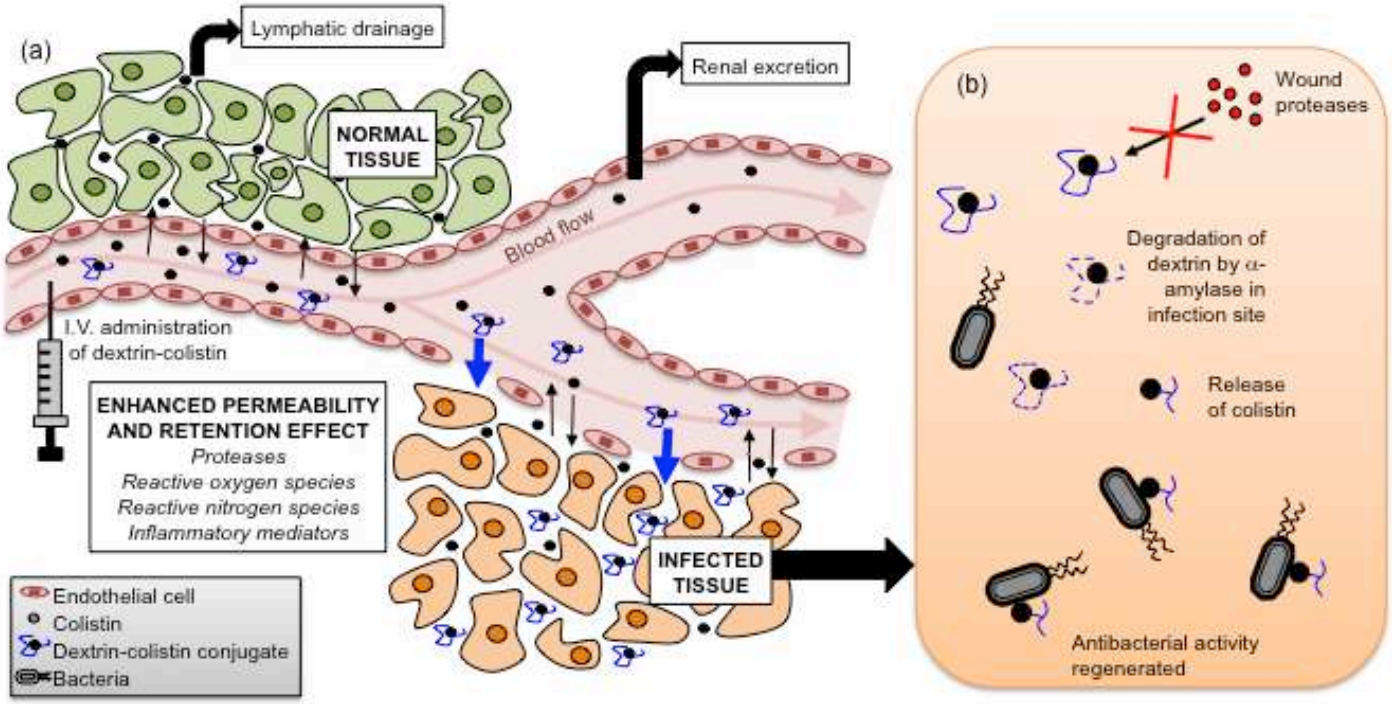



**FIG 1** Schematic showing the proposed mechanism of action of dextrin-colistin conjugates.

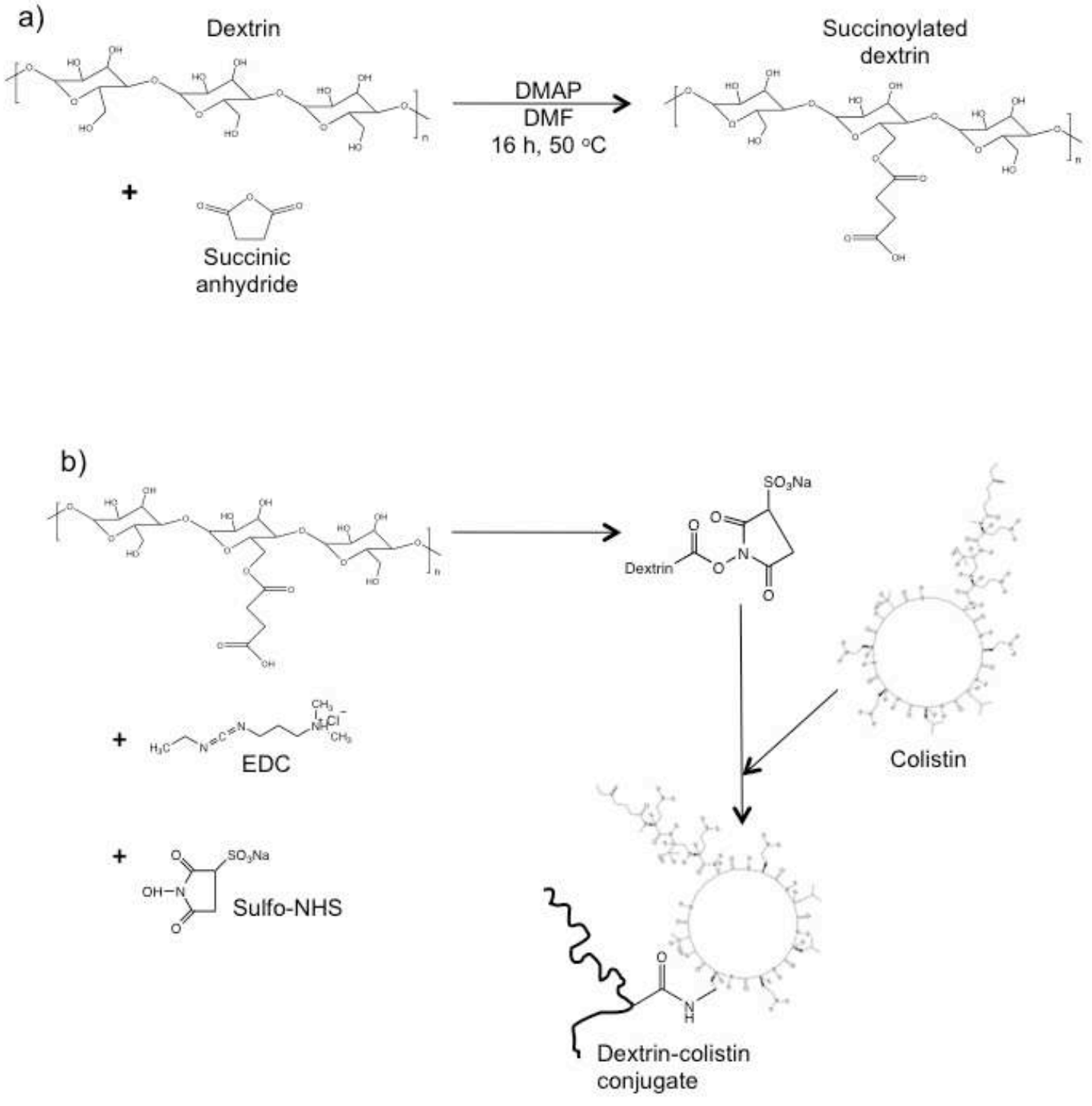



**FIG 2** Synthesis of (a) succinoylated dextrin; and (b) dextrin-colistin conjugates.

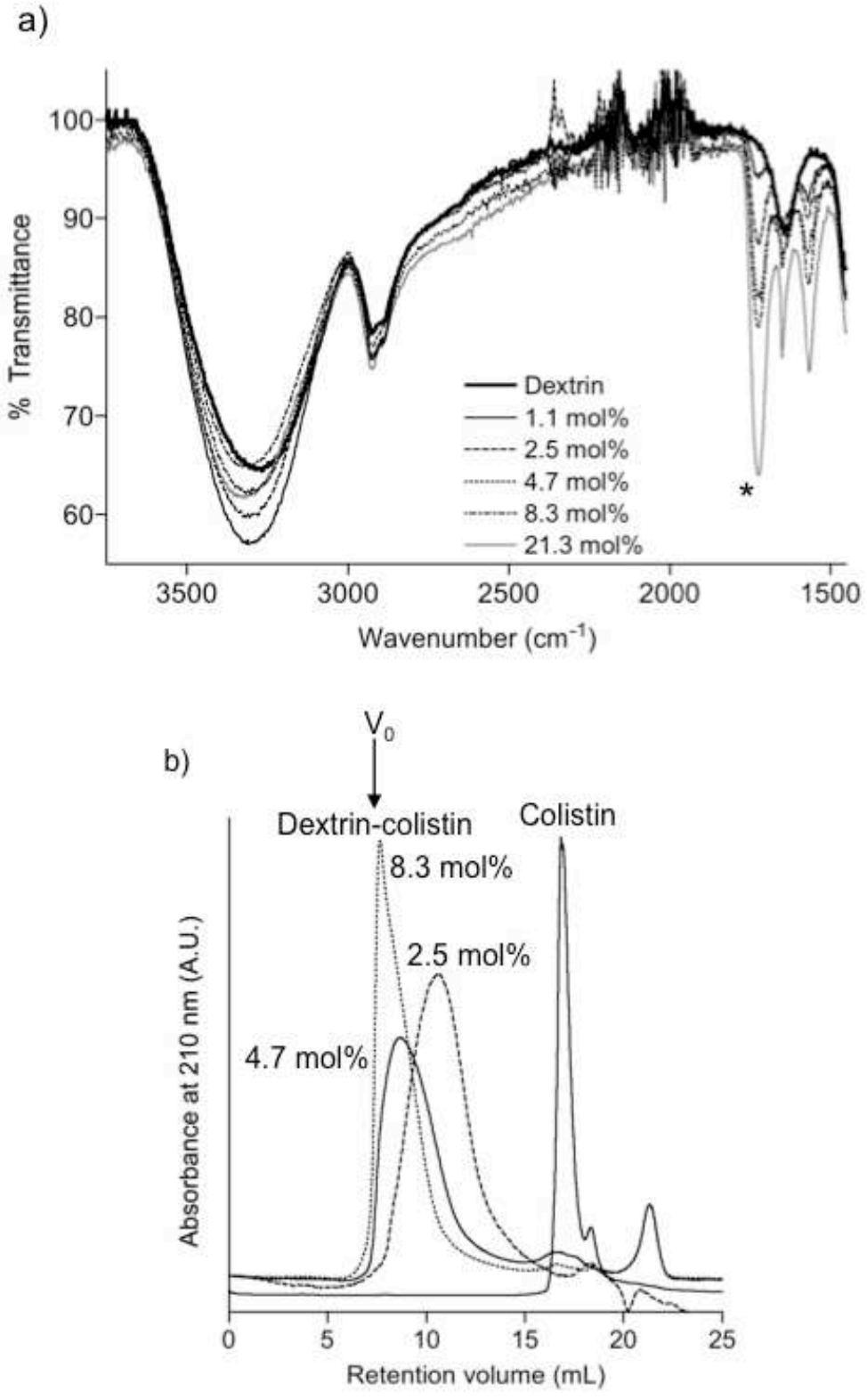



**FIG 3** Characterization of succinoylated dextrin intermediates and dextrin-colistin conjugates. (a) FT-IR spectra showing amplification of peak intensity at 1720 $cm^{-1}$ with increasing incorporation of carboxyl groups; and (b) FPLC chromatogram of dextrin-colistin conjugates containing dextrins (7,500 g/mol) with different degrees of succinoylation ($V_0$ = void volume (7.7 mL)).

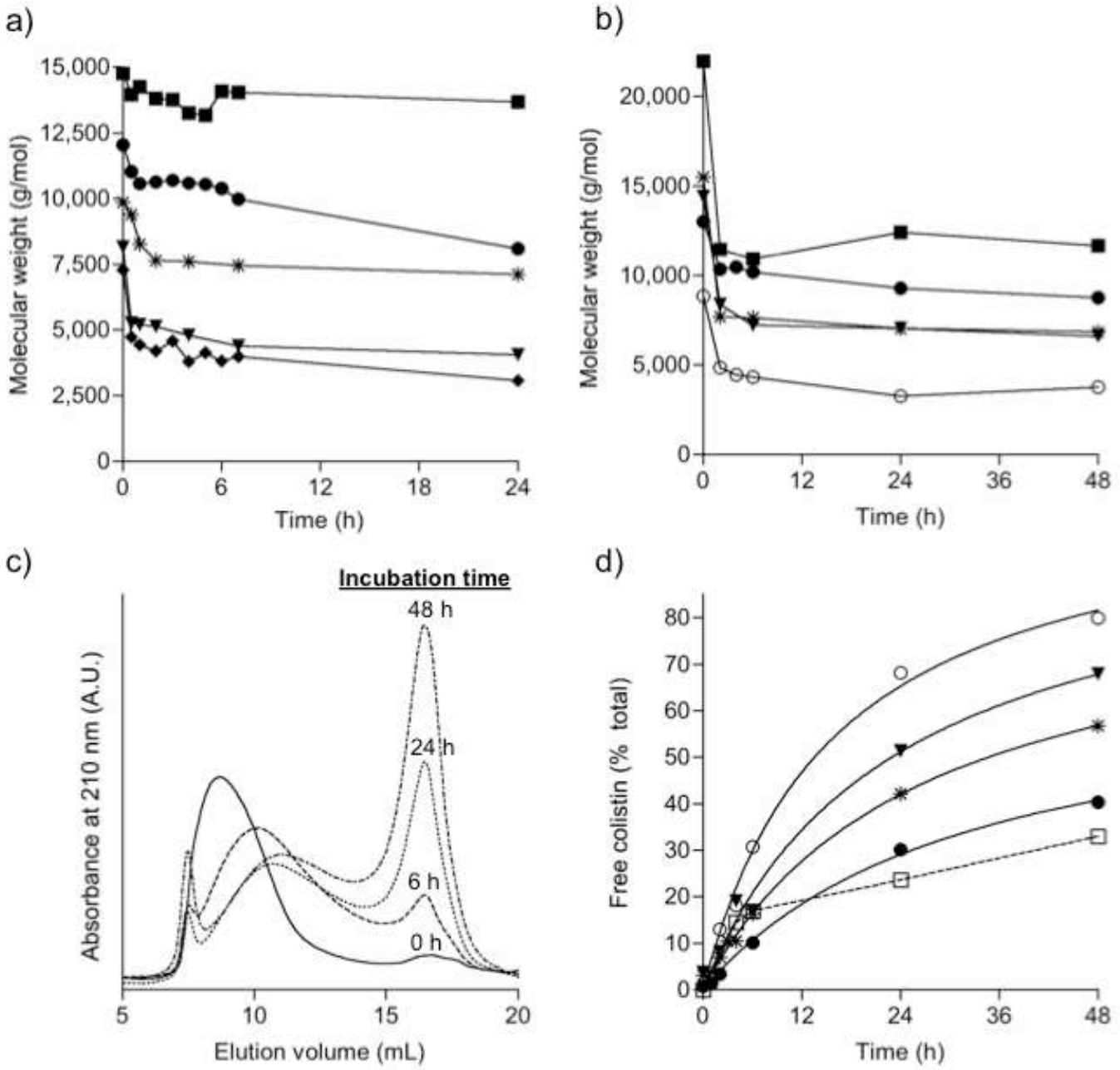



**FIG 4** Characterization of the degradation of dextrin, succinoylated dextrin and dextrin-colistin conjugates (3 mg/mL) in the presence of amylase (100 IU/L in PBS at 37 °C). Panels (a) and (b) show the change in relative molecular weight in the presence of amylase by GPC of (a) native dextrin (7,500 g/mol) and its succinoylated intermediates; and (b) dextrin-colistin conjugates containing dextrin (7,500 g/mol) with different degrees of succinoylation. Panel (c) shows a typical elution profile of dextrin-colistin conjugate (containing 7,500 g/mol dextrin, 4.7 mol% succinoylation) from a Superdex 75 FPLC column, following incubation with amylase ($V_0$ = void volume (7.7 mL)), and panel (d) shows the release of colistin from dextrin-colistin conjugates (containing 7,500 g/mol dextrin; 3 mg/mL) in the presence of amylase (100 IU/L in PBS) and CMS (3 mg/mL) in PBS at 37 °C (measured by FPLC). Data is expressed as the percentage of total colistin. ($n$=1). Where ◆ = dextrin or dextrin-colistin conjugate; ○ = 1.1 mol% dextrin or dextrin-colistin conjugate; ▼ = 2.5 mol% dextrin or dextrin-colistin conjugate; ✳ = 4.7 mol% dextrin or dextrin-colistin conjugate; ● = 8.3 mol% dextrin or dextrin-colistin conjugate; ■ = 21.3 mol% dextrin or dextrin-colistin conjugate and □ = CMS.

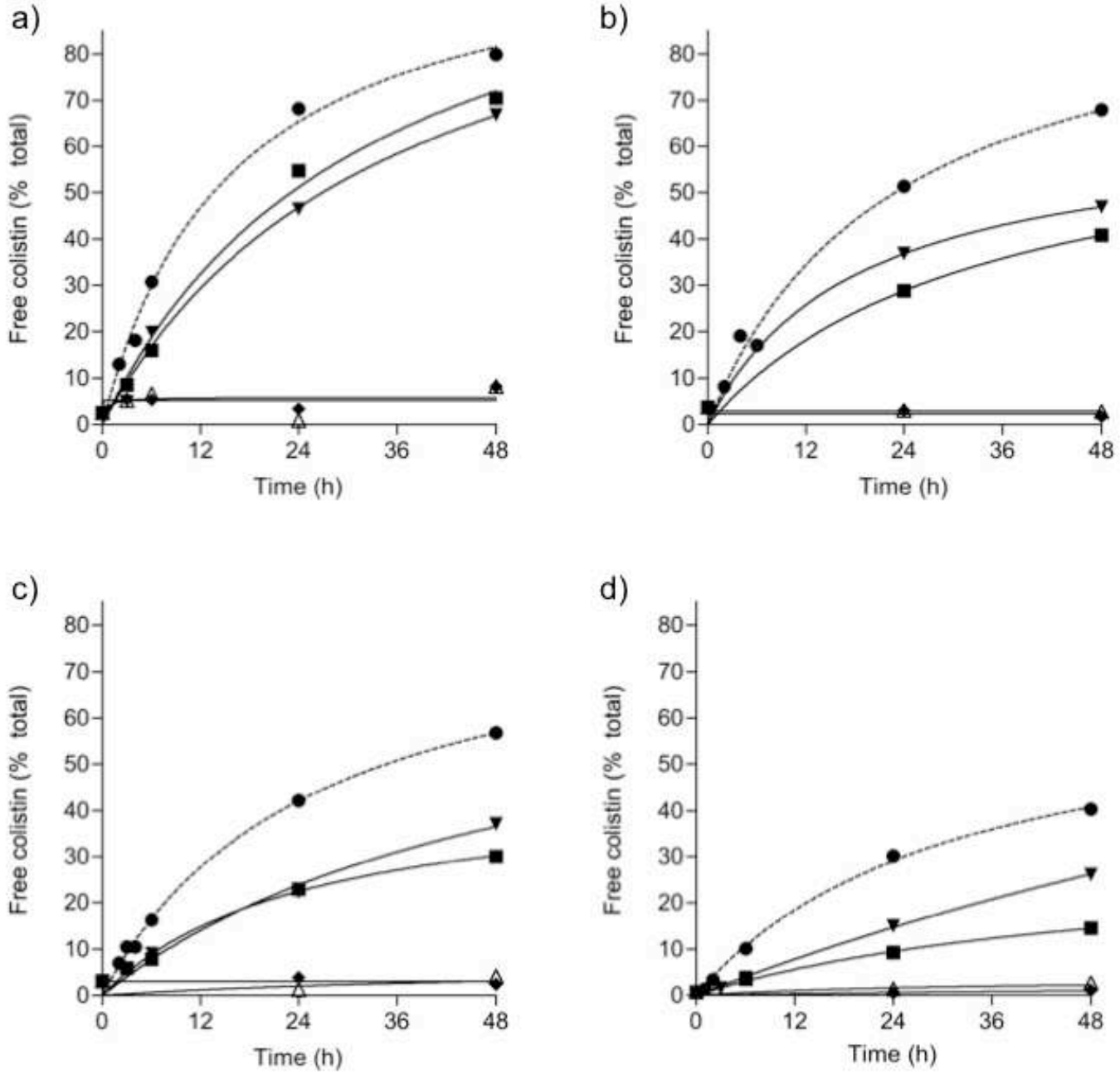



**FIG 5** Stability of dextrin-colistin conjugates (containing 7,500 g/mol dextrin; 3 mg/mL) in $dH_2O$ and PBS at pH 7.4 (37 °C) in the absence of amylase (measured by FPLC), and in comparison with amylase-treated conjugates. Data is expressed as the percentage of total colistin. Panels show conjugates containing (a) 1.1 mol% succinoylation, (b) 2.5 mol% succinoylation, (c) 4.7 mol% succinoylation, and (d) 8.3 mol% succinoylation. Where ● = dextrin-colistin conjugate with amylase (100 IU/L) in PBS at 37 °C; ▼ = dextrin-colistin conjugate in PBS at 37 °C; ◆ = dextrin-colistin conjugate in PBS at 4 °C; ■ = dextrin-colistin conjugate in $dH_2O$ at 37 °C and △ = dextrin-colistin conjugate in $dH_2O$ at 4 °C.

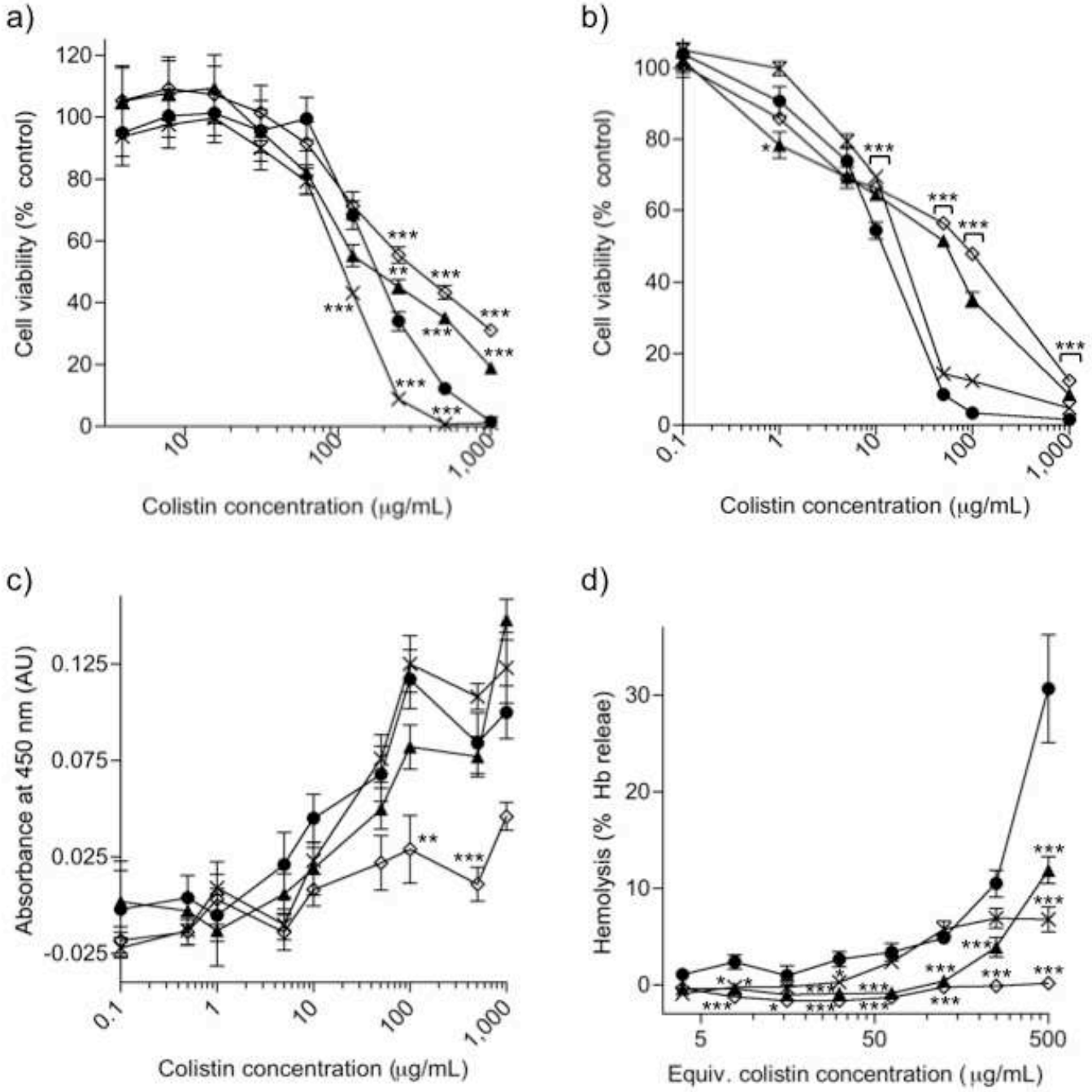



**FIG 6** *In vitro* cytotoxicity of colistin sulfate, CMS and dextrin-colistin conjugates. Panels (a) and (b) show cell viability by MTT assay of RAW 264.7 (24 h incubation) and HK-2 (72 h incubation) cells, respectively, following incubation with colistin sulfate, CMS or dextrin-colistin conjugate with and without amylase (100 IU/L) at 37 °C. Data is expressed as mean % untreated control ± SEM, n=18. Panel (c) shows membrane integrity by LDH assay of HK-2 cells incubated for 24 h with colistin, CMS or dextrin-colistin with and without amylase (100 IU/L) at 37 °C. Data is expressed as mean ± SEM, n=6. Panel (d) shows hemolysis of rat erythrocytes following incubation for 24 h with colistin, CMS or dextrin-colistin conjugate with and without amylase (100 IU/L) at 37 °C. Data is expressed as mean % triton X-100 control ± SEM, n=18. Where ● = colistin sulfate; × = CMS; ◇ = dextrin-colistin conjugate and ▲ = dextrin-colistin conjugate with amylase. * indicates significance ($p$<0.05) compared to colistin sulfate; ** indicates significance ($p$<0.01) compared to colistin sulfate; *** indicates significance ($p$<0.001) compared to colistin sulfate.

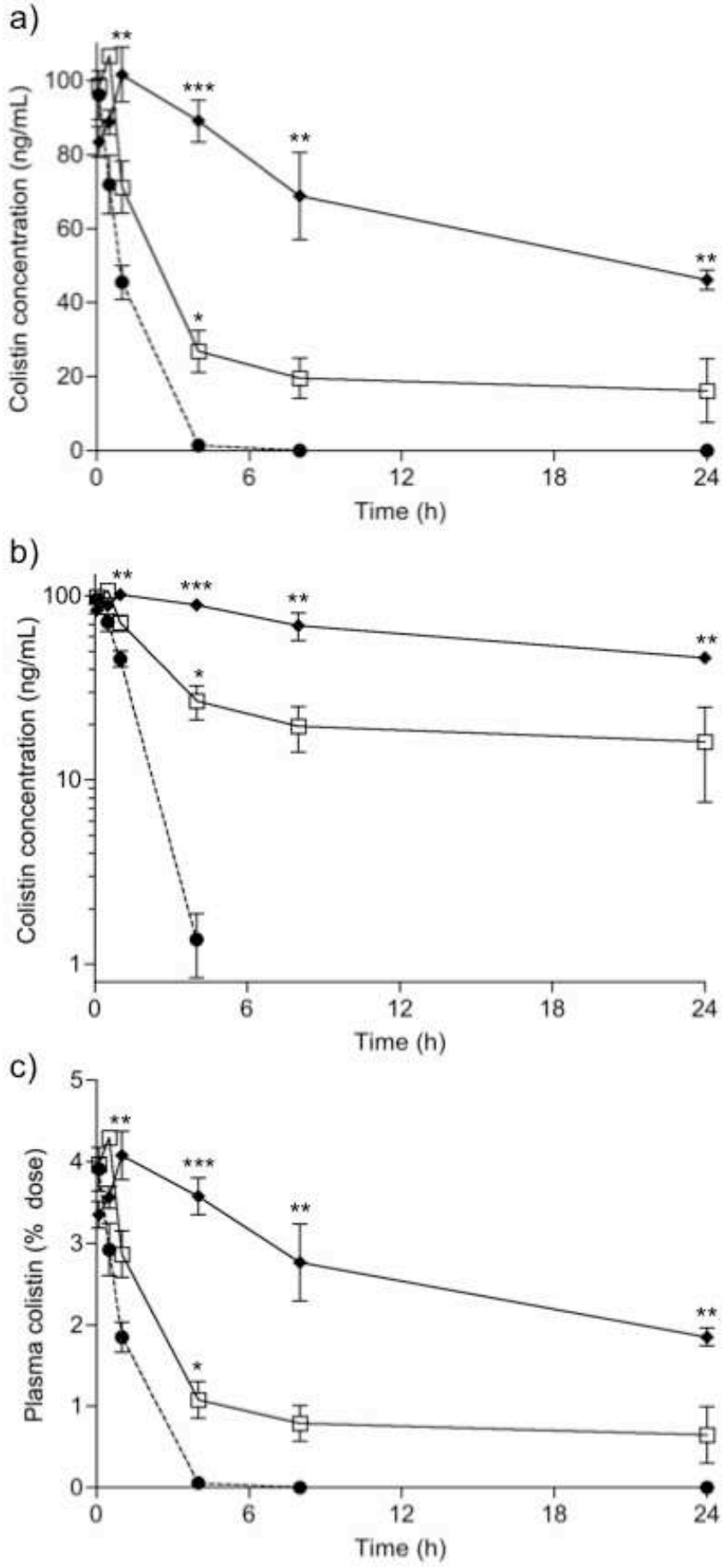
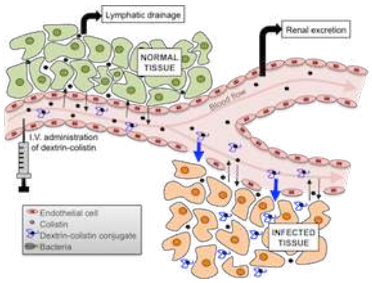



**FIG 7** Mean plasma concentration of colistin following an IV dose of colistin sulfate, dextrin-colistin conjugate (1.4 mol%) and dextrin-colistin conjugate (7.2 mol%) (0.1 mg/kg). Data is expressed as colistin concentration ± S.D. (n=2). Concentrations of colistin following administration of colistin sulfate were not quantifiable beyond 4 h. Where ● = colistin sulfate; □ = dextrin-colistin conjugate (with 1.4 mol% succinoylation) and ◆ = dextrin-colistin conjugate (with 7.2 mol% succinoylation). * indicates significance ($p<0.05$) compared to colistin sulfate; ** indicates significance ($p<0.01$) compared to colistin sulfate; *** indicates significance ($p<0.001$) compared to colistin sulfate.


Lymphatic drainage
Renal excretion
NORMAL TISSUE
Blood flow
I.V. administration of dextrin-colistin
Endothelial cell
Colistin
Dextrin-colistin conjugate
Bacteria
INFECTED TISSUE