

Anaerobic Lignocellulolytic Rumen Bacterium could Reduce Theobromine in Liquid Media Containing Rumen Fluid (Preliminary Study)

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In 2009, Indonesia produced around 195,471.25 tons of cacao and 637,406.25 tons of pod cacao as a waste. Pod cacao contains 50.8% of energy (TDN) and 8% of crude protein, thus it is potential to be used as ruminant feed. However, its utilization as conventional feed resource is limited due to the present of theobromine that is toxic and harmful to the animal even at very low dose. This work was done to evaluate the effect of anaerobic lignocellulolytic rumen bacterium to reduce the theobromine content under liquid media. Anaerobic lignocellulolytic rumen bacterium was provided by the UGM Nutritional Biochemistry Laboratory. Liquid media consisted of rumen liquor (40%), yeast extract (0.5%), sucrose (1%), and mixture of minerals (K_2HPO_4 , KH_2PO_4 , $(NH_4)_2SO_4$, $MgSO_4$, $CaCl_2$). The bacterium was grown in the liquid medium containing 100 ppm of theobromine for 6 days under anaerobic condition. Samples were collected at 0, 3, and 6 days and analyzed for theobromine using HPLC (Lichrosorb RP 18 25cm, eluent MeOH:H₂O 40:60). Results indicated that theobromine content decreased to 68.05 ppm after 3 days incubation. However, further incubation did not reduce the theobromine content in the medium. It showed that the anaerobic lignocellulolytic rumen bacterium could reduce the theobromine content in the medium up to 26%. To confirm whether the reduction of theobromine was due to attachment to the cells or degradation of theobromine, cells of the anaerobic lignocellulolytic rumen bacterium were suspended in liquid medium containing around 150 ppm of theobromine for 10 minutes and then removed by centrifugation. The theobromine content in the medium did not decrease due to attachment to the cells. Further research should be done to evaluate the possibility of those bacteria as the biological agent for detoxifying theobromine present in the pod cacao.

Key Words: Theobromine, Anaerobic lignocellulolytic bacteria, Liquid media, Pod cacao