TwistAmp® exo *Quick Guide*

Part Number: TAEXO01Guide | Revision C

Basic Information

RPA

1) Primers must be 30-35 bases
2) Works best at constant temperature (37-39°C)
3) Amplicons of 80-400bp are preferred
4) TwistAmp® exo Probe required - see overleaf

PCR

1) Primers typically 18-25 bases
2) Thermal cycling required
3) Amplicons of 50bp upwards are typical/optimal

Set-up *(single-plex)*[1]

1) Prepare reaction mix in 1.5ml tube:

Primer A (10µM)	2.1 µl
Primer B (10µM)	2.1 µl
TwistAmp® exo Probe (10µM)	0.6 µl
Rehydration Buffer	29.5 µl
Template and dH2O	13.2 µl
(Total Volume	47.5 µl)

Vortex and spin briefly

2) Add reaction mix to freeze-dried reaction. Pipette to mix.
3) Add 2.5 µl of 280mM MgAc (supplied) and mix well to start reaction.

WARNING: RPA REACTIONS START AT ROOM TEMPERATURE AS SOON AS MAGNESIUM IS ADDED.

4) Place strip in Twista® and start run: 37-39°C, 20 minutes.

4b) For low template copy number, remove strip after 4 minutes, vortex & spin briefly, replace in Twista®.

WARNING: IF TUBES ARE OPENED AFTER AMPLIFICATION THERE IS A GREAT RISK OF CONTAMINATION OF WORK SURFACES WITH AMPLICON! ENSURE THAT APPROPRIATE AVOIDANCE MEASURES ARE TAKEN!

WARNING: SWITCH OFF HEATED LIDS BEFORE STARTING REACTIONS!

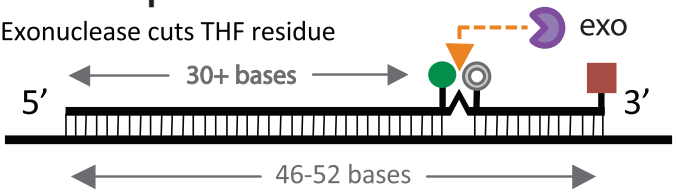
1 See manual for multiplexing

RPA uses TwistDx's proprietary probe systems

RPA does NOT use PCR probe systems

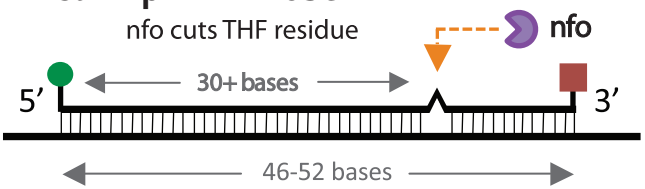
TwistAmp® exo Probe

Exonuclease cuts THF residue



TwistAmp® LF Probe

nfo cuts THF residue



Fluorophore
Quencher
Nuclease
3' block
THF residue

refer to manual for details of probe design

TwistDx

www.twistdx.co.uk