AP Biology DNA Extraction (Gene in a Bottle) Lab

PURPOSE: Extract and examine DNA from human cells.

MATERIALS: (per preparation)

4 – 15ml tubes (containing 3 ml of water)

1 - Pink micro test tube of protease and salt solution

1 - 15 ml tube of lysis buffer

6 - Disposable plastic pipets

1 - Foam micro test tube holder

1 - permanent marker

1 – disposable paper cup

PROCEDURE (Be sure to note any observations in your write-up):

Steps 1 and 2: Collecting and breaking open cells

- 1. Obtain a 15 ml tube containing 3 ml of water, and label it with your initials.
- 2. Chew the insides of your mouth for 30 seconds. Collect as many cheek cells as possible by chewing the insides of your mouth for 30 seconds (do not draw blood) and then rinse your mouth with a small amount of water. Ample cell collection is critical for success. For best results, make sure you spend the recommended amount of time collecting cells.
- 3. Take the 3 ml of water from your tube into your mouth and rinse vigorously for 30 seconds. Don't swallow the water!
- 4. Carefully expel all your water mouthwash back into your 15 ml tube.
- 5. Locate the 15 ml tube at your workstation labeled "lysis". Using a fresh disposable plastic transfer pipet, add 2 ml of lysis buffer to your tube.
- 6. Place the cap back in you tube. Gently invert your tube 5 times to lyse your cells. Don't shake the tube. If you observe any changes to your cells at this time, write them down.

Step 3: Removing proteins

- Obtain the pink tube labeled "prot" and add 5 drops of protease and salt solution to the 15 ml tube containing your cell extract. Cap the cell extract tube and gently invert it 5 times to mix.
- 2. Place your cell extract tube in the beaker or test tube holder in the 50 degrees C water bath for 10 minutes to allow the protease to work.

Steps 4 and 5: Making the DNA visible

- 1. Fill a disposable transfer pipet with cold alcohol.
- 2. Tilt your 15 ml tube at a 45 degree angle and slowly add the alcohol, carefully letting it flow gently down the inside of the tube. Fill the tube with cold alcohol (about 10 ml total). You may need to use several pipets of cold alcohol. You should be able to see two layers (upper and lower) forming. As you add the alcohol, pay close attention to the place where the alcohol and cell extract layers meet. Write down you observations
- 3. Place your 15 ml tube upright either on the cup or a test tube and leave it undisturbed at room temperature for 5 minutes
- 4. After 5 minutes, look again at the contents of your tube, especially in the area where the alcohol and cell extract layers meet. Do you see anything? Write down your observations. Compare your sample with those of your classmates.
- 5. With the cap of your tube tightly sealed, mix the contents of your tube by slowly inverting the tube 5 times. Look for any stringy, white or clear material. THIS IS YOUR DNA!