Lab: Paternity Test Analysis

AP Biology
Mrs. Kymissis and Ms. Martel

You have just been hired by the Jerry Springer show as the in house molecular biologist in charge of paternity testing. During your first day on the job, Bobby B. accuses his wife, Candy C., of having an affair with his best friend (Ernie E.) 9 months prior to the birth of their son. Your task is to find out just who is the baby's father.

Step 1: Making the gel.

1. As a class, you will need to make a large stock solution of TAE buffer. Begin by adding 4ml of concentrated TAE buffer to a beaker containing 800ml distilled water. Mix thoroughly.
2. Each group should obtain 65ml of the diluted buffer and add it to a 100ml beaker that contains .8g agarose powder. Boil this solution for approx. 1 min, continuously stirring to dissolve the agarose powder. (DO NOT overboil)

NOTE: Do not discard the remaining diluted buffer solution! This will be used in a later step.

3. Let the solution cool to 60-70°C. Carefully poor the warm solution into the casting tray and insert the comb.
4. Allow 30 minutes for this solution to solidify. Please read over the remainder of the instructions. If you need to practice pipetting, now would be the time to do so using distilled water.

Step 2: Positioning and loading the gel.

1. CAREFULLY remove the comb from the solidified gel. Place the gel into the gel box, taking care to place the end with the wells towards the negative (black) electrode. Once everyone has their gel in place, pour the remaining diluted buffer solution into the gel box. All gels should be completely submerged in the buffer.
2. If this is your first time loading a gel, you may want to practice using the blank or standard samples (20µl). Gently lower the tip of the pipet into the desired well and SLOWLY release the solution by pushing down on the plunger.
3. Load your gel with 20µl of each of the following DNA samples, taking note of the order that you load them: DNA from mother, DNA from child, DNA from male 1 (Bobby B.), DNA from male 2 (Ernie E.).
4. Cover the gel box with the lid and attach the color coded power leads. Plug in the power supply and set it to 72 volts, 200mA. This will run for approx 1 hour.
5. While the gel is running, make a diluted methylene blue solution using 5ml concentrated methylene blue and 300 ml distilled water.

6. After 1 hour, unplug the power supply and remove the gel from the casting tray using the spatula. Transfer the gel into a container with the diluted methylene blue. The gel will need to sit in this solution overnight in order to clearly see the location of the DNA bands.
7. THE NEXT DAY: Place the gel onto a piece of white paper in order to visualize the DNA bands. Draw what you see on the gel for further analysis.

Questions:

1. Who is the father of the child? How did you come to this conclusion?

2. Given that the overall charge on DNA is negative, why do we choose to position the gel loading wells at the end with the negative electrode?

3. Using your knowledge of genetics and gel electrophoresis, design and describe in detail another experiment in which this experimental method could be put to use.