Salivary Duct Carcinoma and Invasive Ductal Carcinoma of the Breast: A Comparative Immunohistochemical Study

Jalal B. Jalaly¹ · Souzan Sanati¹ · Rebecca D. Chernock¹ · Dikson G. Dibe¹ · Samir K. El-Mofty¹

Received: 26 October 2017 / Accepted: 29 December 2017 / Published online: 4 January 2018
© Springer Science+Business Media, LLC, part of Springer Nature 2018

Abstract

Salivary duct carcinoma (SDC) is a high-grade salivary gland malignancy with great morphological resemblance to invasive ductal carcinoma (IDC) of the breast. Rarely, female patients may have a past history of both SDC and IDC. When these patients present with distant metastasis, accurate identification of the primary tumor is particularly difficult. Additionally, rare metastasis of SDC to the breast and IDC to the salivary (parotid) gland can also present a diagnostic challenge. Our aim was to develop an immunohistochemical panel that reliably distinguishes SDC from IDC. We included all SDCs diagnosed from 1989 to 2016 (23 cases) and 29 treatment naïve and histologically similar IDCs. All cases were stained with androgen receptor (AR), estrogen receptor-alpha (ER- α), progesterone receptor (PR), HER-2, CK5/6, p63, and beta-catenin. The great majority (> 90%) of both SDCs and IDCs reacted positively to AR. The main discrepancy in the immunohistochemical profiles was a distinctly different reactivity to ER- α , PR and HER-2. While 28 IDCs (96.6%) reacted positively to ER- α and/or PR, the majority expressing both (82.8%) with a moderate to strong staining intensity, only 2 SDCs expressed ER- α (8.7%) and 5 others expressed PR (21.7%) with only one case expressing both (P value < 0.05). On the other hand, 8 SDC (34.8%) were positive for HER-2 while none of the IDCs were positive (P value < 0.05). ER- α , PR, and HER-2 may be helpful to distinguish SDC from IDC. Positive reactivity to ER- α , PR or both and negative HER-2 favors a diagnosis of IDC while ER- α , PR negative, HER-2 positive tumors are more likely SDC.

Keywords Salivary glands · Breast · Neoplasms · Immunohistochemistry · Estrogens · Progesterone

Introduction

Salivary duct carcinoma (SDC) is a high-grade salivary gland neoplasm first described by Kleinsasser [1]. It morphologically resembles a subset of invasive ductal carcinoma (IDC) of the breast [2]. This subset of breast carcinoma is ductal and has higher Nottingham histological grade. They may also have apocrine features. When examined by gene expression analysis, these tumors usually fall into luminal-B and luminal/AR positive breast carcinoma [3]. These IDC and SDC are composed of cells with ample eosinophilic cytoplasm that are arranged in nests, ducts, cribriform glands, and infiltrating cords in a desmoplastic stroma (Fig. 1). In addition, both can have in situ (intraductal)

component with or without necrosis and calcifications. SDC most commonly occurs in the parotid gland, followed by the submandibular and minor salivary glands.

Salivary duct carcinoma represents 1–3% of all salivary gland malignancies [4, 5]. A recent review showed that 29% of SDC patients are women [6]. When these patients present with distant metastatic disease, the differential diagnosis may include both SDC and morphologically similar IDC. Rarely, female patients may have a past history of both SDC and IDC and may present with distant metastasis. Such a case prompted this investigation. Moreover, metastasis of SDC to the breast [7] and IDC to the parotid gland has been reported as well and is also a diagnostic challenge [8].

Several authors have studied the morphology and immunohistochemical profiles of these two morphologically overlapping tumors [9–16]. While some immunohistochemical stains may overlap, such as GATA-3 [17], others can be helpful and suggestive of one diagnosis over the other. However, these stains cannot reliably distinguish SDC and IDC as some degree of overlap exists. Our aim was to develop

✉ Samir K. El-Mofty
Elmofty@wustl.edu

¹ Department of Pathology & Immunology, Washington University School of Medicine, 660 S Euclid Ave, Box 8118, Saint Louis, MO 63110, USA

Fig. 1 Salivary duct carcinoma (a) is morphologically similar to invasive breast carcinoma (b)

an immunohistochemical panel to more reliably distinguish SDC from morphologically identical but biologically distinct IDC.

Materials and Methods

Study approval was obtained from the Washington University Human Research Protection Office. The laboratory information system was used to query for all SDC and IDC. All primary SDC with material available for review diagnosed from 1989 to 2016 were included in the study. Metastatic SDC were included only if the primary tumor was a known SDC and in the absence of tissue from the primary site. The diagnosis of SDC was confirmed by two Head and Neck study pathologists (SKE, RDC). Most recent consecutive resections of IDC were included if they were grade 2 or 3 by Nottingham histological grade [18, 19], and morphologically resembled SDC (IDC with apocrine features) whereas those that do not were excluded (high-grade IDC with medullary features). IDC treated with neoadjuvant chemotherapy prior to resection were also excluded. Immunohistochemical stains with adequate controls were performed on representative sections from SDC and IDC. The stains performed were androgen receptor SP107 (AR), estrogen receptor-alpha SP1 (ER- α), progesterone receptor

1E2 (PR), HER-2 4B5, Cytokeratin 5/6 K5/6-D5/16B4 (CK5/6), p63-4A4, and beta-catenin-14 (Table 1). All stains were performed on Ventana Benchmark automated immunostainer (Ventana Medical Systems, Inc., Tucson, AZ) according to standard protocols with positive and negative controls. Antigen retrieval, standard on the machine, utilized the Ventana CC1 buffer at pH 8.0 solution. The percentage of tumor cells staining and intensity (no staining, 1+, 2+, and 3+) were recorded and an Allred score was given for ER and PR stains [20]. Allred score combines proportion (scores from 0 to 5) and intensity (0–3) into a single score ranging from 0 to 8 with results ≥ 3 considered positive ($> 1\%$ nuclear staining of tumor cells per College of American Pathologists guidelines) (Table 2). CAP guidelines were also used to interpret HER-2 stain (Table 3). HER-2 was considered positive if more than 10% of tumor cells showed homogeneous, dark circumferential staining (3+). An equivocal (2+) HER-2 interpretation was given when circumferential membrane staining that is incomplete and/or weak/moderate and within $> 10\%$ of the invasive tumor cells; or complete and circumferential membrane staining that is intense and within $\leq 10\%$ of the invasive tumor cells. IDC equivocal (2+) HER-2 staining was followed by HER-2 FISH analysis for clinical care. Any staining was considered positive for the other stains. Chi square test was used for statistical analysis.

Table 1 Immunohistochemistry antibody summary

Antibody	Clone	Vendor	Dilution	Antigen retrieval (min)	Incubation time (min)
Estrogen receptor- α	SP1	Ventana	Prediluted	CC1-36	12
Progesterone receptor	1E2	Ventana	Prediluted	CC1-64	12
HER-2	4B5	Ventana	Prediluted	CC1-56	4
Androgen receptor	SP107	Ventana	Prediluted	CC1-76	36
CK5/6	D5/16B4	Ventana	Prediluted	CC1-64	36
Beta catenin	(14)	Cell Marque	Prediluted	CC1-36	32
P63	4A4	Ventana	Prediluted	CC1-64	40

Table 2 Allred scoring system for ER- α and PR with total scores (combined proportion and intensity scores) ranging from 0 to 8

Proportion score	Positive cells (%)	Intensity	Intensity score
0	0	None	0
1	< 1	Weak	1
2	1–10	Intermediate	2
3	10–33	Strong	3
4	33–66		
5	≥ 67		

Total scores ≥ 3 are considered positive if the proportion score is at least 2

Results

The total number of SDCs included in this study was 23, five of which were metastatic but with known salivary gland primaries. The parotid gland was the primary site in $n = 16$ (69.6%). Other primary sites were minor salivary glands ($n = 5$, 21.7%) and the submandibular gland ($n = 2$, 8.7%). 5 (21.7%) SDCs included were carcinoma ex-pleomorphic adenoma. The 23 patients had an average age of 60.7 years (range 47–79 years) and 73.9% were males. There were 29 IDCs included in the study, of which 15 cases were grade 2 and 14 were grade 3 by Nottingham histological grade. The mean patient age was 53.6 years (range 25–80 years) and all except one patient were female (96.6%).

SDCs and IDCs shared some similarity in immunohistochemical staining patterns. Both were beta-catenin (membranous) and AR receptor positive in the majority of cases ($> 90\%$) (Table 4). In addition, both showed a subset of cases staining with CK5/6 and p63 in the invasive epithelial component. The triple negative (ER- α , PR, and HER-2 negative) IDC expressed these two stains. In the case of SDC, CK5/6 and p63 were expressed in cases with antecedent pleomorphic adenoma.

The main disparity in the immunohistochemical profiles was a distinctly different reactivity to ER- α , PR, and HER-2 (Fig. 2a–f). While 28 (96.6%) IDCs reacted

Table 4 Immunohistochemical expression profiles of invasive ductal carcinoma (IDC) and salivary duct carcinoma (SDC)

Immunohistochemical stain	IDC	SDC
Estrogen receptor-alpha	82.8% (24)	8.7% (2)
Progesterone receptor	93.1% (27)	21.7% (5)
HER-2	0% ^a	34.8% (8)
Androgen receptor	93.1% (27)	91.3% (21)
p63	27.6% (8)	26.1% (6)
Cytokeratin 5/6	20.7% (6)	39.1% (9)
Beta-catenin	96.6% (28)	100% (23)

^aOne IDC case had HER-2 amplification by FISH analysis

positively to ER- α and/or PR, the majority expressing both ($n = 24$, 82.8%) with a 2–3+ intensity score (median Allred score = 8 for ER- α and 7 for PR), only 2 (8.7%) SDCs expressed ER- α (Allred score = 5 in both cases) and 5 (21.7%) others expressed PR (Allred score ranged from 3 to 4) (P value < 0.05). Only one (4.3%) SDC studied expressed both ER- α and PR. On the other hand, 17 (34.8%) SDCs were positive for HER-2 by immunohistochemistry (3+) while no IDCs were positive (P value < 0.05). SDCs that expressed ER- α had positivity that was either focal or weak. One case was strongly positive (3+) but only in 1% of tumor cells (Allred score is $2 + 3 = 5$) and the other had weak intensity (1+) in a significant (50%) proportion of tumor cells (Allred score is $4 + 1 = 5$). The average PR proportion score in SDCs was 15% (range 2–30%) and all cases had weak intensity score (1+) (Allred score ranging from 3 to 4). Of note, SDCs in women were all ER and PR negative ($n = 6$, 26.1%).

Discussion

Although SDC and a subset of IDC can be morphologically identical, their immunohistochemical profiles are different. ER- α , PR, and HER-2 were especially helpful in our series. Most IDC in our series express both hormone receptors. In contrast, only a minority of SDCs express either marker, with only one case expressing both. IDC and SDC share similar immunohistochemical profiles for AR, beta-catenin,

Table 3 HER-2 results by immunohistochemistry

Result	Criteria
Negative (0)	No staining or incomplete, faint membrane staining in $\leq 10\%$ of invasive tumor cells
Negative (1+)	Incomplete, faint membrane staining in $> 10\%$ of invasive tumor cells
Equivocal (2+)	Incomplete and/or weak to moderate circumferential membrane staining in $> 10\%$ of invasive tumor cells or complete, intense, circumferential membrane staining in $\leq 10\%$ of invasive tumor cells
Positive (3+)	Complete, intense, circumferential membrane staining in $> 10\%$ of invasive tumor cells

Fig. 2 Salivary duct carcinoma (SDC) is more likely to be negative for ER (a) and PR (b) and positive for HER-2 (c). Invasive ductal carcinoma that resembles SDC is more likely to be ER (d) and PR (e) positive and HER-2 (f) negative

CK5/6 and p63. AR status maybe helpful in ER- α and PR negative tumors. In ER- α and PR negative SDC, AR status is universally positive ($n=18$). On the other hand, our only case of ER- α and PR negative IDC in this study was also AR negative. However, it is difficult to draw conclusions based on this observation.

The distinct ER- α and PR profiles have been shown in prior studies [11, 14]. For example, Wick et al. showed that none of the 17 SDC expressed ER- α , and only 17.6% expressed PR [14]. In the same study, they showed that 52.5 and 45.8% of IDC are ER and PR positive, respectively. A more recent study showed that 79.1% of IDC are ER/PR positive [12]. Similar to our findings, Wick et al. showed that 29.4% of SDC expressed HER-2 [14]. Although lower

HER-2 expression has been reported by others [9], its presence in the correct clinical setting can support the diagnosis of SDC [21].

Similar to our study, high AR expression was previously reported in both SDC and IDC [9, 10, 15]. Others have suggested that all SDC express AR, and that AR negative SDC are extremely rare and should be reclassified [22]. In addition, a subset of SDC expresses p63 and CK5/6 [9].

This study has several limitations. First, salivary gland specimens do not follow the same formalin fixation guidelines that are applied to breast specimens. This may affect the way tumor cells express ER- α , PR, and HER-2. Second, our IDC HER-2 expression is significantly lower than the literature [14, 23, 24], and may partially be explained by

a selection bias for cases that morphologically resemble SDC. Another possible explanation is differences in HER-2 antibody clones used in different studies [13]. Two of our cases with equivocal (2+) staining underwent HER-2 FISH analysis for routine patient care with one being positive for HER-2 amplification.

In summary, ER- α , PR, and HER-2 can help distinguish SDC and morphologically similar IDC. ER- α and PR positive tumors are more likely to be from the breast while HER-2 expression is more commonly seen in SDC. However, there is some degree of overlap especially with HER-2 staining, and therefore the immunophenotype cannot be relied upon to definitively separate IDC and SDC.

Funding Washington University Department of Pathology funded this study.

Compliance with Ethical Standards

Conflict of interest The authors declare that they have no conflict of interests.

Ethical Approval All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

References

- Kleinsasser O, Klein HJ. Basal cell adenoma of the salivary glands. *Arch Klin Exp Ohren Nasen Kehlkopfheilkd.* 1967;189(3):302–16.
- El-Naggar AK, Chan JKC, Grandis JR, Takata T, Slootweg PJ. WHO classification of head and neck tumours. Lyon: IARC; 2017.
- Sørli T, Perou CM, Tibshirani R, Aas T, Geisler S, Johnsen H, Hastie T, Eisen MB, Van De Rijn M, Jeffrey SS, Thorsen T. Gene expression patterns of breast carcinomas distinguish tumor subclasses with clinical implications. *Proc Natl Acad Sci.* 2001;98(19):10869–74.
- Jayaprakash V, Merzianu M, Warren GW, Arshad H, Hicks WL Jr, Rigual NR, Sullivan MA, Seshadri M, Marshall JR, Cohan DM, Zhao Y, Singh AK. Survival rates and prognostic factors for infiltrating salivary duct carcinoma: analysis of 228 cases from the Surveillance, Epidemiology, and End Results database. *Head Neck.* 2014;36(5):694–701.
- Salovaara E, Hakala O, Bäck L, Koivunen P, Saarilahti K, Passador-Santos F, Leivo I, Mäkitie AA. Management and outcome of salivary duct carcinoma in major salivary glands. *Eur Arch Otorhinolaryngol.* 2013;270(1):281–5.
- Gilbert MR, Sharma A, Schmitt NC, Johnson JT, Ferris RL, Duvvuri U, Kim S. A 20-year review of 75 cases of salivary duct carcinoma. *JAMA Otolaryngol Head Neck Surg.* 2016;142(5):489–95.
- Guo S, Yao J. Breast metastasis of salivary duct carcinoma in a patient: a case report. *Int J Clin Exp Med.* 2015;8(11):21765–9.
- Ando K, Masumoto N, Sakamoto M, Teraoka K, Suzuki T, Kurihara T, Abe S, Tozaki M, Fukuma E, Hoshi K. Parotid gland metastasis of breast cancer: case report and review of the literature. *Breast Care.* 2011;6(6):471–3.
- Butler RT, Spector ME, Thomas D, McDaniel AS, McHugh JB. An immunohistochemical panel for reliable differentiation of salivary duct carcinoma and mucoepidermoid carcinoma. *Head Neck Pathol.* 2014;8(2):133–40.
- Cochrane DR, Bernales S, Jacobsen BM, Cittelly DM, Howe EN, D'Amato NC, Spoelstra NS, Edgerton SM, Jean A, Guerrero J, Gómez F, Medicherla S, Alfaro IE, McCullagh E, Jedlicka P, Torkko KC, Thor AD, Elias AD, Protter AA, Richer JK. Role of the androgen receptor in breast cancer and preclinical analysis of enzalutamide. *Breast Cancer Res.* 2014;16(1):R7.
- Fan CY, Wang J, Barnes EL. Expression of androgen receptor and prostatic specific markers in salivary duct carcinoma: an immunohistochemical analysis of 13 cases and review of the literature. *Am J Surg Pathol.* 2000;24(4):579–86.
- Onitilo AA, Engel JM, Greenlee RT, Mukesh BN. Breast cancer subtypes based on ER/PR and Her2 expression: comparison of clinicopathologic features and survival. *Clin Med Res.* 2009;7(1–2):4–13.
- Skalova A, Stárek I, Vanecek T, Kucerová V, Plank L, Szépe P, Di Palma S, Leivo I. Expression of HER-2/neu gene and protein in salivary duct carcinomas of parotid gland as revealed by fluorescence in-situ hybridization and immunohistochemistry. *Histopathology.* 2003;42(4):348–56.
- Wick MR, Ockner DM, Mills SE, Ritter JH, Swanson PE. Homologous carcinomas of the breasts, skin, and salivary glands. A histologic and immunohistochemical comparison of ductal mammary carcinoma, ductal sweat gland carcinoma, and salivary duct carcinoma. *Am J Clin Pathol.* 1998;109(1):75–84.
- Williams MD, Roberts D, Blumenschein GR Jr, Temam S, Kies MS, Rosenthal DI, Weber RS, El-Naggar AK. Differential expression of hormonal and growth factor receptors in salivary duct carcinomas: biologic significance and potential role in therapeutic stratification of patients. *Am J Surg Pathol.* 2007;31(11):1645–52.
- Zhu S, Schuerch C, Hunt J. Review and updates of immunohistochemistry in selected salivary gland and head and neck tumors. *Arch Pathol Lab Med.* 2015;139(1):55–66.
- Miettinen M, Cue PA, Sarlomo-Rikala M, Rys J, Czapiewski P, Wazny K, Langfort R, Waloszczyk P, Biernat W, Lasota J, Wang Z. GATA 3—a multispecific but potentially useful marker in surgical pathology—a systematic analysis of 2500 epithelial and non-epithelial tumors. *Am J Surg Pathol.* 2014;38(1):13.
- Bloom HJ, Richardson WW. Histological grading and prognosis in breast cancer; a study of 1409 cases of which 359 have been followed for 15 years. *Br J Cancer.* 1957;11(3):359–77.
- Elston CW, Ellis IO. Pathological prognostic factors in breast cancer. I. The value of histological grade in breast cancer: experience from a large study with long-term follow-up. *Histopathology.* 1991;19:403–10.
- Harvey JM, Clark GM, Osborne K, Allred DC. Estrogen receptor status by immunohistochemistry is superior to the ligandbinding assay for predicting response to adjuvant endocrine therapy in breast cancer. *J Clin Oncol.* 1999;17:1474–81.
- Chandrasekar C, Salati N, Rao L, Radhakrishnan R. Salivary duct carcinoma in the mandibular anterior region: the role of immunohistochemical markers in its definitive diagnosis. *J Oral Maxillofac Pathol.* 2016;20(3):505–9.
- Williams L, Thompson LD, Seethala RR, Weinreb I, Assaad AM, Tuluc M, Ud Din N, Purgina B, Lai C, Griffith CC, Chiosea SI. Salivary duct carcinoma: the predominance of apocrine morphology, prevalence of histologic variants, and androgen receptor expression. *Am J Surg Pathol.* 2015;39(5):705–13.
- Piris A, Peng Y, Boussahmain C, Essary LR, Gudewicz TM, Hoang MP. Cutaneous and mammary apocrine carcinomas have different immunoprofiles. *Hum Pathol.* 2014;45(2):320–6.
- Vranic S, Marchiò C, Castellano I, Botta C, Scalzo MS, Bender RP, Payan-Gomez C, di Cantogno LV, Gugliotta P, Tondat F, di Celle PF, Mariani S, Gatalica Z, Sapino A. Immunohistochemical and molecular profiling of histologically defined apocrine carcinomas of the breast. *Hum Pathol.* 2015;46(9):1350–9.