ABSTRACT

Evelyn Taruna, 24020220130038. Preparation, Characterization, and in vitro Evaluation of Cationic Linoleic Acid Lipopeptide-Based Liposomes (C18:2-CHS-PKKKRKV) as a Transfection Agent Candidate (under the guidance of Nurhayati and Tarwadi).

Transfection is the delivery of genetic material into eukaryotic cells which in its application requires the help of a non-viral vector. In this study, liposomes based on the cationic lipopeptide linoleic acid (C18:2-CHS-PKKKRKV) were formulated with the phospholipid dioleoylphosphatidylethanolamine (DOPE) and cholesterol lipids to increase transfection efficiency with low cytotoxicity. The comparison control used in this study was the commercial transfection agent polyethylenimine (PEI25K). The DNA mobility assay and stability assay of the DNA-liposome complex showed that liposomes could complex and protect DNA starting from DNA/liposome mass ratios of 1:1 (0.5 µg:0.5 µg) and 1:2 (0.5 µg:1 µg). The DNA condensation assay shows that liposomes can condense DNA well, where there is a decrease in fluorescence intensity as the added liposome content increases. The transfection test of the plasmid encoding the Green Fluorescent Protein gene (pCSII-EF-AcGFP) using liposomes in Human Embryonic Kidney (HEK-293T) cells showed expression of the target protein although the efficiency was lower than PEI25K. Lipopeptides formulated with cholesterol have the ability to form complexes with DNA, provide DNA protection from DNAse enzyme degradation, and better transfection efficiency compared to lipopeptides formulated with DOPE or a combination of DOPE and cholesterol. Based on the cytotoxicity test, overall liposomes have a lower level of cytotoxicity compared to PEI25K. Linoleic acid cationic lipopeptide-based liposomes formulated with DOPE phospholipids, cholesterol lipids, or a combination of the two can complex and protect DNA from DNAse enzyme degradation at various DNA/liposome mass ratios (1:1; 1:2; 1:4; 1: 8). Although linoleic acid cationic lipopeptide-based liposomes still had lower transfection efficiency than the commercial transfection agent control PEI25K, they had a lower level of cytotoxicity against HEK-293T mammalian cells.

Key words: transfection agent, DOPE, green fluorescent protein, liposomes, cationic lipopeptides, cholesterol, cytotoxicity.