chemistry we offer market leading performance with minimal or no optimisation. High throughput screening has resulted in a buffer system that allows efficient amplification from GC rich and AT rich templates, under fast and standard cycling conditions.

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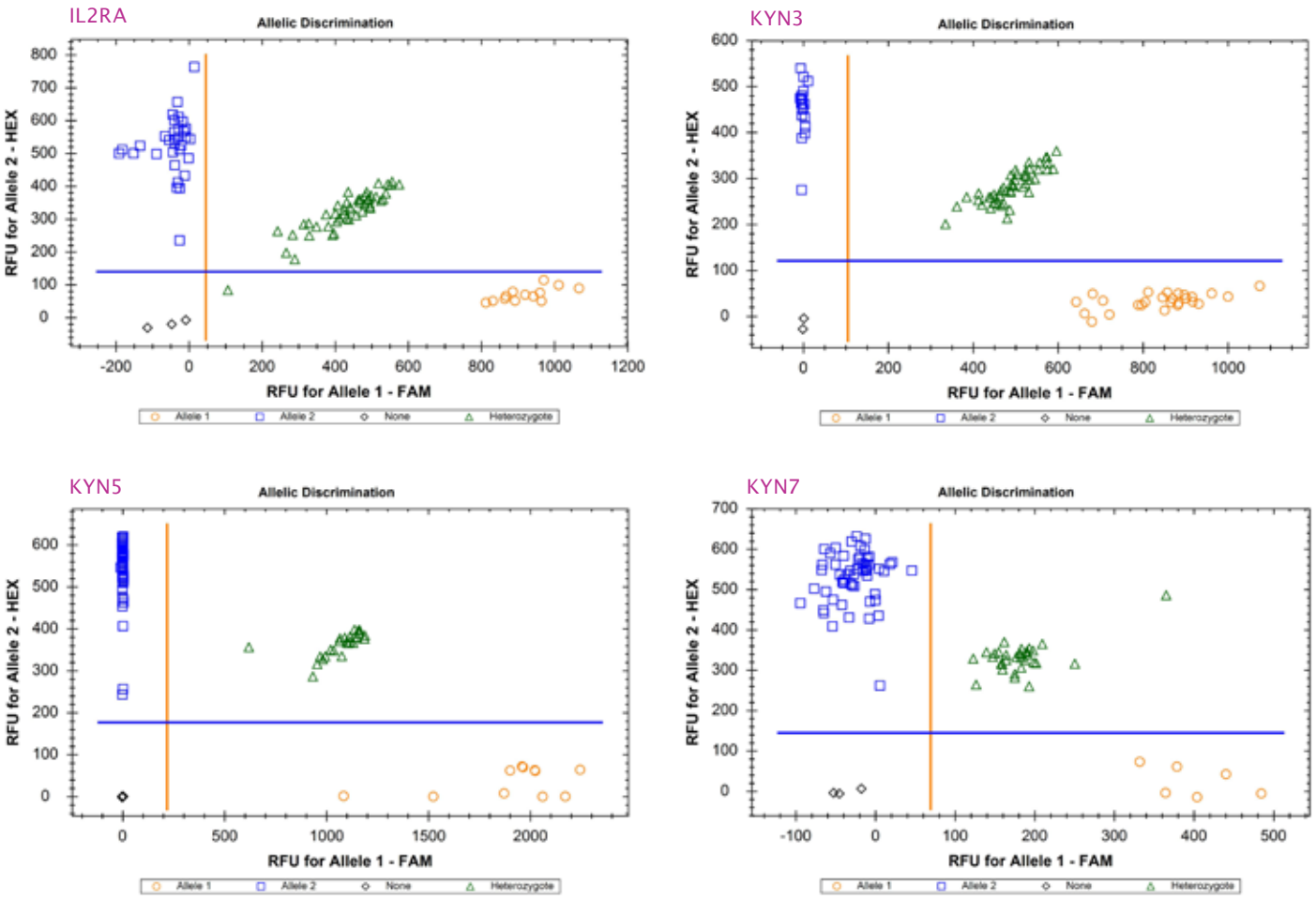

Figure 1.

Extracted genomic DNA from 96 human samples was analysed at 4 loci using TaqMan® probes and qPCRBIO Genotyping Mix. The analysis was performed on a Biorad CFX96 instrument and analysed using the Biorad CFX Manager software version 3.0. Single nucleotide polymorphisms (SNPs) within 4 genes were analysed: IL2RA (A/G), KYN3 (T/G), KYN5 (A/C), KYN7 (A/T). Cycling conditions for this assay were 50 cycles of 95ºC for 10 seconds and 56ºC for 50 seconds.

For each SNP analysed, high confidence, clear clustering of homozygous w/t, homozygous mutant and heterozygous was demonstrated. PCR Biosystems provide a fast, accurate, reliable 2x mix for reproducible allelic discrimination.

Catalogue Number	Product Name	Pack size	Presentation
PB20.43-01	qPCRBIO Genotyping Mix No-ROX	100 x 20µl rxns	1 x 1ml
PB20.43-05		500 x 20µl rxns	5 x 1ml
PB20.43-20		2000 x 20µl rxns	20 x 1ml
PB20.41-01	qPCRBIO Genotyping Mix Lo-ROX	100 x 20µl rxns	1 x 1ml
PB20.41-05		500 x 20µl rxns	5 x 1ml
PB20.41-20		2000 x 20µl rxns	20 x 1ml
PB20.42-01	qPCRBIO Genotyping Mix Hi-ROX	100 x 20µl rxns	1 x 1ml
PB20.42-05		500 x 20µl rxns	5 x 1ml
PB20.42-20		2000 x 20µl rxns	20 x 1ml

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qPCRBIO **SyGreen Mix**

- Sensitive
- Specific
- Fast

Features

- Non-PCR inhibiting intercalating dye, better signal
- Rapid extension rate for early Ct values
- Market leading sensitivity - increased limit of detection
- Compatible on all real-time PCR platforms - standard and fast cycling conditions
- Blue mix available for easy sample visualisation during pipetting

Applications

- Absolute quantification
- Relative gene expression analysis
- High throughput qPCR from genomic, cDNA and viral sequences
- Low copy number target genes

Further Applications

- Crude sample PCR
- Standard and fast PCR conditions
- Specific amplification from complex templates (eg GC/AT rich)
- Compatible with all real-time PCR instruments

PCR Biosystems use a proprietary intercalating dye that does not inhibit PCR, unlike other popular fluorescent dyes. Combined with advanced enzyme, hot start and reaction buffer technology we offer market-leading sensitivity and reproducibility.

qPCRBIO SyGreen Mix can be used to quantify any DNA template including genomic, cDNA and viral sequences. Extremely low copy number targets can be detected specifically and with high efficiency. Antibody-mediated hot start technology prevents the formation of primer dimers and non-specific products leading to improved reaction sensitivity and specificity. Combining the latest developments in polymerase technology and advanced buffer chemistry we offer market-leading performance with minimal or no optimisation.

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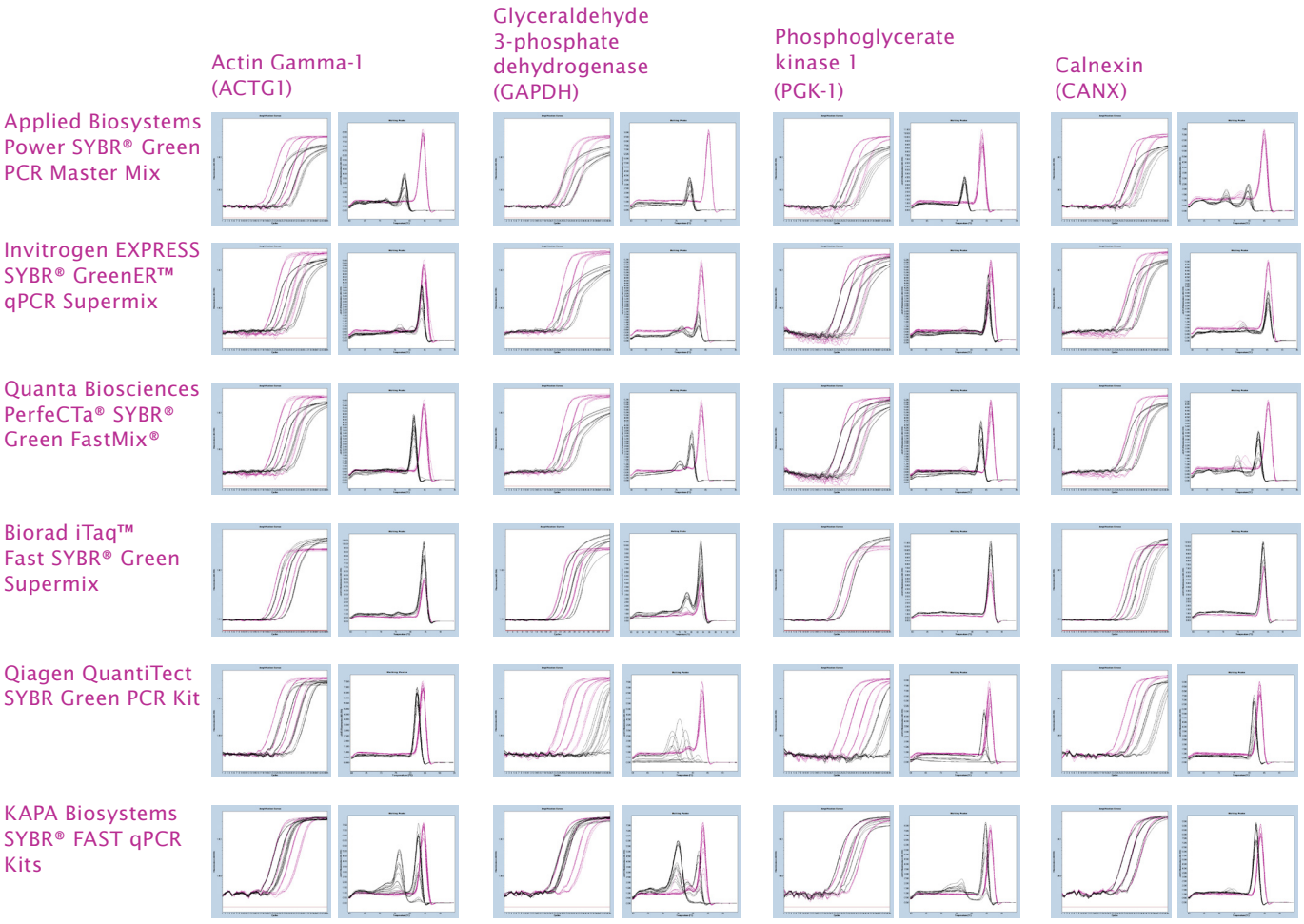

Black trace = Competitor Mix
Purple trace = qPCRBIO SyGreen Mix

Figure 1.

Shows amplification and melt traces of 4 mouse housekeeping genes from a cDNA dilution series. qPCRBIO SyGreen Mix traces (purple) and 6 competitor mixes (black). Cycling conditions were 95˚C 2min, 40 cycles of 95˚C 10sec, 60˚C 15sec on Roche LC480. For ACTG1 amplicon qPCRBIO mix was 2 to 4 Ct values earlier than 5 of 6 competitor mixes. The Ct was equal to that of Kapa Biosystems. The sensitivity of qPCRBIO mix was equal to 5 of 6 competitor mixes, but superior to Kapa Biosystems, demonstrated by absence of primer dimer at low template concentrations. For GAPDH amplicon qPCRBIO mix was 1 to 3 Ct values earlier for 4 of 6 competitor mixes and equal to 2 mixes. The sensitivity of qPCRBIO mix was superior to 4 of 5 competitor mixes, demonstrated by absence of primer dimer. Applied Biosystems mix showed equal sensitivity for this amplicon. For PGK amplicon, qPCRBIO mix had Ct values equal or lower than 5 of 6 competitor mixes. Sensitivity was equal to 4 mixes and superior to 2 mixes. For CANX amplicon, Ct values were 1 to 6 lower than 5 of 6 competitor mixes and equal to Kapa Biosystems mix. Sensitivity was superior to 3 of 6 mixes and equal to the other 3 mixes.

Overall, qPCRBIO SyGreen Mix outperformed each competitor mix on the 4 amplicons tested.

Catalogue Number	Product Name	Pack Size	Presentation
PB20.11-01	qPCRBIO SyGreen Mix Lo-ROX	100 x 20µl rxns	1 x 1ml
PB20.11-05		500 x 20µl rxns	5 x 1ml
PB20.11-20		2000 x 20µl rxns	20 x 1ml
PB20.12-01	qPCRBIO SyGreen Mix Hi-ROX	100 x 20µl rxns	1 x 1ml
PB20.12-05		500 x 20µl rxns	5 x 1ml
PB20.12-20		2000 x 20µl rxns	20 x 1ml
PB20.13-01	qPCRBIO SyGreen Mix with Fluorescein	100 x 20µl rxns	1 x 1ml
PB20.13-05		500 x 20µl rxns	5 x 1ml
PB20.13-20		2000 x 20µl rxns	20 x 1ml
PB20.14-01	qPCRBIO SyGreen Mix Separate-ROX	100 x 20µl rxns	[1 x 1ml mix] & [1 x 200µl ROX]
PB20.14-05		500 x 20µl rxns	[5 x 1ml mix] & [1 x 200µl ROX]
PB20.14-20		2000 x 20µl rxns	[20 x 1ml mix] & [4 x 200µl ROX]

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qPCRBIO **Probe Mix**

- High efficiency in multiplex reactions
- Rapid extension rate for early Ct values
- Market-leading sensitivity

Features

- High efficiency in multiplex reactions
- Rapid extension rate for early Ct values
- Market leading sensitivity - increased limit of detection
- Compatible on all real-time PCR platforms - standard and fast cycling conditions
- Efficient amplification from GC rich and AT rich templates
- Antibody-mediated hot start technology
- Blue mix available for easy sample visualisation during pipetting

Applications

- Absolute quantification
- Relative gene expression analysis
- TaqMan®, Scorpions® and molecular beacon probes
- Low copy number target genes
- Multiplex or singleplex
- Diagnostic real-time PCR

qPCRBIO Probe Mix is a universal probe kit designed for use in all probe-based real-time PCR assays. Whether your application is for a singleplex or multiplex expression study or a diagnostic assay, qPCRBIO Probe Mix is the robust choice for all your probe-based real-time PCR needs.

qPCRBIO Probe Mix can be used to quantify any DNA template including genomic, cDNA and viral sequences. Extremely low copy number targets can be detected specifically with high efficiency.

Combining the latest advancements in polymerase technology and advanced buffer chemistry we offer market-leading performance with minimal or no optimisation and high efficiency in multiplexed reactions.

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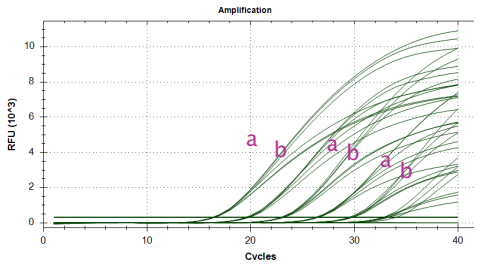
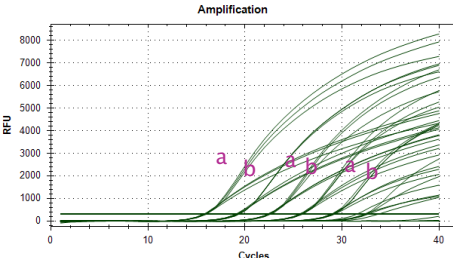
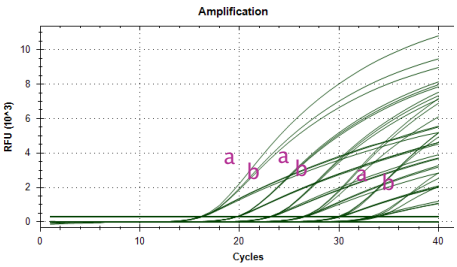
Experiment 1-qPCRBIO Probe Mix
a=Singleplex b=Quadplex



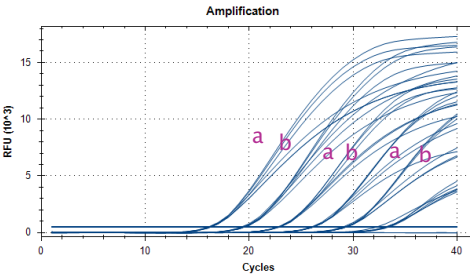
Experiment 2-Invitrogen EXPRESS
a=Singleplex b=Quadplex



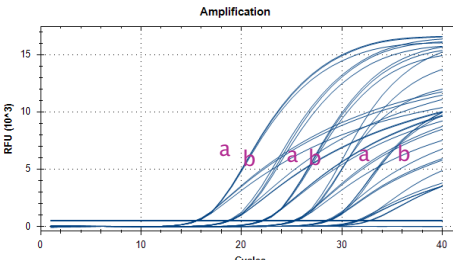
Experiment 3-Biorad Ssofast Probe Mix
a=Singleplex b=Quadplex



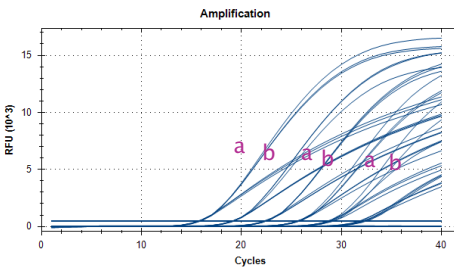
Experiment 4-qPCRBIO Probe Mix
a=Singleplex b=Quadplex



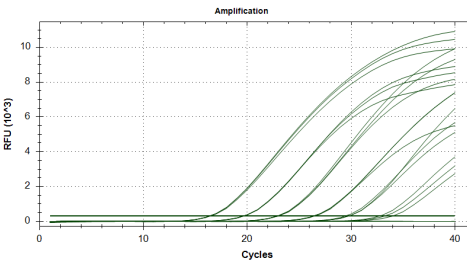
Experiment 5-Invitrogen EXPRESS
a=Singleplex b=Quadplex



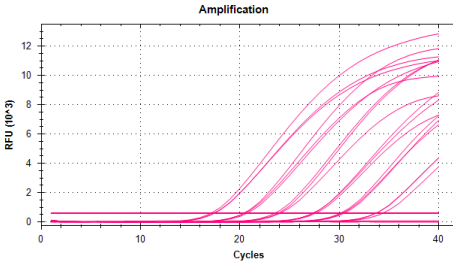
Experiment 6-Biorad Ssofast Probe Mix
a=Singleplex b=Quadplex



Experiment 7-qPCRBIO Probe Mix
Singleplex sensitivity test ACVR2B



Experiment 8-qPCRBIO Probe Mix
Singleplex sensitivity test LIMK1



Experiment 9-qPCRBIO Probe Mix
Singleplex sensitivity test ACVR1B

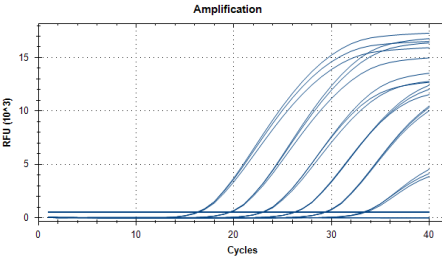


Figure 1.

Experiments 1, 2 and 3 show TaqMan probe amplification traces of human gene ACVR2B in singleplex and in quadplex (ACVR2B, LIMK1, ACVR1B and CDK7) from a cDNA dilution series. a) traces indicate singleplex reactions, b) traces indicate quadplex reactions. qPCRBIO Probe Mix was tested against the latest competitor mixes from Invitrogen (experiment 2) and Biorad (experiment 3). qPCRBIO Probe Mix shows the least PCR inhibition when in multiplex compared to Invitrogen and Biorad mixes. This is evident in more delayed amplification traces in quadplex (b) compared to singleplex (a). Experiments 4, 5 and 6 show TaqMan probe amplification traces of human gene LIMK1 in singleplex and quadplex (ACVR2B, LIMK1, ACVR1B and CDK7). As with experiments 1, 2 and 3 LIMK1 amplification is less inhibited in multiplex in the PCR Biosystems probe mix than the competitor mixes tested. Cycling conditions were 95˚C 2min, 40 cycles of 95˚C 10sec, 60˚C 15sec on Biorad CFX instrument.

Experiments 7, 8 and 9 show TaqMan probe amplification traces from plasmid dilution series of $1x10^6$ copies to 10 copies of DNA. For each gene qPCRBIO Probe Mix amplified with 100% efficiency and detected 10 copies of DNA.

Catalogue Number	Product Name	Pack Size	Presentation
PB20.21-01	qPCRBIO Probe Mix Lo-ROX	100 x 20µl rxns	1 x 1ml
PB20.21-05		500 x 20µl rxns	5 x 1ml
PB20.21-20		2000 x 20µl rxns	20 x 1ml
PB20.22-01	qPCRBIO Probe Mix Hi-ROX	100 x 20µl rxns	1 x 1ml
PB20.22-05		500 x 20µl rxns	5 x 1ml
PB20.22-20		2000 x 20µl rxns	20 x 1ml
PB20.23-01	qPCRBIO Probe Mix No-ROX	100 x 20µl rxns	1 x 1ml
PB20.23-05		500 x 20µl rxns	5 x 1ml
PB20.23-20		2000 x 20µl rxns	20 x 1ml
PB20.24-01	qPCRBIO Probe Mix Separate-ROX	100 x 20µl rxns	[1 x 1ml mix] & [1 x 200µl ROX]
PB20.24-05		500 x 20µl rxns	[5 x 1ml mix] & [1 x 200µl ROX]
PB20.24-20		2000 x 20µl rxns	[20 x 1ml mix] & [4 x 200µl ROX]

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