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Effect of medium supplementation on very high gravity bioethanol fermentation using sago *hampas* hydrolysate as a feedstock

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Abstract. Bioethanol fermentation under very high gravity (VHG) conditions by using sago *hampas* hydrolysate (SHH) supplemented with yeast extract as a fermentation medium has resulted in an incomplete metabolism of glucose, leading to relatively low bioethanol production in comparison to the theoretical titre. Therefore, additional supplementation of the fermentation medium is necessary to increase the yeast tolerance towards inhibitors and high concentration of glucose and bioethanol. This work investigates the effect of supplementing SHH media with various nutrients on bioethanol fermentation under VHG conditions. The nutrients included magnesium sulphate (0.12 g/L), urea (3 g/L), glutamic acid (5 g/L), and peptone (5 g/L). Our results showed that culture supplemented with peptone has significantly improved the yeast growth by 0.9-fold and glucose consumption efficiency by 10% compared to the control cultures. Besides that, the media formulation has also increased bioethanol production by 13%, with a maximum concentration of 126.20 ± 3.0 g/L. In general, the results suggest an improved formulation of fermentation medium consisting of SHH for bioethanol production under VHG conditions. These results will provide useful insights into the development of bioethanol production from sago-based feedstock in the future.

Keywords: bioethanol, sago *hampas*, supplement, very high gravity fermentation, yeast

INTRODUCTION

Very high gravity (VHG) fermentation technology, which uses high concentration of sugar (≥250 g/L), is often selected as the process to generate bioethanol. This is because the VHG technology has several advantages in meeting industrial demand, such as low energy and cost, less likelihood of bacterial contamination, and elevating bioethanol production (Bvochorá *et al.,* 2000; Bayrock & Ingledew, 2001; Bai *et al*., 2008). Numerous studies have highlighted the success of VHG technology in the production of the first generation of bioethanol (Nuanpeng *et al*., 2011; Kawa-Rygielska & Pietrzak, 2012; Khongsay *et al*., 2012). Nevertheless, the application of VHG for the production of second-generation bioethanol remains under-explored.

Second-generation bioethanol, which refers to bioethanol produced from lignocellulosic materials (LCMs), is considered a promising substitute for bioethanol derived from crops such as corn and sugarcane (García *et al.,* 2014). Effective pretreatment of LCM is crucial in achieving an efficient bioethanol production from lignocellulose as this process could enhance the accessibility of cellulose and thus, improves the enzymatic saccharification process (Singh & Bishnoi, 2013; Buruiana *et al*., 2014). Unfortunately, during the pretreatment step,

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