eStain® 2.0 Protein Staining System - Frequently Asked Questions

1. **What's the difference between eStain® 2.0 Protein Staining System and original eStain® System?**
   eStain® 2.0 Protein Staining System is the upgrade product of original eStain™1.0 System. The eStain® 2.0 Protein Staining Device is a totally redesigned electric staining unit. The stylish and durable design delivers a more user-friendly unit with extended service life. A larger LCD screen on the control panel clearly displays all the parameters. A waste tray is added to the right side of eStain® 2.0 Device to collect liquid waste and simplify the cleanup. The optimized circuit and control program increase reliability and reproducibility. The eStain® 2.0 Device uses a new eStain® 2.0 Graphite Electrode (L02017) which cannot be used on original eStain®1.0 Device. The eStain® Protein Staining Pads (R-250, L02011 and G250, L02012) are compatible with both eStain® 2.0 System and original eStain™1.0 System.

2. **Why should I use eStain®2.0 Protein Staining System?**
   The eStain® 2.0 Protein Staining System is an innovation in protein electrophoresis analysis. It is designed to quickly, easily and reliably perform protein gel staining with Coomassie blue dye within 7 minutes or less. This system can greatly shorten your workflow and save your valuable research time for subsequent analysis.

3. **What's the difference between eStain®2.0 Protein Staining System and conventional Coomassie blue staining methods?**
   Conventional Coomassie blue staining methods rely on Brownian motion and require three steps including fixing (or washing), staining and de-staining (or washing), etc., which is a long and tedious process. The eStain®2.0 Protein Staining System applies GenScript's proprietary electric staining technology. A certain definite voltage is applied to the graphite anode and titanium cathode to drive the negatively charged Coomassie blue dye into the gel matrix to stain the proteins and to push the unbound Coomassie blue dye out of the gel matrix to destain the gel. The eStain®2.0 System integrates three steps of traditional tri-step methods into single one step and significantly reduces the time required for protein staining analysis.

4. **What are the functions of eStain® Protein Staining Pads?**
   The eStain® Protein Staining Pads are the consumable part of eStain®2.0 System. Each pack of eStain® Protein Staining Pad contains an eStain® Cathode Pad presoaked with proprietary cathode buffer with CBB dye R-250 or G-250 incorporated, and an eStain® Anode Pad presoaked with proprietary anode buffer. Similar to semi-dry blotting, the eStain® Cathode Pad and eStain® Anode Pad act as ion reservoirs，and the eStain® Cathode Pad also supplies negatively charged CBB dye for protein staining.

5. **What's the difference between eStain® Protein Staining Pads R-250 and G-250?**
   The eStain® Pads (R-250) have incorporated Coomassie blue dye R-250 and make dark blue protein bands. The eStain® Pads (G-250) have incorporated Coomassie blue dye G-250 and deliver bright blue protein bands. Customers can select R-250 or G-250 eStain® Pads according to their color preference.

6. **Can the eStain® Protein Staining Pads be re-used for staining multiple gels?**
   No. For best results, one pack of eStain® Protein Staining Pad is used to stain one gel. Discard used eStain® Pad after each staining.

7. **Can the eStain® Protein Staining Pads be used on blotting device to stain protein gels?**
   No. The eStain® Protein Staining Pads are designed to work with eStain™ 1.0 and eStain®2.0 Protein Staining Devices. If they are used on blotting device, we can't guarantee users satisfactory staining results.

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8. **Why should I replace the eStain® 2.0 Graphite Electrode?**
   During electric staining process, the eStain® 2.0Graphite Electrode will absorb ions from anode pad while lose carbon composition, which will change the conductivity of the graphite electrode, thereby affecting the staining results. For best staining results, after 100 uses, the worn eStain® 2.0 Graphite Electrode should be replaced by a new one; the graphite electrode should also be replaced when staining performance is significantly reduced.

9. **Which types of protein gels are eStain®2.0 Protein Staining System compatible with?**
   The eStain®2.0 Protein Staining System is designed to work with multiple homemade or precast mini polycrylamide gels, including Tris-Glycine, Bis-Tris, Tris-Acetate and Tris-Tricine gels, etc. For specially formulated gels, optimization of the staining time may be needed for best results.

10. **How many gels can be stained at the same time?**
    Using eStain®2.0 Protein Staining System, one piece of gel can be stained for each run.

11. **What is the sensitivity of the eStain®2.0 Protein Staining System?**
    The eStain®2.0 System utilizes Coomassie blue dye R-250 and G-250 as staining reagents. They have similar detection sensitivity down to dozens of ng per protein band.

12. **Is eStain®2.0 Protein Staining System compatible with mass spectrometry and 2D electrophoresis?**
    Yes. It is compatible with mass spectrometry and 2D electrophoresis just like conventional Coommassie blue staining methods.

13. **Can a Western blot be performed on a gel that has been stained with eStain®2.0 Protein Staining System?**
    No. To ensure optimum Western blot results, gels without staining, either conventional Coomassie blue staining or eStaining, should be used.

14. **Can I stain very thin (less than 1 mm) or very thick (more than 1 mm) gels using eStain®2.0 Protein Staining System?**
    Yes. However, the staining time needs to be optimized. For example, 6 -7 minutes are needed to stain a 0.75 mm mini gel and 8-9 minutes are needed to stain a 1.5 mm mini gel.

15. **What is the recommended temperature for using eStain®2.0 Protein Staining System?**
    For best staining results, it is recommended to use eStain® 2.0 Protein Staining System at room temperature (22 - 28 ℃). If temperature is below 22 ℃, users may need to extend the running time to obtain satisfied staining results based on gel thickness.

16. **Why is there a small crack at the titanium cathode side of the lid of the eStain® 2.0 device after a period of use?**
    A tiny crack may develop at one of the two lower corners next to the titanium cathode due to thermal expansion difference between the lid material and the titanium cathode. However, it does not affect the staining performance of the device.

17. **Can I start a run without the assembled staining stack (eStain Anode Pad-Gel-eStain Cathode Pad) in place?**
    No**.** It will **DAMAGE** the device due to short circuit. We have placed a warning sign on the lid of eStain and the cover page of the User Manual.

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