DOI: https://doi.org/10.2298/SARH220607115A
UDC: 616.314.2-006:575.1; 616.314.2-003.4:575.1

ORIGINAL ARTICLE / ОРИГИНАЛНИ РАД

STAT3 gene expression in ameloblastomas and odontogenic keratocysts

Leonardo de Araújo Melo[1], Dandara Andrade de Santana[2], Caroline Brandi Schlaepfer Sales[3], Braúlio Carneiro Júnior[4], Marília Trierveiler Martins[5], Valéria Souza Freitas[6], Flávia Caló Aquino Xavier[1], Patrícia Ramos Cury[2], Clarissa Araújo Silva Gurgel[2], Jean Nunes dos Santos[1,2]

[1]Federal University of Bahia, School of Dentistry, Laboratory of Surgical Pathology, Salvador, Bahia, Brazil;
[2]Federal University of Bahia, Program Dentistry and Health, Salvador, Bahia, Brazil;
[3]Federal University of Bahia, Department of Biomorphology, Salvador, Bahia, Brazil;
[4]South University of Bahia, Department of Oral and Maxillofacial Surgery, Jequié, Bahia, Brazil;
[5]University of São Paulo, Department of Oral and Maxillofacial Pathology, São Paulo, São Paulo, Brazil;
[6]State University of Feira de Santana, Department of Health, Feira de Santana, Bahia, Brazil

SUMMARY

Introduction/Objective *STAT3* (signal transducers and activators of transcription) is involved in different physiological processes, including cell proliferation and survival. High expression of this protein is observed in various types of cancer.

This study aimed to investigate the gene and protein expression of *STAT3* in a series of odontogenic cysts and tumors to provide more information about their biological profile.

Methods The *STAT3* gene expression at mRNA was quantified by real-time quantitative polymerase chain reaction (RT-qPCR) in 23 odontogenic keratocysts (OKCs) and seven ameloblastomas (AMs), and compared to the non-neoplastic oral mucosa. We also assessed the expression of *STAT3* gene at protein levels, using immunohistochemistry, in 43 OKCs and 47 AMs.

Results *STAT3* transcripts were found in 96.6% of the tumors studied; however, the gene was downregulated in OKC and AM compared to the non-neoplastic oral mucosa. The *STAT3* gene expression at mRNA level was higher in sporadic OKC than in syndromic OKC ($p = 0.04$). There was no difference in *STAT3* gene expression at mRNA level between OKCs and AMs ($p = 0.88$). Immunostaining of STAT3 revealed no significant difference between sporadic and syndrome OKC ($p > 0.05$), nor between conventional and unicystic AMs ($p > 0.05$). Ameloblastomas exhibited significantly higher STAT3 immunostaining than OKCs ($p = 0.03$). In OKC and AM, STAT3 immunostaining was predominantly cytoplasmic and no difference in the cellular localization of STAT3 was observed between these lesions ($p = 0.58$).

Conclusion Our findings showed low expression of *STAT3* gene in OKCs and AMs in relation to non-neoplastic oral mucosa. However, higher *STAT3* immunostaining was observed in AMs compared to OKCs.

Keywords: odontogenic cysts; ameloblastoma; STAT3 transcription factor; gene expression; immunohistochemistry

INTRODUCTION

Odontogenic keratocysts (OKCs) and ameloblastomas (AM) are benign heterogeneous lesions of the jaws that arise from disturbances in tooth formation and are characterized by locally aggressive growth and recurrent rates [1]. The fifth edition of the World Health Organization (WHO) Classification of Head and Neck Tumours has considered the advanced molecular investigation, a fact that may cause a clinical impact [2]. As observed during odontogenesis, the development of the two lesions is related to the interaction between the odontogenic epithelium and ectomesenchyme. This process is mediated by signaling pathways forming a complex network [3]. One such pathway is the STAT3 signaling pathway, that has been suggested to be involved in the pathogenesis, progression, and recurrence of odontogenic tumors [4, 5].

STAT3 acts as signal transducers and transcription activators that play key physiological roles, including proliferation, survival, differentiation, and apoptosis [6, 7]. In addition to its participation in developing tooth germs and their disorders, the STAT3 signaling pathway is hyperactivated in most human cancers [8, 9]. It is generally associated with poor clinical prognosis [10, 11].

The STAT3 pathway has been the focus of studies on head and neck tumors. An *in vitro* study concluded that STAT3 is involved in the motility, metastasis, and progression of oral squamous cell carcinoma [12]. Recently, it has been suggested that the phosphorylation of STAT3 by IL-22 is essential for the increased invasion capacity of oral squamous cell carcinoma cell lines [13]. Furthermore, STAT3 activation is associated with the regulation of immunomodulatory proteins in head and neck tumors, and may therefore be a promising target for therapeutic intervention [12, 13].

Few studies have investigated the expression of STAT3 in odontogenic cysts and tumors [4, 5]. STAT3 and other related pathways participate in the epithelial-mesenchymal transition

Received • Примљено: June 7, 2022
Revised • Ревизија: November 10, 2022
Accepted • Прихваћено: November 12, 2022
Online first: December 19, 2022

Correspondence to:
Dandara ANDRADE DE SANTANA
62 Avenue Araújo Pinho
40110-040 Canela – Salvador
Bahia, Brazil
andradedandara@gmail.com;
dandara.andrade@ufba.br

of AM via IL-6 and the acquisition of epithelial stem cell-like properties by this tumor [5]. However, it was not found STAT3 immunostaining in OKCs, only during their malignant transformation to primary intraosseous squamous cell carcinoma [4]. Therefore, the present study aimed to evaluate the STAT3 gene expression (mRNA and protein) in a series of odontogenic cysts and tumors, including sporadic OKC, OKC associated with nevoid basal cell carcinoma syndrome (NBCCS), conventional AM, and unicystic AM, to provide more information about the biological profile of this group of lesions.

METHODS

All procedures performed in studies involving human participants have been in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. The study has been approved by the Ethics Committee of the School of Dentistry, Federal University of Bahia, Salvador, Bahia, Brazil (Protocol Nº 646.051). After approval, 47 cases of AM (30 conventional and 17 unicystic) and 43 cases of OKC (35 sporadic and eight associated with NBCCS) were obtained from the archives of the Surgical Pathology Service of the School of Dentistry, Federal University of Bahia, and University of São Paulo. The histopathological diagnosis was based on the World Health Organization Classification [2].

The samples were submitted to immunohistochemistry using STAT3 antibody. The odontogenic cysts and tumors selected comprised cases collected between 2002 and 2014. Thirty samples including seven AMs (five conventional and two unicystic) and 23 OKCs (14 sporadic and nine associated with NBCCS) were selected for gene expression analysis. Three samples corresponding to non-neoplastic oral mucosa from healthy individuals undergoing surgical excision of third molars, for orthodontic reasons, were included.

The samples were collected between 2005 and 2013 and stored in RNAlater solution (Ambiom®) at -80ºC until the experiments were performed.

RNA extraction and reverse transcription

Total RNA was extracted from 25–30 mg of frozen OKCs and AMs according to manufacturer specifications (RNeasy Mini Kit, Qiagen, Hilden, Germany). Genomic DNA was eliminated with DNase I (DNase I Amplification Grade Kit, Invitrogen, Carlsbad, CA, USA). RNA purity was evaluated by spectrophotometry (NanoDrop, Thermo Scientific, Wilmington, DE, USA) and values of 1.9–2.05 (A260/280) were considered satisfactory. The quantity of total RNA was determined by fluorimetry (QuBit™, Life Technologies, Camarillo, CA, EUA). The integrity of total RNA was confirmed by agarose gel electrophoresis (containing 1% formaldehyde). The cDNA was synthesized from 2 µg total RNA using oligo (dT) primers and the SuperScript II Reverse Transcriptase Kit (Invitrogen) in a reaction volume of 20 µL according to the protocol of the manufacturer. The reaction mixtures were incubated at 42°C for 2 minutes, followed by 65°C for 50 minutes, 42°C for 55 minutes, 70°C for 15 minutes, 37°C for 20 minutes, and 4°C for 5 minutes. The efficiency of reverse transcription was evaluated by amplifying the *GAPDH* and *B2M* reference genes.

Real-time quantitative polymerase chain reaction (RT-qPCR) and analysis of gene expression

The RT-qPCR assays were carried out in duplicate using inventoried TaqMan Gene Expression Assays™ for the *STAT3* gene (Hs00374280_m1), as well as for the *GAPDH* (Hs02758991_g1) and *B2M* (Hs00984230_m1) reference genes. The reactions were run on the ViiA™ 7 Real-Time PCR System (Applied Biosystems™, Foster City, CA, USA) using 96-well plates, in a total volume of 20 µL. Each well contained 2.5 ng/µL cDNA of the sample (8 µL), 1 µL of the assay, 10 µL TaqMan PCR Master Mix (Applied Biosystems™), and 5 µL RNAse-free water. The amplification program consisted of an initial cycle at 50°C for 2 minutes and 95°C for 10 minutes, followed by 40 cycles of 95°C for 15 seconds and 60°C for 1 minute. The calibrator sample (control) of the reactions consisted of a pool of samples of non-neoplastic oral mucosa. After the amplification runs and dissociation, the relative quantification (RQ) values were calculated using the Expression Suite v.1.0.3 (Applied Biosystems™).

Immunohistochemistry

For immunohistochemistry, 4-µm paraffin-embedded tissue sections were deparaffinized and rehydrated using routine methods. The STAT3 antigen epitopes were exposed by immersing the sections in citrate buffer, pH 6.0, in moist heat for 45 min, followed by the blockade of endogenous peroxidase (Peroxidase Blocking Solution™, Dako Corporation, Carpinteria, CA, USA) for 10 minutes protected from light and of tissue proteins (Protein Blocking Solution™, Dako Corporation) for 10 minutes. The sections were incubated with the primary STAT3 antibody (clone F-2, Santa Cruz Biotechnology, Santa Cruz, CA, USA), diluted at 1:100, overnight at 4°C. Next, the HRP Link and HRP Enzyme reagents (Advance™, Dako Corporation) were applied to the histological sections for 20 minutes each. The reactions were developed with 3,3-diaminobenzidine (Dako Corporation) for 5 minutes in a dark chamber, and the slides were counterstained with Harris hematoxylin. Lung squamous cell carcinoma sections were used as a positive control of the reactions. Phosphate-buffered saline was used as negative control in all reactions.

Immunohistochemical analysis

A previously trained examiner performed the immunohistochemical analysis under a light microscope coupled to a digital camera system (Axiocam ICC3; Zeiss, Göttingen, Germany, 2008) using the Axio Vision 4.8 software (Zeiss).

Brown-stained cells were defined as immunopositive and the intensity of staining was classified as follows: 0 = no staining, 1 = mild staining, 2 = moderate staining, and 3 = intense staining. The proportion of positive cells was scored 0–3, where 0 = up to 5% of positive cells, 1 = 6–25% of positive cells, 2 = 26–75% of positive cells, and 3 = > 75% of positive cells. Multiplication of the intensity score (0–3) by the proportion of stained cells (0 to > 75%) resulted in the following final scores: when the product of the two scores was 0, the case was classified as negative; when the product of the two scores ranged 1–3, the case was classified as low immunohistochemical expression; when the product was ≥ 4, the case was classified as high immunohistochemical expression [14]. The distribution of proteins was evaluated particularly in the basal, intermediate, and superficial layers of sporadic and syndromic (NBCCS) OKCs, as well as in the tumor islands, cystic epithelial lining, and areas of squamous metaplasia of AMs.

Statistical analysis

The sample data did not show a normal distribution according to the Gauss curve. Differences between groups were evaluated using the Mann–Whitney and Fisher's exact tests. All statistical calculations and graphics were performed with the GraphPad Prism 5.01 program (San Diego, CA, USA). A p value < 0.05 was considered statistically significant.

RESULTS

Gene expression profile of *STAT3* in odontogenic keratocyst and ameloblastoma

Expression of the *STAT3* gene was found in 29 (96.66%) of the 30 odontogenic cysts and tumors studied. *STAT3* transcripts were detected in all cases of OKC, with RQ values ranging 0.095–19.39 (median = 0.402, SD = 1.807). This gene was downregulated compared to the non-neoplastic oral mucosa (control) (Figure 1). It should be noted that the RQ values deviated from the values of the other samples in two OKC cases (9.021 and 19.394). These cases were sporadic/recurrent OKCs. Sporadic/recurrent OKCs exhibited higher RQ values than syndromic OKCs (p = 0.04; Mann–Whitney test) (Figure 2). In AMs, no *STAT3* transcripts were detected in one case, with RQ values ranging 0–1.315 (median = 0.325, SD = 1.975). The gene was downregulated compared to the to the non-neoplastic oral mucosa (control) (Figure 1). No difference in *STAT3* gene expression was observed between OKCs and AMs (p = 0.88; Mann–Whitney test; Figure 3).

STAT3 protein in odontogenic keratocyst and ameloblastoma

We evaluated STAT3 immunostaining in 90 odontogenic tumors, including 43 OKCs and 47 AMs. The distribution of scores for the proportion of stained cells and staining intensity, the product of scores, and staining pattern are summarized in Table 1.

Immunostaining in OKC was evident mainly in the basal and intermediate layers of the epithelium lining the fibrous cystic wall (Figure 4a). There was no significant difference in the proportion of positive cells, staining intensity, or final STAT3 score between sporadic OKCs and OKCs associated with NBCCS (p > 0.05; Fisher's exact test). Similarly, no difference was observed in nuclear staining of STAT3 between

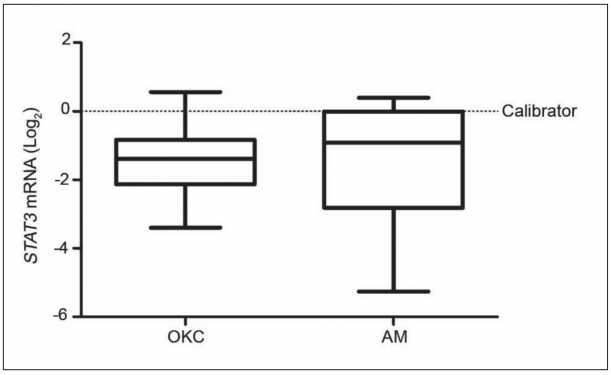

Figure 1. Relative expression (Log2) of the *STAT3* mRNA in odontogenic keratocysts (OKC) and ameloblastomas (AM) compared to the calibrator (control) sample

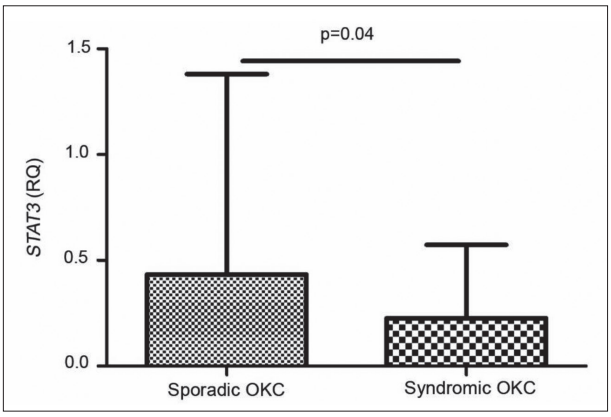

Figure 2. Comparison of *STAT3* mRNA relative quantification (RQ) between sporadic odontogenic keratocysts (OKC) and syndromic OKCs by Mann–Whitney test

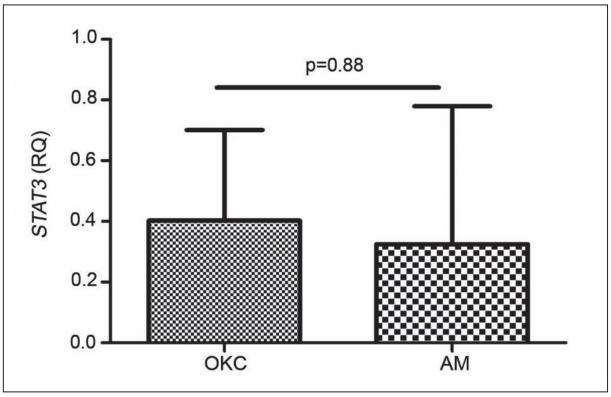

Figure 3. Comparison of *STAT3* mRNA relative quantification (RQ) between odontogenic keratocysts (OKC) and ameloblastomas (AM) by Mann–Whitney test

Table 1. Distribution of scores regarding *STAT3* in odontogenic keratocysts and ameloblastomas

Criteria evaluated	Sporadic OKC (n = 35)	Syndromic OKC (n = 8)	Conventional AM (n = 30)	Unicystic AM (n = 17)
Stained cells				
Score 0	13	1	3	7
Score 1	10	2	8	3
Score 2	4	5	5	2
Score 3	8	0	14	5
Intensity				
Absent	13	1	3	7
Mild	10	6	9	4
Moderate	7	1	7	4
Intense	5	0	11	2
Product of scores				
Negative	13	1	3	7
Low expression	13	6	12	4
High expression	9	1	15	6
Cell staining				
Absent	13	1	3	7
Only cytoplasmic	13	4	17	7
Cytoplasmic and nuclear	9	3	10	2
Only nuclear	0	0	0	1

OKC – odontogenic keratocysts; AM – ameloblastomas

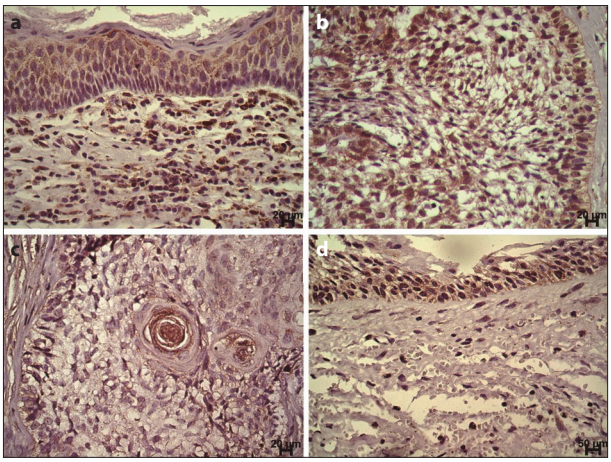

Figure 4. STAT3 immunostaining; (a) cytoplasmic staining in cells of the intermediate layer of the cystic epithelial lining in odontogenic keratocysts; (b) predominantly cytoplasmic and possible nuclear staining in central and peripheral areas of tumor islands in conventional ameloblastoma; (c) immunostained area of squamous metaplasia in conventional ameloblastoma; (d) cytoplasmic staining in all layers of the cystic lining of unicystic ameloblastoma

sporadic and syndromic OKCs (p = 0.66; Fisher's exact test). Immunostaining in AMs was more evident in the tumor parenchyma of epithelial islands and in all layers of the cystic lining. Furthermore, staining was more common in peripheral than in central cells of the tumor islands. STAT3 immunoreactivity was also observed in areas of squamous metaplasia (Figure 4b–d). No significant difference was found in the proportion of positive cells, staining intensity, or final STAT3 score between conventional and unicystic AMs (p > 0.05; Fisher's exact test). There was also no difference in nuclear staining of STAT3 between conventional and unicystic AMs (p = 0.32; Fisher's exact test).

A significant difference in STAT3 immunostaining occurred between OKCs and AMs, with a higher proportion of positive cells in the latter (p = 0.03; Fisher's exact test). However, there was no difference in nuclear staining of STAT3 between OKCs and AMs (p = 0.58; Fisher's exact test).

DISCUSSION

Advances in understanding STAT3 signaling and its role in tumor progression and aggressiveness have rendered this transcription factor a potential target in different studies on head and neck pathologies [4, 5, 12, 13]. Our study aimed to evaluate the *STAT3* gene expression at mRNA and protein levels in OKC and AMs, common lesions with variable degrees of aggressive behavior and different recurrence rates, to gain insight into the molecular profile of this group of odontogenic lesions.

STAT3 transcript in OKC and AM was downregulated compared to the to the non-neoplastic oral mucosa (control). Interestingly, 67.44% of the OKC cases and 78.72% of the AM cases exhibited low or high immunolabelling of STAT3. Thus, *STAT3* gene expression was more evident at protein than at mRNA level in the cases studied. In malignant head and neck tumors, expression of STAT3 protein is related to cell migration, and proliferation and tumor progression [4, 15]. STAT3 exists in two isoforms, α and β; STAT3β is a less abundant isoform that reduces the transcriptional function of α [16]. The primer used in the present study was specific for the α and β isoforms and we can therefore rule out the possibility that the downregulation of the gene had occurred in a specific isoform. We hypothesized that a limited number of available samples for qPCR and standard deviation could explain our results. This aspect represented a limitation of this study. Perhaps a larger number of cases and a laser-microdissection-based analysis could clarify this matter.

In the present study, STAT3 immunostaining in OKC and AM was predominantly cytoplasmic, while nuclear staining was less common. STAT3 is present in the cytoplasm under basal and inactive conditions. The nuclear translocation after activation is fundamental for the function of this protein as a transcription factor and regulator of specific genes [17]. The predominantly cytoplasmic immunostaining of the protein and downregulation of the *STAT3* gene at mRNA level suggest low transcriptional activity of this protein in the odontogenic cysts and tumors studied. In the cytoplasm, unphosphorylated STAT3 interacts with protein kinase R, blocking its enzymatic activity and inhibiting autophagy, and also regulates cell migration through microtubule polymerization [18, 19].

pSTAT3 was not included in the analysis because of technical difficulties with antibody staining. Despite this, the latent cytoplasmic STAT3 protein is activated by tyrosine phosphorylation mediated by Janus or Src kinases. Once phosphorylated (pSTAT3), the protein forms dimers and is translocated to the nucleus, where it binds to specific DNA promoter sequences for transcription of its target

genes [17]. Although the physiological performance of signaling is important for the standard cell response, disordered activation of the STAT3 pathway occurs in many human diseases, especially tumors. Thus, disruption of STAT3 signaling is related to the process of tumorigenesis, inducing the transcriptional activation of various genes that regulate inflammation, angiogenesis, apoptosis resistance, and metastasis [15, 20].

In contrast to the present study, in a series of only three cases analyzed by immunohistochemistry, absence of STAT3 in OKCs was revealed [4]. However, other authors found positive staining for this protein in follicular AM [5]. Given the higher immunostaining of STAT3 in AMs observed in the present study, even considering the lack of a significant difference in mRNA expression between OKC and AM, we suggest that STAT3 may participate at least in the tissue differentiation of these lesions. Regarding the type of keratocyst, we found significantly higher expression of the STAT3 gene at protein levels in sporadic OKCs than those associated with NBCCS. However, it is difficult to explain this difference because of the small number of syndromic cases. It is important to point out that, despite the lack of studies comparing STAT3 between different types of AM and OKC, there is no consensus in the literature regarding the distribution of tumor markers among different odontogenic cysts and tumors. However, this distribution is generally associated with proliferation, recurrence, and tumor aggressiveness, which cannot be inferred here [21, 22].

Immunostaining of STAT3 was observed in stellate reticulum-like cells, in areas of squamous metaplasia. It was also observed in peripheral cells of epithelial islands of the tumor parenchyma in AMs. In OKCs, STAT3 immunostaining was detected in superficial and intermediate layers. In addition, immunostaining was found in the suprabasal layer of the epithelium lining the fibrous cystic wall in unicystic AMs. These findings suggest the participation of STAT3 in the morphogenesis and differentiation of AM and OKC, indicating a role of this protein in physiological processes that are fundamental for tumor development [6, 7, 10, 11]. Activated STAT3 has been reported to participate in the differentiation of the stratified squamous epithelium by regulating the gene expression of cytokeratins [23]. These proteins are mainly found in stellate reticulum-like cells of the enamel organ and areas of squamous metaplasia in AMs, as well as in the intermediate and superficial layers of the cystic epithelial lining of OKCs, suggesting complete differentiation of the epithelial component of OKC [21, 24].

CONCLUSION

STAT3 expression at both mRNA and protein levels was detected in OKCs and AM regardless of their clinical presentation. This protein participates in the differentiation and maintenance of the cytoarchitectural pattern of these odontogenic lesions. This study provided insight into the role of STAT3 in OKC and AM. However, further studies investigating the Janus kinase / STAT3 signaling pathway, especially the phosphorylated form of the protein, would be useful to elucidate other aspects related to the pathogenesis of odontogenic tumors and cysts.

ACKNOWLEDGMENT

This study was funded by the National Council for Scientific and Technological Development (CNPq) and Coordination for the Improvement of Higher Education Personnel (CAPES).

The authors are grateful to CAPES and CNPq for student scholarship. Patrícia Ramos Cury, Clarissa Araújo Gurgel, and Jean Nunes dos Santos are research fellows funded by the Brazilian National Council for Scientific and Technological Development (CNPq).

Conflict of interest: None declared.

REFERENCES

1. Rioux-Forker D, Deziel AC, Williams LS, Muzaffar AR. Odontogenic Cysts and Tumors. Ann Plast Surg. 2019;82(4):469–77. [DOI: 10.1097/SAP.0000000000001738] [PMID: 30856625]
2. Vered M, Wright JM. Update from the 5th Edition of the World Health Organization Classification of Head and Neck Tumors: Odontogenic and Maxillofacial Bone Tumours. Head Neck Pathol. 2022;16(1):63–75. [DOI: 10.1007/s12105-021-01404-7] [PMID: 35312978]
3. Duarte-Andrade FF, Vitório JG, Pereira T dos SF, Gomes CC, Gomez RS. A review of the molecular profile of benign and malignant odontogenic lesions. Oral Surg Oral Med Oral Pathol Oral Radiol. 2020;129(4):357–68. [DOI: 10.1016/j.oooo.2019.12.017] [PMID: 32035859]
4. Zhang L, Liu SL, Ye WM, Zheng J. STAT3 is a key pathway in primary intraosseous squamous cell carcinoma arising from an odontogenic keratocyst. Am J Transl Res. 2015;7(10):1860–9. [PMID: 26692930]
5. Jiang C, Zhang Q, Shanti RM, Shi S, Chang TH, Carrasco L, et al. Mesenchymal Stromal Cell-Derived Interleukin-6 Promotes Epithelial–Mesenchymal Transition and Acquisition of Epithelial Stem-Like Cell Properties in Ameloblastoma Epithelial Cells. Stem Cells. 2017;35(9):2083–94. [DOI: 10.1002/stem.2666] [PMID: 28699252]
6. Gharibi T, Babaloo Z, Hosseini A, Abdollahpour-Alitappeh M, Hashemi V, Marofi F, et al. Targeting STAT3 in cancer and autoimmune diseases. Eur J Pharmacol. 2020;878:1–32. [DOI: 10.1016/j.ejphar.2020.173107] [PMID: 32278856]
7. Sadrkhanloo M, Entezari M, Orouei S, Ghollasi M, Fathi N, Rezaei S, et al. STAT3-EMT axis in tumors: modulation of cancer metastasis, stemness and therapy response. Pharmacol Res. 2022;182(106311):1–24. [DOI: 10.1016/j.phrs.2022.106311] [PMID: 35716914]
8. Fan L, Ou YJ, Zhu YX, Liang YD, Zhou Y, Wang YN. Lif Deficiency Leads to Iron Transportation Dysfunction in Ameloblasts. J Dent Res. 2022;101(1):63–72. [DOI: 10.1177/00220345211011986] [PMID: 34034544]
9. Liao C, Ou Y, Wu Y, Zhou Y, Liang S, Wang Y. Sclerostin inhibits odontogenic differentiation of human pulp-derived odontoblast-like cells under mechanical stress. J Cell Physiol. 2019;234(11):20779–89. [DOI: 10.1002/jcp.28684] [PMID: 31025337]

10. Tang L, Liu JX, Zhang ZJ, Xu CZ, Zhang XN, Huang WR, et al. High expression of Anxa2 and Stat3 promote progression of hepatocellular carcinoma and predict poor prognosis. Pathol Res Pract. 2019;215(6):152386. [DOI: 10.1016/j.prp.2019.03.015] [PMID: 30935762]
11. Fei Y, Yu J, Li Y, Li L, Zhou S, Zhang T, et al. Plasma soluble PD-L1 and STAT3 predict the prognosis in diffuse large B cell lymphoma patients. J Cancer. 2020;11(23):7001–8. [DOI: 10.7150/jca.47816] [PMID: 33123290]
12. Wang Y, Wu C, Zhang C, Li Z, Zhu T, Chen J, et al. TGF-β-induced STAT3 overexpression promotes human head and neck squamous cell carcinoma invasion and metastasis through malat1/miR-30a interactions. Cancer Lett. 2018;436:52–62. [DOI: 10.1016/j.canlet.2018.08.009] [PMID: 30118844]
13. Komine-Aizawa S, Aizawa S, Takano C, Hayakawa S. Interleukin-22 promotes the migration and invasion of oral squamous cell carcinoma cells. Immunol Med. 2020;43(3):121–9. [DOI: 10.1080/25785826.2020.1775060] [PMID: 32546118]
14. Dultra FKAA, Barros AC, Schaer-Barbosa H, Figueiredo AL, Gurgel CAS, Ramos EAG, et al. Immunohistochemical assessment of CD1a-positive Langerhans cells and their relationship with E-cadherin in minor salivary gland tumors Immunohistochemical assessment of CD1a-positive Langerhans cells and their relationship with E-cadherin in minor salivar. J Oral Pathol Med. 2012;41(1):47–53. [DOI: 10.1111/j.1600-0714.2011.01052.x] [PMID: 21627694]
15. Jiang X, Huang Z, Sun X, Zheng X, Liu J, Shen J, et al. CCL18-NIR1 promotes oral cancer cell growth and metastasis by activating the JAK2/STAT3 signaling pathway. BMC Cancer. 2020;20(1):1–13. [DOI: 10.1186/s12885-020-07073-z] [PMID: 32641093]
16. Tolomeo M, Cascio A. The multifaced role of stat3 in cancer and its implication for anticancer therapy. Int J Mol Sci. 2021;22(2):1–25. [DOI: 10.3390/ijms22020603] [PMID: 33435349]
17. Johnson DE, O'Keefe RA, Grandis JR. Targeting the IL-6/JAK/STAT3 signalling axis in cancer. Nat Rev Clin Oncol. 2018;15(4):234–48. [DOI: 10.1038/nrclinonc.2018.8] [PMID: 29405201]
18. You L, Wang Z, Li H, Shou J, Jing Z, Xie J, et al. The role of STAT3 in autophagy. Autophagy. 2015;11(5):729–39. [DOI: 10.1080/15548627.2015.1017192] [PMID: 25951043]
19. Yan B, Xie S, Liu Z, Luo Y, Zhou J, Li D, et al. STAT3 association with microtubules and its activation are independent of HDAC6 activity. DNA Cell Biol. 2015;34(4):290–5. [DOI: 10.1089/dna.2014.2713] [PMID: 25621430]
20. Mohassab AM, Hassan HA, Abdelhamid D, Abdel-Aziz M. STAT3 transcription factor as target for anti-cancer therapy. Pharmacol Reports. 2020;72(5):1101–24. [DOI: 10.1007/s43440-020-00156-5] [PMID: 32880101]
21. Alsaegh M, Altaie A, Zhu S. Expression of keratin 15 in dentigerous cyst, odontogenic keratocyst and ameloblastoma. Mol Clin Oncol. 2019;(8):377–81. [DOI: 10.3892/mco.2019.1802] [PMID: 30847177]
22. Embaló B, Parize HN, Rivero ERC. Evaluation of cell proliferation in cystic lesions associated with impacted third molars. Microsc Res Tech. 2018;81(11):1241–5. [DOI: 10.1002/jemt.23128] [PMID: 30295367]
23. Steinbichler TB, Savic D, Dejaco D, Romani A, Kofler B, Skvortsova II, et al. Pleiotropic Effects of Epithelial Mesenchymal Crosstalk on Head and Neck Cancer: EMT and beyond. Cancer Microenviron. 2019;12(2–3):67–76. [DOI: 10.1007/s12307-019-00228-y] [PMID: 31297730]
24. Akhila CHB, Sreenath G, Prakash AR, Kanth MR, Simha Reddy AV, Kumar SN. Expression of stem cell marker cytokeratin 19 in reduced enamel epithelium, dentigerous cyst and unicystic ameloblastoma – A comparative analysis. J Oral Maxillofac Pathol. 2021;25(1):136. [DOI: 10.4103/jomfp.JOMFP_316_19] [PMID: 34349424]

Експресија гена *STAT3* код амелобластома и одонтогених кератоциста

Леонардо де Араухо Мело[1], Дандара Андраде де Сантана[2], Каролине Бранди Шлепфер Салес[3], Браулио Карнеиро Жуниор[4], Марилија Триервеилер Мартинс[5], Валерија Соуза Фреитас[6], Флавија Кало Акино Хавијер[1], Патриција Рамос Кари[2], Клариса Араухо Силва Гургел[2], Жан Нунес дос Сантос[1,2]

[1]Федерални универзитет Баије, Стоматолошки факултет, Лабораторија за хируршку патологију, Салвадор, Баија, Бразил;
[2]Федерални универзитет Баије, Програм стоматологије и здравља, Салвадор, Баија, Бразил;
[3]Федерални универзитет Баије, Одељење за биоморфологију, Салвадор, Баија, Бразил;
[4]Јужни универзитет у Баији, Одељење за оралну и максилофацијалну хирургију, Жекије, Баија, Бразил;
[5]Универзитет у Сао Паулу, Одељење за оралну и максилофацијалну патологију, Сао Пауло, Сао Пауло, Бразил;
[6]Државни универзитет у Феири де Сантани, Здравствени програм, Феира де Сантана, Баија, Бразил

САЖЕТАК

Увод/Циљ *STAT3* (претварачи сигнала и активатори транскрипције) укључен је у различите физиолошке процесе, укључујући пролиферацију и преживљавање ћелија. Висока експресија овог протеина се примећује код различитих врста рака.

Ова студија је имала за циљ да истражи експресију гена и протеина *STAT3* у серији одонтогених циста и тумора како би пружила више информација о њиховом биолошком профилу.

Методе Експресија гена *STAT3* на мРНК је квантификована квантитативном ланчаном реакцијом полимеразе у реалном времену (*RT-kPCR*) у 23 одонтогене кератоцисте (ОКЦ) и седам амелобластома (АМ) и упоређена са ненеопластичном оралном слузницом. Такође смо проценили експресију гена *STAT3* на нивоима протеина, користећи имунохистохемију, у 43 ОКЦ и 47 АМ.

Резултати *STAT3* транскрипти су пронађени у 96,6% проучаваних тумора; међутим, ген је смањен у ОКЦ и АМ у поређењу са ненеопластичном оралном слузокожом. Експресија *STAT3* гена на нивоу мРНК била је већа код спорадичног ОКЦ него код синдромског *STAT3* ($p = 0,04$). Није било разлике у експресији *STAT3* гена на нивоу мРНА између ОКЦ и АМс ($p = 0,88$). Имунобојење *STAT3* није открило значајну разлику између спорадичног и синдрома ОКЦ ($p > 0,05$), нити између конвенционалних и уницистичних АМ ($p > 0,05$). Амелобластоми су показали значајно веће *STAT3* имунобојење од ОКЦ ($p = 0,03$). Код ОКЦ и АМ, *STAT3* имунобојење је било претежно цитоплазматско и није примећена разлика у ћелијској локализацији *STAT3* између ових лезија ($p = 0,58$).

Закључак Наши налази су показали ниску експресију гена *STAT3* у ОКЦ и АМс у односу на ненеопластичну оралну слузокожу. Међутим, примећено је веће *STAT3* имунобојење код АМ у поређењу са ОКЦ.

Кључне речи: одонтогене цисте; амелобластом; фактор транскрипције *STAT3*; експресија гена; имунохистохемија