Color Compensation Set 3
Ready Reference Guide

Revision A, December 2023

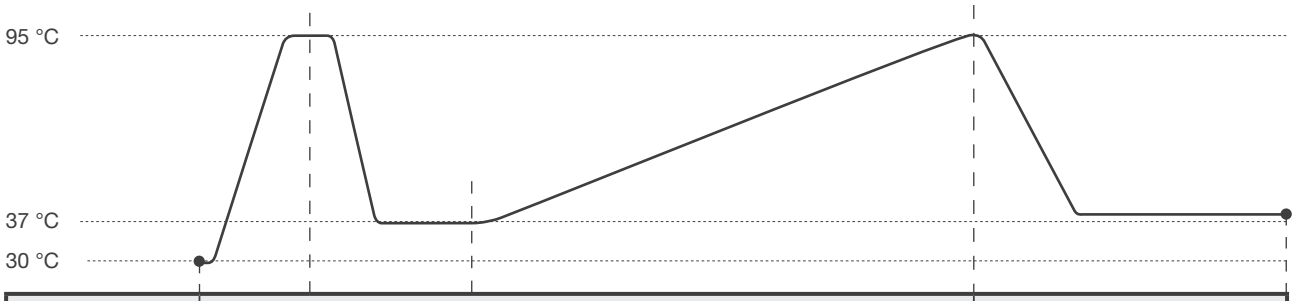
Product No. KIT230005

Color compensation set for the generation of color compensation objects for the LightCycler® 480 System. The set is designed for 5 calibration runs.

Before starting, it is strongly recommended to read the entire product manual available on our website.

PROGRAM SETUP

Program the LightCycler® 480 instrument as shown below before preparing the calibration mixes.

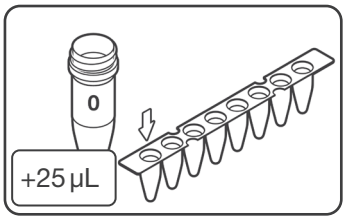


	Temperature Gradient			Cooling
Cycles	1			1
Analysis Mode	Color Compensation			None
Steps	1	2	3	1
Target Temp	95 °C	37 °C	95 °C	40 °C
Acquisition Mode	None	None	Continuous	None
Hold	1 s	10 s	-	30 s
Ramp Rate	4.4	2.2	-	2.2
Acquisition/°C	-	-	1	-

Parameter	Setting	Name	Melt Factor	Quant Factor	Max Integration Time (s)
Detection Format	465 - 510	FAM	1	10	2
	533 - 580	HEX	1	10	2
	533 - 610	ROX	1	10	2
	618 - 660	Cy5	1	10	2
Block Size	96				
Reaction Volume	25 µL				

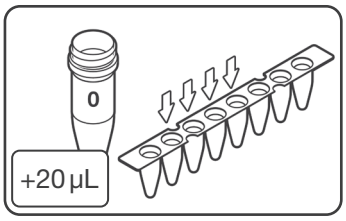
PREPARATION OF THE PCR MIX

Take appropriate precautions to prevent contamination, e.g., by using filter tips and wearing gloves. Thaw reagents, mix (do not vortex!), and briefly spin vials before opening. Use tube strips or plates matching Hygiena Diagnostics' kit consumables. The tube strips and caps supplied with the Color Compensation Kits are identcal to those found in the foodproof LyoKits.



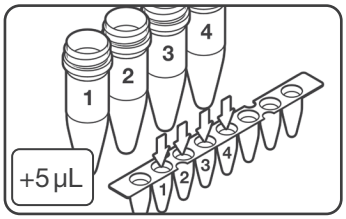
1. ADD BLANK TO FIRST WELL

Pipet 25 µL of Blank (vial 0, white cap) into first well.



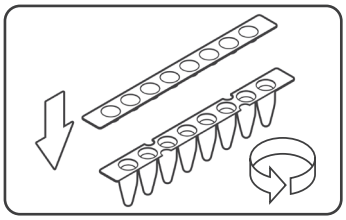
2. ADD BLANK TO ADDITIONAL WELLS

Pipet 20 µL of Blank (vial 0, white cap) into second, third, fourth and fifth well.



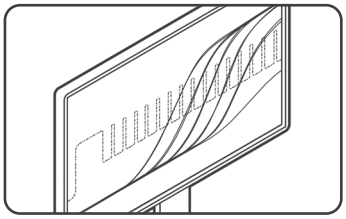
3. ADD CALIBRATOR DYES

Pipet 5 µL of Calibrator 1 (yellow cap), Calibrator 2 (red cap), Calibrator 3 (purple cap) and Calibrator 4 (blue cap) into second, third, fourth and fifth well, respectively.



4. SEAL & CENTRIFUGE

Seal strips accurately and briefly spin strips/plate in a suitable centrifuge.



5. CYCLE SAMPLES

Insert the PCR strips into a support frame matching LightCycler® plates and load to the instrument.

Cycle the samples as described.

Vials	Dominant Channel
0	water
1	465 - 510 (FAM)
2	533 - 580 (HEX)
3	533 - 610 (ROX)
4	618 - 660 (Cy5)

6. ANALYSIS

In the Subset Editor, create a new subset for the Color Compensation reactions. In the Sample Editor, define the dominant channel for each position.

Select 'Color Compensation' from the analysis menu, click 'Calculate' and then click the 'Save CC Object' button.

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Kit for 5 runs
Store kit in the dark
at -15 to -25 °C

FOR IN VITRO USE ONLY
Made in Germany

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