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Isolation of an invertebrate-type lysozyme from the nephridia of the echiura, *Urechis unicinctus*, and its recombinant production and activities

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Isolation of an invertebrate-type lysozyme from the nephridia of the echiura, Urechis unicinctus, 1 2 and its recombinant production and activities 3 Hye Young Oh*, Chan-Hee Kim*, Hye-Jin Go, and Nam Gyu Park† 4 Department of Biotechnology, College of Fisheries Sciences, Pukyong National University, 45 5 6 Yongso-ro, Nam-gu, Busan, 48513, Korea 7 8 * These authors contributed equally to this work. 9 10 † Corresponding author: Nam Gyu Park, Department of Biotechnology, College of Fisheries 11 Sciences, Pukyong National University, 45 Yongso-ro, Nam-gu, Busan 48513, Korea. 12 Tel: +82 51-629-5867 13 Fax: +82 51-629-5863 14 E-mail: ngpark@pknu.ac.kr 15 16 17 Keywords: Urechis unicinctus, invertebrate-type lysozyme, protein isolation, recombinant protein 18 production, non-enzymatic antibacterial activity 19 20 Abbreviations: AA, amino acid; CFU, colony forming unit; HEWL, hen egg white lysozyme; HPLC, high performance liquid chromatography; IPTG, isopropyl β -D-1-thiogalactopyranoside; MEC, 21 22 minimal effective concentration; NAG, N-acetylglucosamine; NAM, N-acetylmuramic acid; nano 23 LC-MS/MS, nanoscale liquid chromatography coupled to tandem mass spectrometry; ORF, open 24 reading frame; PBS, phosphate-buffered saline; PCR, polymerase chain reaction; Q-TOF, quadrupole 25 time-of-flight; RACE, rapid amplification cDNA ends; RT-qPCR, real-time quantitative PCR; SDS-26 PAGE, sodium dodecyl sulfate-polyacrylamide gel electrophoresis; TFA, trifluoroacetic acid; TSB, 27 tryptic soy broth; URDA, ultrasensitive radial diffusion assay; UPLC, ultraperfomance liquid chromatography; UTR, untranslated region 28

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