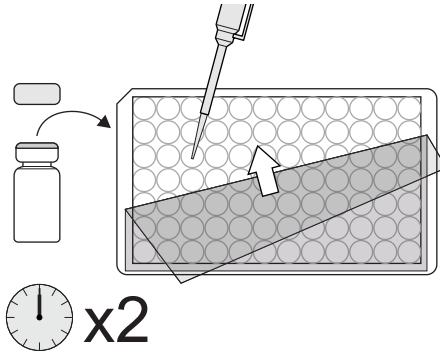


# Anatoxin-a RBA Kit, Detailed Procedure

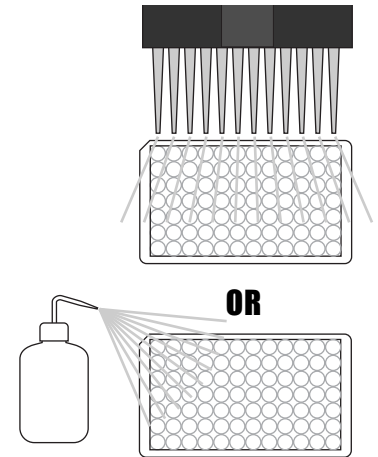
## 1. Addition of Standards, Samples

Add 100  $\mu$ L of the standard solutions and/or samples into the wells of the test strips according to the working scheme given. Analysis in triplicate is recommended. Cover the wells with adhesive plate cover and mix the contents by moving the strip holder in a rapid circular motion on the benchtop for 1 minute. Be careful not to spill contents. Incubate the strips for 2 hours at 37°C.



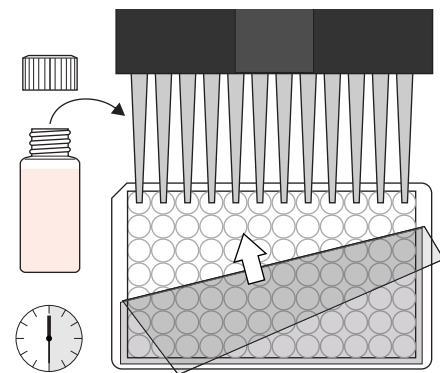
## 5. Washing of Plates

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 or wash bottle using the diluted 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.



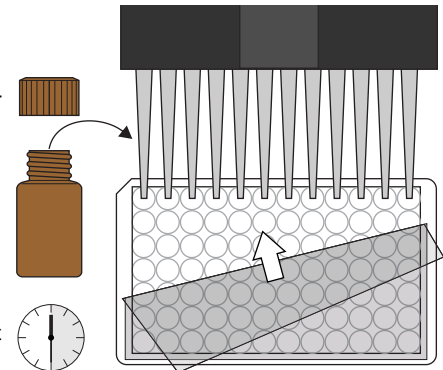
## 2. Addition of Biotinylated Alpha-Bungarotoxin Solution

Add 50  $\mu$ L of the biotinylated alpha-bungarotoxin solution to the individual wells successively using a multi-channel pipette. Cover the wells with adhesive plate cover and mix the contents by moving the strip holder in a rapid circular motion on the benchtop for 1 minute. Be careful not to spill contents. Incubate the strips for 30 minutes at 37°C.



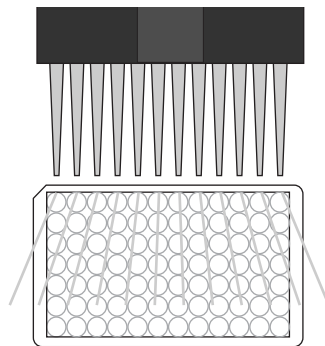
## 6. Addition of Substrate/Color Solution

Add 150  $\mu$ L 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 for 1 minute. Be careful not to spill contents. Incubate the strips for 30 minutes at room temperature.



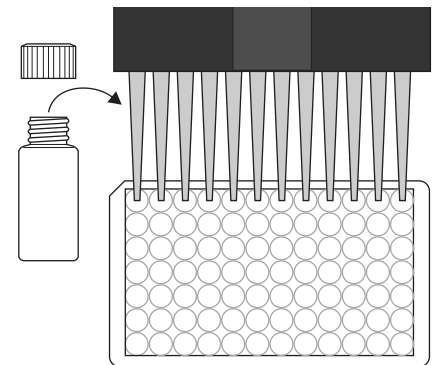
## 3. Washing of Plates

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 diluted 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.



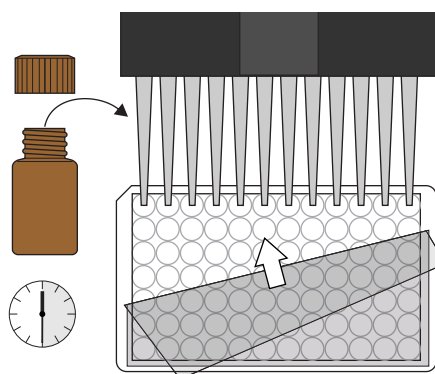
## 7. Addition of Stopping Solution

Add 100  $\mu$ 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.



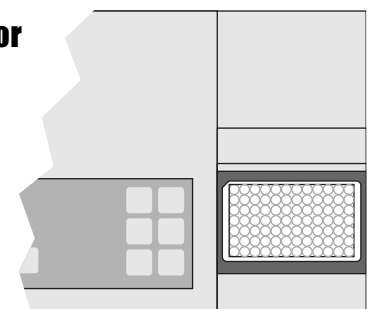
## 4. Addition of Streptavidin-HRP Conjugate

Add 150  $\mu$ L of the streptavidin-HRP conjugate to the individual wells successively using a multi-channel pipette or a stepping pipette. Cover the wells with adhesive plate cover and mix the contents by moving the strip holder in a rapid circular motion on the benchtop for 1 minute. Be careful not to spill contents. Incubate the strips for 30 minutes at 37°C.



## 8. Measurement of Color

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

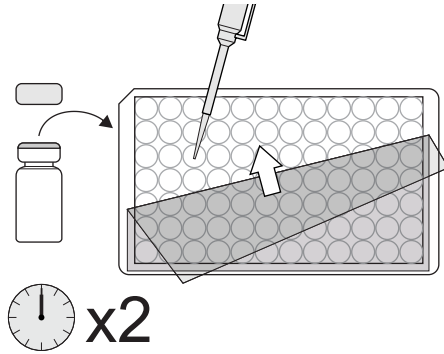


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 124 Railroad Drive, Warminster, PA 18974  
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 www.abraxiskits.com

# Anatoxin-a RBA Kit, Concise Procedure

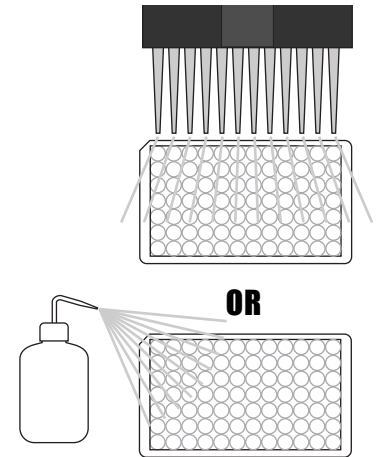
## 1. Addition of Standards, Samples

Add 100  $\mu$ L of the standard solutions and/or samples. Cover and mix for 1 minute. Incubate the strips for 2 hours at 37°C.



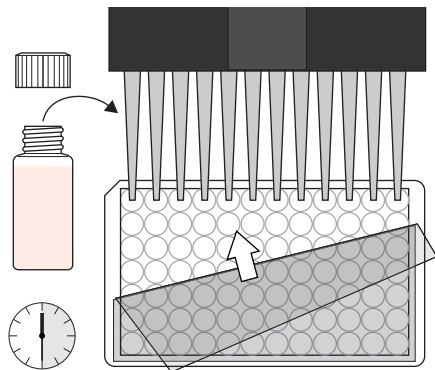
## 5. Washing of Plates

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



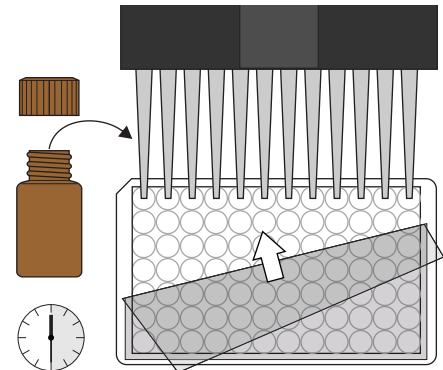
## 2. Addition of Biotinylated Alpha-Bungarotoxin Solution

Add 50  $\mu$ L of the biotinylated alpha-bungarotoxin solution. Cover and mix for 1 minute. Incubate the strips for 30 minutes at 37°C.



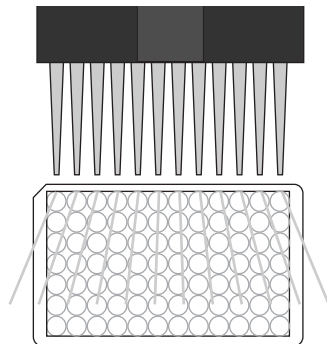
## 6. Addition of Substrate/Color Solution

Add 150  $\mu$ L of substrate/color solution. Incubate 30 minutes at room temperature and away from direct sunlight.



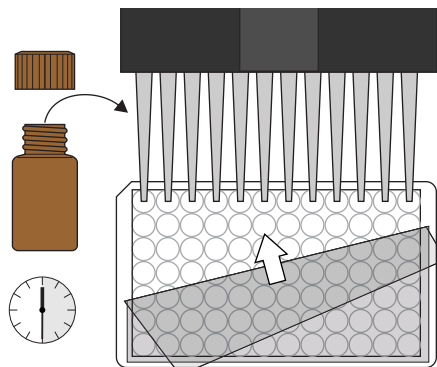
## 3. Washing of Plates

Wash the strips three times with 250  $\mu$ L of diluted 1x washing buffer.



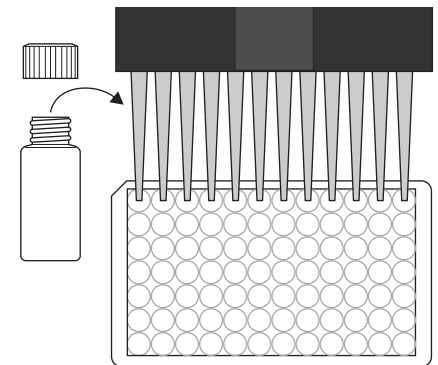
## 4. Addition of Streptavidin-HRP Conjugate

Add 150  $\mu$ L of the streptavidin-HRP conjugate. Cover and mix for 1 minute. Incubate the strips for 30 minutes at 37°C.



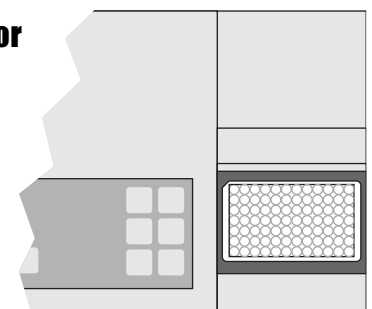
## 7. Addition of Stopping Solution

Add 100  $\mu$ L of stop solution.



## 8. Measurement of Color

Read the absorbance at 450 nm. Calculate results.



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