

**Crystal Violet (CV) Staining of Cells and Clone counting**

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Stocks.

3.7% Paraformaldehyde (PFA) or 10% Formalin

0.05% Crystal Violet in Distilled Water (Filter at 0.45um before use)

Fix the cells for 5 min. with 3.7% PFA

STAIN them 30 minutes with 0.05%CV

Wash 2x with tap water,

Drain them inverted for a couple minutes.

The dishes can then be photographed or scanned for colony counts

READING CV STAIN (OD 540)

add a third to half the total well volume of straight methanol to solubilize s the dye,

Read the plates (OD 540) either directly or by taking an aliquot to another plate.

The cells never lyse and morphology is extremely well preserved by the combined formalin/methanol fixations.

ALTERNATIVE PROTOCOL:

USE 0.1 % CV in 2 % ethanol in 0.1 M borate ph9

NO NEED TO PRE-FIX