Fermentasi Semisolid Sampah Sisa Makanan untuk Produksi Biogas dan Analisis Biodiversitas Mikroba yang Berperan Berdasarkan Profil DGGE (*Denaturing Gradient Gel Electrophoresis)*

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ABSTRAK

Produksi sampah Kota Bandung mencapai 7.500 m^3 setiap hari dengan kandungan sampah organik mencapai 63,56%. Bagian penting dari sampah organik tersebut adalah sampah sisa makanan yang secara rutin diproduksi di wilayah pemukiman dan sentra kuliner di perkotaan. Sampah organik menyumbang beban cemaran terhadap lingkungan karena mengandung COD (*chemical oxygen demand)* yang tinggi. Fermentasi anaerob dilaporkan mampu mengolah beban cemaran organik hingga 80 kgCOD/m^3-hari dibandingkan dengan proses aerob yang hanya mampu mengolah beban cemaran organik kurang dari 1 kgCOD/m^3-hari. Disamping itu, fermentasi anaerob juga berpotensi menghasilkan biogas. Pada penelitian ini dilakukan fermentasi anaerob sampah sisa makanan dengan penambahan inokulum yang berasal dari keluaran reaktor biogas pengolah sampah pasar organik. Fermentasi anaerob dioperasikan pada suhu ruang secara *batch* dan semikontinyu satu tahap, yaitu semua tahapan hidrolisis-asidogenesis-asetogenesis dan metanogenesis berlangsung dalam satu reaktor. Fermentasi *batch* menggunakan substrat sampah sisa makanan dengan kandungan padatan total sebesar 20%. Fermentasi *batch* dioperasikan selama 91 hari dengan variasi penambahan zeolit serbuk sebanyak 0%, 10% dan 15% (v/v) berturut-turut pada reaktor (SI), (SIZ-10%) dan (SIZ-15%). Reaktor kontrol positif berisi inokulum (I) dan kontrol negatif berisi substrat (S). Produksi biogas total terbanyak yaitu 25.820 mL dan laju 11,84 mL/jam dengan kandungan metana tertinggi sebesar 33% pada hari ke-42 dihasilkan oleh reaktor (SIZ-15%). Peningkatan kandungan metana didahului oleh adanya kandungan hidrogen yang tinggi, diduga metanogen hidrogenotrofik berperan penting dalam fermentasi semisolid sampah sisa

makanan. Degradasi bahan organik total dari reaktor (I), (SI), (SIZ-10%), (SIZ-15%), (S) berturut-turut sebesar 39%, 24%, 37%, 41% dan 32%. Kondisi pH reaktor dengan penambahan zeolit relatif lebih stabil dibandingkan reaktor tanpa penambahan zeolit. Reaktor semikontinyu satu tahap dioperasikan dengan substrat sampah sisa makanan yang mengandung padatan total 14% dengan penambahan zeolit berukuran -10/+18 *mesh* sebanyak 15% (v/v) reaktor. Proses fermentasi diawali tahap aklimatisasi selama 58 hari dan dilanjutkan tahap semikontinyu dengan pemberian umpan harian sampah sisa makanan sebanyak 60,5 kgCOD/m^3-hari (OLR) dan HRT 20 hari. Pada tahap aklimatisasi, dihasilkan biogas total tertinggi yaitu 8 liter/hari dengan kandungan metana 54% pada hari ke-10 fermentasi. degradasi bahan organik total dan COD terlarut sebesar 30% dan 47%. Setelah dioperasikan semikontinyu, padatan total yang tersisihkan sebanyak 2% per hari dan bahan organik total tersisihkan 4% per hari. Terjadi penumpukan kandungan bahan organik yang disertai penurunan kandungan metana hingga menjadi 2% pada akhir operasional semikontinyu. Analisis mikroba yang berperan selama proses fermentasi menggunakan metode molekular yang tidak bergantung kultivasi. Fragmen gen 16S rRNA dari sampel (I) *fresh* dan t_{23}, (SIZ-15%) t_{23} t_{42} t_{86} dan reaktor semikontinyu (SK) t_{11} t_{55} t_{80}, diamplifikasi dari DNA total menggunakan sepasang primer yang diinkorporasi dengan 40 basa GC *clamp*, sehingga diperoleh amplikon ± 450 *bp* yang telah dikonfirmasi dengan gel agarosa. Pita-pita fragmen DNA tersebut selanjutnya dipisahkan dengan metode DGGE. Diperoleh sejumlah 12, 10, 8,7,8, 9, 11 dan 10 pita DGGE berturut-turut dari sampel (I) *fresh*, (I) t_{23}, (SIZ-15%) t_{23}, (SIZ-15%) t_{42}, (SIZ-15%) t_{86}, (SK) t_{11}, (SK) t_{80} dan (SK) t_{55} yang cenderung jelas terlihat secara visual. Perbedaan pola migrasi pita menunjukkan adanya perbedaan biodiversitas mikroba pada tiap tahapan fermentasi dan perubahan kelimpahan mikroba terlihat dari perubahan ketebalan pita DGGE. Selanjutnya perlu dilakukan penentuan urutan nukleotida dari masing-masing pita fragmen gen 16S rRNA tersebut untuk mengetahui jenis-jenis mikrobanya. Sampah sisa makanan dapat dijadikan substrat pembuatan biogas melalui fermentasi semisolid. Penambahan zeolit dapat mempertahankan kondisi pH reaktor sehingga berpengaruh dalam peningkatan produksi biogas total dan kandungan metana. Penambahan zeolit 15% (v/v) relatif lebih baik dibandingkan 10% (v/v). Metanogen hidrogenotrofik diduga berperan penting dalam produksi metana dengan substrat sampah sisa makanan. Perubahan profil pita DGGE menggambarkan adanya perubahan biodiversitas mikroba pada tahap-tahap fermentasi semisolid sampah sisa makanan.

Kata kunci: fermentasi semisolid, sampah sisa makanan, *batch*, semikontinyu satu tahap, zeolit alam, HRT, OLR, gen 16S rRNA, analisis DGGE

Semisolid Fermentation Of Food Waste for Biogas Production and Analysis of Involved Microbial Biodiversity Evaluated by DGGE *(Denaturing Gradient Gel Electrophoresis)* Profiles

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ABSTRACT

Garbage production in Bandung city is 7.500 m^3 each day with organic waste content about 63.56%. One of an important part of organic waste is food waste which routinely produced from resident and culinary area in cities. Organic waste gives pollution load to environment because it contains high COD (chemical oxygen demand). Anaerobic fermentation was reported has the ability to digest organic waste up to 80 kgCOD/m^3-day while aerobic process abilities only less than 1 kgCOD/m^3-day. Besides, anaerobic fermentation is potential for biogas production. In this research, organic waste from food waste was fermented anaerobically with inoculum addition from effluent biogas reactor which proceed organic waste from traditional market. Anaerob fermentation is operated in room temperature, using batch and one stage semicontinuous, which all steps (hydrolysis, acidogenesis, acetogenesis and methanogenesis) took place in one reactor. Batch fermentation utilized food waste as substrate with total solid content 20% for 91 days. It used 3 kinds variation of zeolite powder addition (v/v), i.e.: 0% in reactor (SI), 10% in reactor (SIZ-10%) and 15% in reactor (SIZ-15%). The positive control reactor was filled with inoculum (I) and the negative control reactor was filled with substrate (S). The highest total biogas production was 25.820 mL, rate 11.84 mL/hour, with the highest methane content 33% in day 42 (SIZ-15% reactor). The hydrogenotrophic methanogens were assumed have an important role for methane production. This is supported by the fact that hydrogen content increased before methane production. The degradation of total organic content from reactor (I), (SI), (SIZ-10%), (SIZ-15%) and (S) were 39%, 24%,

37%, 41% and 32% respectively. pH measurement showed that reactor with zeolite addition has more stabile pH than without zeolite one. The one stage semicontinuous reactor was operated with food waste substrate containing total solid 14% and zeolite addition size -10/+18 mesh as much as 15% (v/v) reactor. It was initiated with acclimatization for 58 days before continued by semicontinuous operation for 25 days with OLR 60,5 kgCOD/m^3-day and HRT 20 days. In acclimatization process, the highest total biogas was 8 L/days with methane content 54% in the 10^{th} day fermentation. The average of soluble COD removal was 47% and degradation of organic content was 30%. During semicontinuous operation, the removal of total solid and organic content were 2% and 4% per day. In the end of operation, organic material accumulated with methane decrease until 2%. Microbe that involved in the fermentation characterized with molecular method. The 16S rRNA gene fragment from sample ((I), (SIZ-15%) t_{23} t_{42} $t_{86)}$ and (SK) t_{11} t_{55} t_{80}) were amplified from total DNA. This was conducted using a pair primer which incorporated with 40 base GC clamp, until ± 450 bp amplicon was obtained (confirmed by agarose gel). The DNA fragment bands was subsequently separated with DGGE method. The result visually showed numbers of band from sample (I) *fresh*, (I) t_{23}, (SIZ-15%) t_{23}, (SIZ-15%) t_{42}, (SIZ-15%) t_{86}, (SK) t_{11}, (SK) t_{55} dan (SK) t_{80} were 12, 10, 8, 7, 8, 9, 11 and 10, respectively. The difference in band migration pattern indicated the change of microbe diversity while the thickness indicated its abundance. Subsequently, it was necessary to determine the nucleotides sequence from each bands in 16S rRNA gene fragment to identify the microbes. As conclusion, biogas from food waste was successfully produced through semisolid fermentation. Zeolite addition can maintaining pH so reactor with zeolite can produced more biogas and methane content. Addition of 15% zeolite (v/v) was better than 10%. The hydrogenotrophic methanogens assumed have an important role for methane production from semisolid fermentation of food waste.The difference of DGGE band indicated the change of microbial diversity at semisolid fermentation phase.

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Key words: semisolid fermentation, food waste, batch, one stage semicontinuous, natural zeolite, HRT, OLR, 16S rRNA gene, DGGE analysis