

Selection of Reference Genes for Quantitative RT-PCR Studies in Beluga Whale

(*Delphinapterus leucas*) Blood

陳怡樺¹、李宗賢²、詹孟矜³、張文炳²、王建雄¹、周世認¹、吳永惠⁴、楊瑋誠¹

I-Hua Chen¹, Tsung-Hsien Li², Molly Zhan³, Wen-Been Chang², Jiann-Hsiung Wang¹, Shih-Jen Chou¹, Yeong-Huey Wu⁴, Wei-Cheng Yang¹

¹ 國立嘉義大學獸醫學系 ² 國立海洋生物博物館 ³ 海景世界企業股份有限公司

⁴ 國立屏東科技大學獸醫學院

¹Department of Veterinary Medicine, National Chiayi University, Chiayi, 600, Taiwan, ROC.

²National Museum of Marine Biology and Aquarium, Pingtung, 944, Taiwan, ROC.

³Hi-Scene World Enterprise Co., LTD., Pingtung, 944, Taiwan, ROC.

⁴Collage of veterinary medicine, National Pingtung University of Science and Technology, Pingtung, 912, Taiwan, ROC.

Quantitative RT-PCR is often used for research in gene expression, and it is vital to choose appropriate housekeeping genes (HKGs) as reference genes to obtain correct result. To date, there is no study on selection of reference genes in cetacean blood. The purpose of this study is to determine stably-expressed HKGs in blood, which can be the appropriate reference genes in relative quantification in gene expression research. Thirty-two EDTA-anticoagulated blood samples were taken monthly from 4 beluga whales (*Delphinapterus leucas*) in National Museum of Marine Biology and Aquarium and preserved in RNAlater[®] (Ambion) immediately. Total RNA was extracted then following by cDNA synthesis and qPCR for candidate genes (ACTB, B2M, GAPDH, HPRT1, LDHB, PGK1, RPL4, RPL8, RPL18, RPS9, RPS18, TFRC, YWHAZ). And the stability values of the HKGs were determined by *geNorm* and *NormFinder* software. The results revealed that ACTB and RPL4 are the most stable HKGs in beluga whale blood. Blood can serve as an indication of health status in cetaceans because changes of gene expression in blood is prior to hematology and chemistry findings. This research provides recommendation of reference gene selection, which may contribute to further mRNA relative quantification research in the peripheral blood leukocytes in belugas.