

Olink Target 48 High Multiplex Immunoassay Panels Instructions

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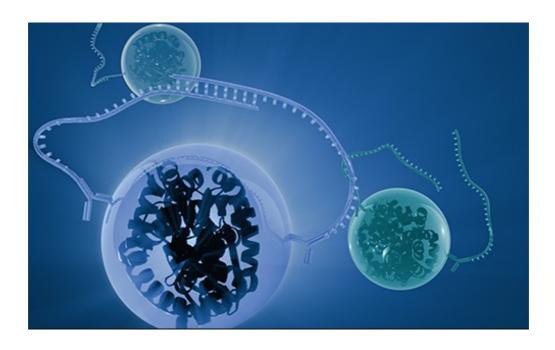


Contents

- 1 Olink Target 48 High Multiplex Immunoassay **Panels**
- 2 Incubation
- 3 Extension
- **4 Detection**
- 5 Documents / Resources
 - **5.1 References**
- **6 Related Posts**



Olink Target 48 High Multiplex Immunoassay Panels

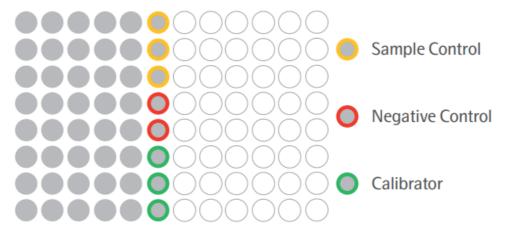


Incubation

1. Prepare the Incubation mix in a microcentrifuge tube according to the table below.

| Incubation mix | per ½ 96-well plate (μL) |
|--------------------------------------|--------------------------|
| Olink® Target 48 Incubation Solution | 168 |
| Olink® Target 48 Frw-probes | 21 |
| Olink® Target 48 Rev-probes | 21 |
| Total | 210 |

- 1. Vortex and spin down the Incubation mix. Transfer 23 μ L of the Incubation mix to each well of a new 8-well strip.
- 2. Transfer 3 μ L of Incubation mix to each well of the first 6 columns of a 96-well plate by reverse pipetting and name the plate Incubation Plate.
- 3. Add 1 μ L of each sample using a multi-channel pipette to the bottom of the well, 1 μ l of Sample Control to the three top wells (yellow), 1 μ L of Negative Control to two wells (red), and 1 μ L of Calibrators to three wells (green), according to the plate layout.



4. Seal the plate with an adhesive plastic film, spin at $400 - 1000 \times g$, 1 min at room temperature. Incubate

Extension

1. Prepare an extension mix according to the table below.

| Extension mix | per ½ 96-well plate (µL) |
|-------------------------------|--------------------------|
| High Purity Water (+4 °C) | 4350 |
| Olink® Target 48 PEA Enhancer | 580 |
| Olink® Target 48 PEA Solution | 580 |
| Olink® Target 48 PEA Enzyme | 58 |
| Total | 5 568 |

- 2. Bring the Incubation Plate to room temperature, spin at $400 1000 \times g$ for 1 min. Preheat the PCR machine.
- 3. Vortex the Extension mix and pour it into a multichannel pipette reservoir.
- 4. Start a timer for 5 min and transfer 96 μL of Extension mix to the upper parts of the well walls of the Incubation Plate by using reverse pipetting.
- 5. Seal the plate with a new adhesive plastic film, use the MixMate® to vortex the plate at 2500 rpm for 30 sec, ensuring that all wells are mixed, and spin down.
- 6. Place the Incubation Plate in the thermal cycler and start the PEA program. (50 °C 20 min, 95 °C 5 min (95 °C 30 sec, 54 °C 1 min, 60 °C 1 min) x 17, 10 °C hold)

Detection

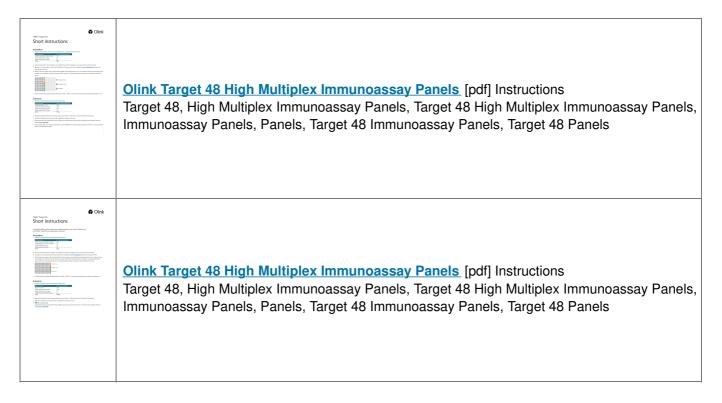
- 1. Prepare and prime an Olink® 48.48 IFC for Protein Expression. Briely, inject one control line fluid syringe into each accumulator on the chip, and then prime the IFC on the Olink® Signature Q100 following the instructions on the instrument screen.
- 2. Thaw the Primer Plate, vortex and spin briefly.
- 3. Prepare a Detection mix according to the table below.

| Detection mix | per ½ 96-well plate (µL) |
|-------------------------------------|--------------------------|
| Olink® Target 48 Detection Solution | 275.0 |
| High Purity Water | 116.0 |
| Olink® Target 48 Detection Enzyme | 3.9 |
| Olink® Target 48 PCR Polymerase | 1.5 |
| Total | 396.4 |

- 4. Vortex the Detection mix and spin briefly and add 46 μL of the mix to each well of an 8-well strip.
- 5. Transfer 7.2 μL of the Detection mix to each well of column 1-6 in a new 96-well plate by reverse pipetting, and name it Sample Plate.
- 6. Remove the Incubation Plate from the thermal cycler, spin down the content and transfer 2.8 μL to the Sample Plate, using forward pipetting.
- 7. Seal the plate with an adhesive film, vortex and spin both at $400 1000 \times g$, 1 min at room temperature.
- 8. Transfer 5 μ L from each well of the Prime Plate and 5 μ L of the Sample Plate into the primed 48.48 IFC left and right inlets, respectively. Use reverse pipetting and change tips after each primer or sample. Do not leave any

- inlets empty.
- 9. Remove bubbles and load the chip in the Olink Signature Q100 and follow the instructions on the instrument screen.
- 10. Run the IFC on the Olink Signature Q100.
- 11. Carefully remove the adhesive film from the Primer Plate to avoid contamination between wells.
- 12. Transfer 5 μL of each primer using reverse pipetting from each well in position 1 A-H (green) to the inlets in the first column on the left side of the IFC (green). Change pipette tips after each column. When using an eight-channel pipette every other inlet will be filled according to the image.
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 - 1126, v1.1, 2022-05-05

Documents / Resources



References

Mean - Olink