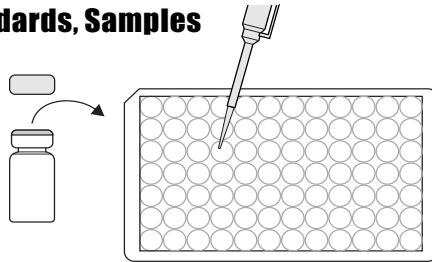


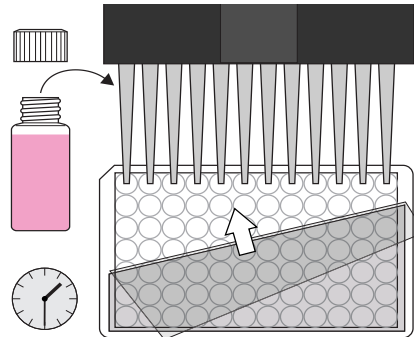
## 1. Addition of Standards, Samples

Add 50  $\mu$ L of the standard solutions, control, or samples into the wells of the test strips according to the working scheme given. We recommend using duplicates or triplicates.



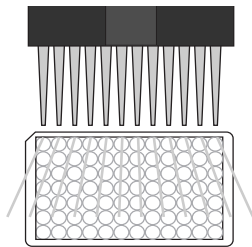
## 2. Addition of Antibody Solution

Add 50  $\mu$ L of the antibody 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 for 30 seconds. Be careful not to spill contents. Incubate for 90 minutes at room temperature.



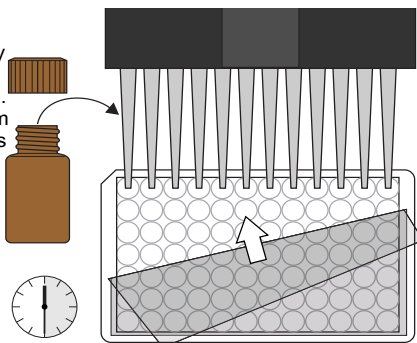
## 3. Washing of Plates

After incubation, remove the covering and vigorously shake the contents of the wells into a sink. Wash the strips three times with a multi-channel pipette using the 1X washing buffer solution. Please use at least a volume of 250  $\mu$ L 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.



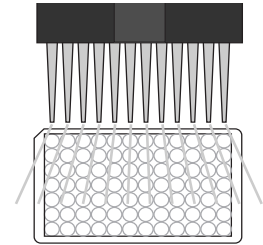
## 4. Addition of Enzyme Conjugate

Add 100  $\mu$ L of enzyme conjugate 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. Incubate for 30 minutes at room temperature.



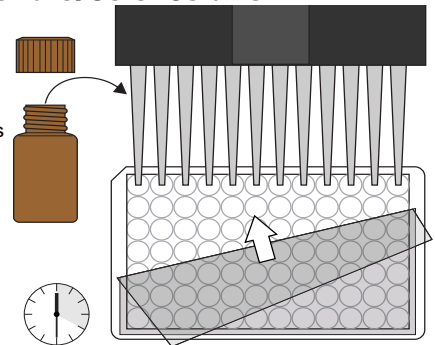
## 5. Washing of Plates

After incubation, remove the covering and vigorously shake the contents of the wells into a sink. Wash the strips three times with a multi-channel pipette using the 1X washing buffer solution. Please use at least a volume of 250  $\mu$ L 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.



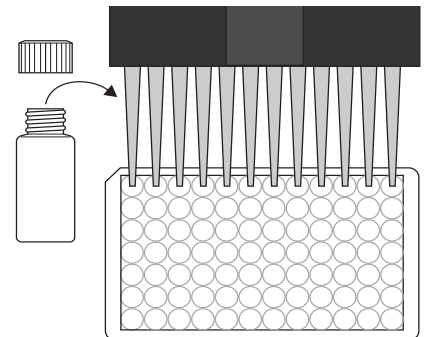
## 6. Addition of Substrate/Color Solution

Add 100  $\mu$ L of substrate/color solution to the wells using a multi-channel pipette or a stepping pipette. The strips are incubated for 20-30 minutes at room temperature. Protect the strips from direct sunlight.



## 7. Addition of Stopping Solution

Add 50  $\mu$ L of stop solution to the wells in the same sequence as for the substrate solution using a multi-channel pipette or a stepping pipette.



## 8. Measurement of Color

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

