

# Effect of adding Agricultural and Organic Lime on Survival Rate of *E. coli* and General Bacteria in Farmland Soils in Kampong Cham Province, Cambodia

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**Abstract:** The farmers of Kampong Cham Province applied manures and compost to their farmlands soil as fertilizer or soil amendment. The application of fresh manures or immature compost to farmlands have been notified as one of the main sources that spread pathogenic bacteria *Escherichia coli* (*E. coli*) to the soil. *E. coli* is recognized as a hazardous microorganism in the environment and for public health. It is an important pathogen associated with several foodborne and waterborne outbreaks of gastrointestinal illness, which has been widely reported in Cambodia and other countries around the world in the last decades. Therefore, the contamination of farmlands' soil, surface water, irrigation water and fresh vegetables, can become a reservoir of infection. However, most of the previous study has shown that soil pH is one of the dominant factors affecting the survival rate of microorganisms in the soil.

## INTRODUCTION

In agriculture, lime is usually defined as calcium or calcium-magnesium containing compounds capable to reduce harmful effects of acid soil by neutralizing soil acidity and raising soil pH.



Fig.1 Application of agricultural lime to farmlands' soil

Lime and fertilizers are common amendments that are routinely applied to farmlands' soil. Their effects on soil fertility and plant nutrient are well known but their application may well have beneficial effect on other soil properties such as physical, chemical and biological condition.

Manure or compost manure are working as the sources of organic fertilizer and were used by farmers in Kampong Cham Province. Manures were generated by cattle and stored in piles for composting. However, in raw or fresh manure may contain pathogens and this pathogen can be spread to the soil with improper treated or well decomposed.

## Objectives

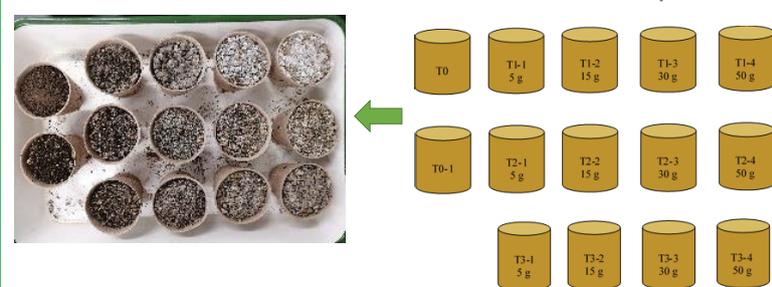
To investigate the effect of different concentration of  $\text{CaCO}_3$  added to the soil

To analyze the relation of  $\text{CaCO}_3$  with the survival of *E. coli* and other microorganisms in the soil

## MATERIALS AND METHODS

### Experimental designed

Pots experiment were conducted in the experimental room at Tokyo University of Agriculture from November to December in 2020 (winter season). Agricultural limestone powder, eggshell, and clam shell were used as the sources of  $\text{CaCO}_3$ . Eggshell and clam shell were crashed into powder by using grinder and added to the soil with the ratio of 0, 5, 15, 30 and 50 g in 200 g of soil in 4.1% of water content.



## RESULTS AND DISCUSSION

### The effect of $\text{CaCO}_3$ on soil chemical properties

The effectiveness of agricultural and organic lime ( $\text{CaCO}_3$ ) on soil pH has been observed by applying 0, 5, 15, 30 and 50 g of crushed  $\text{CaCO}_3$ . From the laboratory works, the increasing amount of  $\text{CaCO}_3$  had given an effect on soil pH for each sample. From the observation, the amount of  $\text{CaCO}_3$  that has been applied from 5 g to 50 g has shown some increase on soil pH ( $P < 0.01$ ) and soil EC was decreased ( $P < 0.05$ ) with the increase of  $\text{CaCO}_3$  concentration (Fig. 2).

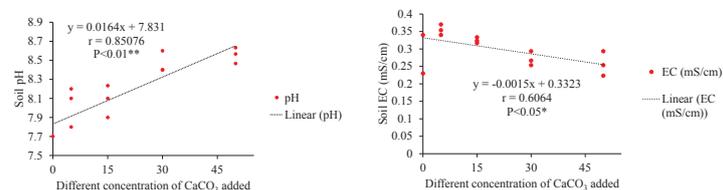


Fig.2 Effect of  $\text{CaCO}_3$  on soil pH and EC

### The relation of $\text{CaCO}_3$ with soil chemical properties and the survival of microorganisms

The correlation of agricultural and organic lime ( $\text{CaCO}_3$ ) on soil chemical properties and the survival rate of coliform and *E. coli* was observed, the result was indicated that the amount of agricultural and organic lime ( $\text{CaCO}_3$ ) has a positive effect on the increase of soil pH ( $P > 0.01$ ),  $\text{K}^+$  and the survival rate of coliform ( $P > 0.05$ ), however, for the survival rate of *E. coli* was not observed with different concentration of  $\text{CaCO}_3$  added. In addition to the survival of coliform in (Fig. 3) showed that the coliform was increased with the amount of agricultural lime added and soil pH. Due to this result, it was discussed that in the acidic soils, coliform or *E. coli* could not be detected by 7 days after inoculation and in neutral soils, *E. coli* declined much slowly than in acidic soil and the survival for up to 38 days (Taoning et al., 2013).

	Lime ( $\text{CaCO}_3$ )	pH	EC (mS/cm)	$\text{K}^+$	$\text{Na}^+$	$\text{NO}_3^-$	Coliform	<i>E. coli</i>
Lime ( $\text{CaCO}_3$ )	1							
pH	0.819955**	1						
EC (mS/cm)	-0.25302	-0.13324	1					
$\text{K}^+$	0.523772*	0.516752*	0.561051*	1				
$\text{Na}^+$	0.018821	0.121048	0.705255**	0.729967**	1			
$\text{NO}_3^-$	0.024293	0.127166	0.271418	0.393883	0.670483*	1		
Coliform ( $10^4$ cfu/g)	0.615472*	0.505633*	-0.028769	0.238222	0.038135	0.078039	1	
<i>E. coli</i> ( $10^4$ cfu/g)	0.003116	-0.224447	0.2357958	0.400165	0.487927	0.273015	-0.24922	1

Fig.3 Correlation matrix of  $\text{CaCO}_3$  and soil properties

n = 14

$r > 0.497309$  significance level is 0.05

$r > 0.622591$  significance level is 0.01

\*\* $P < 0.01$ , \* $P < 0.05$

## CONCLUSION

The probability of *E. coli* outbreaks due to food contamination is directly linked to its survival in secondary environments such as soil, which is in turn dependent on soil characteristics (Zhang et al., 2013). In this study the attention was focused on the effect of adding different rate of  $\text{CaCO}_3$  on the soil properties and the survival rate of *E. coli*. The results were showed that soil pH was increased with different concentration of  $\text{CaCO}_3$ , and the survival rate of coliform was detected with the increased of soil pH in a short time. However, the study will continue to investigate the survival times of *E. coli* and the times that soil pH decreasing after added  $\text{CaCO}_3$ .