

## Optimasi produksi enzim mannanase dari *kitasatospora sp.* pada limbah biomassa palm kernel meal (PKM) = Production optimization of mannanase enzyme from *kitasatospora sp.* through palm kernel meal (PKM) waste biomass.

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### Abstrak

Palm Kernel Meal (PKM) merupakan by-product pengolahan minyak kernel sawit yang jarang dimanfaatkan. Penggunaan masih terbatas sebagai campuran pakan ternak dengan komposisi 3-5% dari total pakan. Hal ini disebabkan tingginya serat kasar mencapai kisaran 20-40% dari berat kering PKM. Padahal, PKM mengandung komposisi mannan dengan kandungan total 45-56% pada jaringan hemiselulosa. Pemanfaatan lebih lanjut biomassa PKM sebagai substrat produksi -mannanase dan menghasilkan turunan manooligosakarida (MOS) sangat menjanjikan. Konsentrasi PKM sebagai substrat, pH, suhu inkubasi, dan penambahan mikropartikel Al<sub>2</sub>O<sub>3</sub> sebagai faktor penentu simulasi RSM. Kondisi operasi terpilih menjadi basis scale-up produksi fermentor batch kapasitas 1,5 L dengan pengaruh laju aerasi 1,0 vvm. Disamping, itu dilakukan estimasi parameter kinetika pertumbuhan *Kitasatospora sp.* produksi aktivitas -mannanase dan substrat PKM terkonsumsi dengan asumsi model Logistik, Luedeking-Piret dan Modified Luedeking-Piret. Titik optimum yang diperoleh dilanjutkan dengan purifikasi parsial. Setelah itu, hidrolisis PKM dilakukan untuk mengamati sinergisitas pelarut NaOH-HCl dengan enzim sebagai ekstraktor turunan mannan. Simulasi menunjukkan 3% (w/v) PKM, pH 6,5, suhu 34 °C, dan 0,2% (v/v) Al<sub>2</sub>O<sub>3</sub> merupakan kondisi terpilih untuk scale-up. Pada fermentor 1,5 L setelah melalui pemurnian parsial, diperoleh aktivitas tertinggi 44,34 U/mL, laju aktivitas 0,302 U/mL-1 jam-1, konsentrasi delta gula total (S) 39,17 gr/L, dan SUY 78,15%. Estimasi kinetika dari fitting model terukur  $\mu_{max}$ ,  $X_0$ ,  $X_{max}$ ,  $\lambda$ ,  $Z$ , dan secara berurutan adalah 0,0492 jam-1; 0,435 gr/L; 8,93 gr; 0,085 U/mgX.jam; 4,467 U/mgX; 0,026 grS/grX.jam; dan 4,328 grS/grX. Adapun hasil hidrolisis zona bening dan TLC menunjukkan kemampuan -mannanase yang disintesis *Kitasatospora sp.* menghasilkan turunan MOS yang didominasi mannobiosa (M<sub>2</sub>), dengan 72 jam pembentukan dari bantuan pelarut.

.....Palm Kernel Meal (PKM), called as by-product from palm kernel oil processing, which is rarely being utilized. The usage is limited as livestock feed's blend with composition accounted only 3-5% off from total feed. The problem lies on the high content of crude fibres, up to 20-40% of PKM's dry matter. Meanwhile, PKM contains relatively high mannan comprises around 45-56% from hemicellulose's tissue. Further application of PKM biomass as substrate for -mannanase production and resulting any derivatives of manooligosaccharides (MOS) are very promising. Substrate, initial pH, incubation temperature, and additional microparticle Al<sub>2</sub>O<sub>3</sub> were determined as independent factors for RSM simulation. The chosen condition was used for scaled-up through 1,5 L stirred tank-bioreactor batch, 1,0 vvm aeration rate. The kinetics parameters of *Kitastospora sp.* growth, enzyme production and substrate consumption were estimated through Logistic, Luedeking-Piret, and Modified Luedeking-Piret model assumption. The optimal point obtained was continued by partial purification. Subsequently, PKM hydrolysis was also done to observe synergistic enzyme effect with NaOH-HCl solvent-assisted for mannan's derivative produced. The evidence showed 3% (w/v) PKM, pH 6.5, 34 °C, and 0.2% (v/v) Al<sub>2</sub>O<sub>3</sub> were the best operating for -

mannanase production. Further confirmation in scale-up phase indicated the highest enzyme activity, rate of production, total sugar concentration, and SUY were calculated as 44.34 U/mL, 0.302 U/mL-1 hr-1, 39.17 g/L and 78.15%, respectively. Kinetics production parameter components, comprised as  $\mu_{max}$ ,  $X_o$ ,  $X_{max}$ ,  $Z$ , and  $\lambda$ , were expected around 0,0492 hr-1; 0,435 g/L; 8,93 g/; 0,085 U/mgX.hr; 4,467 U/mgX; 0,026 gS/gX.hr; dan 4,328 gS/gX, respectively. From clear zone and TLC experimental, it proved that the enzyme was capable to produce MOS from PKM, mainly mannobiose (M2) with extension of 72 hours duration by solvent-assisted enzymatic reaction.