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PM Abiotic Color

From Dye Reduction

Biolog employs a series of redox dyes in the PM Protocols. These dyes are all reduced to form a purple color, but the redox threshold at which they get reduced is different. At one end of the spectrum, Dye A is most difficult to reduce. At the other end of the spectrum, Dye G is most easily reduced. The order is: A>B>F>D>H>G.

Some of the chemicals used in the Biolog PM MicroPlates are reducing chemicals that can directly reduce the dye, even when no cells are added to the wells. This occurs with the more easily reduced dyes (F, D, H, G) resulting in the formation of purple color. Without cells present, these wells will give abiotic "false-positive" reactions because the dye is being chemically reduced. In PM1 for example these dyes will get abiotically reduced in the wells with the 4 pentose sugars: arabinose, ribose, xylose, lyxose. Here is a partial list:

PM1: A-2 L-arabinose, B-8 D-xylose, C-4 D-ribose, H-6 L-lyxose
PM2: B-5 D-arabinose, B-9 2 deoxy-D-ribose, B-12 3-O-B-D-galactopyranosyl-D-arabinose, C-12 palatinose, E-5 D-glucosamine, E-12, 5-keto-D-gluconic acid, F-5 oxalomalic acid, F-9 sorbic acid, H-9 dihydroxyacetone.
PM3: A-11 L-cysteine, G-3 uric acid, G-10 D,L-a-amino-caprylic acid
PM6: D-4 cys-gly, E-4 gly-cys

It is not possible to give a definitive list of abiotic "false-positive" wells because the dye reduction is increased by variable test parameters, particularly high temperature, high pH, the duration of incubation, and others.

The best way to test for and detect abiotic reactions is to run a set of PM MicroPlates using the exact assay protocol but leaving out the cells. Another way to detect abiotic reactions is to examine the dye reduction kinetics. Biological reduction will be gradual over time, whereas abiotic reduction usually occurs rapidly, more like a step function.

From Chemical Color

The inhibitory chemicals in PM11-25 are provided in a titrated series of 4 wells with increasing concentrations going from left to right. Some of these chemicals have inherent color (yellow, orange, brown) that can be visually seen to increase with the titration series. This color should be subtracted away and ignored as the relevant color in the PM assays is the formation of the purple reduced form of the redox dye.