Gel Electrophoresis Protocol

Materials:

1. TAE Buffer
   Purchased from Sigma
   10X Tris-Acetate
   EDTA Concentrate
   Biotechnology Performance Certified
   pH approx. 8.3

2. Agarose, Standard, Low Electroendosmosis (EEO)
   Purchased from VWR International
   Molecular Biology Grade

3. Horizontal Gel Chamber with gel tray and well combs

4. 100mL Erlenmeyer flask

Procedure:

1. Dilute 10X Tris-Acetate 10 times for a final concentration of 1X. Dilute enough Tris-Acetate for a 40mL gel and to completely fill the gel chamber. Set aside.

2. Prepare the gels such that it is 1-1.5% agarose. Weigh out approximately 0.4 – 0.6 g of the agarose powder and transfer to 100 mL Erlenmeyer flask.

3. Add 40 mL 1X TAE buffer to the 100 mL Erlenmeyer containing the agarose powder. Swirl the flask to ensure homogeneous distribution of the agarose powder in the TAE buffer.

4. Heat the agarose-TAE buffer mixture until it begins to boil. Swirl the flask very well and make sure all agarose is dissolved in the buffer (no solid powder form should be visible).

5. Continue heating the agarose-TAE buffer mixture in intervals to ensure the agarose is completely dissolved in the buffer without the mixture boiling over.

6. The agarose is completely dissolved in the buffer when a clear translucent solution is formed. Set aside to cool down just enough to where the solution will not melt the gel tray.

7. While the gel solution is cooling fill the gel chamber with enough 1X TAE buffer (pH>7 for nano crystals with COOH and pH <7 with –NH2) to fill the chamber half way.

8. Once the agarose buffer has cooled slightly (enough to hold in your hand and not feel burned) pour the solution in to the gel tray with well comb fixed properly.

9. After the gel has cooled completely and solidified the combs can be removed and the tray inserted properly into the gel chamber.

10. Pour enough 1X TAE buffer into the chamber to cover the gel and fill the wells.

11. Pipette 5µL of iron oxide nanoparticles into the appropriate well of the gel.

    NOTE: It may be necessary to increase the density of the particles by the addition of a loading buffer.

12. The gel should be run at 100V for 30 minutes.

13. In general, unconjugated nanocrystals will migrate faster than the conjugated nanocrystals. Also, it is noteworthy to know that theoretically, the positively charged conjugated nanocrystals will migrate towards the negative electrode while the negatively charged nanocrystals will migrate towards the positive electrode.