

### Summary Points

**Sterilization without autoclave can be controlled by  $\text{Ca}(\text{ClO})_2$**

- Treatment with 30ppm  $\text{Ca}(\text{ClO})_2$  had complete sterilization effects
- Y1 can grow at 12h after treatment with  $\text{Ca}(\text{ClO})_2$

**Y1 was inoculated in 500L incubator for investigation of growth conditions.**

**Y1 can grow in BB medium which was already treated with 2%  $\text{H}_2\text{O}_2$  without autoclave.**

### •Motivation/Introduction

•The consumption of many microbial pesticides is increasing to replace the problems of chemical pesticides but, there are various types of limitations for the mass production of the biocontrol agents such as the expensive incubating facilities, the difficulties to the supplement of the microbial products and application to the field. The cost-effective incubator for the mass production of bacteria should be considered to replace the expensive incubating facilities.

### •Objectives

•To develop the low-cost incubator for large scale cultivation of bacteria and to replace expensive media without autoclave by using low-cost media with  $\text{Ca}(\text{ClO})_2$  for sterilization.

### •Methodology

• $\text{Ca}(\text{ClO})_2$  was used for sterilization of soil bacteria without autoclaving.

•Two type of mass cultivation experiments were done by using BlueBrown (BB) medium treated with 2%  $\text{H}_2\text{O}_2$  and BB medium treated with 40ppm  $\text{Ca}(\text{ClO})_2$  in 500L incubator under non autoclave condition.

•Co-inoculation of *B. amyloliquefaciens* Y1 and *B. thuringiensis* KKY were done in 500L incubator containing BB medium treated with  $\text{H}_2\text{O}_2$ .

•Mass cultivations in other fields (Hwasun and Yeonggwang) were conducted with 500L rubber incubator in and with 15,000L iron incubator in Gwangju Agriculture Technology Center, South Korea.

### Results

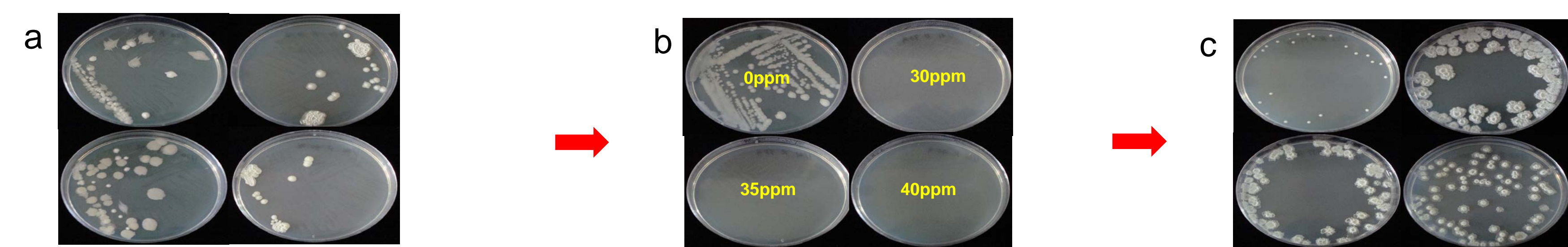


Fig 1. Growth of soil bacteria before sterilization with  $\text{Ca}(\text{ClO})_2$  (a), effect of different  $\text{Ca}(\text{ClO})_2$  concentration (0, 30, 35 and 40 ppm) on the growth of soil bacteria (b) and *B. amyloliquefaciens* Y1 inoculation after sterilization with  $\text{Ca}(\text{ClO})_2$  (c).

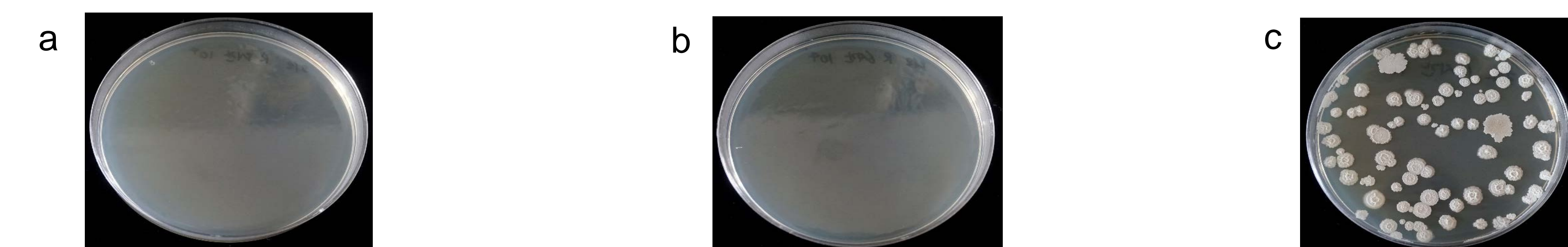


Fig 2. Growth of Y1 at different time after sterilization with  $\text{Ca}(\text{ClO})_2$  3h (a), 6h (b), 12h (c).

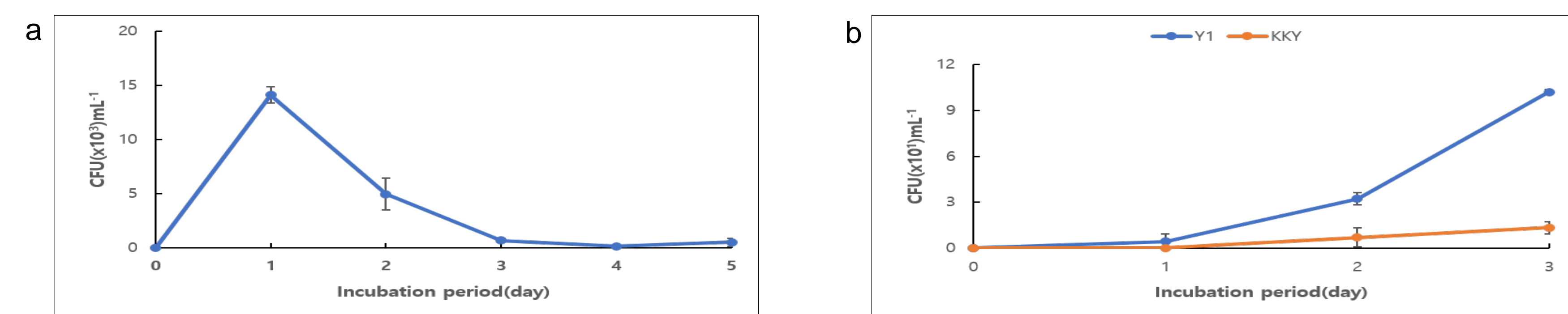


Fig 3. Growth curve of *B. amyloliquefaciens* Y1 (a) and Co-inoculation of *B. amyloliquefaciens* Y1 and *B. thuringiensis* KKY (b) in 500L rubber incubator at 40°C with non-autoclaved BB medium.



Fig 4. Investigation of Y1 growth from mass cultivation of 15ton L incubator in Gwangju Agriculture Technology Center, South Korea.



Fig 5. Investigation of Y1 growth from mass cultivation of 500L incubator in Hwasun (a) and Yeonggwang (b), South Korea.

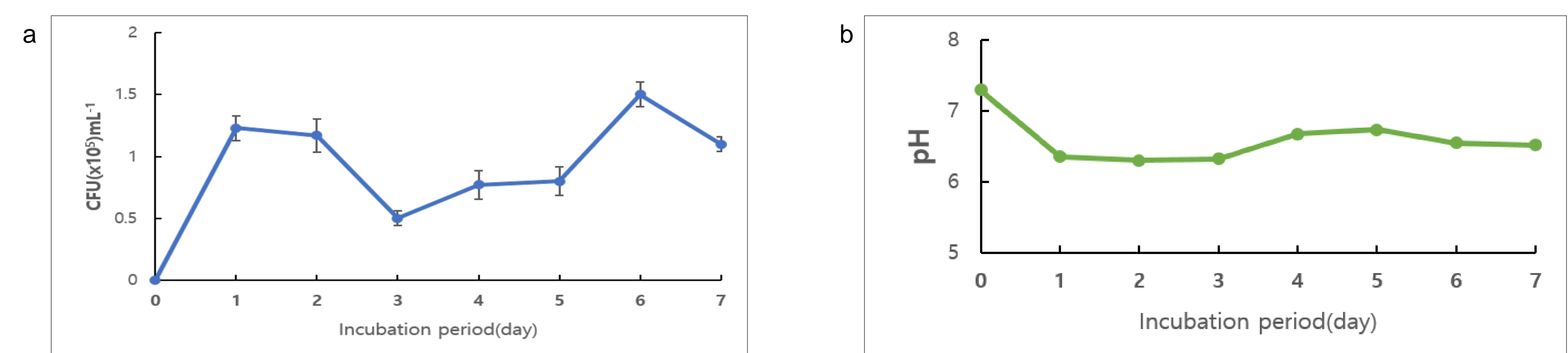


Fig 6. Growth curve (a) and changes of pH (b) of *B. amyloliquefaciens* Y1 in 500L Incubator for at 45°C 7days, by using non autoclaved BlackWhite (BW) medium

### •Conclusion

•This study demonstrated that the low-cost incubator and medium can be used for Y1 culture without autoclaving at the agricultural fields and  $\text{Ca}(\text{ClO})_2$  can be used for sterilization.

### •Acknowledgment

•This research was supported by iPET (Korea Institute of Planning and Evaluation for Technology in Food, Agriculture, Forestry and Fisheries) through Agri-Bio industry Technology Development Program, funded by Ministry of Agriculture, Food and Rural Affairs (MAFRA) (316032-5).

