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TwistDx™
Unwind DNA's possibilities

# TwistAmp™ Liquid exo

## Quick Guide

Part number: INLQEXO
Revision 3

# TwistAmp™ Liquid exo Quick Guide

**Please see instruction and assay design manuals at twistdx.co.uk for information regarding components and storage, assay design, and detailed use.**

**Instructions are based on 50 µl reaction volumes; if using a different volume, quantities should be adjusted appropriately.**

## Primer screen set-up (single-plex)[1]

1. Add 2.1 µl of each primer and 0.6 µl of exo probe at 10µM concentration to 0.2 ml PCR tubes.

2. Prepare a pre-master mix (per reaction) in the order below:

| | |
|---|---|
| 2x Reaction Buffer | 25 µl |
| dNTPs[2] + water[3] to | 8.2 µl |
| 10x Probe E-mix | 5 µl |

Vortex and spin briefly.

3. To the pre-master mix, add 2.5 µl 20x Core Reaction Mix[4] (per reaction) to tube lid. Mix by 10x full inversions and spin briefly.

4. Add 1 µl 50x exo (per reaction) to tube lid. Mix by 10x full inversions and spin briefly. Master mix is now complete[5]. Pipette mix before use.

5. Add 41.7 µl[3] master mix to primers and probe prepared in tubes (step 1) and pipette mix.

6. Add 2.5 µl of 280mM MgOAc (supplied) and 1 µl template to tube lids[3]. DNA and MgOAc should be kept separate in the tube lid prior to spin-down. Spin in MgOAc/template and mix well (6x inversions) to start reaction. Spin briefly.

**Warning:** RPA reactions start as soon as MgOAc is added.

7. Place reactions in a fluorometer and start run: 37-42°C, 20 minutes. For low template copies, remove strip after 4 mins, mix by 6x full inversions and spin briefly, replace in fluorometer.

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## Template screen set-up (single-plex)[1]

1. Prepare a primer and probe premaster mix (per reaction) in the order below:

| | |
|---|---|
| 2x Reaction Buffer | 25 µl |
| dNTPs[2] + water[3] to | 8.2 µl |
| 10x Probe E-mix | 5 µl |
| Primer A (10µM) | 2.1 µl |
| Primer B (10µM) | 2.1 µl |
| Probe (10µM) | 0.6 µl |

Vortex and spin briefly.

2. Add 2.5 µl 20x Core Reaction Mix[4] (per reaction) to tube lid. Mix by 10x full inversions and spin briefly.

3. Add 1 µl 50x Exo (per reaction) to tube lid. Mix by 10x full inversions and spin briefly. Master mix is now complete[5]. Pipette mix before use.

4. Add 46.5 µl[3] master mix to 0.2 ml PCR tubes.

5. Add 2.5 µl of 280mM MgOAc and 1µl template to tube lid3. DNA and MgOAc should be kept separate in the tube lid prior to spin-down. Spin in MgOAc/template and mix well (6x inversions) to start reaction. Spin briefly.

**Warning:** RPA reactions start as soon as MgOAc is added.

6. Place reactions in fluorometer and start run: 37-42°C, 20 minutes. For low template copies, remove strip after 4 mins, mix by 6x full inversions and spin briefly, replace in fluorometer.

1 See manual for multiplexing.
2 Suggested final concentration of 1.8mM (total) dNTPs. Optimisation is recommended.
3 Volumes should be adjusted if adding more/less template and/or MgOAc.
4 Warm to room temperature and pipette mix slowly to ensure homogeneity.
5 Master mix may appear cloudy, this is normal.

TwistDX
Liquid exo QG

Size:
8.268 in x 5.827 in

PMS 185 C
Red

Black

PMS 7541 C
Gray 1 - 10%

Part Number: INLQEXO
Revision 3

Date of Revision:
3.2 2019/01