Catalog #	Aliquot Size
TQ03-E011-500	500 reactions

Taq Pro Universal SYBR qPCR Master Mix

Catalog # TQ03-E011
Lot # 1L2208-7

Product Description

This product is a special premix for qPCR using the SYBR Green I chimeric fluorescence method. This product contains a 2X master mix to which primers and template can be added.

Formulation

This master mix contains dNTP, Mg^{2+}, Taq Pro DNA Polymerase, SYBR Green I, Specific ROX Reference Dye, and an optimized buffer in a proprietary formulation.

Storage and Stability

For longer term storage, keep at -30°C to -15°C protected from light. After thawing, product can be stored stably at 2°C to 8°C for 6 months if protected from light. To avoid repeated handling and multiple freeze/thaw cycles aliquot product into smaller quantities.

Scientific Background

The core component of Taq Pro Universal SYBR qPCR Master Mix, Taq Pro DNA Polymerase, has many advantages such as strong specificity, high detection sensitivity and high amplification yield compared to previous generations of polymerases. Together with qPCR optimized buffer and specific enhancer, it is very suitable for qPCR reactions with high specificity and sensitivity. This kit contains Specific ROX Reference Dye, which is suitable for all qPCR instruments without adjusting the concentration of ROX on different instruments.

This product is manufactured in an ISO 9001 and ISO 13485 certified facility.

Taq Pro Universal SYBR qPCR Master Mix

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Stability	Once thawed, 6 months if stored at 2°C to 8 °C and protected from light.
Storage & Shipping	Store at -30 to -15°C and protect from light. Transport at ≤0 °C. To avoid repeated handling and multiple freeze/thaw cycles aliquot product into smaller quantities.

To place your order, please contact us by phone 1-778-326-0223 or 1-888-606-3424 (Toll free)
or by email: orders@signalchemdx.com or info@signalchemdx.com - www.signalchemdx.com

qPCR Protocol

Notes:

1. If white precipitate is found in the Master Mix after thawing, please place it at room temperature for a short while and invert it upside down several times to dissolve the precipitate before use.
2. Avoid repeated freezing and thawing, so as not to cause a decrease of enzyme activity. If the amount of each use is small, it is recommended to aliquot the Master Mix into smaller portions.
3. Please invert the Master Mix upside down to mix thoroughly. Do not vortex to avoid air bubbles, which will affect the quantitative results. The Master Mix is ready to use after mixing and centrifuging briefly. Mix gently by pipetting. If air bubbles are generated, please centrifuge again before use.
4. As this product contains the fluorescent dye SYBR Green I, it should be stored protected from light. Avoid strong light when preparing the reaction solution.
5. Because detection sensitivity of this product is very high, aerosols in the air can easily cause contamination. Therefore, the preparation of the reaction system should be carried out on a clean bench. Sterile tips and reaction tubes should be used in the preparation process. If laboratory conditions permit, special pipettes and tips with filtering elements are recommended.

Protocol:

1. Prepare the following mixture in a qPCR tube

Component	Volume
2X Taq Pro Universal SYBR qPCR Master Mix	10.0 µl
Primer 1 (10 µM)	0.4 µl
Primer 2 (10 µM)	0.4 µl
Template DNA/cDNA	x µl
ddH_2O	To 20.0 µl

The amount of each component in the reaction system can be adjusted according to the following principles:

- In general, a better amplification effect can be obtained when the final concentration of primers is 0.2 µM. When the reaction performance is poor, the primer concentration can be adjusted in the range of 0.1 - 1.0 µM.
- Due to the high sensitivity of qPCR, the accuracy of the amount of template added to the reaction system will greatly affect the final quantitative results. It is recommended to dilute the template and add it to the reaction system, which can effectively improve reproducibility of the experiment.
- The volume of undiluted cDNA template should not exceed 1/10 of the total volume of the qPCR reaction.
- If white precipitate is found in the Master Mix after thawing, place it at room temperature for a short while and invert it upside down to dissolve the precipitate before use.

2. Perform the qPCR reaction according to the following conditions

Stage 1	Initial Denaturation[a]	Rep:1	95 °C	30 secs
Stage 2	Cycling Reaction	Reps:40	95 °C 60 °C	3-10 sec[b] 10-30 sec[c]
Stage 3	Melting Curve[d]	Rep:1	95 °C 60 °C 95 °C	15 sec 60 sec 15 sec

a. The initial denaturation condition is suitable for most amplification reactions. If the template structure is complex, the initial denaturation time can be extended to 3 min to improve the initial denaturation effect.
b. For a standard program, select 10 sec. For a fast program select 3 sec.
c. For a standard program, select 30 sec. For a fast program: if amplicons are within 200 bp, the shortest extension time can be set to 10 sec; if amplicons are over 200 bp, the recommended extension time is 30 sec.
d. The melting curve acquisition procedures of different qPCR instrument types are not the same, and please select the instrument default melting curve acquisition procedures.

FAQ & Troubleshooting

Abnormal shape of amplification plot

- Rough amplification plot: The signal is too weak and generated after system correction. Increase template concentration and retry.
- Broken or downward amplification plot: The template concentration is too high and the baseline endpoint value is greater than C_T value. Reduce the baseline endpoint (CT value - 4) and repeat data analysis.
- Amplification plot goes downward suddenly: There are bubbles remaining in the reaction tube. Pay attention to centrifugation when processing samples and carefully check the reaction tube for any remaining bubbles before performing reaction.

No amplification plot

- Insufficient number of reaction cycles: In general, the number of cycles is set to 40, but it should be noted that too many cycles will increase background signals and reduce the reliability of the data.
- Confirm whether the signal acquisition step is set up in the program: The two-step amplification program generally sets the signal acquisition at the annealing and extension stage, while the three-step amplification program should set the signal acquisition at the 72℃ extension stage.
- Confirm whether the primers are degraded: Primers that have not been used for a long time should be tested for integrity using PAGE electrophoresis before use to rule out the possibility of degradation.
- Low template concentration: Reduce the dilution factor and repeat the test. In general, samples with unknown concentration should be started at the highest concentration.
- Template degradation: Prepare new template and retry.

C_T value appears too late

- Low amplification efficiency: Optimize the PCR system, then try the three-step amplification program or redesign and synthesize primers.
- Low template concentration: Reduce the dilution factor and repeat the test. In general, samples with unknown concentration should be started at the highest concentration.
- Template degradation: Prepare new template and retry.
- Long PCR products: The recommended length of PCR products is 80 - 150 bp.
- PCR inhibitors in the system: They are usually introduced along with the template. Increase the dilution factor or prepare new template and retry.

Amplification observed in negative control

- Contaminated reaction system: Replace with new mix, ddH2O and primers and repeat the experiment. The reaction system should be prepared on a clean bench to reduce aerosol contamination.
- Primer dimer: Carry out analysis in association with the melting curve.

The linear relationship of standard curve is not satisfactory in absolute quantification

- Deviations of pipetting volume: Increase the dilution factor of template and increase the pipetting volume accordingly.
- Degradation of standards: Prepare new standards and retry.
- High template concentration: Increase the dilution factor.

Multiple peaks in the melting curve

- Inappropriate primer design: Design and synthesize new primers according to the primer design principles.
- High primer concentration: Decrease the primer concentration.
- cDNA template with contamination of genomic DNA: Prepare new cDNA template.

Poor experiment repeatability

- Inaccurate pipetting volume: Use higher performance pipette; increase the template dilution factor, and increase the sample loading volume.
- Differences in temperature control between wells in qPCR instrument: Calibrate the instrument regularly.
- Low template concentration: The lower the template concentration, the worse the repeatability. Reduce the template dilution factor or increase the volume of sample added.

SAFETY DATA SHEET

Article 1 - Product Identification

Product Name: Taq Pro Universal SYBR qPCR Master Mix **Catalog # TQ03-E011**

This product is sold only for research use by qualified laboratory personnel, and is not to be used as a drug, medical device, food additive, cosmetic, nor household chemical. It is not to be used in diagnostic, therapeutic, consumer, agricultural, nor pesticidal applications.

Supplier of Datasheet:	SignalChem Diagnostics Inc.
Street Address:	*190-13160 Vanier Place*
City, Prov. Postal Code:	*Richmond, BC, V6V 2J2*
Country:	*Canada*
Emergency Phone:	1-888-606-3424 (Toll free) 1-778-326-0223 (local)

Article 2 - Hazard Identification

- **WHMIS Classification:** Not WHMIS controlled.
- **GHS classification:** Not GHS classified.
- **Hazard Pictograms:** No labeling applicable.
- **Signal words:** None.
- **Hazard statements:** None.
- **Precautionary statements:** Wear protective gloves/protective clothing/eye protection/ face protection. Avoid breathing dust. IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
- **Other hazards:** May cause eye and skin irritation. May cause respiratory and digestive tract irritation.

Article 3 – Composition/Information on Ingredients

Chemical Characterization: Mixture.
Description: The components of this product which may be hazardous are listed below.

Common name	Chemical name	CAS-No.	Concentration
Glycerol	Glycerol	56-81-5	≤ 50%

Article 4 – First-aid Measures

- **General information:** Consult a physician by providing the SDS.
- **After inhalation:** In case of irritation by inhaling this product, move affected person to fresh air and await recovery. If irritation persists, seek immediate medical attention. If casualty cannot breathe, give artificial respiration and seek immediate medical attention.
- **After skin contact:** Immediately wash with soap and plenty of water and rinse thoroughly. Consult a physician.
- **After eye contact:** Rinse opened eyes with plenty of water for at least 15 minutes. Remove contact lenses, if present and easy to do so. Consult a physician.
- **After swallowing:** Not expected to present a significant ingestion hazard under anticipated conditions of normal use. If you feel unwell, seek medical advice.

Article 5 - Fire-fighting Measures

- **Suitable extinguishing media:** Use water spray, extinguishing powder, carbon dioxide, or other appropriate measure that is suitable to the environment.
- **Specific hazards arising from the substance or mixture:** None known.
- **Special protective equipment and precautions for fire-fighters:** Self-contained breathing apparatus if necessary.

SAFETY DATA SHEET

Article 6 – Accidental Release Measures

- **Personal precautions, protective equipment, and emergency procedures:** Apply standard laboratory practices and personal protective equipment. Avoid breathing vapors, mist, or gas. Ensure adequate ventilation.
- **Environmental precautions:** Do not allow to enter drains.
- **Methods and materials for containment and cleaning up:** Absorb on sand or vermiculite and place in closed containers for disposal.

Article 7 - Handling and Storage

- **Precautions for safe handling:** Wear chemical safety goggles and compatible chemical-resistant gloves. Avoid inhalation, contact with eyes, skin or clothing.
- **Conditions for safe storage:** Store according to product label instructions. Keep container upright and tightly closed.

Article 8 - Exposure Controls/Personal Protection

- **Components with limit monitoring values at workplace:**
  Glycerol (CAS-No: 56-81-5)

Values	Control parameters	Regulations
TWA	10 mg/m³ for mist	British Columbia, Canada
TWA	3 mg/m³ for respirable mist	British Columbia, Canada
TWA	10 mg/m³	Alberta, Canada
TWAEV	10 mg/m³	Ontario, Canada
TWAEV	10 mg/m³	Quebec, Canada
TWA	10 mg/m³	USA

- **Appropriate engineering controls:**
  Apply adequate ventilation including mechanical exhaust or laboratory fume hood. Follow standard laboratory practices.
- **Individual protection measures:**

Respiratory protection:
Use appropriate respirator if there is inadequate ventilation by following the government standards.

Hand protection:
Wear gloves and use proper glove removal technique to avoid skin contact. Discard gloves after use by following the applicable laboratory regulations. Wash and dry hands.

Eye/face protection:
Safety goggles with side-shields approved under appropriate government standards.

Skin/body protection:
Use appropriate clothing, footwear and any additional protection measures to protect from splashing or contamination.

Article 9 – Physical and Chemical Properties

Appearance: Colorless fluid.	**Danger of explosion:** Product does not present an explosion hazard.
Odour/Odour Threshold: Not determined.	**Explosion limits:** Lower: Not determined
pH: ~8.2	**Decomposition temperature:** Not available.
Melting point/freezing point: Not determined.	**Vapor pressure at 20 °C:** Not determined
Boiling point/Boiling range: 106 °C.	**Density:** ~1.10g/cm³.
Flash point: Not determined.	**Relative density:** Not determined.
Flammability (solid, gaseous): Not determined.	**Vapor density:** Not determined.
Ignition temperature: Not determined.	**Evaporation rate:** Not determined.
Auto-igniting: Product is not self-igniting.	**Solubility in / Miscibility with Water:** Fully miscible.

Article 10 - Stability and Reactivity

- **Reactivity:** Stable under recommended transport and storage conditions.
- **Chemical stability:** Stable under recommended transport and storage conditions.
- **Possible hazardous reactions:** No dangerous reactions known.
- **Conditions to avoid:** Heat and moisture.
- **Incompatible materials:** Not determined.
- **Hazardous decomposition products:** Not determined.

SAFETY DATA SHEET

Article 11 - Toxicological Information

- **Acute toxicity:** Not available.
- **LD/LC50:** Not available.
- **Skin corrosion/irritation:** Not available.
- **Serious eye damage/eye irritation:** Not available.
- **Respiratory or skin sensitization:** Not available.
- **Germ cell mutagenicity:** Not available.
- **Carcinogenicity:** No components are listed in IARC, or NTP, or OSHA, or ACGIH.
- **Reproductive toxicity:** Not available.
- **Teratogenicity:** Not available.
- **Specific target organ toxicity - single exposure/ - repeated exposure (GHS):** Not available.
- **Aspiration hazard:** Not available.
- **Potential health effects:**
  Inhalation: No data available
  Ingestion: No data available
  Skin: No data available
  Eyes: No data available
- **Signs and Symptoms of Exposure:** No data available
- **Synergistic effects:** Not available.

Article 12 - Ecological Information

- **Eco-toxicity:** No data available.
- **Biodegradability:** Not applicable.
- **Bio-accumulative potential:** Not applicable.
- **Mobility in soil:** Not applicable.
- **PBT and vPvB assessment:** Not applicable.
- **Other adverse effects:** Not applicable.

Article 13 - Disposal Considerations

- **Disposal methods:** In accordance to applicable national, regional, or local laws and regulations. For additional handling information and protection of employees please refer to Article 7 and 8.
- **Contaminated packaging:** Disposal should be made in accordance to official regulations. Use water or cleansing agents to clean the area.

Article 14 - Transport Information

- **DOT:** Not dangerous goods.
- **IMDG:** Not dangerous goods.
- **IATA:** Not dangerous goods.

Article 15 – Regulatory Information

- **WHMIS Classification:** Non-hazardous.
- **GHS label elements:** Not applicable.
- **Signal word:** Not applicable.
- **Hazard statements:** Not applicable.

Article 16 - Other Information

The above information is believed to be correct but does not purport to be all-inclusive and shall be used only as a guide. SignalChem shall not be held liable for any damage resulting from handling or from contact with the above product. See the Technical Specification, Packing Slip, Invoice, and Product Catalog for additional terms and conditions of sale.