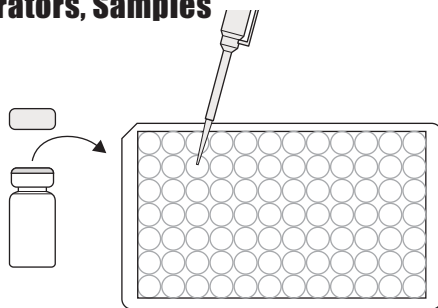


# 2,4-D Plate, Detailed ELISA Procedure

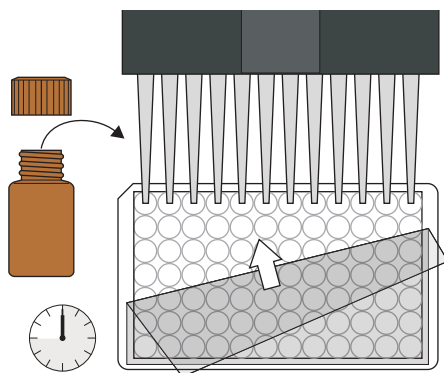
## 1. Addition of Calibrators, Samples

Add 50 uL of calibrators or sample into the wells of the test strips according to the working scheme given. Be sure to use a clean pipet tip for each.



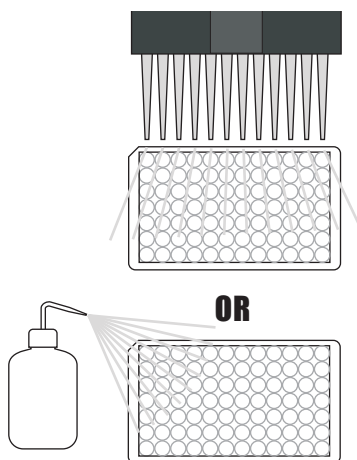
## 2. Addition of Enzyme Conjugate

Add 50 uL of the Enzyme Conjugate to the individual wells successively using a multi-channel pipette or a stepping pipette. Cover the wells with parafilm or tape and mix the contents by moving the strip holder in a rapid circular motion on the benchtop for 60 seconds. Be careful not to spill contents. Incubate the strips for 60 min at room temperature.



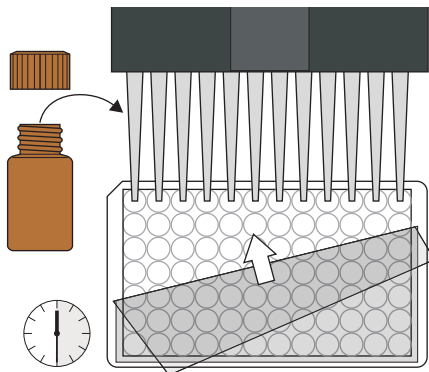
## 3. Washing of Plates

After incubation, remove the covering and vigorously shake the contents of the wells into a sink. Wash the strips five times with a multi-channel pipette or wash bottle using the diluted 5X washing buffer solution. Please use at least a volume of 250 uL of washing buffer for each well and each washing step. Remaining buffer in the wells should be removed by patting the plate dry on a stack of paper towels.



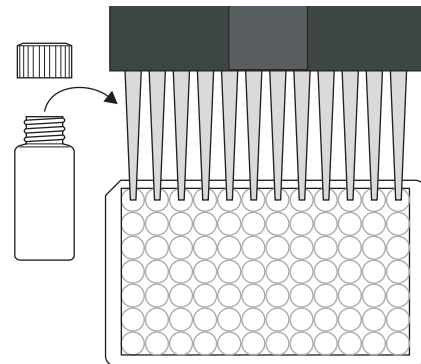
## 4. Addition of Substrate/Color Solution

Add 100 uL of substrate/color solution to the individual wells successively using a multi-channel pipette or a stepping pipette. Cover the wells with parafilm or tape and mix the contents by moving the strip holder in a rapid circular motion on the benchtop. Be careful not to spill contents. Incubate the strips for 30 min at room temperature.



## 5. Addition of Stopping Solution

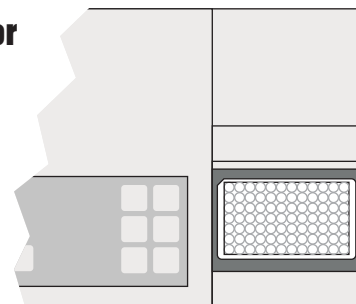
Add 100 uL of stop solution to the wells in the same sequence as for the substrate solution using a multi-channel pipette or a stepping pipette.



## 6. Measurement of Color

Read the absorbance at 450 nm using a microplate ELISA reader. Calculate results.

NOTE: Multiply ELISA results by appropriate dilution factors.

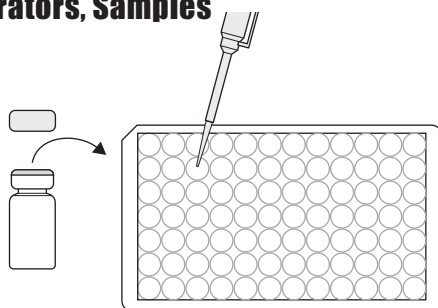


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# 2,4-D Plate, Concise ELISA Procedure

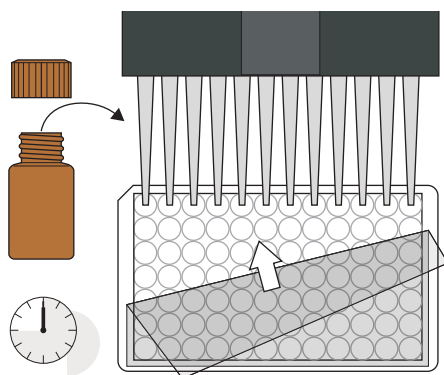
## 1. Addition of Calibrators, Samples

Add 50  $\mu$ L of calibrators or diluted sample extracts.



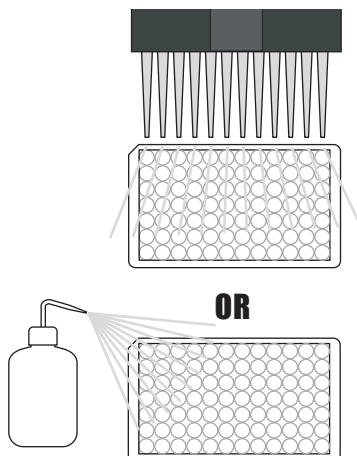
## 2. Addition of Enzyme Conjugate

Add 50  $\mu$ L of the Enzyme Conjugate. Cover and mix for 60 seconds by rotating on benchtop. Incubate for 60 minutes at room temperature.



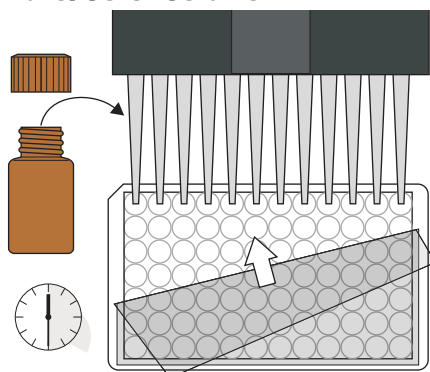
## 3. Washing of Plates

Wash the plates five times with 250  $\mu$ L of diluted 1X washing buffer.



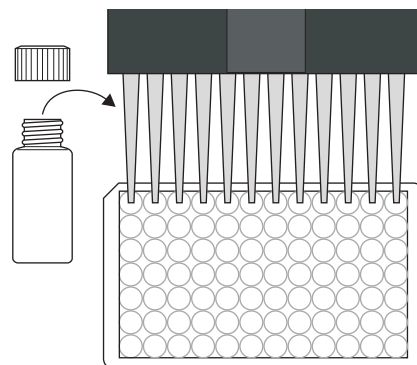
## 4. Addition of Substrate/Color Solution

Add 100  $\mu$ L of substrate/color solution. Incubate 30 minutes at room temperature and away from direct sunlight.



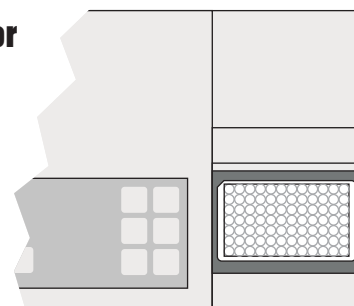
## 5. Addition of Stopping Solution

Add 100  $\mu$ L of stop solution.



## 6. Measurement of Color

Measure color at 450 nm.  
Calculate results.



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