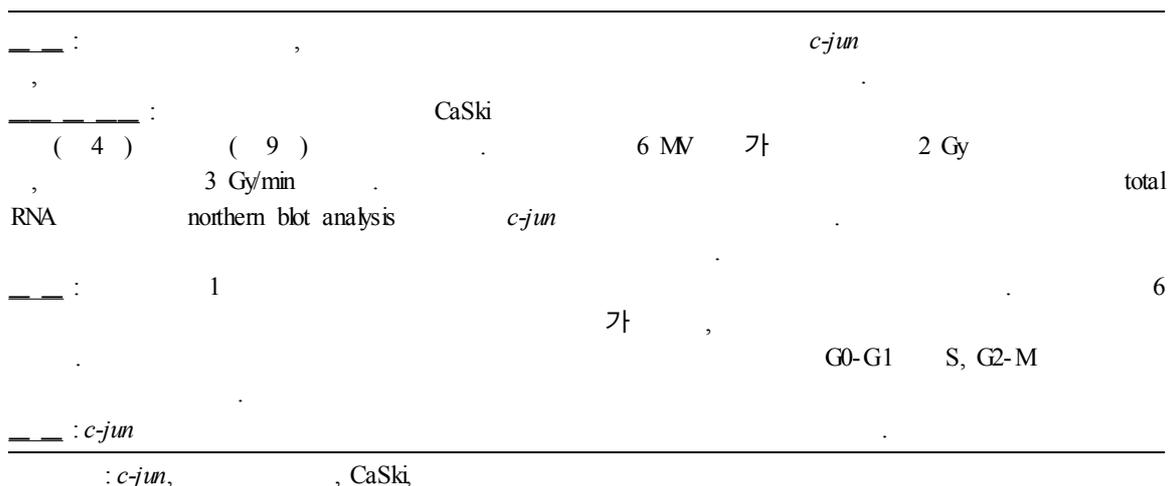


1)

CaSki

X-

c-jun



DNA 가 tyrosine kinase, protein kinase C (PKC), Raf-1 kinase, mitogen activated protein kinase (MAPK)

Datta ⁶⁾ TPA N-acetyl-L-cysteine reactive oxygen intermediates (ROI) Chae

PKC *cjun* tyrosine kinase herbimycin

cytokine ¹⁾ *cjun* etoposide, ara-C, cisplatin

leucine ^{2, 3)} *cjun* jun fos jun + jun homodimer activator protein-1 (AP-1) AP-1 DNA promoter element (5'TGAC/GTCA3') cytokine

1999 6 9 1999 8 13

Tel: 043 1)269- 6376 Fax : 043 1)269- 6387
 E- ma il : wypark@med.chungbuk.ac.kr

medium liquid holding medium , complete 가

c-jun 가 2 3 (β, 8) 10%
cjun 가 24 10%
 가 1 RNA (cs).
cjun 가 (ci)
 (co) total RNA
cjun PBS RNazol B (Tel-Test, USA)
 guanidine isothiocyanate
cjun RNA 20 μg RNA 12
 % agarose/formaldehyde gel (Amresco, USA)
 nylon membrane (Hybond-N+, Amersham, USA) 50% formamide
 가 Hybrisol I (Oncor, USA) 45 4
 prehybridization ³²P-labeled c-DNA probe 12
 16 hybridization SSPE, 0.1% SDS
 membrane *cjun* Phosphor imager
 (Biorad, USA) membrane GAPDH
 RNA loading
 3 *cjun* probe
 EcoR1 1.1kb
 3
 0, 1, 3, 5, 7, 9
 PBS 2 trypsin
 10% trypsin
 PBS 70% cold ethanol
 Propidium iodide (Sigma, USA) 37 30
 filter (aggregates)
 (flow cytometry) (facscalibur, Becton Dickinson, USA)
 3
 ANOVA (G0G1S;G2M)
 9 (%)
 2 Gy 3 Gy
 2. Northern blot analysis
 Northern blot RNA 15, 30,
 45, 1, 1.5, 2, 3, 6

49.7% , S 35.3% 33.2% G2-M
 13.3% 17.1%
 가

(cell loss)

G0-G1 S, G2-

1.

가
 (Fig. 1), 가 Sigmoid
 4 가
 9 , 4 70% M
 (confluency) , 6 95%
 . 95% 가

2. *c-jun*

cjun (co)
 가 1 15
 (Fig. 2). 가

(Fig. 3).

1 6
 6
 가 ,
 가 가
 가 가
 가 가

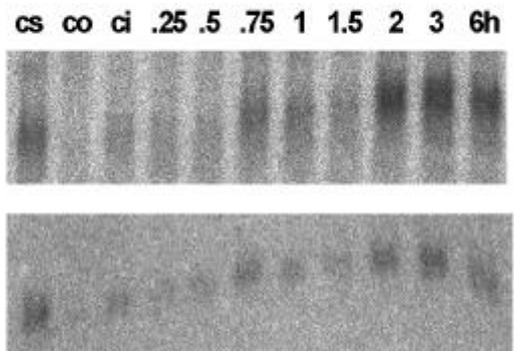


Fig. 2. Expression of *cjun* in exponential growth phase. Northern blot analysis of total cellular RNA levels was

3.

, G0-1 48.9%

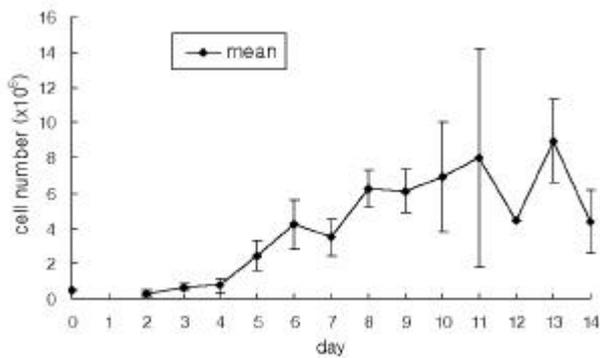


Fig. 1. Growth curve of CaSki cells according to the elapse of days.

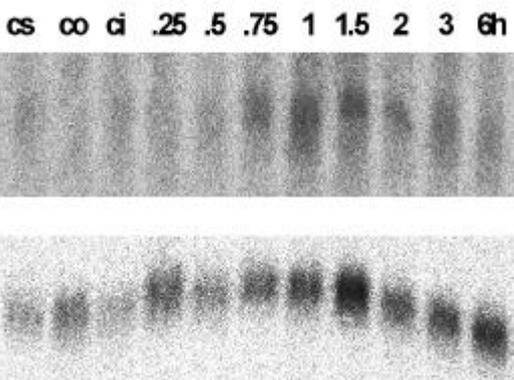


Fig. 3. Expression of *cjun* in stationary phase. Northern blot analysis of total cellular RNA levels was performed in CaSki cells after treatment with 2 Gy of ionizing radiation. Hybridization was performed using a ³²P-labeled *cjun* or GAPDH DNA probe. There were three nonirradiated control groups; cs (serum activation), co (room temperature during irradiation), ci (inside the CO₂ incubator during irradiation).

c-jun

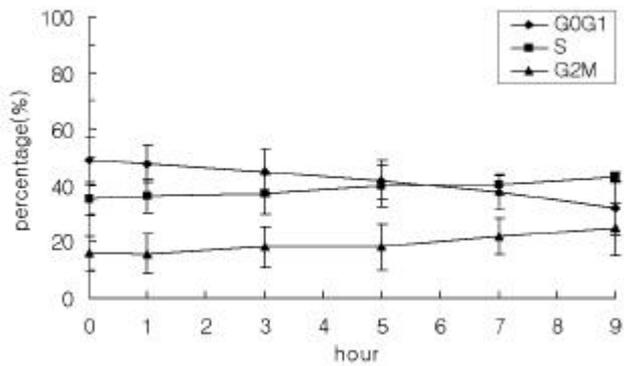


Fig. 4. Cell cycle distribution in exponential growth phase. Cell cycle distribution was analyzed using flow cytometry according to time course after 2 Gy irradiation.

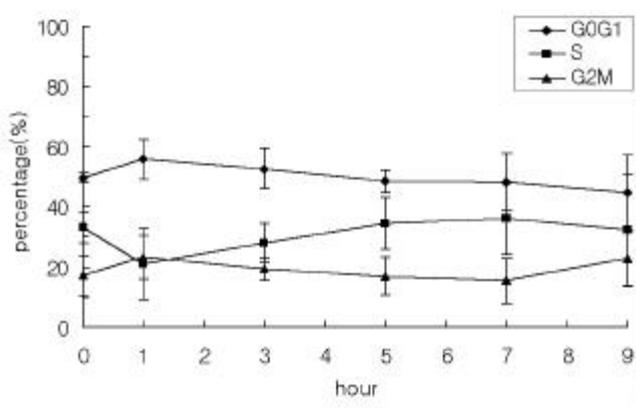


Fig. 5. Cell cycle distribution in stationary phase. Cell cycle distribution was analyzed using flow cytometry according to time course after 2 Gy irradiation.

($p=0.5161$).

cjun 가 Sherman ²⁾
HL-60 AG-1522 20 Gy
, 14.3 Gy/min 3
24
, (2, 5, 10, 20, 50 Gy) 3
가 가
10 Gy 20 Gy 14.3
Gy/min 0.67 Gy/min 가
. Manome ³⁾

20 Gy, 14.3 Gy/min 3
24
. Hallahan ⁹⁾
RIT-3 STSAR-5 20 Gy 3
1 3 가
. Prasad ¹⁰⁾ 244B
(0.25 2 Gy) *cjun* 0.5 Gy,
1.17 Gy/min 1
12
, (0.25,
0.5, 0.75, 1, 2 Gy) 1 0.5 Gy
. Collart ¹¹⁾
, , , , 50 Gy,
1.5 Gy/min 2
cjun , ,
cjun 0.25 50 Gy
, 1 3
1
2 24 가 , 2, 3, 9, 10)
1
6 가 ,
가 가
cjun ,
(limited nut-
rient supply) (density-inhibition) 가 ¹²⁾
, contact inhibition G0-1
(unfed
culture) ^{13, 14)}
2 3

contact inhibition
 plateau 가 cell loss DNA가 check-
 가 (cell loss factor)가 70% , point 가
 CaSki , DNA가 p53 G1 가 PLD
 가 (EF-1 delta) 가, cyclin B , elongation factor-1
 3 G2 checkpoint ,
 (1), 1 ,
 (2), (3)
 12)
 PBS 2 , Abate 21) fos-jun heterodimer DNA
 , fos jun DNA cy-
 steine ,
 Price 22) G1 gadd45
 , gadd153, glucose-regulated protein 78 (GRP 78) mRNA
 가 가 ,
 cytokine ,
 , gadd45 ,
 16, 17) 가 가 가 gadd45
 , cytokine ,
 , G1 DNA ,
 가 Hallahan 15) ,
 , *cjun* ,
 dominant negative(delta 9) , G1 가 *cjun*
 G1/S ,
 , *cjun*
 가 *cjun*
 G0-G1 S, G2-
 M , *cjun*
 ,
 ,
jun ,
 ,
 8)

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Abstract

Expression of *c-jun* by X-ray According to Cell Growth State in CaSki Cell Line

Seong Sun Jang, M.D., Woo Yoon Park, M.D.

Department of Therapeutic Radiology, College of Medicine, Chungbuk National University, Cheongju, Korea

Purpose :The expression pattern of *c-jun* by ionizing radiation according to cell growth state (exponential growth phase vs. stationary phase) and its relationship with cell cycle redistribution were investigated.

Materials and Methods :The exponential growth phase (day 4) and stationary phase (day 9) cells were determined from cell growth curve according to the elapse of days in CaSki. The cells were irradiated using 6 MV X-ray with a dose of 2 Gy at a fixed dose rate of 3 Gy/min. Northern blot analysis was performed with total cellular RNA and cell cycle distribution was analyzed using flow cytometry according to time-course after irradiation.

Results :The maximum expression of *c-jun* occurred 1 hour after irradiation in both exponential growth and stationary phase cells. After then *c-jun* expression was elevated upto 6 hours in exponential growth phase cells, but the level decreased in stationary phase cells. Movements of cells from G0-G1 to S, G2-M phase after irradiation were higher in exponential growth phase than stationary phase.

Conclusion :*c-jun* may be involved in the regulation of cellular proliferation according to the growth states after irradiation.

Key Words :*c-jun*, Cell growth state, CaSki, Cell cycle distribution