

GBI-Emerald Chromogen for HRP Use in Immunohistochemistry

Organic Resistant GBI-Emerald Chromogen for Horseradish Peroxidase Detection

Storage: 2-8°C

Catalog No: C12-18 18 mL

C12-120 120 mL

Intended Use:

GBI-Emerald Chromogen is provided in ready to use solution. GBI-Emerald Chromogen produces an emerald green color precipitate at the site of reaction when it reacts with horseradish peroxidase. GBI-Emerald chromogen is resistant to organic solvent which allows the user to dehydrate their slides in graded alcohols and xylene. However it is water soluble so we recommend you limiting the wash steps after emerald staining.

Kit Components:

This product is supplied as ready to use product. Slide tests number based on using 100µL per tissue section.

Product	Catalog No.	Emerald Chromogen Size	Number of Slide Tests
GBI-Emerald (RTU)	C12-18	18mL	180 slides
GBI-Emerald (RTU)	C12-120	120mL	1200 slides

Recommended Protocol:

We strongly recommend that you counter-stain slides before adding emerald chromogen especially for weak signal to reduce signal loss in wash steps since emerald chromogen is water soluble.

1. To counter stain before GBI-Emerald Chromogen. We recommend the following protocol
 - a) **Counter stain with Hemotoxylin 10 - 30 seconds.**
 - b) **Wash in 3 changes of dH₂O 1 minute each.**
 - c) **Blue with PBS or Tris pH7.6 +/- 0.2 for 30 seconds.**
 - d) **Wash in 3 changes of dH₂O 1 minute each go to step 2.**

Warning! Do not increase incubation times for any steps listed above for counter staining protocol as this will increase the chance of reducing the HRP enzyme activity.

2. Add one or two drops of **GBI-Emerald** chromogen to completely cover tissue section and incubate for 5 minutes. Color development may be monitored under microscope.
3. After proper color development, wash with distilled water 10 seconds for 2-3 times.
4. **Skip this step if you counterstained before GBI-Emerald Chromogen!** To counter stain after the application of GBI-Emerald Chromogen step follow protocol below.
 - a) **Counterstain with Hemotoxylin 30 seconds.** (We find filtering Hemotoxylin just prior to counter staining with 3MM paper removes precipitant which allows for shorter wash times after counter stain.)
 - b) **Rinse with distill water for 30 seconds 3 times to clear slides.**
 - c) **Blue with PBS or Tris pH 7.6 +/- 0.2 for 30 seconds.**
 - d) **Wash in 3 changes of dH₂O 30 to 60 seconds each then go directly to the dehydration steps in #5. (Note: We recommend 30 seconds for wash since GBI-Emerald Chromogen is soluble in water)**
5. **GBI-Emerald** Chromogen is insoluble in organic solvent; however the dehydration steps must be shorter for optimal tissue structure and chromogen signal maintenance.
 - a) **1x 80% Ethanol 20 seconds**
 - b) **1x 95% Ethanol 20 seconds**
 - c) **3x 100% Ethanol 20 seconds each**
 - d) **1x 100% Xylene 20 seconds**
 - e) **Add 1drop of xylene based mountant and coverslip**

Note: If there is green background on the slide, then concentration of primary antibody is too high.

Remarks:

For research use only.

Larger sizes of chromogen available upon request.