INTISARI

Penyakit *Newcastle Disease* (ND) atau tetelo merupakan permasalahan utama dalam industri peternakan unggas di Indonesia. Meskipun program vaksinasi telah dilaksanakan, *outbreak* penyakit ND masih terus terjadi. Pengembangan vaksin rekombinan ND berbasis isolat lokal masih perlu dilakukan. Protein *Fusion* (F) virus ND diketahui dapat menginduksi imunitas protektif terhadap infeksi virus ND dan berpotensi menjadi agen vaksin rekombinan. Penelitian ini dilakukan untuk mengekspresikan protein rekombinan F virus ND isolat lokal pada *Escherichia coli* BL21(DE3). Sampel penelitian berupa genom RNA virus ND yang diisolasi dari ayam broiler asal Kartasura, Karanganyar (0627/04/2013), ayam kampung asal Galur, Kulon Progo (0663/04/2013), dan ayam broiler asal Sukomoro, Magetan (0819/05/2013). *Full length* gen F diamplifikasi menggunakan metode *One Step* RT-PCR dengan primer spesifik gen F, kemudian disekuensing dengan metode Sanger. Sekuen gen F dianalisis variasi genetik dan filogenetik terhadap beberapa virus ND menggunakan *software* MEGA 6.0. Selanjutnya sekuen gen F isolat Kulon Progo (0663/04/2013) sebesar 600 bp (sekuen nukleotida no. 181-780) disintesis dan dikloning pada plasmid pBT7-N-His oleh Perusahaan Bioneer, Korea. Plasmid rekombinan kemudian ditransformasi pada *E. coli* BL21(DE3) untuk diekspresikan. Ekspresi protein rekombinan F dioptimasi dengan variasi waktu induksi dan kadar IPTG. Hasil penelitian menunjukkan variasi nukleotida gen F dan asam amino protein F ketiga sampel terhadap strain vaksin B1 dan La Sota lebih tinggi dibandingkan terhadap beberapa isolat Indonesia. Analisis filogenetik menunjukkan ketiga sampel termasuk dalam genotipe VII kelas II, berbeda dengan strain vaksin B1 dan La Sota yang termasuk genotipe II kelas II. Hal ini menunjukkan adanya perbedaan antigenik dan genotipe antara virus ND ketiga sampel dengan strain vaksin B1 dan La Sota. Plasmid rekombinan pBT7-N-His yang diinsersi gen F dapat ditransformasi di *E. coli* BL21(DE3). Protein rekombinan F dengan ukuran 25,6 kDa dapat terekspresi optimal dengan induksi IPTG 0,8 mM saat kultur bakteri mencapai OD_{600}=0,6 dan dengan waktu induksi selama 8 jam.

Kata kunci: virus *Newcastle disease*, protein *Fusion*, analisis filogenetik, ekspresi protein

ABSTRACT

Newcastle disease (ND) is the main problem in Indonesia poultry industry. Although vaccination programs have been implemented, Newcastle disease outbreaks still occur. Recombinant vaccine development based on local isolates needs to be performed. Fusion (F) protein could induce protective immunity of ND virus infection so that it is potential to be an agent of recombinant vaccine. The study was conducted to express F recombinant protein of local isolate ND virus in *Escherichia coli* BL21(DE3). The samples were ND virus RNA genomes isolated from broiler chicken from Kartasura, Karanganyar (0627/04/2013), layer chicken from Galur, Kulon Progo (0663/04/2013), and broiler chicken from Sukomoro, Magetan (0819/05/2013). Full length F gene was amplified using One Step RT-PCR with F gene specific primer, and then sequenced using Sanger method. Genetic variation and phylogenetic analysis of F gene sequences were performed against ND viruses using MEGA 6.0 software. F gene of Kulon Progo isolate with size of 600 bp (the nucleotide sequence numbers of 181-780) was synthesized and cloned in pBT7-N-His by Bioneer Corporation, Korea. The recombinant plasmid was transformed in *E. coli* BL21(DE3) to express F protein. F protein expression was optimized by induction time and IPTG concentration variation. The result showed that nucleotide and amino acid variation between F gene of samples and vaccine strain (B1 and La Sota) was higher compared to Indonesia isolates. Phylogenetic analysis showed that all samples were grouped as genotype VII class II, different with B1 and La Sota that included in genotype II class II. It revealed that there was antigenic and genotypic differences between samples and B1 and La Sota vaccine strains. Recombinant plasmid pBT7-N-His inserted with F gene was transformed in *E. coli* BL21(DE3). F recombinant protein of 25.6 kDa was expressed optimally by induction of 0.8 mM IPTG when the bacterial culture reached OD_{600}=0.6 and the induction time for 8 hours.

Keywords: Newcastle disease virus, F protein, phylogenetic analysis, protein expression