**MCB2010L –Microbiology Lab**

**Exercise 15: Bacterial Identification of Enterobacteriaceae Using EnteroPluri System**

* + The EnteroPluri is a biochemical identification system for the identification of members of the Enterobacteriaceae family.
		- Enterobacteriaceae are gram negative, oxidase negative
		- Include *E.coli, Salmonella, Yersinia, Klebsiella, Pseudomonas, Shigella*
	+ It is self-contained, compartmented plastic tube containing twelve different media that allow determination of 15 biochemical reactions.



* + Work in pairs
		- Obtain one EnteroPluri tube.
		- Inoculate the tube with unknown bacteria*.*
		- Incubate overnight and interpret all reactions in the next period- page 96-98.



**Exercise 16: Antibiotic Sensitivity Using the Kirby-Bauer Method**

* + Once the pathogen has been identified, the physician will select an appropriate drug for the effective treatment of the disease.
	+ The Kirby-Bauer is a highly standardize disc diffusion method that is approved for wide use in clinical laboratories.
	+ Work in pair; procedure – page 102
		- Obtain two Muller-Hinton agar plates.
			* Why Muller-Hinton? This agar grows a variety of microbes. Also detoxifies toxins that the bacterial release which might alter antibiotic results.
		- Inoculate one plate with *Pseudomonas aeruginosa* and the other plate with *Staphylococcus aureus.*
		- Place BBL discs using dispenser.
		- Incubate the plates overnight
		- Examine plates and measure zones of inhibition in mm and compare with the chart provided – page 104-105
			* Just because there is a zone of inhibition does not mean that the bacteria is sensitive to the antibiotic – it all depends on the species and the antibiotic (that’s why it’s important to measure the zone and compare to the chart)



**Exercise 17: Differentiation of *Staphylococcus* Species**

* + *S. aureus* is the most common species to cause *Staph* infection.
	+ *S. aureus* is catalase positive which distinguishable from *Enterococci* and *Streptococci*.
	+ Disease associated strains produce toxins, and express cell-surface proteins (Protein A) that bind and inactivate antibodies.
	+ *S. aureus* also produces coagulase, an enzyme that is a clotting factor. The fibrin clot allows *S. aureus* to wall itself off and making it harder to detect by immune system.
	+ Coagulase and Staphyloslide test are performed to differentiate S. aureus from other Staphylococci
	+ Work in pairs; procedure – page 108-109
		- Staphyloslide test - Inoculate the *S. aureus* and *S. epidermidis* by mixing with test latex reagent*.*
			* If agglutination occurs, that means that Protein A is present on the bacteria
		- Coagulase test - Inoculate the *S. aureus* and *S. epidermidis* into rabbit plasma tubes.
			* The plasma will coagulate if the bacteria is producing coagulase