

Collagen

I. collagen
 가 , 20 -
 30% , ,
 , , 16,17),
 Collagen , 가
 , , 18 - 21),
 , 22), 23),
 , 19),
 가 24 - 26)
 가 ,
 1 - 3) 27,28),
 4,5) 29,30),
 6 - 8) 31,32)
 , 8,12,15,33 - 35)
 , gel³⁶⁾, sponge³⁷⁾,
 solutions³⁸⁾, membrane^{29,30)}
 가 ,
 collagen ,
 가
 , Rovira ³⁹⁾ cal -
 cium phosphate, elastin, collagen

가 , Mizuno collagen

40) type I collagen matrix가

collagen

, Marchand 41) collagen matrix 가 scaffold

Blumenthal 33), Yaffe 38) collagen gel II. 3 solution collagen

1. , Bell 34) tricalcium phosphate, hydroxyapatite collagen 2 200g Sprague - Dawley 24 , collagen , Helm 35) 가 12 collagen gel

Collagen 2. , Nevins 15) BMP collagen collagen 32.5mg/kg sodium pentobar - bital(,)

Horisaka 42) BMP collagen solution zephiran . 1:80,000 collagen 2% , # 15

, Mattsson 43) collagen . 5mm trephine bur

, Solheim 44) collagen paste가 collagen (COLLATAPE , COLLA - TEC. INC. U.S.A.) , 5 - 0 vicryl

12) collagen

3. collagen

1.5ml/kg ampicillin 1 1 ,
 3, 7, 14, 21
 3 paraformaldehyde(PFA)
 4% PFA 가
 10% EDTA 2
 가 ,
 4 μ m
 , hematoxylin - eosin(H - E)
 ,
 tartrate
 resistant acid phosphatase(TRAP)
 .
 4.
 Immunohistochemical stain
 collagen
 immuno - histochemical
 stain ABC kit(VECTASTAIN , Vector
 Laboratory Inc. U.S.A) avidin -
 biotin - peroxidase complex(ABC
 method⁴⁵)
 ethanol xylene deparaffinization
 rehydration , phosphate buffered
 saline(PBS) 10
 endogeneous peroxidase activity
 3% peroxide 10
 PBS 2 3 1
 normal serum block prima -
 ry antibody 4 16
 . 2 3 PBS
 biotinylated secondary anti -
 body 30 . PBS
 2 3 10 enzyme
 conjugate PBS 2
 3 substrate -
 chromogen mixture(3,3 - diaminobenzidine

in Tris - HCl buffer containing 6% hydrogen peroxide)

1% methyl green

III.

1. (H - E)

(1) 3

, 가

collagen

가

(Figure 1, 2)

(2) 7

, 가

(Figure 3, 4) .

(3) 14

7

가 , collagen
 (Figure 5, 6) .

(4) 21

collagen 가
(Figure 7, 8)

2.

(Figure 9, 10).

3. immunohistochem -
ical stain

14
11).

IV.

가

9 - 15)가

가 ,
가

가

가

가

gen

ion

acid

가

glutaraldehyde, formaldehyde, metal

20,47), hyaluronic

colla -

48).

가

collagen

9,49)

collagen

가

collagen

collagen
aldehyde

bovine type I collagen
47)

(Figure

collagen

가

lysine -

19 - 21,32)

collagen

가

가

12,19,20,32)

Ratner⁵⁰⁾

4가

foreign - body reaction

46)

6 - 8)

가

가

가

가

Collagen collagen

telopeptide , collagen collagen

19,20,51), 가

19,52), DeLustro 21,24)

bovine collagen 14

collagen 14

col - 21

lagen - BB collagen

collagen 14

가 25,51) 12)

Collagen 21

, Doillon 18), Chvapil Solheim 44) collagen paste

20) collagen 4

(porosity) collagen , Mattsson

가 53) , Deporter 43) col -

collagen lagen 1 2

, Marchand 41)

collagen matrix scaffold collagen

. DeLustro collagen

21) type I collagen 90% collagen

가 scaffold 가

collagen 7 가

DeLustro 21)

collagen , 21

가

12), Mattsson 43), Solheim 44) colla -

gen colla - 가

gen , collagen

Sprague - Dawley 24
 12
 5mm trephine bur
 collagen
 (COLLATAPE, COLLA - TEC. INC.
 U.S.A.) 3, 7, 14, 21
 3
 H - E
 TRAP
 immunohistochemical stain

1. 14
 14
 21

2. 7 가
 , 21

3. collagen
 14
 collagen
 collagen

가 ,

VI.

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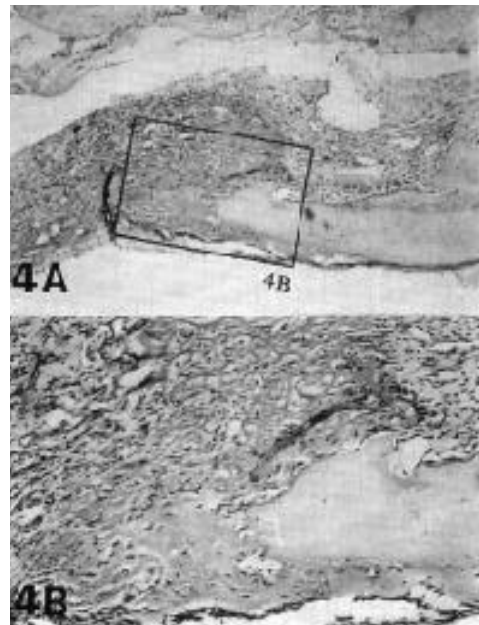
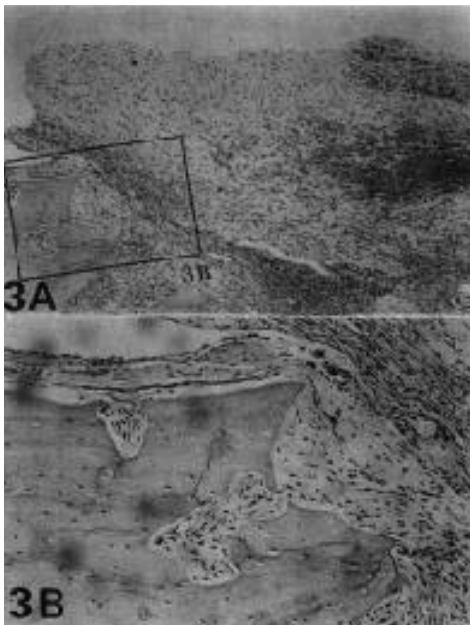
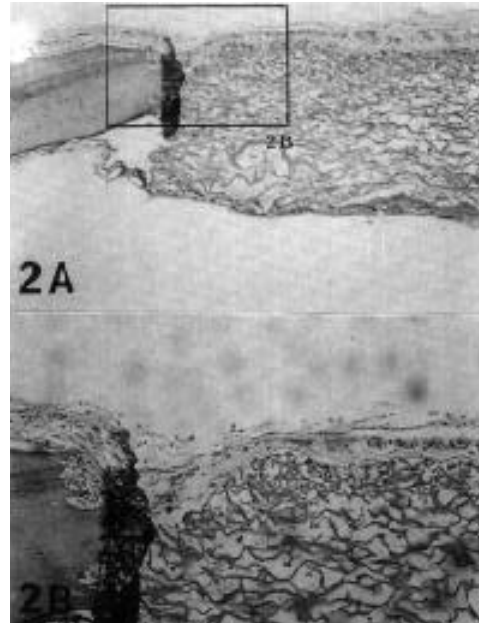
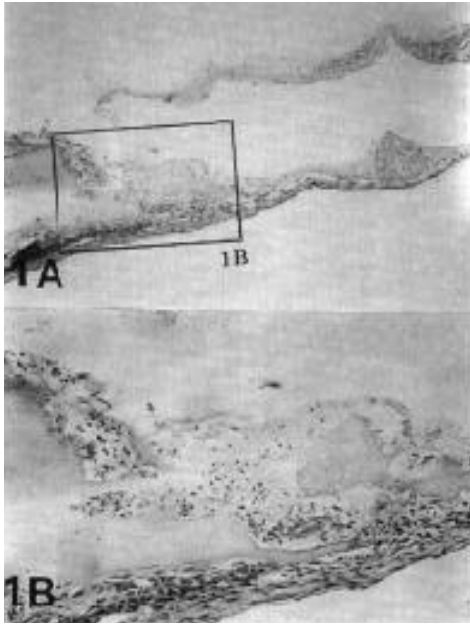
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12. , , :
13. , , , :
Fibronectin
가
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14. , , :
(TGF -
1)가
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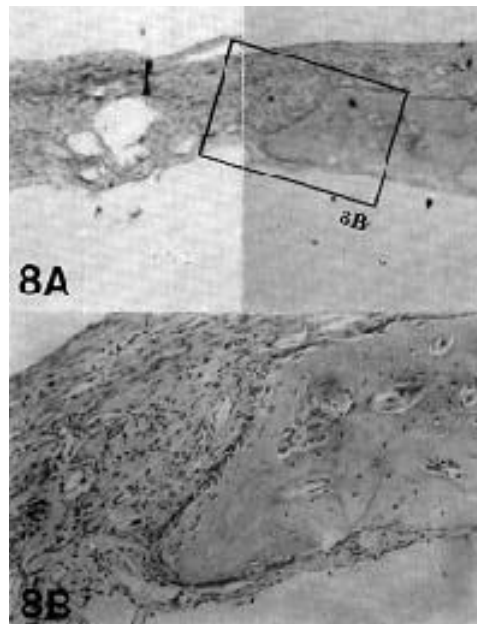
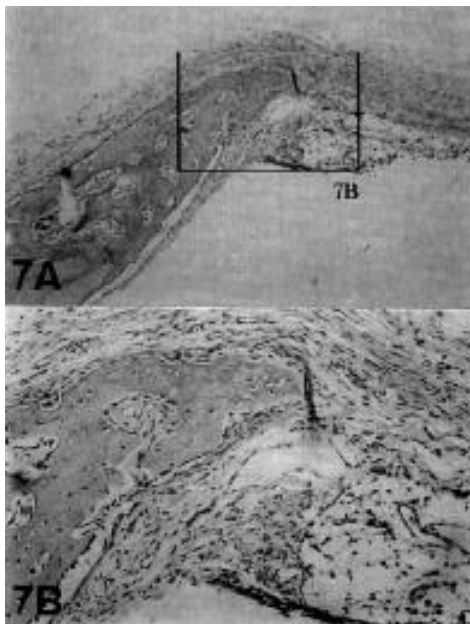
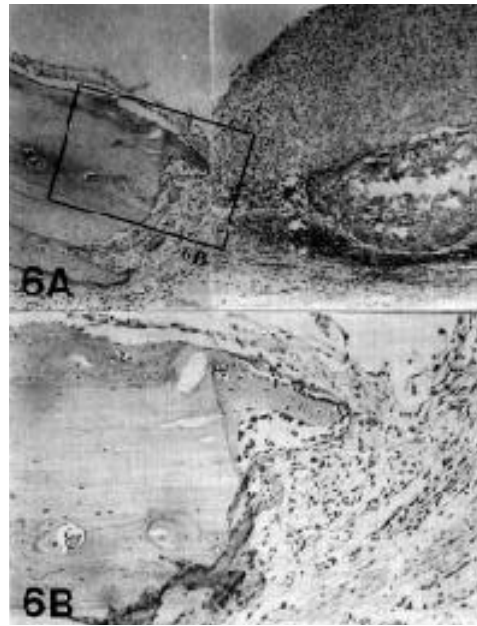
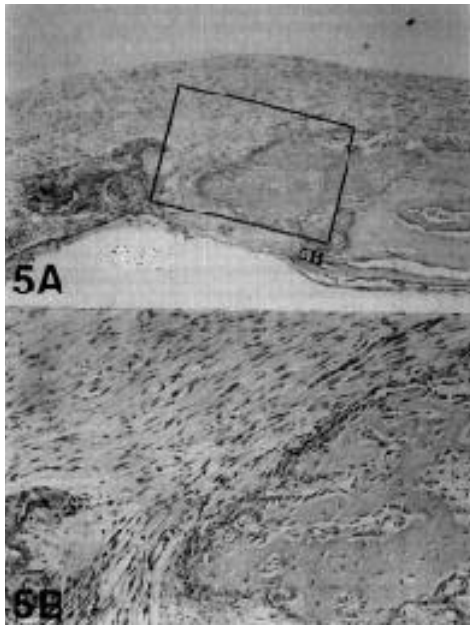
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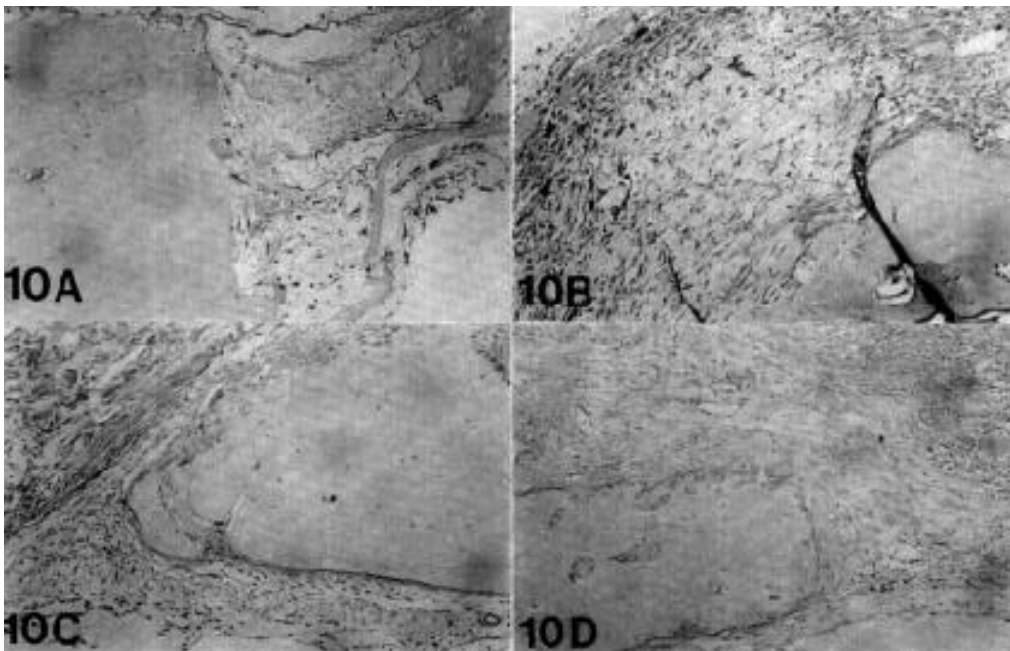
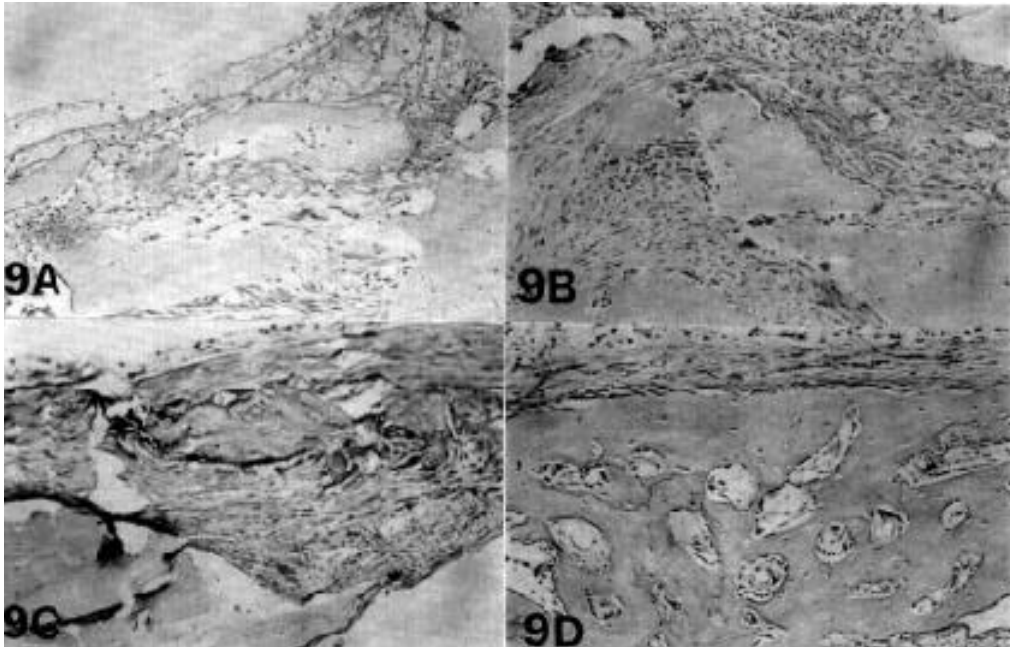
(I)



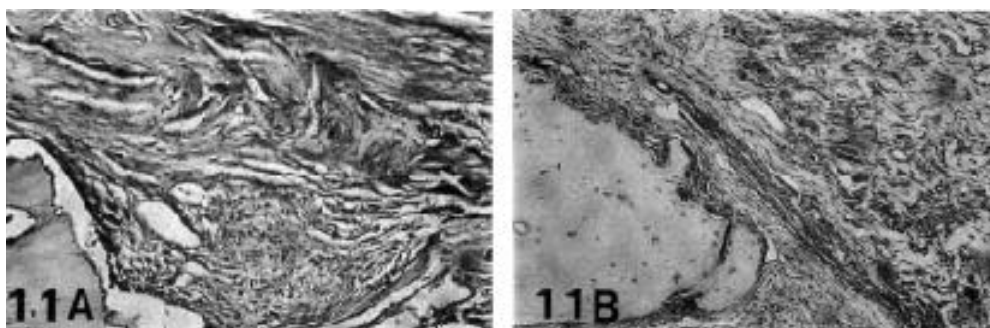
(II)



(III)



(IV)



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Figure 1. Control Group(3 days), (H&E, 1A × 40, 1B × 100)

Photomicrographs show fibrous connective tissue throughout most of the defect and infiltrated inflammatory cells on the periphery of the defect.

Figure 2. Experimental group(3 days), (H&E, 2A × 40, 2B × 100)

Photomicrographs show fibrillar collagen membrane of the defect and remarkable inflammatory cells infiltrated on the defect.

Figure 3. Control group(7 days), (H&E, 3A × 40, 3B × 100)

Photomicrographs show infiltrated inflammatory cells and some appositional growth of new bone on the periphery of the defect.

Figure 4. Experimental group(7 days), (H&E, 4A × 40, 4B × 100)

Photomicrographs show infiltrated inflammatory cells on the implanted collagen and edge of defect. Some appositional growth of new bone on the periphery of the defect is observed.

Figure 5. Control group(14 days), (H&E, 5A × 40, 5B × 100)

Inflammatory cells are reduced compare to day 7, but, most area of defect is filled with fibrous connective tissue. Note that appositional growth of new bone on the periphery of the defect.

Figure 6. Experimental group (14 days), (H&E, 6A × 40, 6B × 100)

Infiltrated inflammatory cells are still observed and some appositional growth of new bone on the edge of the defect is showed. Note that resorption of implanted collagen

Figure 7. Control group(21 days), (H&E, 7A × 40, 7B × 100)

Inflammatory cells are almost disappeared and trabecular bone formation is observed.

Figure 8. Experimental group(21 days), (H&E, 8A × 40, 8B × 100)

Photomicrographs show reduced inflammatory cells and trabecular bone formation. Note that biodegradation of implanted collagen.

Figure 9. Control group, TRAP × 100 (A : 3 days, B : 7 days, C : 14days, D : 21 days)

Photomicrographs show osteoclasts associated with remodeling of newly formed bone at day 14 and day 21.

Figure 10. Experimental group, TRAP × 100

(A : 3 days, B : 7 days, C : 14days, D : 21 days)

Photomicrographs show osteoclasts on the implanted collagen and newly formed bone, but specific relationship between implanted collagen and osteoclasts activity is not observed.

Figure 11. Immunohistochemistry for macrophages, × 100

(A : control group, 14 days B : Experimental group, 14 days)

Photomicrographs show aggregated macrophages on the implanted collagen.

- Abstract -

The Effect of Fibrillar Collagen on Bony Healing of Calvarial Defect in Rats

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Department of Periodontology School of Dentistry, Kyungpook National University

Many researches have been reported that collagen as cellular stroma, matrix of grafting materials, mediator of agents for the purpose of promoting healing process in vivo, but the responses in vivo were seen various. The goal of this experiment is to assess the effect of collagen on bony healing, through histological evaluation of implanted collagen on the calvarial defect in rats. 2 - month - old Sprague - Dawley, 24 rats were used and 12 rats assigned to each group of control and test. Defect of 5mm in diameter was made on the calvarial bone with trephine bur. Following thorough saline rinse, defect of control group was left in empty and that of experimental group was filled with fibrillar collagen(COLLATAPE , COLLA - TEC. INC. U.S.A.) soaked in saline.

3 rats in each group were sacrificed at 3, 7, 14, 21 days after operation respectively, and the tissue blocks were prepared for light microscope with H - E for evaluation of overall healing, with TRAP(tartrate resistant acid phosphatase) for evaluation of osteoclastic activity and with immunohisto -

chemical staining for macrophages.

The results were as follows :

1. In the control group, inflammatory responses were disappeared at day 14, but, in the experimental group inflammatory infiltrates were reduced at day 21. Thus, the experimental group showed more severe soft tissue inflammation than control group.
2. Both control and experimental group showed slight appositional growth at day 7 and gradual bony growth to 21th day. But, complete bony healing of the defect was not shown. There was no significant difference in bony healing between control and experimental group
3. Specific response of macrophages for implanted collagen was observed at day 14 in the experimental group.

In conclusion, although fibrillar collagen caused inflammation of soft tissue during initial healing period, inflammatory responses by fibrillar collagen didn't inhibit bony regeneration and implanted collagen was biodegradaded by macrophages. Thus, we expect that fibrillar collagen can be used for useful mediator of graft materials or growth factors.