

산소자유기의 심근독성에 대한 Glutathione의 영향

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= Abstract =

Effect of Glutathione on Oxidant-induced Cardiotoxicity

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Background : In order to elucidate toxic mechanism of the oxygen radicals on cultured rat myocardial cells, cytotoxic effect of oxygen radicals was evaluated by MTT assay. In addition protective effect of glutathione(GSH) on oxidant-induced cardiotoxicity was investigated on these cultures.

Methods : Myocardial cells derived from neonatal rats were cultured for 12 hours in the medium containing various concentrations of glucose oxidase(GO). Cell viability was measured by MTT assay and morphological changes of the myocardial cells were observed by light microscope.

Results : GO-mediated oxygen radicals remarkably decreased cell viability of cultured myocardial cells in a dose-and time-dependent manner. And also, GSH blocked GO-induced cardiotoxicity in these cultures.

Conclusion : These results suggest that the oxygen radicals are toxic and the selective anti-oxidants such as GSH are effective in blocking against the oxidant-induced cardiotoxicity in cultures of the myocardial cells of neonatal rats.

KEY WORDS : Cultured myocardial cells · Oxygen radicals · Cardiotoxicity · Antioxidant.

서 론

Superoxide(O_2^-) hydroxyl radicals
(OH $^-$) hydrogen peroxide(H_2O_2)
citotoxic amino acid, EAA)
1). , 5,6).
2) 1,7), (ex -
3), 8).
4) EAA
5), 9).

가 phosphate buffered saline(PBS) 3
 10,11), 30mU/ml GO가
 12 GO가

(in vitro) 5. 심근세포의 생존율 분석
 Mosmann¹⁴⁾ MTT
 (3 - (4,5 - dimethylthiazol - 2yl) - 2, 5 - dipheylte -
 trazolium bromide(MTT, Sigma))

재료 및 방법

1. 세포배양 (Nikon)
 Kasten¹³⁾
 1 3 Sprague - Dawley
 Phosphate buffered saline(PBS) 3
 Ea -
 gle's minimum essential medium(MEM, Gibco)
 1 × 10⁵/well 96 - mult -
 iwell 37 , 5% CO₂/95% air
 3

7. 통계처리 Student t - test
 P - value가 0.05

결 과

2. 약제제조 glucose oxidase(GO, Sigma)
 glutathione(GSH, Sigma)

3. 산소자유기의 처리 가
 12 PBS 3 30mU/ml GO 가

4. 항산화제 처리 glutathione(GSH)
 GO system 3
 GSH가

1. 심근세포에 대한 산소자유기의 독성 효과
 1 50mU/ml GO가
 12 MTT
 1mU/ml GO

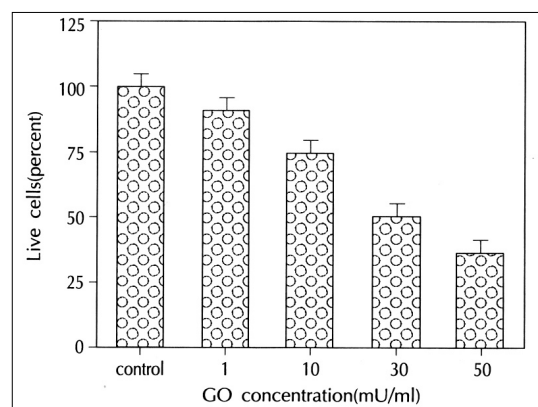


Fig. 1. Dose-response relationship of glucose oxidase (GO) concentration on cultured rat myocardial cells. cytotoxicity was measured by MTT assay. Cultures were exposed to 1, 10, 30 and 50mU/ml for 12 hours. The results indicate the mean ± SEM (n=6).

(100%) 89% 10,
30 50mU/ml GO 76%, 48%
32% (Fig. 1).
GO
30mU/ml GO가
1 24 MTT
1
(100%) 84% 6
65% 12 24
42% 21% (Fig. 2).

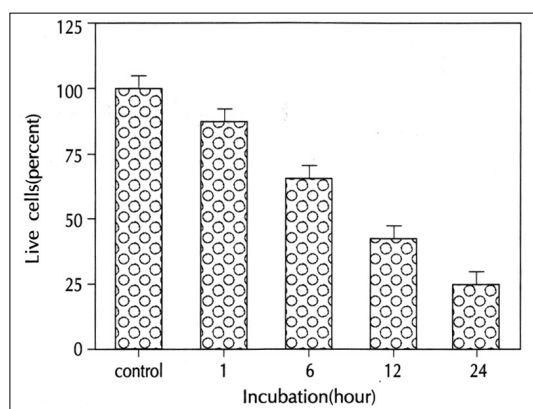


Fig. 2. Time-dependency of glucose oxidase(GO)-induced cardiotoxicity as measured by MTT assay in cultured rat myocardial cells. Cultures were exposed to 30mU/ml GO for 1, 6, 12 and 24 hours. The results indicate the mean \pm SEM(n=6).

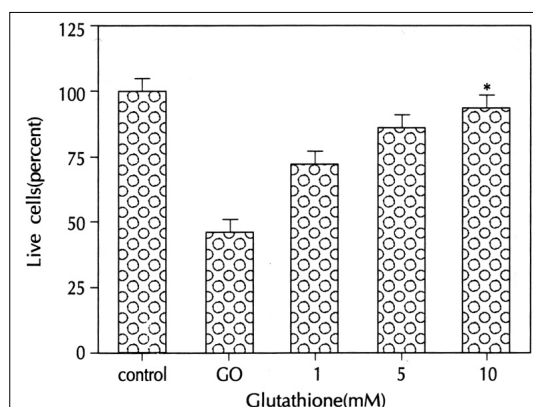


Fig. 3. Dose-response relationship of glutathione(GSH) for its protective effect on glucose oxidase(GO)-induced cardiotoxicity by MTT assay. Cultured rat myocardial cells were preincubated for 3 hours before treatment with 30mU/ml GO. Cultures were exposed to GSH at the concentrations of 1, 5 and 10mM for 2 hours, respectively. The results indicate the mean \pm SEM(n=6). *p<0.05.

2. 산소자유기에 대한 GSH의 영향

GSH
GO
3 GSH GO
12 MTT
GO (100%)
46% 1mM GSH
71% 5mM 10mM GSH
87% 94%(p<0.05)
(Fig. 3).

3. 형태적 관찰

(cluster)
(Fig. 4A). GO

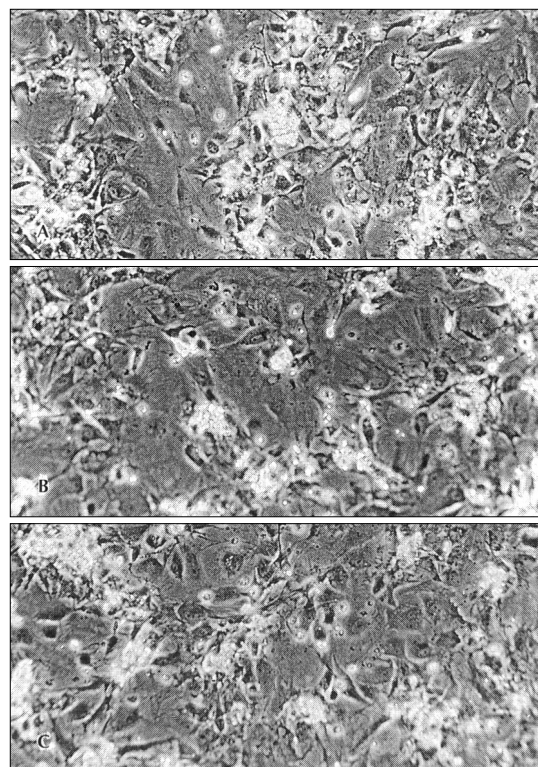


Fig. 4. Dissociated myocardial cells from neonatal rat in vitro.
A : Control myocardial cells for 5 days in vitro(\times 125).
B : A sister culture exposed to 30mU/ml glucose oxidase(GO) for 12 hours(\times 125).
C : A sister culture preincubated with 10mM glutathione for 3 hours before treated with GO(\times 125).

myocardium

(Fig. 4B). , 10mM

GSH (Fig. 4C).

고 안 8,19 가

1,15 , 16)

요 약

가 가

연구배경 :
Superoxide($O_2^{\cdot -}$) hydroxyl ($OH^{\cdot -}$)
hydrogen peroxide(H_2O_2)

4) 가 (in vitro)

glutathione

(GSH)

방 법 :
glucose oxidase(GO) 12
가 GSH가

GSH , MTT

GSH (46%) (100%)
10mM GSH GO system 3
(94%) GSH가 MTT

GSH GSH가

결 과 :
GO 가 GO

GSH가

GSH GO

결 론 :
Myers 17) GSH 가
Gross 18) stunned

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