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Stop Solution

Stop and stabilize TMB microwell substrate reactions.

Stop Solution is a proprietary acidic formulation used to stop TMB microwell substrate reactions. It is suitable for all endpoint ELISAs using a tetramethylbenzidine (TMB) substrate reaction for color development.

TMB substrates (e.g., catalog# LS-M36, LS-M37, LS-M28) are oxidized by peroxidase enzymes to yield a soluble blue-green reaction product. In endpoint assays, the reaction can be stopped by adding equal volumes of Stop Solution for TMB Substrates. Addition of Stop Solution changes the chromagen color from blue-green to yellow, where it can be read at 450 nm, and concurrently stabilizes the yellow TMB product for one hour. Stopping the reaction will increase the sample absorbance value up to 3-fold. To avoid overdeveloping the TMB substrate reaction, the blue-green reaction product should be periodically monitored on an ELISA plate reader using 620-650 nm absorbance filter settings. When OD values reach approximately 0.7 units, the reaction should be stopped with Stop Solution.

For best results, the absorbance should be monitored and read before values exceed 2.5 OD units. If the yellow reaction product yields OD values above 2.5 units, it is recommended to dilute the antibodies/conjugates or shorten the incubation period.

Stop Solution is ready to use at 1X. Stop the reaction by adding an equal volume of Stop Solution to the substrate in every well of the plate.

STOP SOLUTION FOR TMB SUBSTRATES

 Size
 Catalog#

 100 mL
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 1 L
 LS-M43-1000

INSTRUCTIONS:

- 1. Run ELISA according to the specific protocol through the conjugate incubation step.
- 2. Wash the wells three or four times with 1X ELISA Wash Buffer (catalog# LS-M27) to remove any residual HRP-conjugate.
- 3. Add TMB substrate to each well of the plate. For example, use 100 μ L/well; protect from light.
- 4. Incubate the substrate 10-60 minutes. Monitor the color intensity.
- 5. Stop the reaction by adding an equal volume of Stop Solution to the substrate in every well of the plate, e.g., if each well contains $100~\mu L$ substrate, add $100~\mu L$ /well Stop Solution.
- 6. Read the plate at 450 nm within 1 hour.

For more ELISA protocols and information, please visit www.LSBio.com.

SPECIFICATIONS:

- · Clear, colorless liquid
- pH ≤ 2
- 1X ready to use
- Read absorbance at 450 nm

STORAGE:

- 25°C
- Refrigerated temperatures will not harm the reagent

SAFETY & USAGE:

- Contains Maleic acid at 3-7%
- · SDS available upon request
- Not for human or drug use
- For research use only



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www.LSBio.com