

c-myc
 (5) TGF TGF
 (6-9) TGF TGF 1 jun, fos, src, abl, ras TGF 1 TGF 1
 p53 TGF DNA G1 (apoptosis)
 (11) p53 (12,13) 40 80% p53 44.6% (14) K-ras 70 95% (15,16) K-ras codon 12
 (17) TGF 1, T IIR, p53 K-ras
 1) 31 5 26
 61.4±10.5 , 17 9
 65.6±9.9 , 3 2
 (1989 UICC)

2)
 (1) TGF 1, T RII p53 :
 TGF 1, T RII 2 TGF 1 TGF 1
 328 353 (Santa Cruz Biotechnology, Inc., Santa Cruz, CA, USA) TGF 1 (isoform) T RII T R II 246 266 (Santa Cruz Biotechnology, Inc.) TGF 1 p53 DO7 (Novo-Castra, Manhasset, NY, USA)
 (2) : 4µ m 30 30 4°C
 avidin-biotin complex biotin 2 IgG

Table 1. Clinical characteristics of the patients with pancreatic cancer (n=26)

Parameters	Number (%)
Sex	
Male	17 (65.4)
Female	9 (34.6)
Location of tumor	
Head	21 (80.8)
Body/tail	5 (19.2)
Size of tumor	
< 4.0 cm	13 (50.0)
≥ 4.0 cm	13 (50.0)
Cellular differentiation of tumor	
Well	3 (11.5)
Moderate	13 (50.0)
Poor	10 (38.5)
Lymph node metastasis	
Without	11 (42.3)
With	15 (57.7)
Tumor stage	
I	11 (42.3)
II	0 (0.0)
III	13 (50.1)
IV	2 (7.6)

45	streptoavidin	TGF 1, T R II	p53
peroxidase 30	PBS 3		1
	PBS 2 mM H ₂ O ₂	0.06 mM	100
3,3'-diaminobenzidine tetrachloride			10%
hematoxylin	80% glycerol		
gelatin	TGF 1, 2	(3)	DNA :
	kit		
		30	0.3% H ₂ O ₂
p53	TGF 1, T R II	mg 14,000 rpm 10	1
		200μl	proteinase K (10 mg/ml) 10μl
	p53	55°C 4	
		phenol-chloroform	DNA
		3 M sodium acetate (pH 5.2)	100%
		- 20°C	14,000 rpm 10

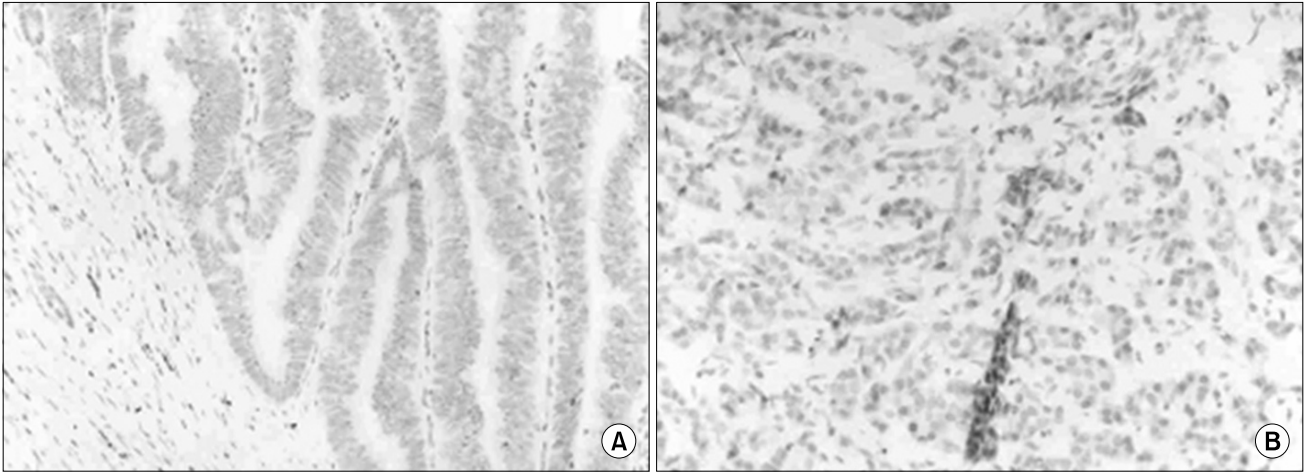


Fig. 1. Immunohistochemical staining of TGF 1. (A) Brownish positive TGF 1 cells are strongly expressed in the cytoplasm of pancreatic cancer cell. (B) However, in the ductal epithelial cell of normal pancreas, TGF 1 is faintly expressed (ABC stain, ×100).

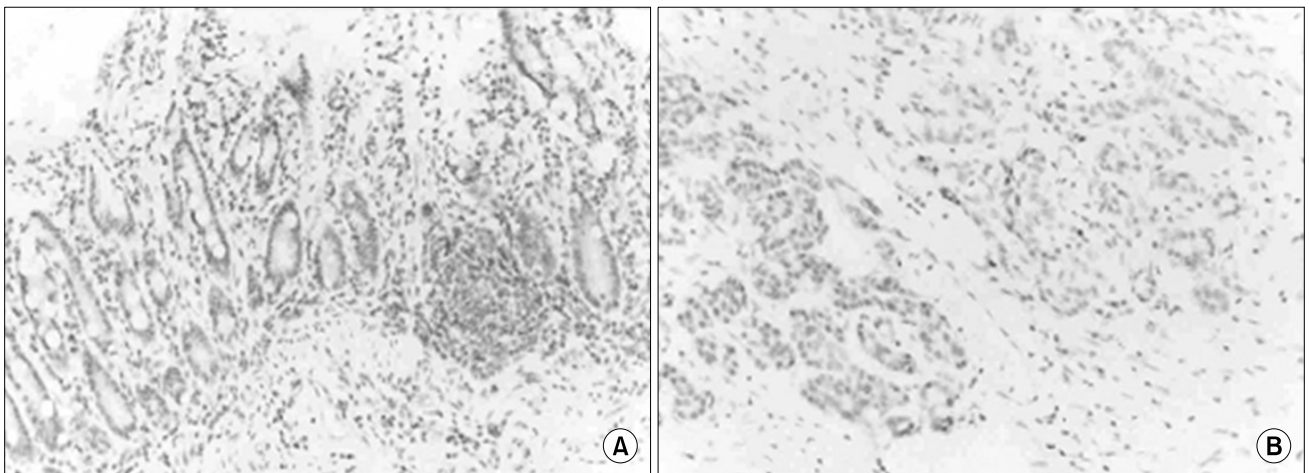


Fig. 2. Immunohistochemical staining of T R II. (A) Diffuse, brownish stained T R II cells are found in the pancreatic cancer cell. (B) However, in the normal pancreas, T R II is faintly expressed (ABC stain, ×100).

100µl DNA 70% 30 73°C 7 . K-ras primer
 - 20°C PCR 157 bp PCR 10µl
 (4) K-ras : (poly-merase chain reaction-restriction fragment length polymorphism, PCR-RFLP)
 K-ras codon 12 PCR 143 bp Mval
 10X PCR buffer 5µl, 25 mM MgCl₂ 3 10µl PCR template DNA
 µl, 10 mM dNTP 1µl, 100µM primer 2.5µl, PCR 40
 Taq polymerase (Promega, USA) 2.5 unit 3 PCR Mval 20 unit
 40µl template DNA 37°C 6 10% agarose gel
 10µl 50µl primers ras codon 12 Mval
 Sense primer A-ACTGAATATAAACTTGTGGTAGTTG- ras Mval 135 bp
 GACCT PCR K-ras PCR-RFLP
 antisense primer B-TCAAAGAATGGTCCTGGACC , SW

Table 2. TGF 1, T R II, p53 protein expressions and K-ras mutation in pancreatic cancer

	Positive immunoreactivity (%)			K-ras mutation rate (%)
	TGF 1*	T R II†	p53	
Pancreatic cancer (n=26)	19/26 (73.1)	20/26 (76.9)	16/26 (61.5)	20/26 (76.9)
Normal pancreas (n=5)	2/5 (40.0)	2/5 (40.0)	0/5 (0)	0/5 (0)
P-value	0.008	0.008	0	0

* = transforming growth factor 1; † = transforming growth factor receptor II.

antisense primer C-TAATATGTCGACTA-
 AAACAAGATTTACCTC PCR
 94°C 5 , 94°C 1 , 55°C 1 , 73°C 30

480 , HT 29
 (5) : TGF 1, T RII p53
 K-ras chi-square
 test Kaplan-Meier
 Log-Rank test
 P 0.05

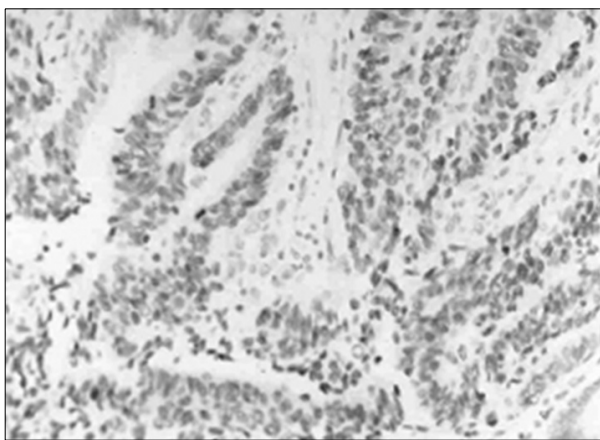


Fig 3. Immunohistochemical staining of p53 protein. p53 protein is strongly expressed in the pancreatic cancer cell (ABC stain, ×100).



Fig 4. Detection of K-ras point mutation. K-ras mutation is very commonly observed in the pancreatic cancer tissue. M, 20 bp ladder; P, mutant control (SW 480); N, wild type control (HT 29); lane 1-3, wild type K-ras at 106 bp in normal pancreas; lane 4-6, mutant K-ras at 135 bp in pancreatic cancer.

p53 16 61.5% (Fig 3), K-ras 76.9% (20/26) (Fig 4) p53 K-ras (P 0.01)(Table 2).

1) 26 21 (80.8%), 5 (19.2%) 4 cm 13 (50.0%), 4 cm 3 (11.5%), 13 (50.0%), 10 (38.5%) 11 (42.3%) 15 (57.7%), 11 (42.3%) 1989 UICC 1 11 (42.3%), 3 13 (50.1%), 4 2 (7.6%) (Table 1).

2) TGF 1, T RII, p53 K-ras TGF 1 T RII TGF 1 T RII 73.3% (11/15), 66.7% (10/15) TGF 1 T RII 36.4% (4/11), 27.3% (3/11) (P 0.01). UICC stage I TGF 1 36.3% (4/11), T RII 27.3% (3/11) stage III+ TGF 1 73.3% (11/15), T RII 66.7% (10/15) TGF 1 T RII (P 0.01)(Fig 1, 2). p53 26 (Table 3).

Table 3 Relation between the expression of TGF 1 and T RII and clinical characteristics in pancreatic cancer (n=26)

Parameters	TGF		P	T R II		P
	Negative (%)	Positive (%)		Negative (%)	Positive (%)	
Sex						
Male (n=17)	4 (64.7)	13 (76.4)	0.73	6 (35.3)	11 (64.7)	0.72
Female (n=9)	3 (33.3)	6 (66.7)		2 (22.2)	7 (77.8)	
Size of tumor						
4.0 cm (n=13)	3 (23.1)	10 (76.9)	0.81	4 (30.8)	9 (69.2)	0.82
4.0 cm (n=13)	3 (23.1)	10 (76.9)		3 (23.1)	10 (76.9)	
Differentiation						
Well (n=3)	0	3 (100)	0.75	1 (33.3)	2 (66.7)	0.68
Moderate(n=13)	4 (30.8)	9 (69.2)		2 (15.4)	11 (84.6)	
Poor (n=10)	2 (20.0)	8 (80.0)		3 (30.0)	7 (70.0)	
Lymph node metastasis						
Without (n=11)	7 (63.6)	4 (36.4)	0.008	8 (72.7)	3 (27.3)	0.007
With (n=15)	4 (26.7)	11 (73.3)		5 (33.3)	10 (66.7)	
Tumor stage						
I (n=11)	7 (63.6)	4 (36.4)	0.008	8 (72.7)	3 (27.3)	0.007
III+IV (n=15)	4 (26.7)	11 (73.3)		5 (33.3)	10 (66.7)	

Table 4. Relation between the expression of p53, K-ras mutation and clinical characteristics in pancreatic cancer (n=26)

Parameters	p53 protein		P	K-ras mutation		P
	Positive (%)	Negative (%)		Positive (%)	Negative (%)	
Sex						
Male (n=17)	9 (52.9)	8 (47.1)	0.95	13 (76.4)	4 (23.5)	0.94
Female (n=9)	6 (66.7)	3 (33.3)		7 (77.8)	2 (22.2)	
Size						
4.0 cm (n=13)	8 (61.5)	5 (38.5)	0.88	10 (76.9)	3 (23.1)	0.95
4.0 cm (n=13)	7 (53.8)	6 (46.1)		10 (76.9)	3 (29.1)	
Differentiation						
Well (n=3)	2 (66.7)	1 (23.3)	0.77	1 (23.3)	2 (66.7)	0.85
Moderate (n=13)	8 (61.5)	5 (38.5)		11 (84.5)	2 (15.4)	
Poor (n=10)	6 (60.0)	4 (40.0)		8 (80.0)	2 (20.0)	
Tumor stage						
I (n=11)	7 (63.6)	4 (36.4)	0.83	8 (72.7)	3 (27.3)	0.87
III+IV (n=15)	9 (60.0)	6 (40.0)		11 (73.3)	4 (26.7)	
Mean survival (months)	13.5±3.4	19.8±8.1	0.03	15.9±5.5	17.4±6.7	0.55

4) p53 K-ras

p53 9 (52.9%), 6 (66.7%)
4.0 cm

p53 8 (61.5%), 4.0 cm 7 (53.8%)

8 (61.5%), 2 (66.7%),
6 (60.0%) p53

I 7 (63.6%), III+IV 9 (60.0%)
p53

K-ras 13 (76.4%),
7 (77.8%)

4 cm 4 cm
10 (76.9%) K-ras

K-ras

I K-ras 8 (72.7%)
III+IV 11 (73.3%)
(Table 4).

5) TGF 1, T R II p53 K-ras

TGF 1 12.6±5.8
, TGF 1 18.5±6.4
(P=0.03) T R II

Table 5. Correlation of Postoperative Survival with Expression of TGF 1, T R II, p53 Protein and K-ras Mutation in Pancreatic Cancer (n=26)

Markers (number of tumors)	Postoperative survival period	
	Mean±SD (months)	Wilcoxon test
TGF 1		
Positive (n=19)	12.6±5.8	0.03
Negative (n=7)	18.5±6.4	
T R II		
Positive (n=20)	11.8±5.8	0.03
Negative (n=6)	17.9±5.2	
p53		
Positive (n=16)	13.5±3.4	0.03
Negative (n=10)	19.8±8.1	
K-ras		
Positive (n=20)	15.9±5.5	0.55
Negative (n=6)	17.4±6.7	

11.8±5.8, T R II
17.9±5.2 TGF 1 T R II
(p=0.03). p53
16 13.5±3.4
10
p53
(P=0.03). K-ras
(Table 5).

5 0.4%

EGF (epidermal growth factor) TGF (transforming growth factor) (18)

ras (15) p53 (19, TGF

(89) c-myc (20)

TGF (transformation)

collagen T-

B- (chemotactic) (4)

TGF T R II

TGF T R II

(9,21-24) TGF 12.6±5.8

18.5±6.4 T R II

TGF T R II

(22) p53 TGF T R II

p53

13.5±3.4 p53

19.8±8.1 p53

p53 (25)

(26) (25)

(27) p53

p53

(14) p53

III IV
70%

I

K-ras

K-ras

(28) K-ras

76.9%

TGF , T R II p53

northern blot , in

situ hybridization

(89,24)

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