



REAGENTS FOR RESULTS

## MyGo SNP Probe Kit No ROX

Cat. No. 6489 | 3605 | 2585

Pack Size	Format	Presentation
100 x 20µl rxns (6489)	2x Ready Mix	1 x 1ml
500 x 20µl rxns (3605)	2x Ready Mix	5 x 1ml
2000 x 20µl rxns (2585)	2x Ready Mix	20 x 1ml

This product is for research use only

### 1. STORAGE

Store all components at -20°C with minimal exposure to light. If stored correctly the kit will retain full activity for 12 months. The kit may be stored at 4°C for short term use (1 month). The kit can go through up to 30 freeze/thaw cycles with no reduction in performance.

### 2. TECHNICAL ASSISTANCE

If you have any questions, or experience any difficulties with MyGo SNP Probe Kit, please email [reagentsupport@mygopcr.com](mailto:reagentsupport@mygopcr.com), providing full details including amplicon size, reaction setup, cycling conditions and screen shots of amplification traces and melt profiles.

### 3. DESCRIPTION

MyGo SNP Probe Kit is designed for fast, accurate and reproducible allelic discrimination for use in TaqMan® and other dual-labelled probe-based genotyping assays. The 2x mix combines MyGo HS Taq DNA Polymerase, dNTPs, and MgCl<sub>2</sub> in an optimised buffer to allow clear allele calling and tight fluorescence clusters.

MyGo SNP Probe Kit uses antibody-mediated hot start to give fast, highly specific PCR. The enzyme is inactive at ambient temperatures, preventing the formation of primer-dimers and mis-primed products with the convenience of room temperature setup. The enzyme is activated at the start of a reaction with a 95°C incubation step. The enhanced efficiency and specificity of MyGo SNP Probe Kit make it the ideal choice for difficult SNP loci or low amounts of starting template.

The kit is fully compatible with ABI TaqMan® pre-designed SNP Genotyping Assays. MyGo SNP Probe Kit No ROX is designed for use with real-time PCR instruments that do not require a passive reference.



## 4. IMPORTANT NOTES

### 4.1 Instrument compatibility

Some real-time PCR instruments require the use of ROX as a fluorescent passive reference to correct for optical artefacts. Generally, modern instruments do not require the use of passive fluorescent references. The MyGo instruments do not require the use of ROX as a passive reference. This kit contains ROX in case you want to use the kit with instruments that do require the use of ROX. Please check our ROX Selection Table at [mygopcr.com](http://mygopcr.com) to determine which ROX concentration your instrument requires.

### 4.2 Amplicon length and primer design

Amplicon lengths of between 80bp and 200bp should be used for the highest efficiency under fast cycling conditions. Amplicons should not exceed 400bp. The shorter the amplicon length, the faster the reaction can be cycled. We recommend using primer design software Primer 3 (<http://frodo.wi.mit.edu/primer3/>). Primers should have a melting temperature ( $T_m$ ) of approximately 60°C. For TaqMan® probes choose probe close to 5' primer and avoid terminal guanosine residues. The probe  $T_m$  should be approximately 10°C higher than the primer  $T_m$ .

### 4.3 Template

Use between 1pg and 20pg human genomic DNA per reaction. Use similar amounts of template in all wells of the same run to ensure accurate allele calling.

## 5. REACTION SETUP

5.1 Gently vortex 2x MyGo SNP Probe Kit then prepare a master mix as follows:

Component	20µl reaction	Final concentration	Notes
2x MyGo SNP Probe Kit	10µl	1x	
Forward primer (10µM)	0.5µl	250nM	See 4.2 above
Reverse primer (10µM)	0.5µl	250nM	
Probe (10µM)	0.4µl	200nM	
Template DNA	1-20pg human gDNA	Variable	See 4.3 above
PCR grade water	Up to 20µl total volume		

5.2 Program the instrument as follows, acquiring data on the appropriate channel:

Cycles	Temperature	Time	Notes
1	95°C	2min	Polymerase activation: 2 minutes for cDNA, or 3 minutes for gDNA
40	95°C 60°C to 65°C	10 seconds 20-30 seconds	Denaturation Anneal/Extension: Do not exceed 30 seconds or use temperatures below 60°C