STRUCTURE-ACTIVITY RELATIONSHIP OF LIPOPEPTIDE FROM OUTER MEMBRANE OF *ESCHERICHIA COLI* AND SYNTHESIS OF HIGHLY IMMUNOPOTENTING LIPOPEPTIDE DERIVATIVES WITH AN ACHIRAL LIPO-PART

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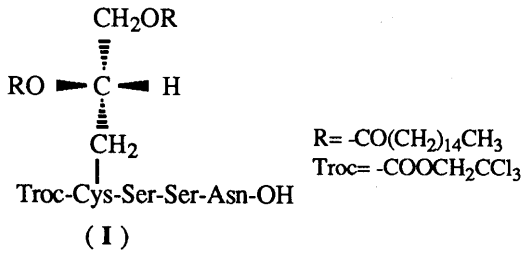
For outstanding the structure-activity relationship of lipopeptide derivatives, high biologically active lipopeptide derivatives with an achiral lipo-part were newly synthesized

KEYWORDS lipoprotein; lipopeptide derivative having an achiral lipo-part; mitogenic activity

It has been known that a lipopeptide from the outer membrane of *Escherichia coli* is an active mitogen and polyclonal activator for B lymphocytes.[1~4] In the preceding paper,[5~8] we reported a new synthesis of chiral lipopeptides, their derivatives(I) with higher activity than the native lipopeptide and the structure-activity relationship between number of amino acids and biological activity. Now we focused our attention on the lipo-part; to investigate the influence of the length and number of the aliphatic chain and its mitogenic activity, and also to find high biologically active lipopeptide derivatives with an achiral lipo-part, we have synthesized lipopeptide derivatives with one aliphatic chain linked to the propane skeleton **1**,[9] **2**[10] linked to the ethane **3**,[11] **4**[12] by an ether bond, linked to the propane skeleton **5**,[13] **6**,[14] **7**[15] by an ester bond and with two aliphatic chain linked to the propane or the ethane skeleton and cystein *N*-terminal **8**,[16] **9**,[17] 2-cysteinylmethyl glycerol type **10**,[18] **11**,[19] and 2-cysteinyl glycerol type **12**[20](shown in Fig.2).



(I)

Fig. 1. Structure of Lipopeptide Derivative with High Activity

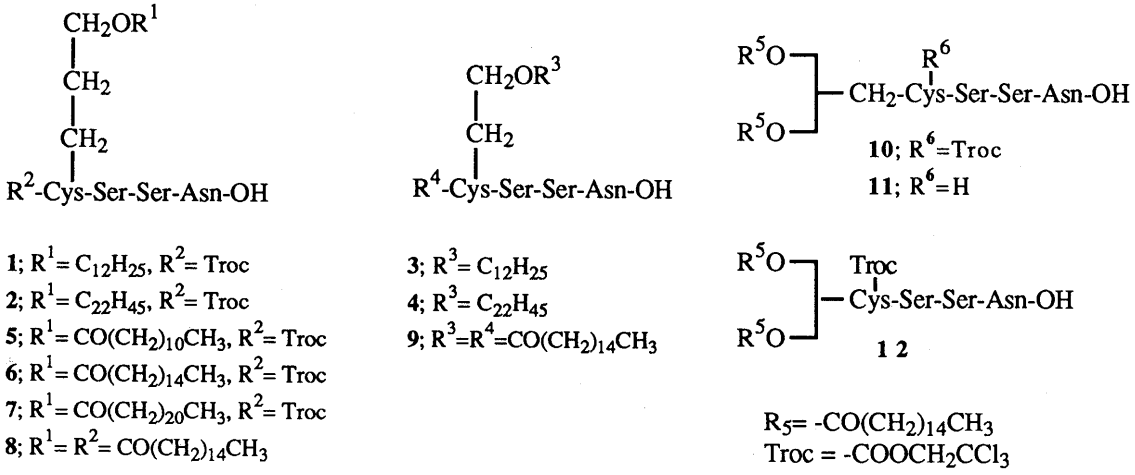
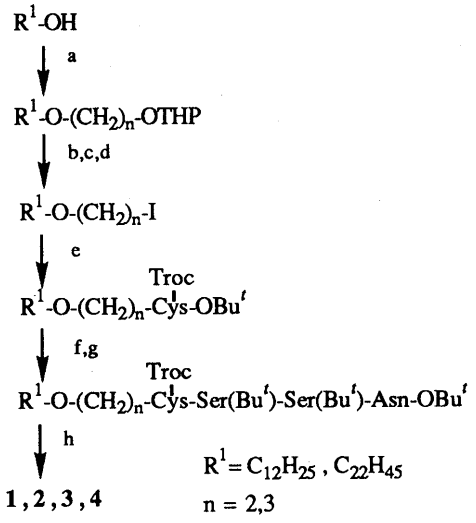


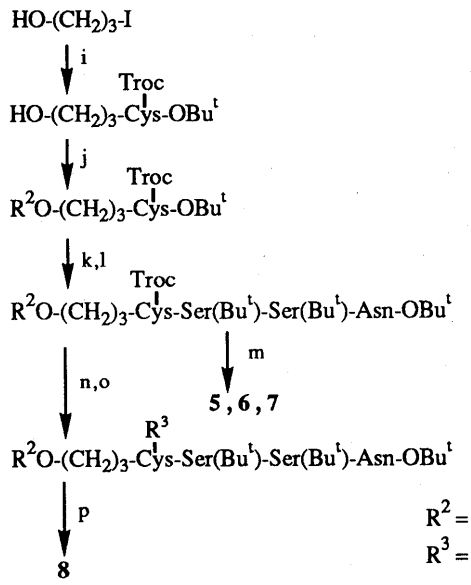
Fig. 2. Structure of Lipopeptide Derivatives



a; NaH, $I(CH_2)_nOTHP$, 8~18%
b; *p*-TsOH, MeOH, 75~77%
c; *p*-TsCl, pyridine, 50~70%
d; NaI, acetone,77~80%
e; Troc-Cys-OBu^t, *i*-Pr_2NEt, DMF 80~90%,
f; TFA, 70~80%
g; DEPC, H-Ser(Bu^t)-Ser(Bu^t)-Asn-OBu^t
Et_3N, CH_2Cl_2, 78~86%
h; TFA, 66~82%.

THP = tetrahydropyranyl
Ts= *p*-toluenesulfonyl
DEPC = diethyl phosphorocyanidate
TFA = trifluoloacetic acid

Chart 1. Synthesis of Ether Type Lipopeptide Derivatives

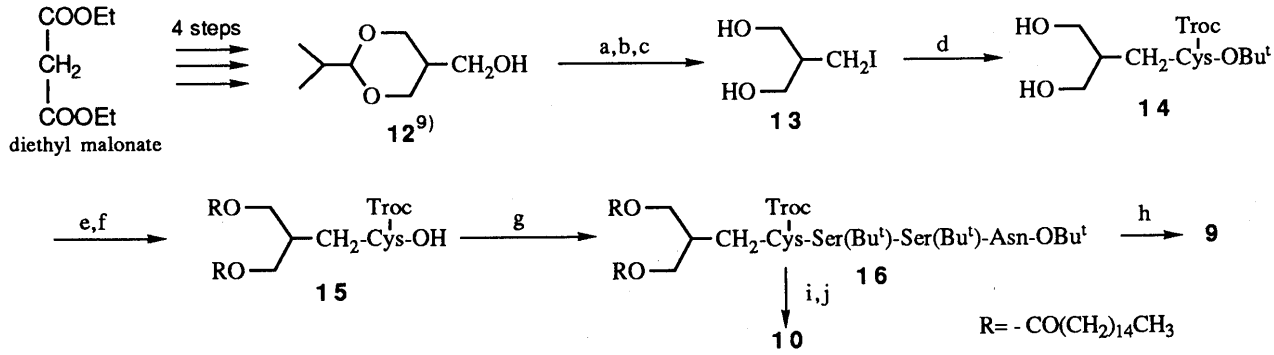


i; Troc-Cys-OBu^t, *i*-Pr_2NEt, DMF, 86%
j; R^2-Cl, *i*-Pr_2NEt, DMAP, CH_2Cl_2 or
R^2-OH, DCC, DMAP, DMF, 62~83%
k;TFA, 80~90%
l; BOP reagent, H-Ser(Bu^t)-Ser(Bu^t)-Asn-OBu^t,
Et_3N, CH_2Cl_2 , 73~81%,
m; TFA, 72~92%,
n; Zn, CH_3COOH, 87%
o; R^3-Cl, *i*-Pr_2NEt, DMAP, CH_2Cl_2 , 83%
p; TFA, 92%

DCC = dicyclohexylcarbodiimido
DMAP = 4-dimethylaminopyridine

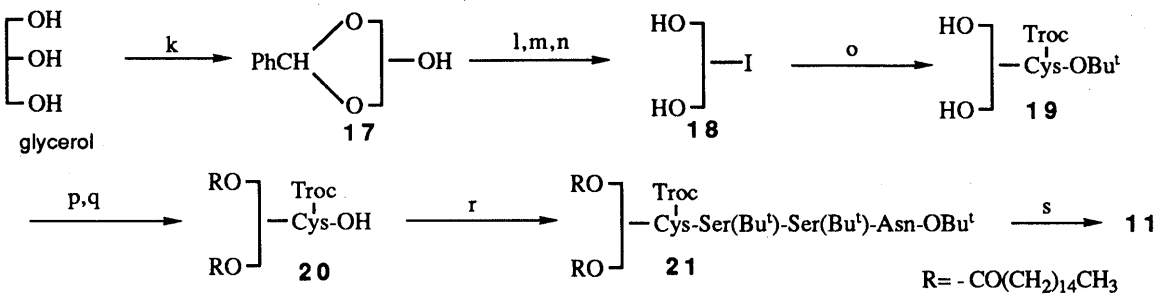
R^2 = $-CO(CH_2)_{10}CH_3$, $-CO(CH_2)_{14}CH_3$, $-CO(CH_2)_{20}CH_3$
R^3 = $-CO(CH_2)_{14}CH_3$

Chart 2. Synthesis of Ester Type Lipopeptide Derivatives



a; Et_3N , CH_2Cl_2, TsCl, 60% b; MeOH, 1N HCl, 85% c; NaI, acetone, 40% d; Troc-Cys-OBu^t, *i*-Pr_2NEt, DMF, 86% e; $CH_3(CH_2)_{14}COCl$, *i*-Pr_2NEt, CH_2Cl_2 DMAP, 87% f; TFA, 64% g; BOP reagent, Et_3N, H-Ser(Bu^t)-Ser(Bu^t)-Asn-OBu^t, CH_2Cl_2, 56% h; TFA, 53% i; Zn, AcOH, 85% j; TFA , 80%.

Chart 3



k; PhCHO, benzene, sulfosalicylic acid, 18% l; Et_3N, CH_2Cl_2, TsCl, 63% m; MeOH, 1N HCl, 82% n; NaI, acetone, 46% o; Troc-Cys-OBu^t, *i*-Pr_2NEt, DMF, 28% p; $CH_3(CH_2)_{14}COCl$, DMAP, *i*-Pr_2NEt, CH_2Cl_2, 65% q; TFA, 81% r; BOP reagent, Et_3N, H-Ser(Bu^t)-Ser(Bu^t)-Asn-OBu^t, CH_2Cl_2, 65% s; TFA, 65%.

Chart 4

The mitogenic activity of all compounds **1~12** were measured and compared with the natural lipopeptide derivative[22] (**I**). The activities of **1~9** were greatly reduced, and compound **10,11** did not show activity as high as that of compound (**I**), while compound **12** had the same degree of activity as compound (**I**). These results indicate that the lipopeptide derivatives with one aliphatic chain cannot show a high activity and that glyceryl part is necessary for lipopeptide derivatives to show a higher activity.

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9) mp 164~166°C, $[\alpha]_D^{22}$ -5.4°(*c* 0.36, $CHCl_3$), FABMS(*m/z*) 810$(M+H)^+$.
10) mp 163~165°C, $[\alpha]_D^{22}$ -8.4°(*c* 0.21, $CHCl_3$), FABMS(*m/z*) 952$(M+H)^+$.
11) mp 167~169°C, $[\alpha]_D^{22}$ -15.3°(*c* 0.19, MeOH), FABMS(*m/z*) 796$(M+H)^+$.
12) mp 168~170°C, $[\alpha]_D^{22}$ -3.5°(*c* 0.22, $CHCl_3$:MeOH=1;1), FABMS(m/z) 936$(M+H)^+$.
13) mp 161~164°C, $[\alpha]_D^{22}$ -3.8°(*c* 0.40, $CHCl_3$), FABMS(*m/z*) 824$(M+H)^+$.
14) mp 169~171°C, $[\alpha]_D^{22}$ -4.2°(*c* 0.33, $CHCl_3$), FABMS(*m/z*) 880$(M+H)^+$.
15) mp 170~173°C, $[\alpha]_D^{22}$ -4.3°(*c* 0.32, $CHCl_3$), FABMS(*m/z*) 964$(M+H)^+$.
16) mp 204~206°C, FABMS(*m/z*) 945$(M+H)^+$.
17) mp 205~207°C, FABMS(*m/z*) 931$(M+H)^+$.
18) mp 170~172°C, $[\alpha]_D^{22}$ +10.4°(*c* 0.20, DMF), FABMS(*m/z*) 1149$(M+H)^+$.
19) mp 238~240°C(dec), $[\alpha]_D^{22}$ +7.0°(*c* 1.44, $CHCl_3$), FABMS(*m/z*) 975$(M+H)^+$
20) mp 192~194°C, $[\alpha]_D^{22}$ -10.0°(*c* 0.32, $CHCl_3$), FABMS(*m/z*) 1135$(M+H)^+$.
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22) The detailed paper will be published elsewhere.

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