A-12. Inducible nitric oxide synthase(iNOS) is synthesized by human gingival tissue and cultured human gingival fibroblasts

지 숙[1], 김흥중[2], 국중기[3], 김병옥[1]
[1]조선대학교 치과대학 치주과학교실
[2]조선대학교 치과대학 구강해부학교실
[3]조선대학교 치과대학 생화학교실

Background

Nitric oxide is known to be an important inflammatory mediator, and is implicated in the pathophysiology of a range of inflammatory disorders. Although nitric oxide(NO) synthesis is increased in periodontal disease, little is known about the possible sources of production by gingival tissue. Transcription of the iNOS gene is activated by pro-inflammatory cytokines such as interleukin-1(IL-1), tumor necrosis factor alpha(TNF-α), interferon gamma (IFN-γ) and bacterial lipopolysaccharide(LPS).

Object

The aim of this study was to determinethe localization and distribution of inducible nitric oxide synthase(iNOS) in human gingival tissue, and to ascertain if human gingival fibroblasts express NOS-II when stimulated with IFN-γ and LPS.

Method

The distribution of NOS-II in inflammed and non-inflammed specimens and young(〈20 year-old) and old(〉55-year-old) specimens of human gingiva was studied using a polyclonal antibody against iNOS. Cultures of fibroblasts derived from healthy human gingiva were used for the cell culture experiments. In vivo aging of human gingival fibroblast cells was prepared from gingiva of old(〉55-year-old) and young(〈20-year-old) patients. In vitro aging of human gingival fibroblast cells was prepared by sequential subcultivations (3 to 4 passages as young, 15 to 16 passages as old). LPS of P.gingivalis was extracted by the hot phenol-water method and was analyzed by SDS-PAGE.

Results

The results from immunohistochemical staining of the tissue indicated an upregulation of NOS-II expression in inflamed compared to non-inflamed gingival tissue. Fibroblasts and inflammatory cells within the inflammed connective tissue were positively staining for iNOS. In addition, basal keratinocytes also stained strongly for iNOS in inflammed tissue specimens. There is no significant differ-

ence in between young and old tissues. When cultured human gingival fibroblasts were stimulated by IFN-γ and Porphyromonas gingivalis LPS, iNOS was more strongly expressed than when the cells were exposed to LPS or IFN-γ alone.

Conclusions

The data suggest that, as for other inflammatory diseases, NO plays a role in the pathophysiology of periodontitis