SHORT CHAIN FATTY ACID (SCFA) PROFILE PRODUCED BY Clostridium butyricum GROWN ON MEDIUM CONTAINING TYPE 3 RESISTANT STARCH (RS3) OF SWEET POTATO

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ABSTRACT

RS type 3 (RS3) is retrograded starch, which is not digested by human starch degrading enzyme and will undergo bacterial fermentation in the colon. The main fermentation products are the short chain fatty acid (SCFA) acetate, propionate and butyrate. Butyrate arising from microbial fermentation is important for the energy metabolism of colonic epithelial cells and has a mainly protective role in relation to colonic disease. The purpose of this research was to study short chain fatty acid profile produced by Clostridium butyricum BCC B2571 isolated from human faeces during fermentation in medium containing RS3 from sweet potato (Ipomoea batatas). Pullulanase enzyme (34,779 U/mg protein) at 5% starch weight was used to hydrolyze α-1,6-glucosidic bonds and the incubation was carried out at 24 hours at 50°C, stored at 4°C at 24 hours to trigger retrogradation, and then dried by spray drier to produce resistant starch type 3. RS3 was fermented in reinforced clostridial medium by Clostridium butyricum BCC B2571 isolated from human faeces. In vitro fermentation resulted in short chain fatty acid (SCFA) at different levels depending on the RS and the concentrations of glucose in the medium. Fermentation of 20 g/l (RS3) and 1 g/l (glucose) resulted in SCFA with the molar ratio of acetate, propionate, and butyrate of: 50 mM : 37 mM : 68 mM (1.3:1:1.8) at 12 h fermentation whereas fermentation of 10 g/l (RS3) and 5 g/l (glucose) resulted in SCFA with the molar ratio of acetate, propionate, and butyrate of: 453 mM : 248 mM : 225 mM (2:1.1:1) at 48 h fermentation. This result indicated that longer fermentation time and higher glucose concentration could increase butyric acid level but there was metabolic shift to higher formation of acetic acid.

Keywords: resistant starch, butyrate, short chain fatty acid, Clostridium butyricum
Gastrointestines is an important part of body metabolism which affects human health. One of the diseases which attack gastrointestines is colorectal cancer (CRC) which can be prevented by proper lifestyle and consumption of healthy functional food. SCFA are the main products of anaerobic microbial fermentation in the large intestine and affect colonic health by providing energy to the epithelial cells. The organic acids produced in the colon are acetic acid, propionic acid, and butyric acid. Interest in butyrate role as a possible protective agent has arisen from its anti-proliferative effects on cells in vitro including colon tumour cell lines.

Resistant starch (RS), is retrogradaded starch that escapes digestion in the small intestine and can be fermented in colon by anaerobic bacteria producing SCFA which contributes to the gastrointestinal health, and thus can be classified as functional food. The crystalline non-granular starch (RS3) can be obtained from many starchy sources, such as rice, corn, maize, wheat, sago, banana, potato, and sweet potato. In this study, jago sweet potato used contain 31% starch content. Jago sweet potato is usually consumed indirectly as the taste is not preferable.

The objective of the research was to study short chain fatty acid profile produced by Clostridium butyricum BCC B2571 isolated from human faeces during fermentation of type 3 resistant starch of sweet potato (Ipomoea batatas). This research was divided into two stages, (1) chemical analysis and (2) fermentation of type three resistant starch by Clostridium butyricum.

The result of proximate analysis showed that jago sweet potato starch contained water 13.73%, protein 0.44%, ash 0.23%, fat 0.56%, and carbohydrate 85.04%. Amylose content of jago sweet potato starch was 25.83%. The resistant starch was made by applying pullulanase enzyme (34,779 U/mg protein) to hydrolyze the α-1,6-glucosidic bonds and the incubation was carried out at 24 hours at 50°C, stored at 4°C at 24 hour to trigger retrogradation, and then dried by spray drier to produce resistant starch type 3. Resistant starch content of jago sweet potato treated with pullulanase enzyme was 28.15%. In vitro fermentation of jago sweet potato resistant starch by Clostridium butyricum BCC B2571 resulted in short chain fatty acid (SCFA) at different levels depend on the RS and the concentration of glucose in the medium. Fermentation of 20 g/l (RS3) and 1 g/l (glucose) resulted in SCFA with the molar ratio (acetate:propionate:butyrate) 50 mM : 37 mM : 68 mM (1.3:1:1.8) at 12 h fermentation, whereas fermentation of 10 g/l (RS3) and 5 g/l (glucose) resulted in SCFA with the highest molar ratio (acetate:propionate:butyrate) 453 mM : 248 mM : 225 mM (2:1.1:1) at 48 h fermentation. This result indicated that longer fermentation time and higher glucose concentrations could increase butyric acid levels but there was metabolic shift to higher formation of acetic acid.