CHAPTER V

SUMMARY AND FURTHER STUDIES

SUMMARY

It was found from the study that the maximum level of SLO production by C 203 S Streptococcus pyogenes strain of the A group in Todd Hewitt broth was achieved when

1. the age of the starter was about 10 - 12 hr
2. the size of the starter was 15 % of fresh medium
3. the initial pH of the medium in the step of SLO preparation was 7.8 which is the approximate pH of this medium without adjusting
4. the incubation temperature in the step of SLO preparation was 37°C
5. the incubation period in the step of SLO preparation was 4 hr
6. the culture in the step of SLO preparation was incubated in atmospheric carbondioxide at standstill.

The optimum conditions found from this study which are different from the conditions in the method proposed by Vejjajiva in 1970 (68, 69) are the size and age of starter, and the incubation period in the step of SLO preparation. These various conditions have been changed from 10 % of fresh medium, 18 hour-preculture and 18 hour-incubation-period to 15 % of fresh medium, about 10 - 12 hour-preculture and 4 hour-incubation-period, respectively.

The improvement of the method for SLO production, in Todd Hewitt broth by Streptococcus pyogenes (group A) C 203 S, according to various optimum conditions found from this study is satisfactory. It not only increases the level of SLO gained but also saves time, is economical and is still quite easy to carry out at any average laboratory.

Further Studies

1. Several investigators had stressed the importance of reducing agents for SLO formation, such as Slade and Knox (42) who suggested that an optimum reducing potential was required for maximum lysin production, and Fuvessy,et al. (43) who found that SLO production was markedly increased by the combination of cysteine and ascorbic acid. It would be interesting to study the effects of various kinds and amounts of reducing agents for SLO production by the strain C 203 S

2. It was proved by Boszormenyi,et al. (44) that a glucose concentration of about 1.0 % and neutralized pH, maintained during the entire cultivation period, were found to be optimum for SLO production by H 64 A Streptococcus pyogenes strain of the C group. It would be of great interest to study in the same way, but using the strain C 203 S of the A group, possibly in continuous culture, in which it is easier to control pH and glucose concentration, instead of in a batchwise culture.

3. Studying the cellular location of SLO by Calandra and Theodore (38) revealed that most of the SLO activity in strain

C 203 S was localized in the periplasm (the region between cell wall and cell membrane) and did not differ in several characteristics from that of extracellular SLO. Grinding or sonic treatment in order to lyse streptococcal cells after the final step of incubation might result in a higher level of SLO in the cultural filtrate. This matter would be of great interest to analyze.