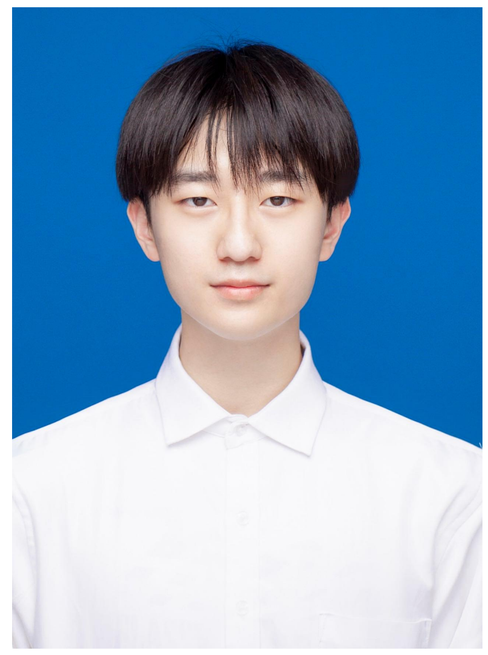
P4-3

Single-cell analysis reveals that cancer-associated fibroblasts promote oral squamous cell carcinoma invasion through TGF-β1/Smad pathway



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Background

- The prognosis of **oral squamous cell carcinoma (OSCC)** is not satisfactory mainly because of local tumor invasion.
- The tumor microenvironment (TME) is a potential target, in which **cancer-associated fibroblasts (CAFs)** are of great significance.
- However, the interaction between CAFs and cancer cells that promotes OSCC invasion is still unclear.
- Here, we investigated the two subtypes of CAF (**iCAF and mCAF**) and their protumor role in OSCC at single-cell resolution.

Results

CAFs promoted OSCC invasion through TGF-β1/Smad2/3 pathway

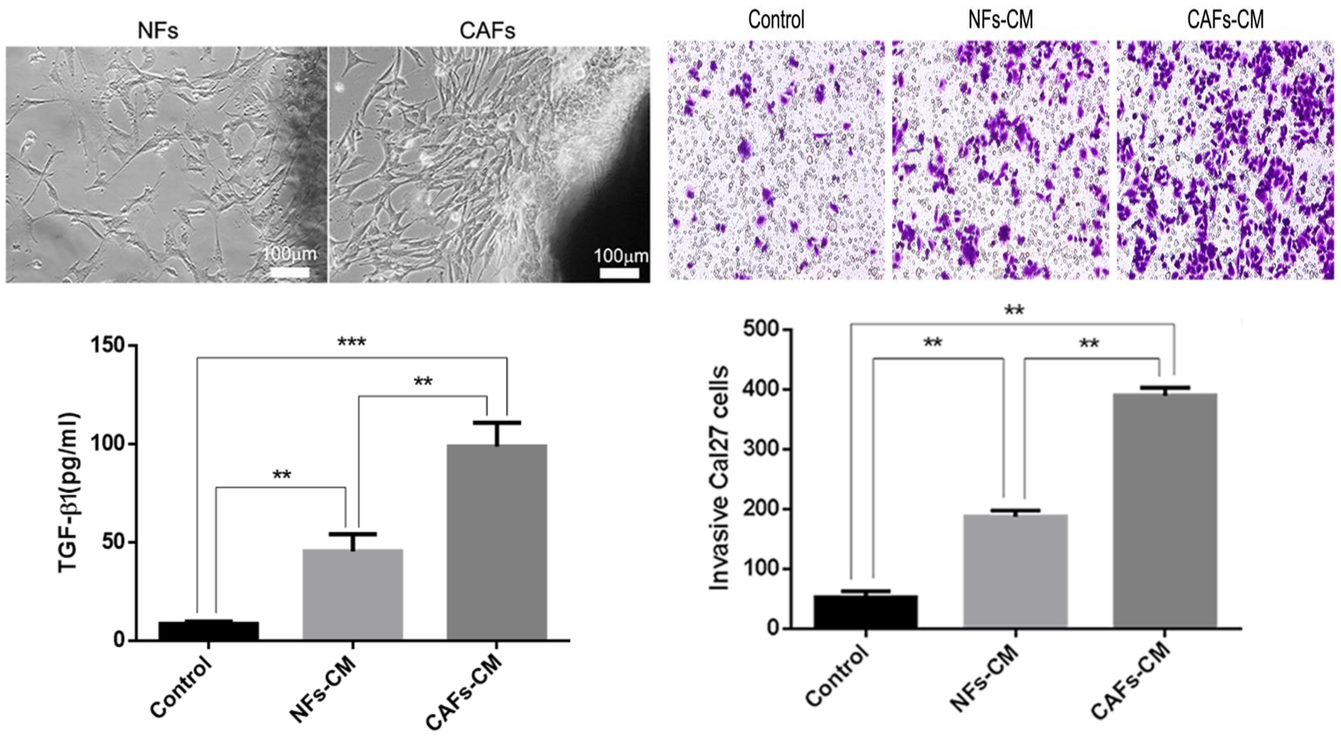


Fig.1 The amount of secreted TGF-β1 from control, normal fibroblast (NF) and CAF in conditioned medium (CM) after 24 hours was detected.

Fig.2 Crystal violet staining of invaded Cal27 cells in control, NFs-CM and CAFs-CM. And histogram showed that CAFs promoted Cal27 cells invasion eminently.

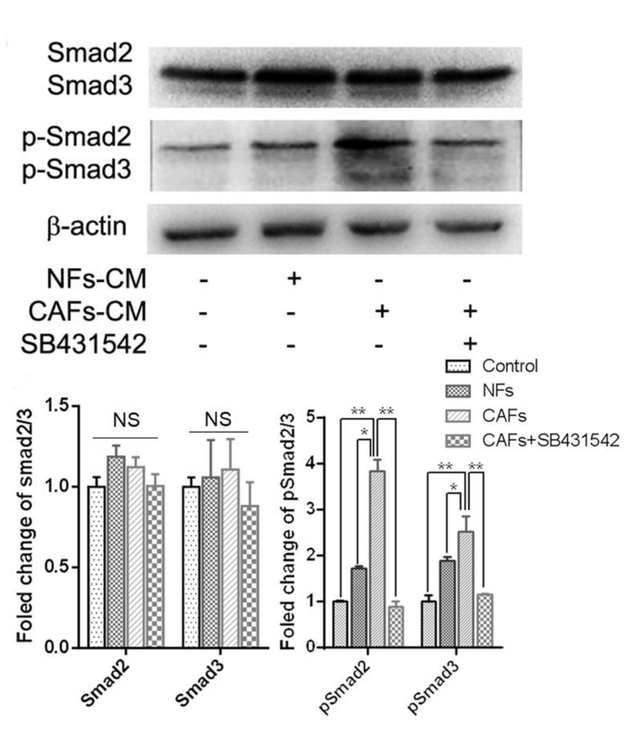


Fig.3 Western blot analysis of Smad2/3 and p-Smad2/3 proteins in Cal27 cells with different treatments. SB431542 was used to inhibit TGF-β type I receptor.

High iCAF and mCAF predicted poor OS

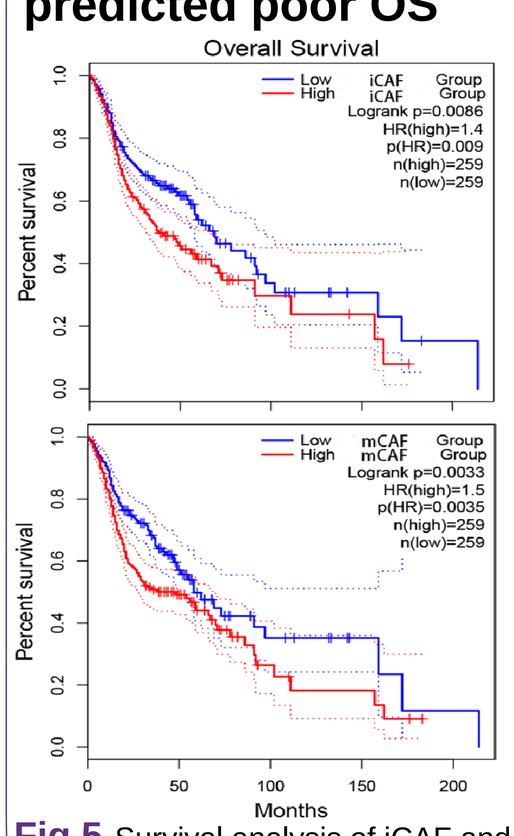


Fig.5 Survival analysis of iCAF and mCAF.

iCAF and mCAF potentially originated from epithelial through EMT

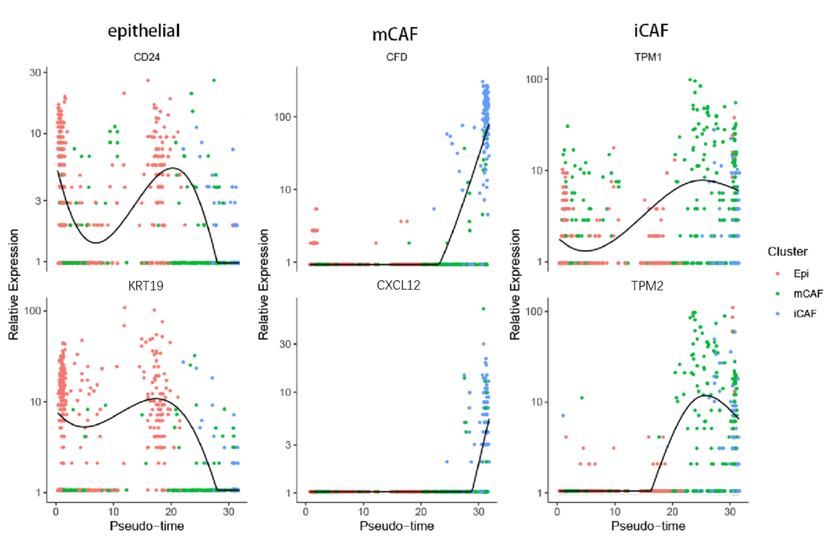


Fig.6 Relative expression alteration of marker genes in epithelial, iCAF and mCAF population across pseudotime.

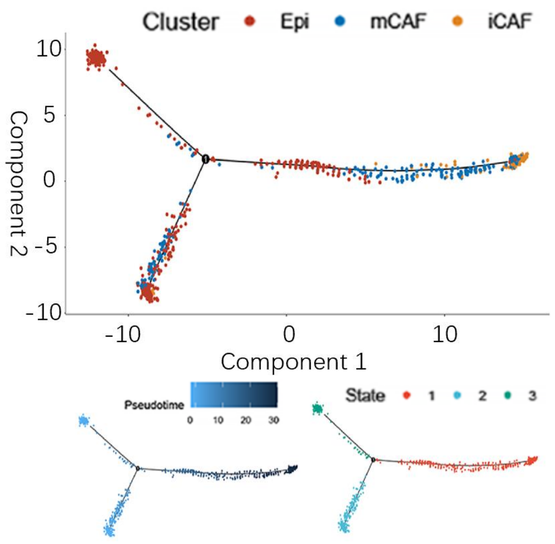
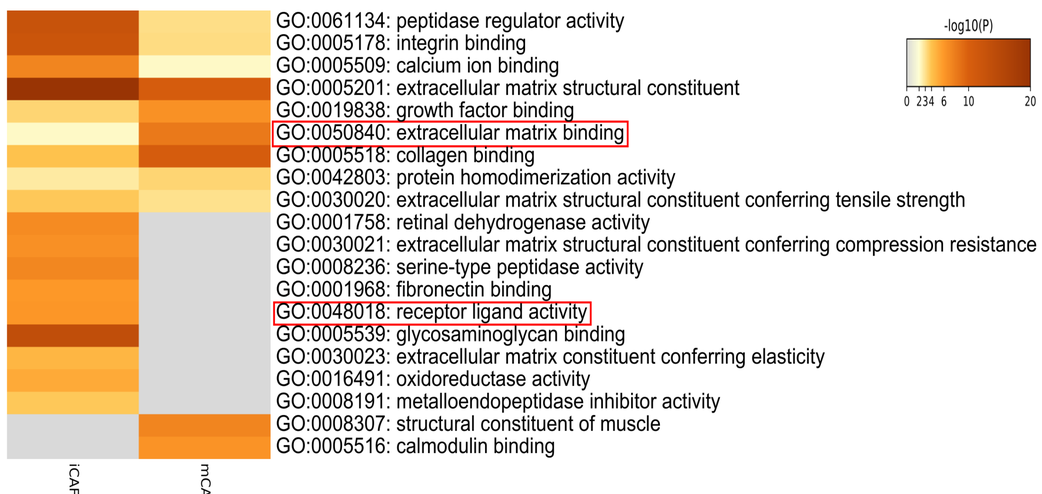


Fig.7 Trajectory order of epithelial, iCAF and mCAF population colored by cell type, pseudotime value and state.

Identification of iCAF and mCAF in OSCC

Fig.4 iCAF and mCAF were distinguished in OSCC at single-cell resolution and their proportion in different tissue types and stages were showed.



GO enrichment analysis of iCAF and mCAF

Fig.8 Receptor ligand activity and extracellular matrix binding were significantly enriched in iCAF and mCAF respectively, revealing their different roles in OSCC progression.

Conclusions

- CAF activated TGF-β1 pathway to promote OSCC invasion.
- iCAF and mCAF, which originated from epithelial, were correlated with survival and played distinct roles in OSCC.