INVESTIGATION 5.4.1 continued

16. Disinfect your laboratory bench using the bleach solution.
17. Wash your hands thoroughly with soap and water.

Once the experiment has been completed, flood plates with bleach to kill the bacterial colonies that have been cultured. Alternative is to place plates in an autoclave before they are disposed of.

Analysis
(b) Copy Table 1 in your notebook and use it to record your observations.

<table>
<thead>
<tr>
<th></th>
<th>MM294/pAmp + pAmp</th>
<th>MM294 − pAmp</th>
</tr>
</thead>
<tbody>
<tr>
<td>LB/amp plate</td>
<td></td>
<td></td>
</tr>
<tr>
<td>LB plate</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

(c) Compare your results to your prediction. Explain any possible causes for variation.
(d) What evidence is there to indicate that protein was synthesised by the bacteria?
(e) Why was it important to streak out both types of bacteria on both types of plates?
(f) This experiment contains both positive and negative controls. Identify them. What information do the controls provide in this experiment?
(g) Why was it important to cool the inoculating loop before obtaining a bacterial colony from a stock plate?
(h) Why was it important to resterilize the inoculating loop between transfers of bacteria?

Evaluation
(i) Suggest possible sources of error in this procedure and indicate their effect on the results.
(j) Using standard scientific format, prepare a written report.

Synthesis
(k) E. coli strains containing the genetic sequence pAmp are resistant to ampicillin. Research how the ampicillin can be deactivated by β-lactamase, the protein that the ampicillin-resistance gene codes for.
(l) Predict what would happen if there was an error in the genetic sequence that codes for β-lactamase.

ACTIVITY 5.4.1

Synthesis of a Protein: A Simulation Activity

In this activity, you will be provided with the DNA nucleotide sequence that codes for a hypothetical protein. The code will be provided to you in three fragments. You will have to transcribe the code into mRNA, remove an intron segment, and translate the mRNA into the protein. In addition, you will have to identify the beginning fragment, the middle fragment, and the end fragment.

Procedure
1. Copy each of the following sequences onto a separate piece of paper.

Sequence A
TCTTCCCTCCTAAACGTTCAACCGGTTCCTTAATCCGC
CGCCAGGGCCCGCCCTCAAGAATTTGGT

Sequence B
TCAGACGTGTGGCCCGTAACAAACTTGTTACAA
CATGGTCATAAACGTCAGAGATGGTCAATCTCTTAAT
GACT

Sequence C
TACAAACATGTAACACACCCCTCAGTGAGATTCAAC
CGCAACATAAACCACACCCGTGCAGCCGCGAAAA
GATATGG

2. Divide the sequences into triplets (codons) by putting a slash between each group of three bases.
**Protein Synthesis: A Very Close Look**

By studying electron micrographs, scientists have been able to obtain even more valuable information about numerous biochemical cellular processes. In this activity, you will examine electron micrographs that illustrate different aspects of protein synthesis. Using your knowledge of the process of protein synthesis, you will identify organelles and enzymes involved in protein synthesis.

**Procedure**
1. Examine each electron micrograph (Figures 1 to 5).

**Analysis**
(a) The electron micrograph in Figure 1 depicts an mRNA strand as it undergoes posttranscriptional modification in a eukaryotic cell. Given your knowledge of this process, identify the enzyme depicted by the large white spot.

(c) Codons 24 to 66 represent an intron. At what point in the process of protein synthesis are introns removed? What is the name of the enzyme responsible for this excision?

(b) Explain the function of the enzyme in Figure 1.

(d) The electron micrograph in Figure 2 depicts ribosomes translating an mRNA sequence. Identify the ribosomes. Can you distinguish the two subunits of the ribosome?

(e) Is this genetic sequence eukaryotic or prokaryotic? How do you know?

(f) If you worked backward, starting with the amino acid sequence of the protein, would you obtain the same DNA nucleotide sequence? Why or why not?

(g) Provide the anticodon sequence that would build this protein.

3. Transcribe the DNA into mRNA.

4. Identify the middle, end, and beginning sequence. Use your knowledge of start and stop codons to help you figure it out.

5. Remove codons 24 to 66, including codon 66.

6. Translate the mRNA into protein using the genetic code.

**Analysis**
(a) Which fragment was the beginning fragment? How do you know?

(b) Which fragment was the end fragment? How do you know?

(d) Figure 3 depicts the process of transcription. What enzyme is represented by the dark spots? Why does more than one enzyme exist on the strand of DNA?