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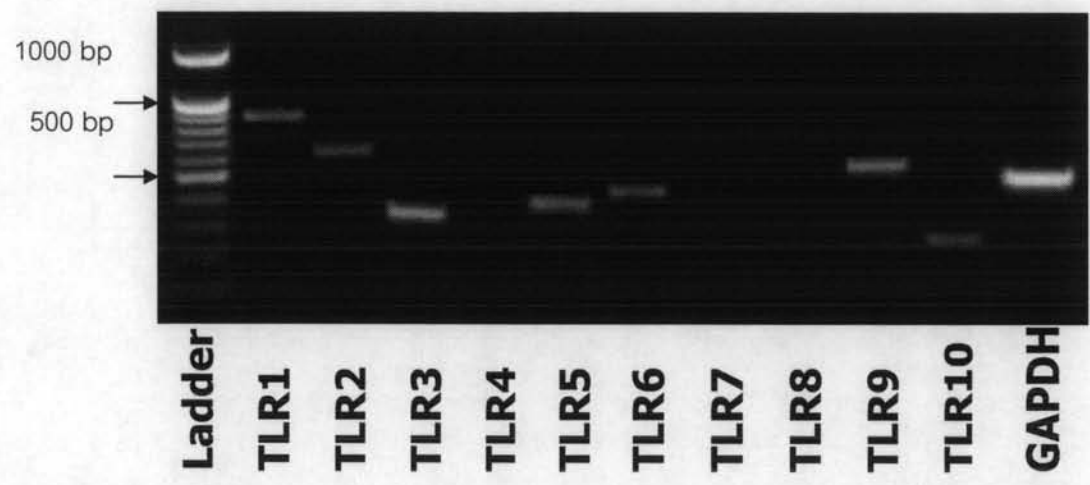
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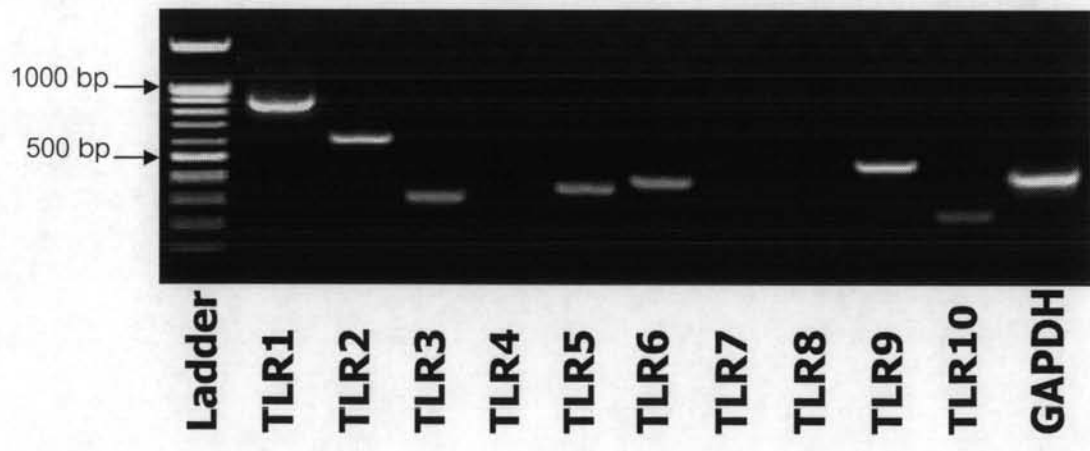
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## APPENDICES

7A)



7B)



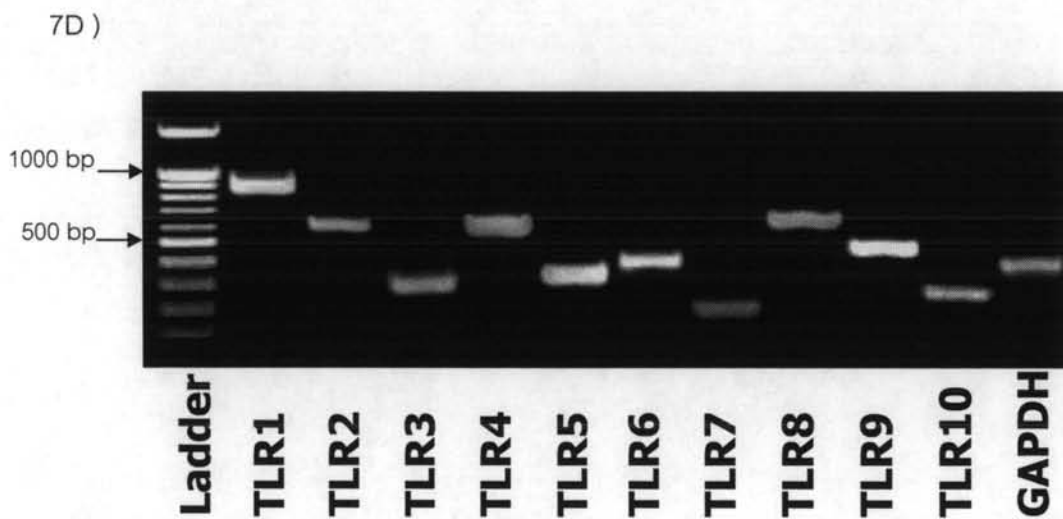
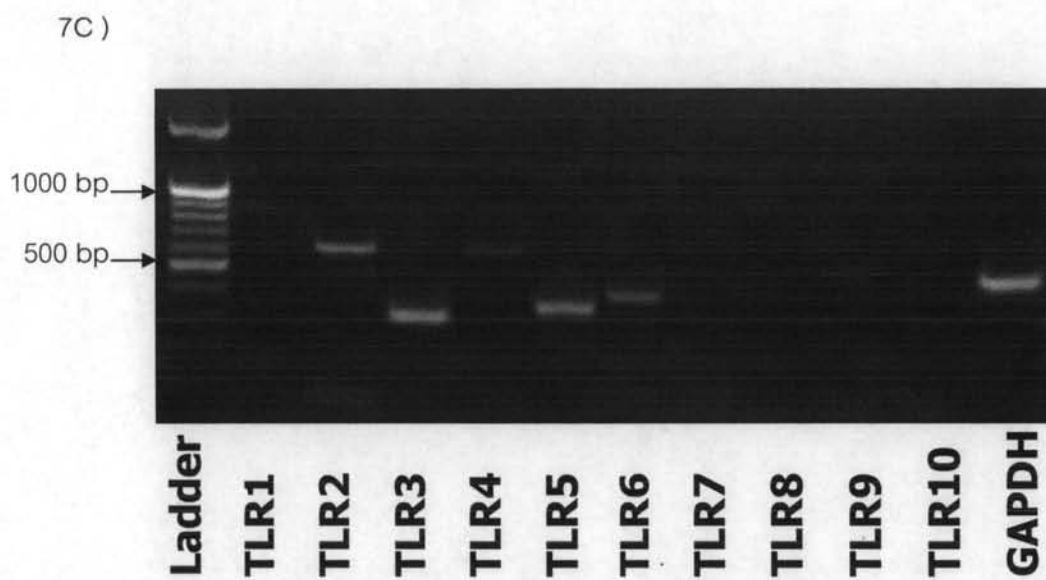
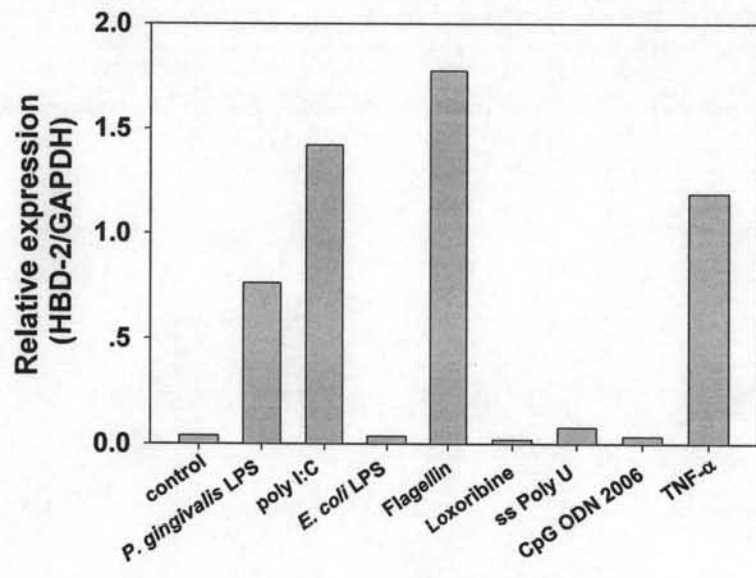
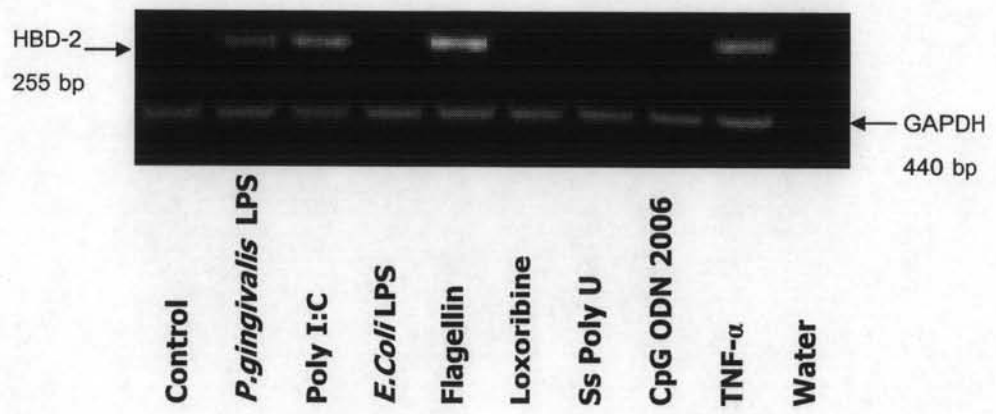
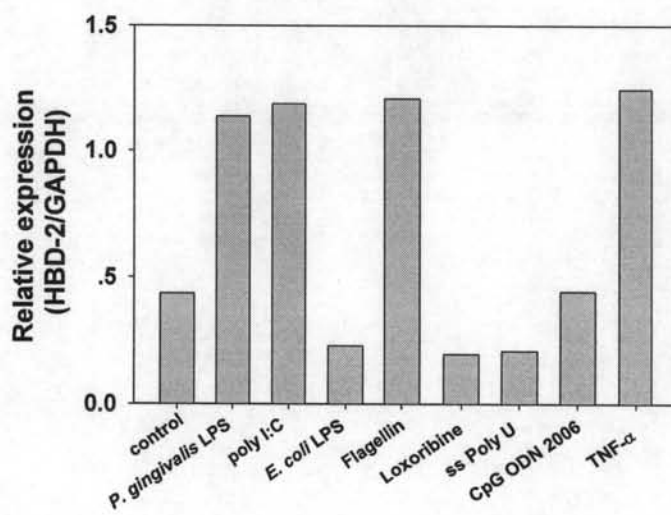
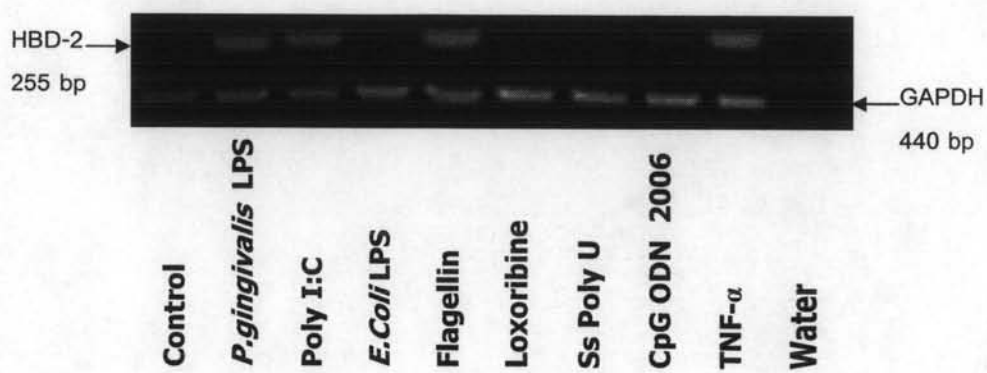


Figure 7. TLR expression in HGECS. TLRs1-10 mRNA was measured in cultured HGECS by RT-PCR (A,B,C). PBMC mRNA was used as positive control (D). GAPDH mRNA was used as an internal control.

8A)

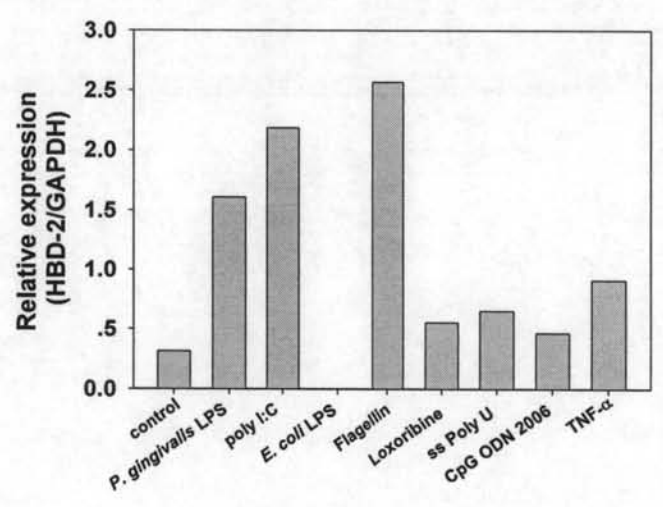
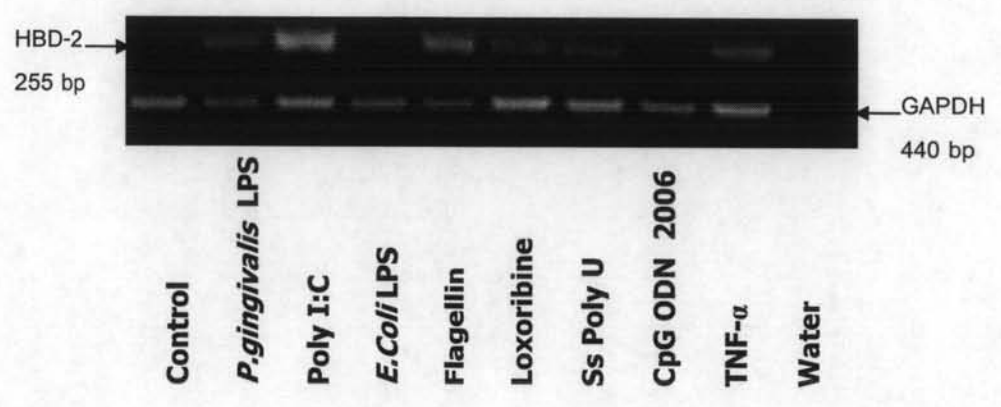


8B )





8C)



8D)

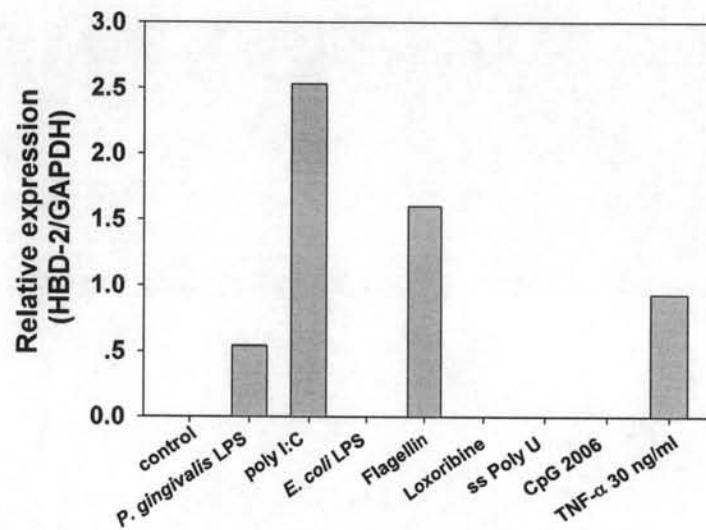
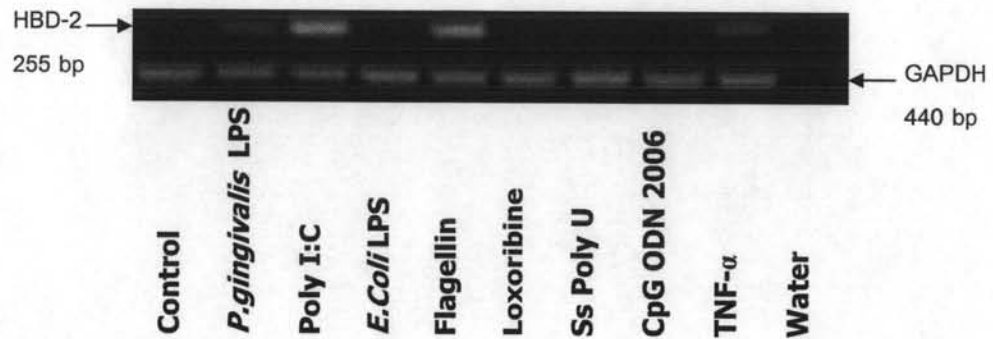
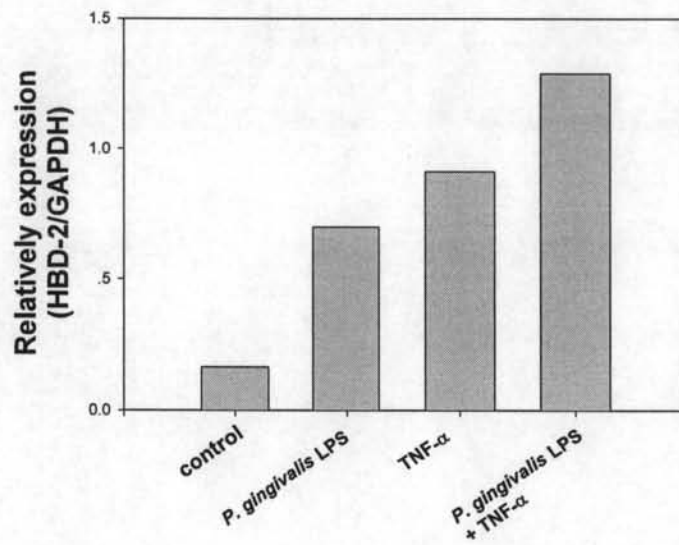
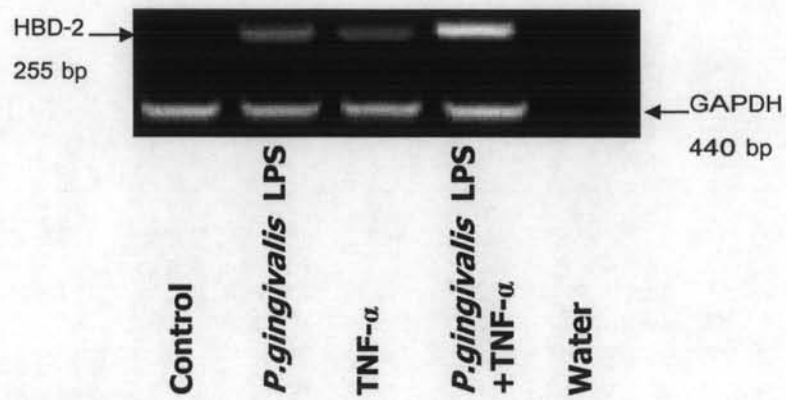
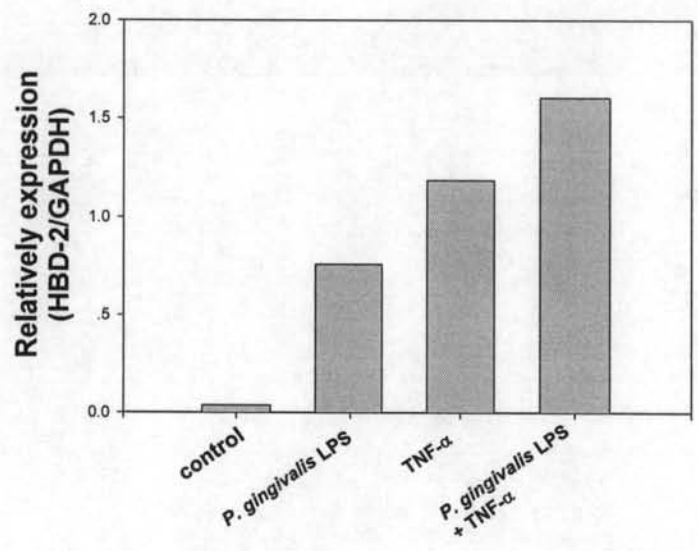
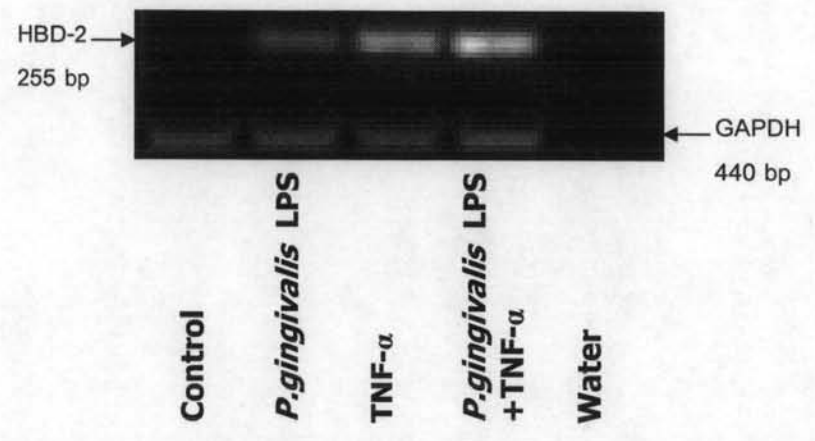


Figure 8A, B, C, D. Expression of HBD-2 in HGECs after stimulation with various TLR ligands. HGECs were cultured in 48-well plates, and stimulated with the following ligands: *P. gingivalis* LPS (TLR2 ligand), poly I:C (TLR3 ligand); *E. coli* LPS (TLR4 ligand); *S. typhimurium* (TLR5 ligand); Loxoribine (TLR7 ligand); polyU (TLR8 ligand); CpG ODN 2006 (TLR9 ligand). Culture medium was used as a control. After 24 h incubation, stimulated cells were harvested and mRNA expression of HBD-2 was analyzed by RT-PCR. TNF- $\alpha$ -stimulated HGECs were used as positive controls. GAPDH mRNA was used as an internal control. Semiquantitative analysis of HBD-2 expression is shown as relative expression (HBD-2:GAPDH).

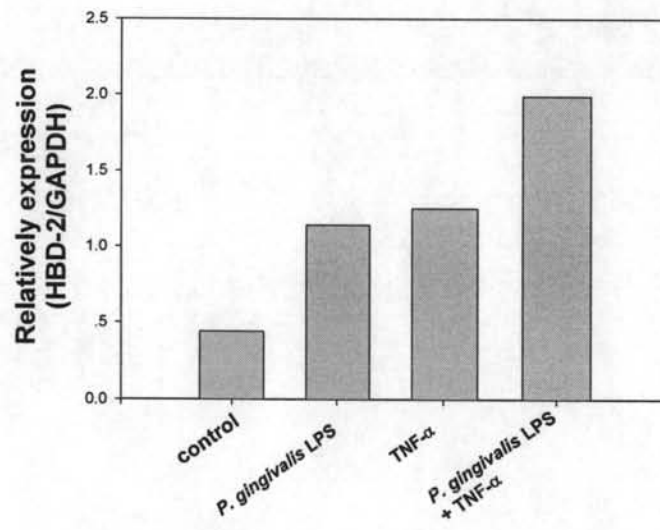
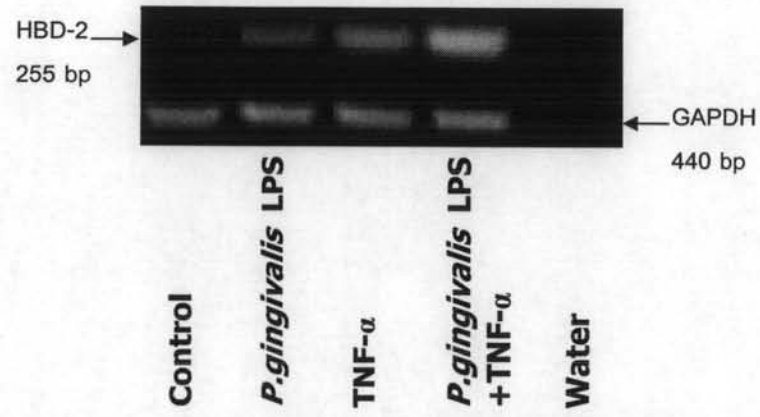
9A)



9B )



9C)



9D)

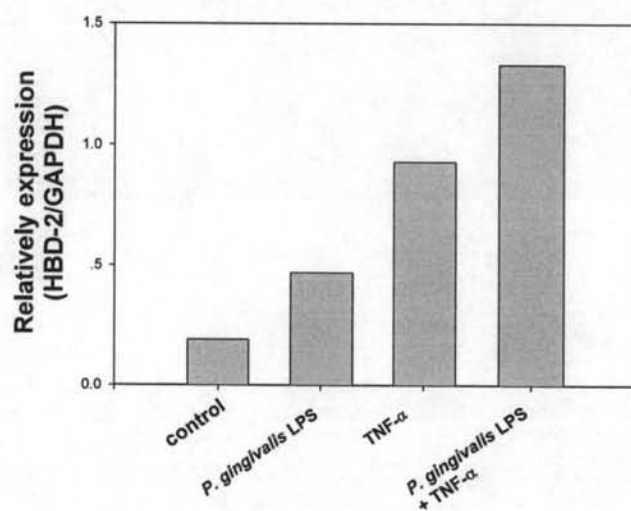
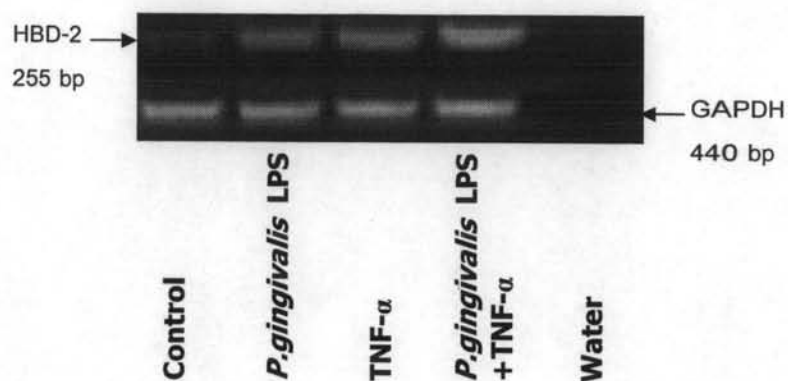
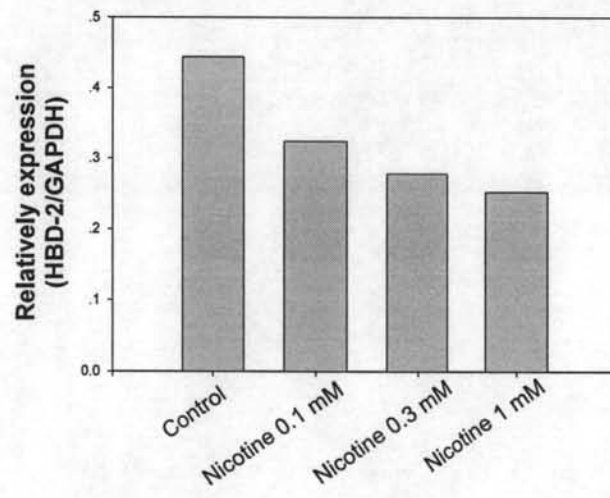
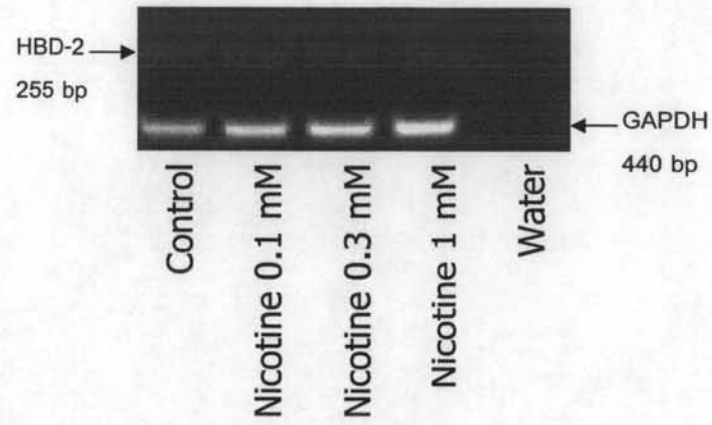


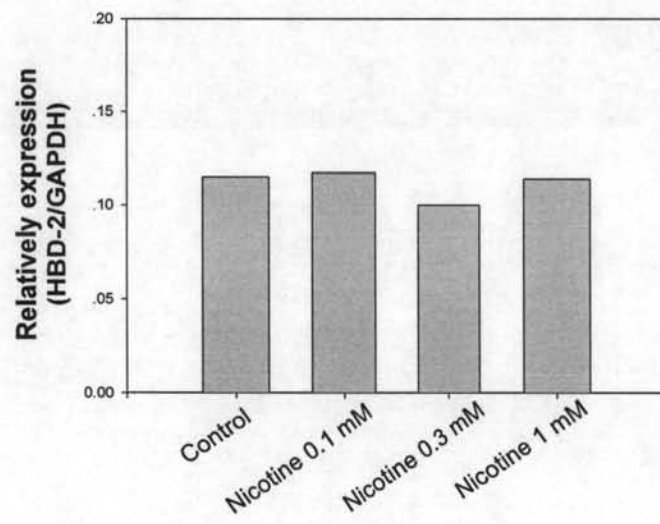
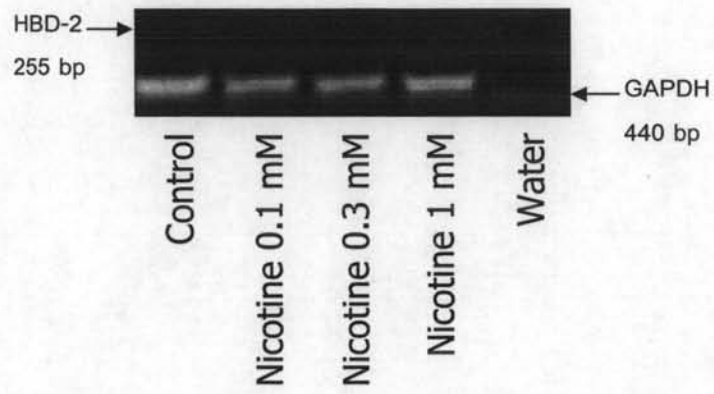
Figure 9A, B, C, D. Expression of HBD-2 in HGECS after stimulation with *P. gingivalis* LPS and TNF- $\alpha$  combination. HGECS were cultured in 48-well plates and stimulated with *P. gingivalis* LPS, TNF- $\alpha$ , or *P. gingivalis* LPS and TNF- $\alpha$ . Culture medium was used as a control. GAPDH mRNA was used as an internal control. Semiquantitative analysis of HBD-2 expression is shown as relative expression (HBD-2:GAPDH).



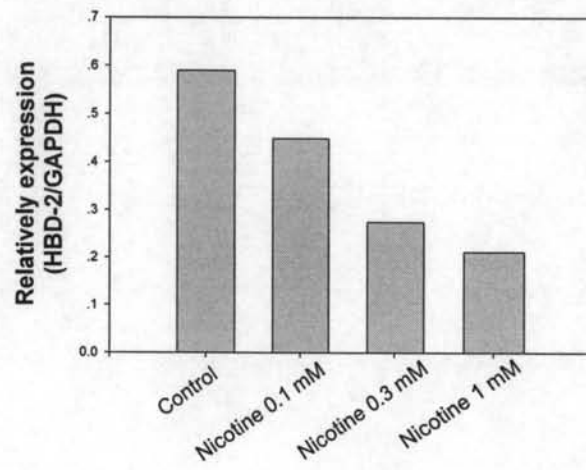
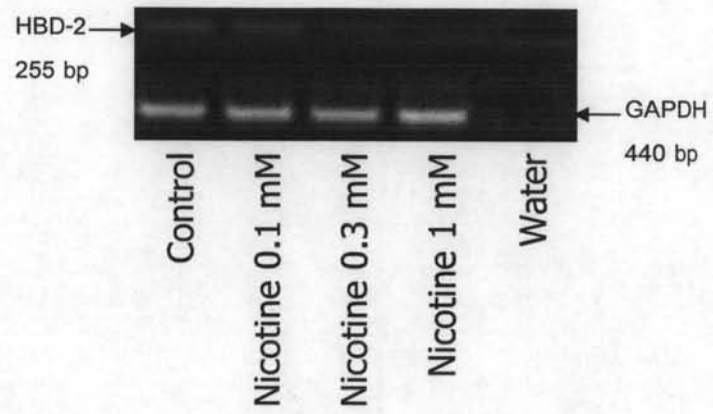
10A)



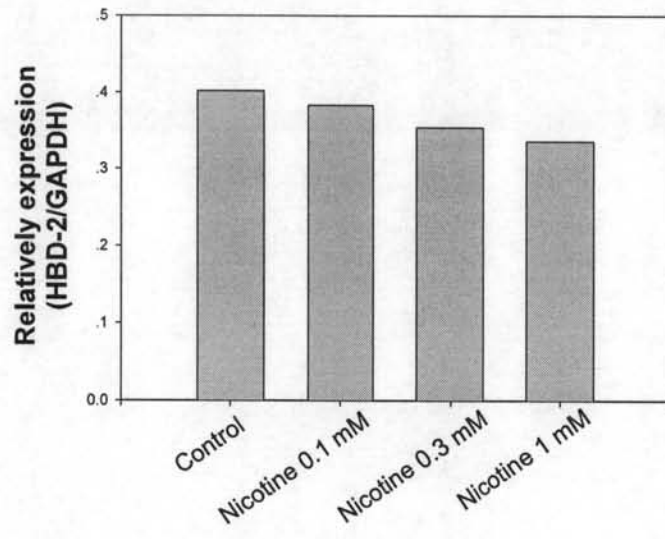
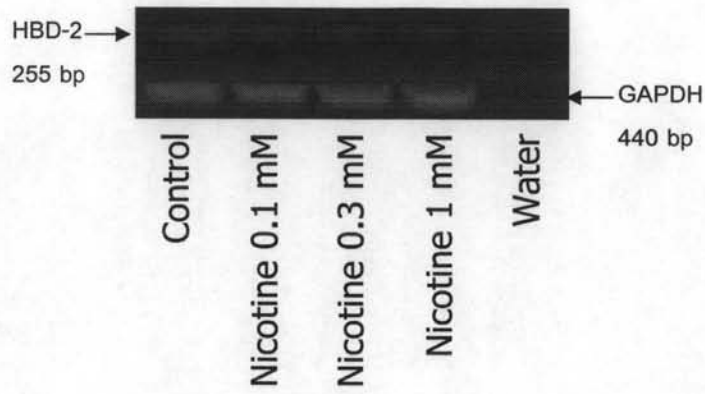
10B )



10C)



10D )



10E)

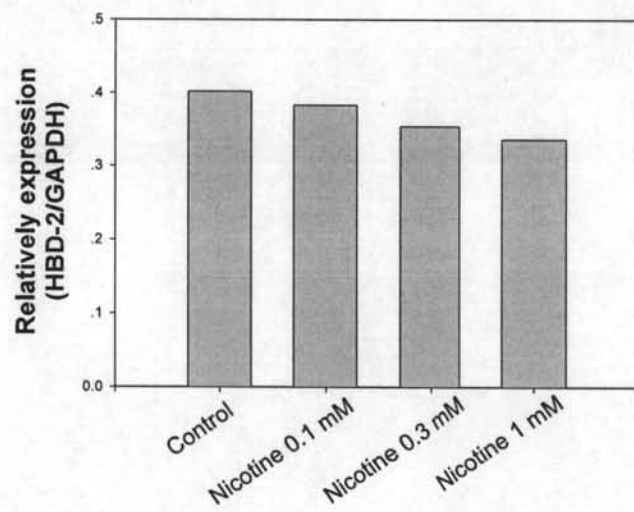
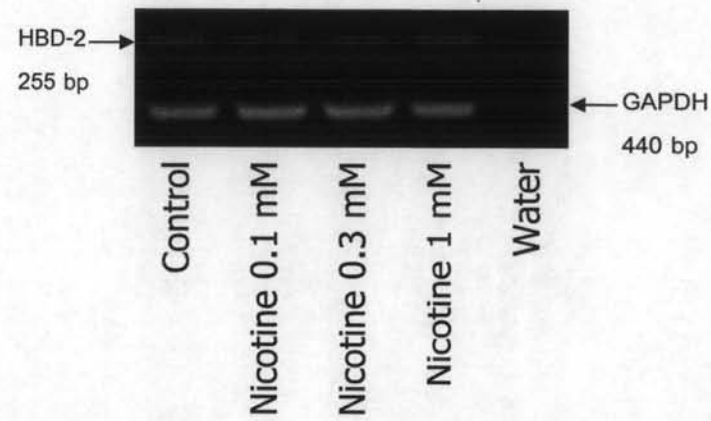
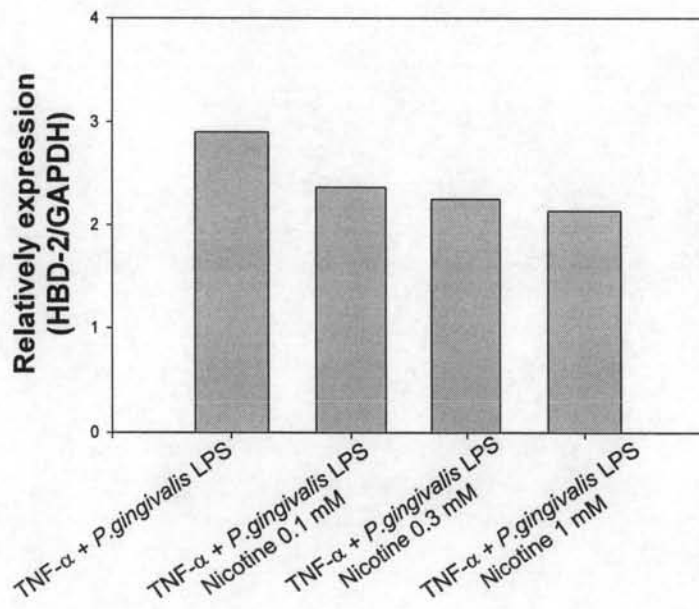
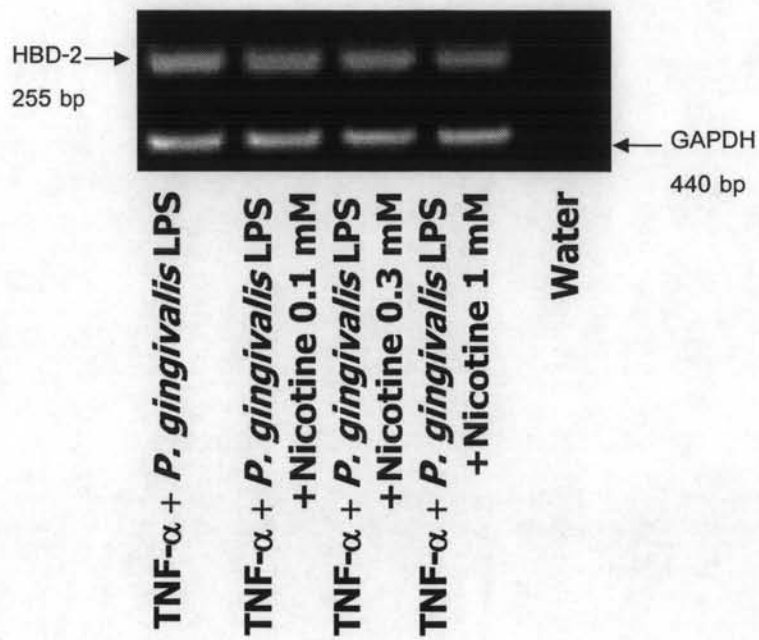
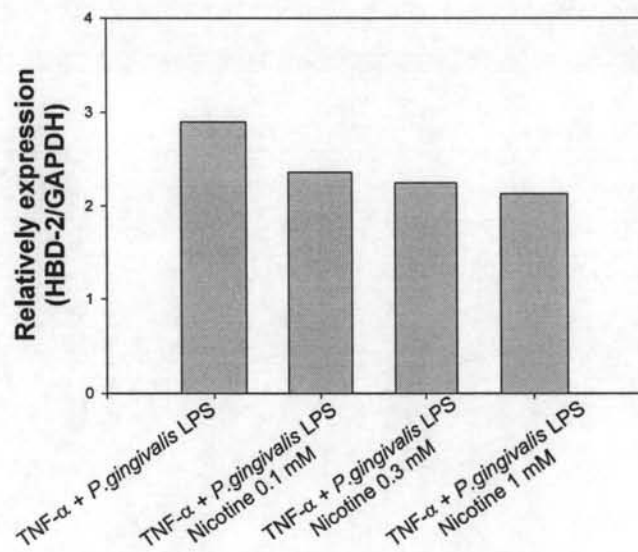
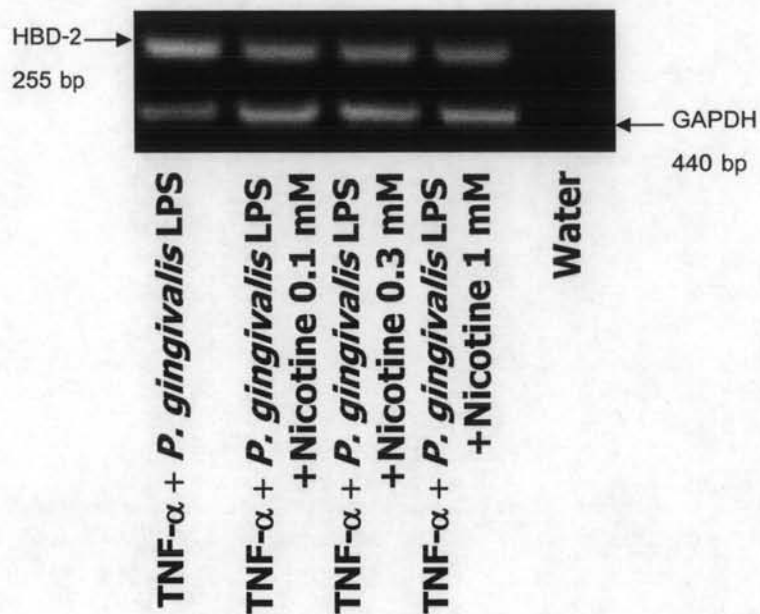


Figure 10A, B, C, D, E. The effect of nicotine on epithelial HBD-2 expression in unstimulated HGECs. HGECs ( $1.2 \times 10^5$  cells/ml) were treated with different concentrations of nicotine (0.1, 0.3, and 1 mM), Culture medium was used as a control. After 24 h incubation, treated cells were harvested and mRNA expression of HBD-2 was analyzed by RT-PCR. GAPDH mRNA was used as an internal control. Semiquantitative analysis of HBD-2 expression is shown as relative expression (HBD-2:GAPDH).

11A)

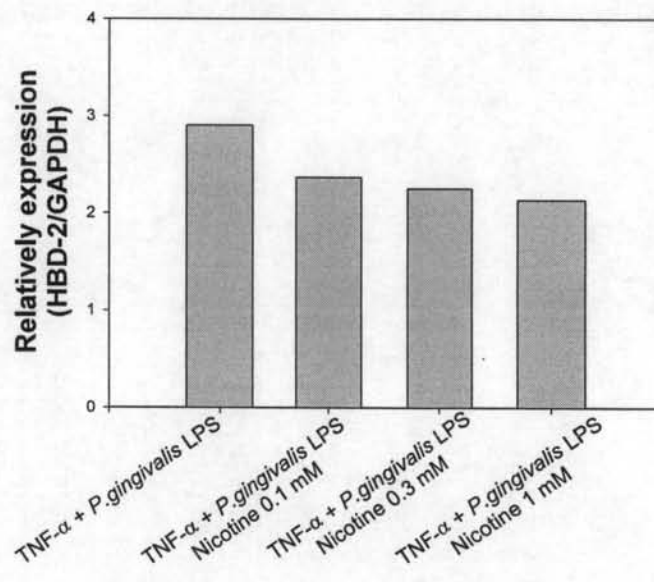
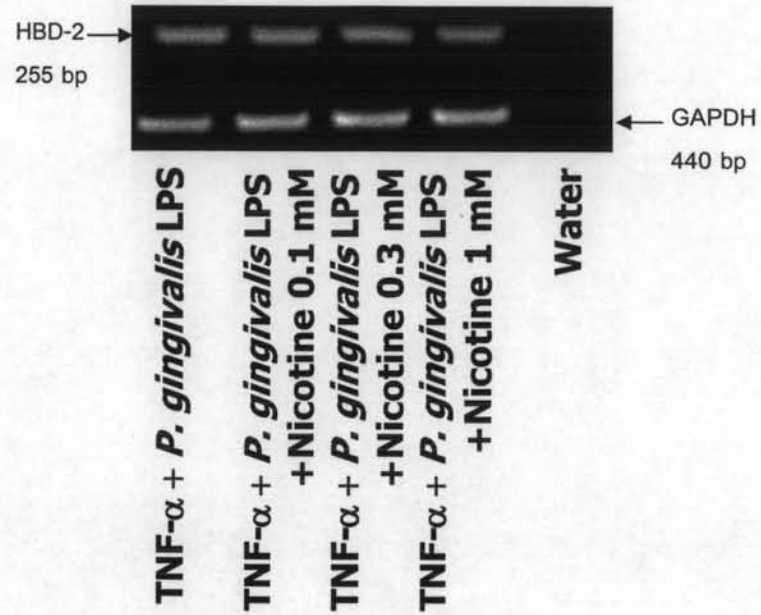


11B)

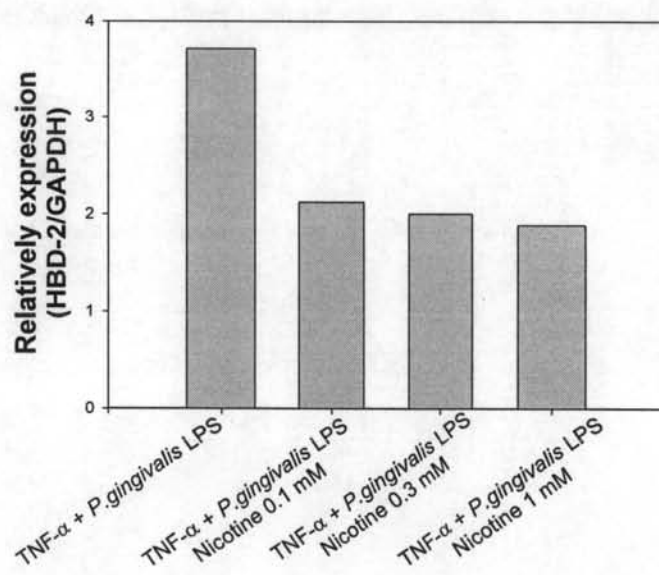
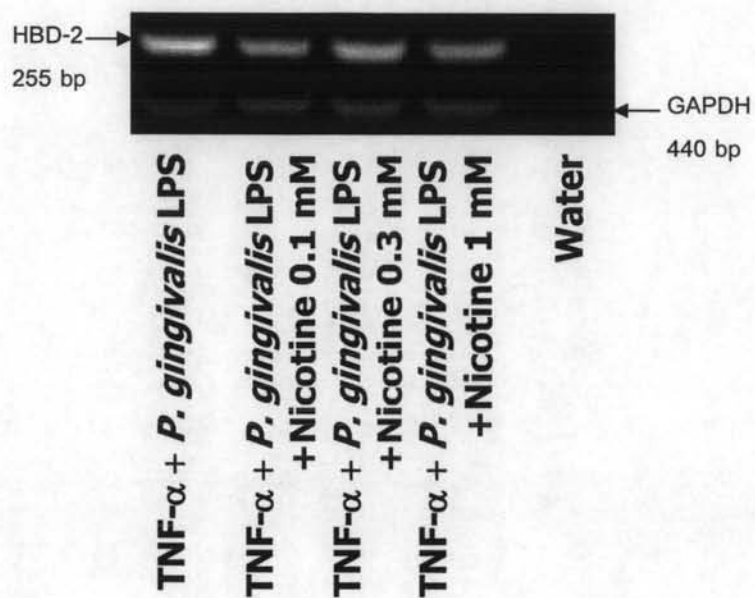




11C)



11D)



11E)

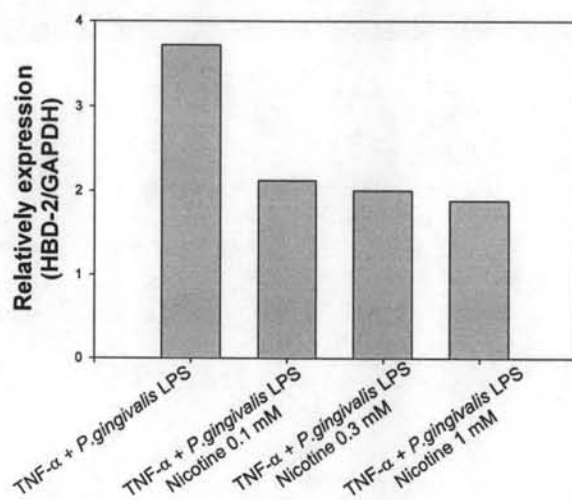
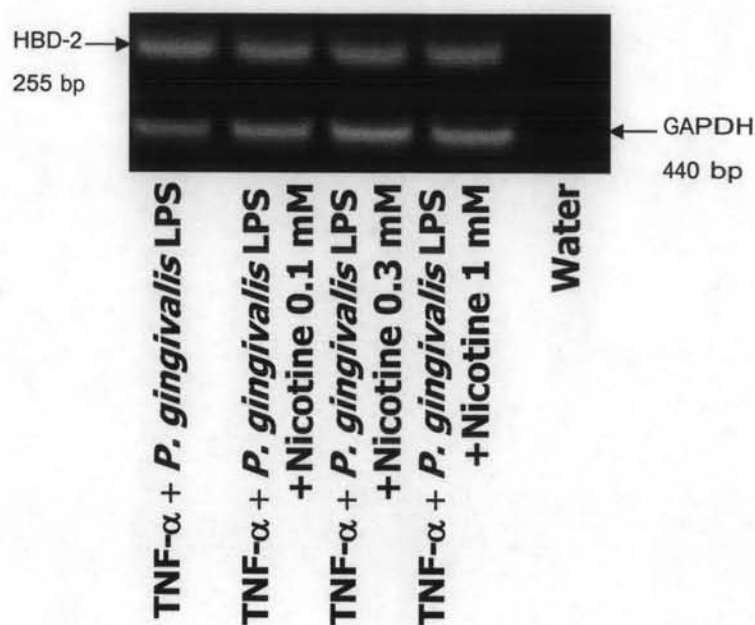


Figure 11A, B, C, D, E. The effect of nicotine on HBD-2 expression in stimulated HGECs. Nicotine (0, 0.1, 0.3, and 1 mM) was added into HGECs ( $1.2 \times 10^5$  cells/ml) which were stimulated with *P.gingivalis* LPS and TNF- $\alpha$  combination. After 24 h incubation, stimulated cells were harvested and mRNA expression of HBD-2 was analyzed by RT-PCR. GAPDH mRNA was used as an internal control. Semiquantitative analysis of HBD-2 expression is shown as relative expression (HBD-2:GAPDH).

## BIOGRAPHY

Miss Mutita Eksomtramate was born on 25<sup>th</sup> of April 1979 in Nakorn Sri Thammarat province. She graduated with D.D.S. (Doctor of Dental Surgery) from the Faculty of Dentistry, Prince of Songkhla university in 2003, and became a staff member of the Faculty of Dentistry, Prince of Songkhla university. She studied in Master degree program in Periodontology at Graduate School, Chulalongkorn University in 2005.