P8-4

Deletion of *actJ* gene affects growth and biofilm formation of *Streptococcus mutans*

Yang Chen[1, 2], Yangyang Pan[1, 3] , Qizhao Ma[1, 2], Shuxing Yu[1, 2], Yuqing Li[1*], Jing Zou[1, 2*]

1 State Key Laboratory of Oral Diseases, National Clinical Research Center for Oral Diseases, West China Hospital of Stomatology, Sichuan University, Chengdu, China

2 Department of Pediatric Dentistry, West China Hospital of Stomatology, Sichuan University, Chengdu, China

3 Department of Pediatric Dentistry, School and Hospital of Stomatology, Wenzhou Medical University, Wenzhou, China

Objective

GCN5-related N-acetyltransferases (GNAT) family proteins are a diverse group of lysine acetyltransferases (KATs) widespread in bacteria. However, the function of acetyltransferases in *S. mutans* remains unknown. The purpose of this study was to analyze the influence of gene *actJ* on growth and biofilm formation in *S. mutans*.

Material & Methods

We constructed the *actJ* in-frame deletion strains of *S. mutans* UA159 and the complement and overexpression strains of *actJ*. The effects of gene *actJ* on planktonic cultures were assessed by growth curve assay. The biofilm biomass and the synthesis of exopolysaccharides (EPS) was measured with crystal violet staining and the anthrone-sulfuric method. Biofilm analysis and structural was imaging by confocal laser scanning microscope (CLSM) and scanning electron microscope (SEM).

Result

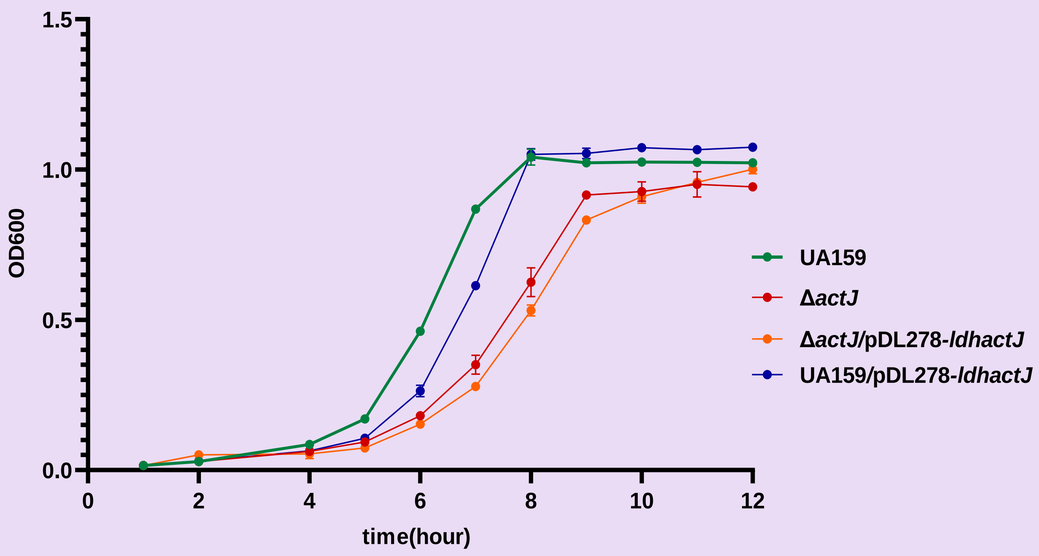

Fig.1 Growth curve of UA159, Δ*actJ*, Δ*actJ*/pDL278-*ldhactJ* and UA159/pDL278-*ldhactJ*. The cell growths of *S. mutans* strains were studied using the optical density (OD) at 600 nm and detected per hour for 12 h. Growth delay was observed in *actJ* mutant and complement strain.

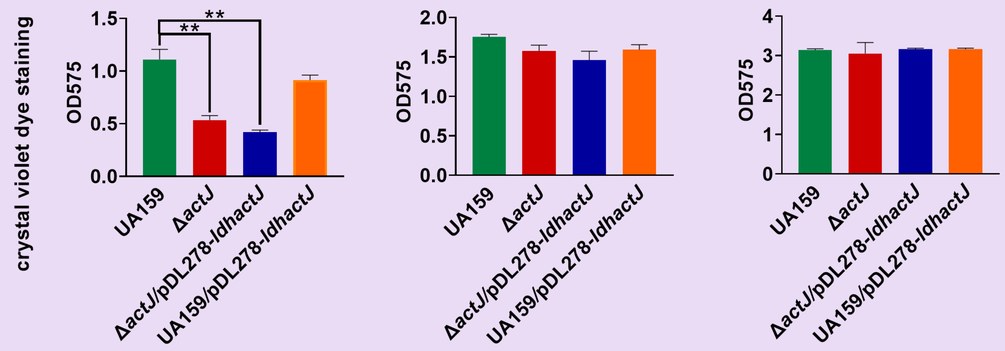

Fig.2 The biomass in the biofilm of *S. mutans* strains were evaluated by crystal violet staining. A significant decrease in biofilm formation was observed in 6-h biofilms of *actJ* mutant and the complement strain.

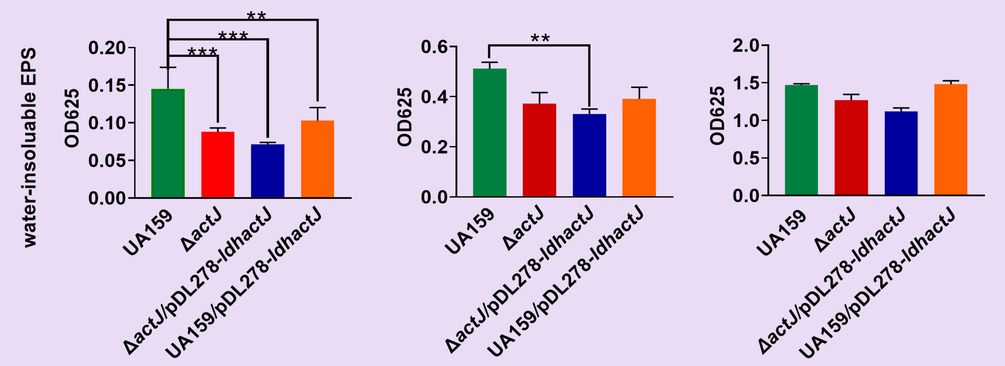

Fig.3 The amounts of water-insoluble glucans in the biofilm of different *S. mutans* strains were evaluated by enthrone -sulfuric acid method. The *actJ* mutant, complement, and the overexpression strain has significantly less water-insoluble glucans accumulation in 6-h biofilm.

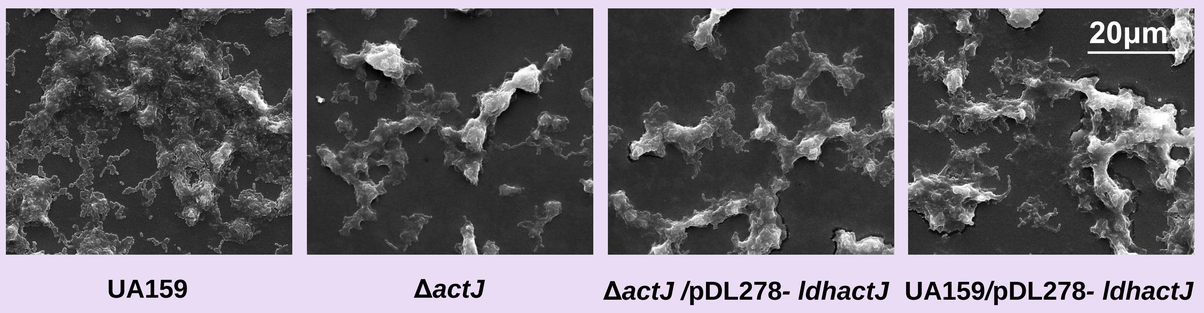

Fig 4. Scanning electron microscopy analysis of biofilms formation. The biofilm was cultured for 6-h, then SEM images were taken at ×5000 magnification.

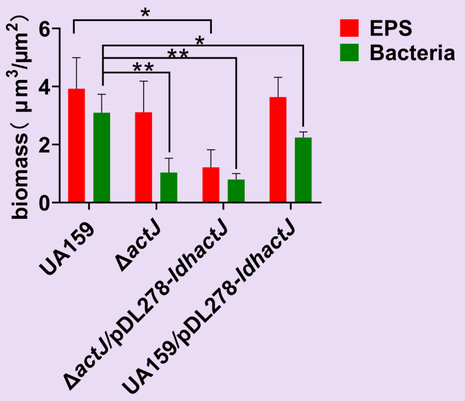

Fig.5 Confocal laser scanning microscope analysis of biofilms formed by UA159, Δ*actJ*, Δ*actJ*/pDL278-*ldhactJ* and UA159/pDL278-*ldhactJ*.

Conclusion

This study suggested that ActJ played a significant role in the growth and biofilm formation in *S. mutans*. Deletion of *actJ* led to growth delay and accompanied with reduction of biofilm formation and EPS synthesis, which could not be restored in complement strain.