**Literature Assignment 4 Due: Tuesday, Nov. 26, 2013**

Fast, cheap sequencing techonologies have made it possible to apply sequencing to solve clinical problems that previously were solved using other technologies. In addition, new scientific areas have opened up that were simply not approachable before the advent of next-generation sequencing technology. Some of the applications of next-generation sequencing include metagenomics, cancer biology, identification of rare polymorphisms through deep sequencing, discovery of small RNAs, investigating protein-DNA interactions when combined with chromatin immunoprecipitation (CHiP), sequencing DNA from fossils, and transcriptome sequencing.

The article you will read for this assignment discusses applications of next-gen sequencing in diagnostic medicine:

Didelot, X., Bowden, R., Wilson, D.J., Peto, T.E.A., Crook, D.W. Transforming clinical

microbiology with bacterial genome sequencing. Nature Reviews Genetics 13, 601-612

(2012).

Read this article and briefly answer the following questions. You may read additional materials, if you wish. If you do, you must cite your sources. You may not quote verbatim without attribution.

1. When a patient presents with a bacterial infection, the identification of the infecting species and testing for its antibiotic susceptibility is very slow. Why is this the case?

2. Mass spectrometry is a relatively new, and much more rapid, method being used to quickly

identify species. How is mass spec used for this purpose? What information must already

exist for this approach to work for species identification? This approach also has drawbacks.

Describe one.

3. Explain the basic experiment currently used to determine which antibiotics to use for a particular infection.

4. The authors mention two ways in which toxin production can be analyzed, serotyping and PCR. Explain how each can be used to determine toxin production.

5. What information will fast, cheap whole genome sequencing provide that can be used to determine which pathogenic species are present in an infection? How will it be used? Be sure to mention any information that must already be available for this approach to work.

6. What are two of the remaining challenges to using whole genome sequencing as a species identification strategy? For each, can next-generation sequencing help to overcome this challenge? If so, how?

7. What information will fast, cheap whole genome sequencing provide that can be used to determine the antibiotic susceptibility of an infecting bacteria? How will it be used? Be sure to mention any information that must already be available for this approach to work.

8. Why is it essential to create and maintain an up-to-date database containing genotypic de- terminants of antibiotic susceptibility and resistance?

9. What is the benfit of continuing to use phenotypic methods to determine antibiotic susceptibility, if we can sequence a bacterial infection and find genotypic determinants?

Give two reasons to support your answer.

10. How can whole genome sequence data be used to augment patient interviews in public

health studies of disease outbreak and tracking?

11. Describe how the availability of inexpensive sequencing machines in individual lab will affect

the use of whole genome sequence data for diagnostic use.