SZABO SCANDIC

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Produktinformation

- Forschungsprodukte & Biochemikalien
- Zellkultur & Verbrauchsmaterial
- Diagnostik & molekulare Diagnostik
- Laborgeräte & Service

Weitere Information auf den folgenden Seiten!
See the following pages for more information!

Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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Description

The WaveQuant™ 2x qPCR Master Mix is designed to provide a high-quality and reliable real-time qPCR (quantitative polymerase chain reaction)-based platform for sensitive detection of gene expression. It comes conveniently pre-mixed, allowing for a quick experimental setup, and optimized for use with double-stranded DNA. This mix contains Taq polymerase, Mg^{2+}, and the fluorescent dye Thiazole green, which is structurally identical to SYBR® Green I. The dye can bind multiple times to a single DNA strand to enhance sensitivity.

This product has been validated using two housekeeping genes as controls.

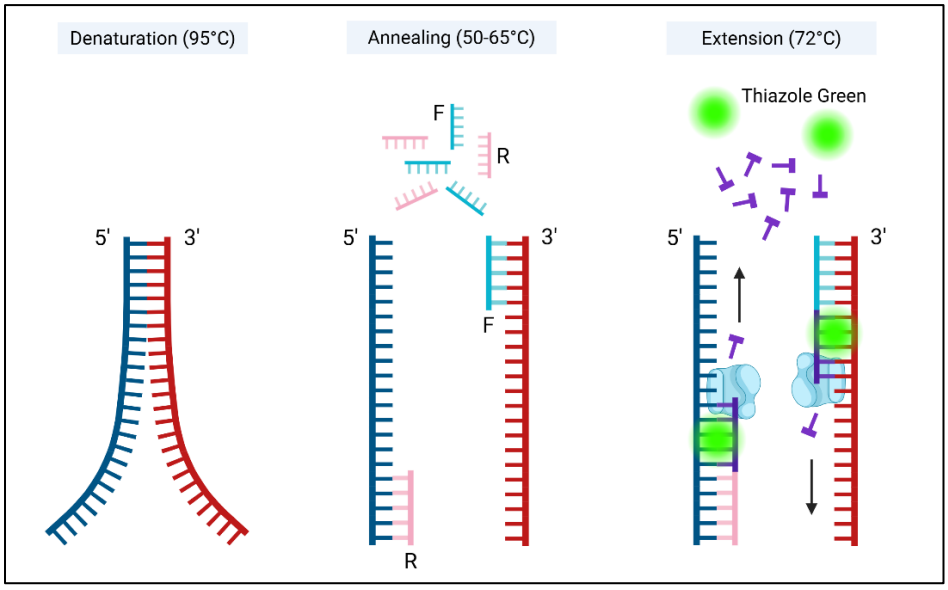

Figure 1. Principle of dye-based qPCR.
Thiazole Green binds to newly formed dsDNA during the qPCR cycles, allowing for real-time monitoring and quantification.

Background

Quantitative PCR (qPCR), also known as real-time PCR, monitors the amplification of a DNA fragment throughout the reaction using a fluorescent dye. It is a tool with high sensitivity, fast turnaround time, and minimal risk of contamination. Commonly used dyes include Thiazole green, which binds to the minor groove of double-stranded DNA (dsDNA) and fluoresces over 1000-fold. This fluorescence allows to quantify the amount of product during any given cycle. The baseline threshold, which is usually set above background fluorescent noise levels, is used to calculate the cycle threshold (Ct or Cq value) and quantitatively evaluate the results. The Ct/Cq values can be used to determine the starting amounts of DNA using a standard curve, and the melting curves can help assess quality of the amplified product. qPCR is a standard technique in gene expression analysis, mRNA analysis, copy number variation (CNV) analysis, and pathogen detection, among others. Its value in medicine and biology makes it crucial in any research workflow**.**

Applications

It can be used for quantification of DNA.

Supplied Materials

Catalog #	Name	Amount	Storage
82942	WaveQuant™ 2x qPCR Master Mix	2.5 ml	-20°C

Materials Required but Not Supplied

- CFX Connect Real-Time PCR Detection System from Bio-Rad Laboratories (no probes required) or similar system
- 0.2ml qPCR tubes or 96 well qPCR plate
- Adjustable micropipette and sterile tips
- Microcentrifuge for qPCR tubes

Storage Conditions

This mix will perform optimally for up to **6 months** from date of receipt when the materials are stored as directed.

Safety

This product is for research purposes only and not for human or therapeutic use. This product should be considered hazardous and is harmful by inhalation, in contact with skin, eyes, clothing, and if swallowed. If contact occurs, wash thoroughly.

qPCR Protocol

- We recommend setting up all reactions on ice and running them in duplicate.
- To generate a standard curve, use a dsDNA reference standard and a specific primer pair with an amplicon size of 70-200 bp.
- Limit light exposure of the premix by keeping it in its dark tube and covered when not in use.
- Set up your reactions and a no-template control in qPCR tubes using the following volumes:

Component	**Test**	**No-Template Control**
Template (50 ng)	2 µl	-
25 µM 5' Primer	2 µl	2 µl
25 µM 3' Primer	2 µl	2 µl
WaveQuant™ 2X qPCR Master Mix	10 µl	10 µl
Distilled water	4 µl	6 µl
Total	**20 µl**	**20 µl**

- We recommend using 20 µl as the total volume, but the qPCR mix has been validated for use down to a final volume of 10 µl (keeping all component ratios the same).

- After samples are prepared, run qPCR reactions using the following parameters:

Step	Temperature	Duration	Cycles
Initial Denaturation	94°C	3 min	1x
Denaturation	94°C	45 sec	40x
Annealing	50-65°C	45 sec	
Extension	72°C	5 min	
Final Extension	72°C	7 min	1x

Note: These parameters have been optimized for use with CFX Connect Real-Time PCR Detection System from Bio-Rad. Further optimization may be required for other applications, instruments, and primer sets.

- Set your baseline threshold to a constant value above the background noise or use the auto-baseline feature to obtain Ct values.

Validation

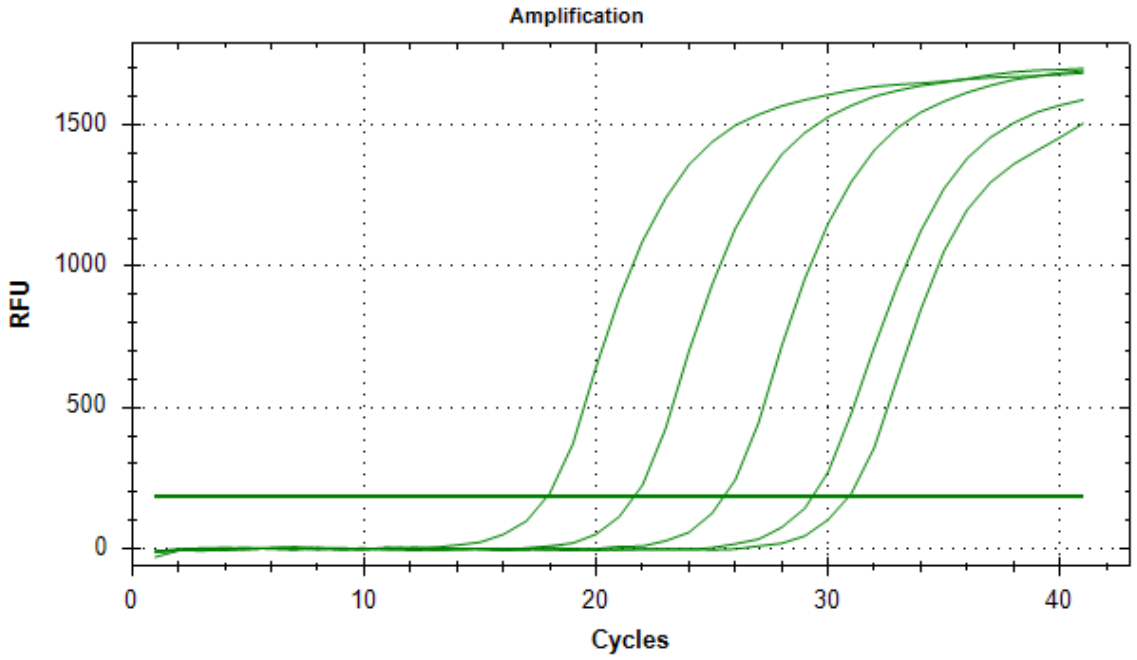

Figure 2. Human cDNA amplification curves.
Human cDNA (Takara #639653) was amplified with GAPDH primers. Primer sequences used: Fw: 5' AGTATGACAACAGCCTCAAG and Rv: 5' TCATGAGTCCTTCCACGATA.
WaveQuant™ 2x qPCR Master Mix was validated in the 10 ng to 1 pg range.

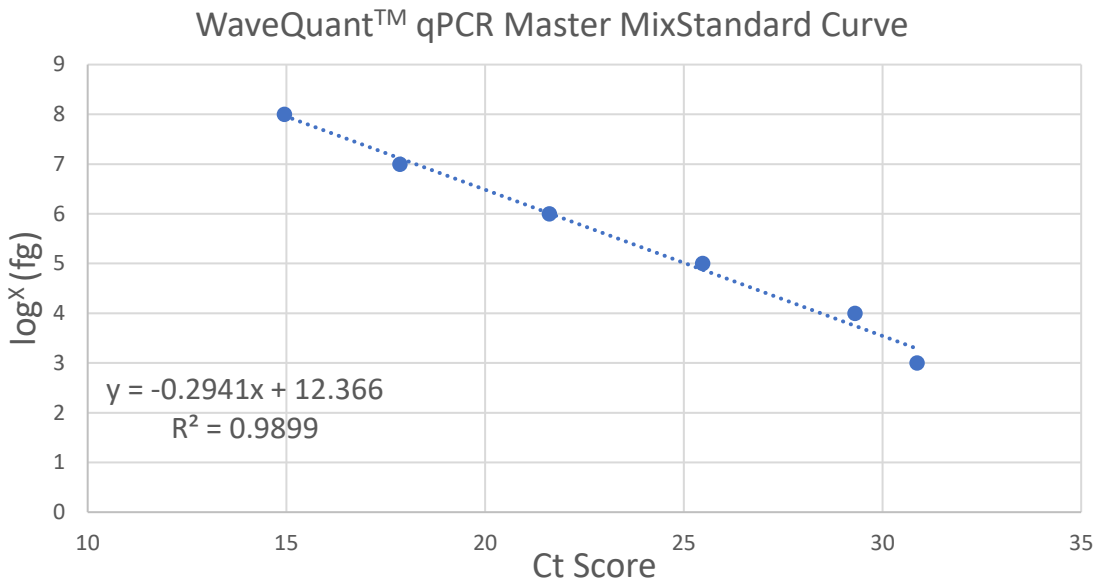

Figure 3. Human cDNA standard curve using WaveQuant™ qPCR Master Mix.
A standard curve for human cDNA was generated using the Ct values from the serial dilution. The curve was generated in the 10 ng-1 pg range (or 9.04 x 10^9 to 9.04 x 10^6 copy numbers).

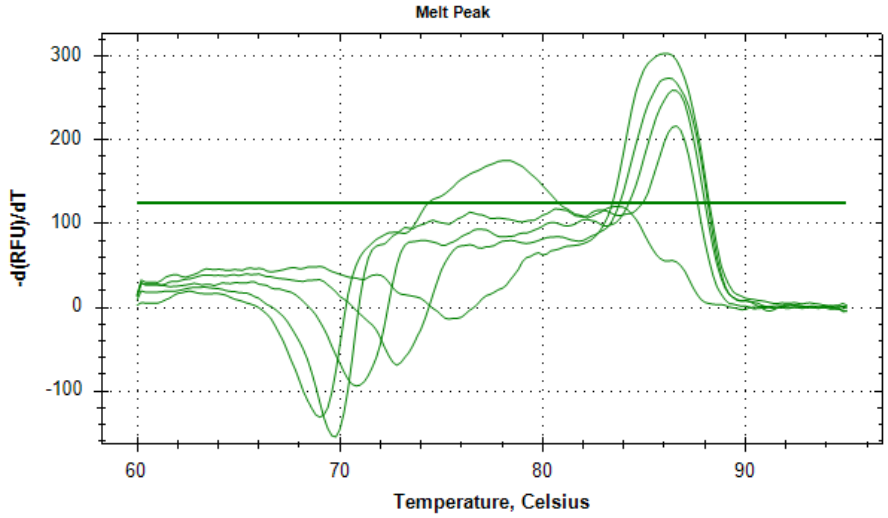

Figure 4. Human cDNA melting curves.
A serial dilution of human cDNA was amplified with GAPDH primers and melting curves were generated (10 ng to 1 pg).

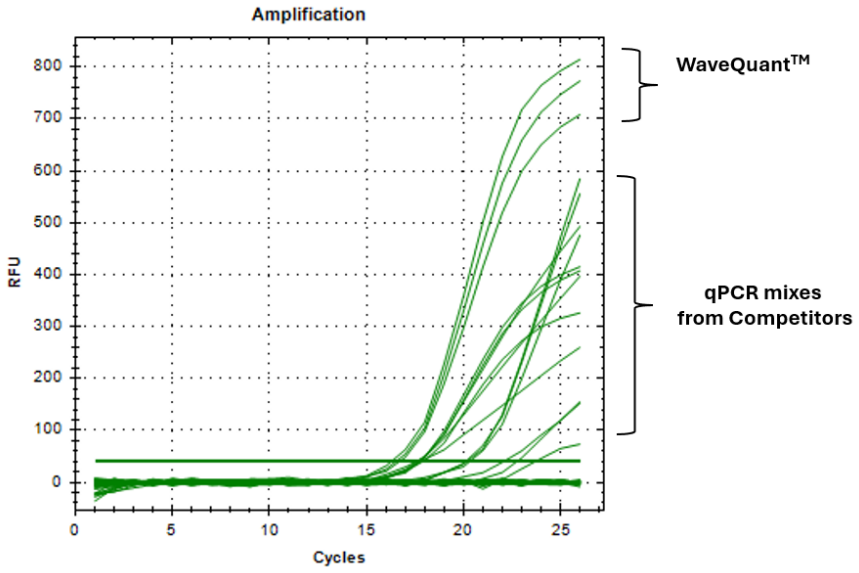

Figure 5. WaveQuant™ qPCR Mix has higher sensitivity versus other competitor qPCR mixes. WaveQuant™ qPCR Mix and other competitors were used with human cDNA.

Data shown is representative.

Troubleshooting Guide

Visit bpsbioscience.com/faqs for detailed troubleshooting instructions. For lot-specific information and all other questions, please email visit https://bpsbioscience.com/contact.

Related Products

Products	*Catalog #*	*Size*
AAV qPCR Titration Kit	82812	1 Kit
Quick PCR™ Plus Assembly Kit	78531	10 reactions/ 50 reactions

Version 060625