Int J Clin Exp Med 2019;12(5):5590-5597
www.ijcem.com /ISSN:1940-5901/IJCEM0079643

Original Article

Co-existing adenoid cystic carcinoma and invasive squamous cell carcinoma of the uterine cervix: a rare case report with immunohistochemical study and literature review

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Received May 14, 2018; Accepted January 6, 2019; Epub May 15, 2019; Published May 30, 2019

Abstract: Adenoid cystic carcinoma (ACC) is a rare neoplasm of the cervix, and its coexistence with other malignancies in the uterine cervix is extremely rare. Here, a case of coexistence of ACC and invasive squamous cell carcinoma (SCC) of the uterine cervix, as well as a literature review are presented. A 71-year-old Chinese woman had abnormal vaginal bleeding for more than 20 days, and watery vaginal discharge for more than half a year. The cervical cytology screening result was highly suspicious for high-grade squamous intraepithelial lesion (ASC-H). This patient underwent cervical biopsy, and finally extensive hysterectomy plus bilateral salpingectomy and oophorectomy, as well as bilateral pelvic lymph node dissection. Subsequent pathological diagnoses of coexisting ACC and invasive SCC were rendered with a negative margin without metastasis of pelvic lymph nodes. Microscopically, ACC showed an obvious cribriform growth pattern. Immunohistochemical studies of the surgical specimen revealed that the ACC component exhibited a pattern distinct from the SCC component. ACC component was characterized by the expression of adeno-epithelial markers (CK8/18, CK7), and myoepithelial markers (P63, Calponin, CK5/6), Vimentin, CD117 patchy positive; Bcl-2, Ki-67 (about 70% positive). SCC component was p63 and CK5/6 strongly and diffusely positive, CK8/18 and CK7 patchy positive, while negative for Calponin, CD117, and Vimentin. This case was among very few previously described cases of coexisting ACC with invasive SCC in the uterine cervix.

Keywords: ACC, invasive SCC, uterine cervix, immunohistochemical stain

Introduction

Adenoid cystic carcinoma (ACC) of the uterine cervix is a rare and peculiar variant of adenocarcinoma. ACC of the uterine cervix was first described by Paalman RJ et al. in 1949 as cylindroma [1]. Currently, there have been more than 140 cases documented in the literature, and most have been published as case reports. ACC accounts for less than 1% of all cases of primary cervical adenocarcinoma [2]. The tumor has been most often seen in patients between sixth and seventh decades (mean age of 71 years in the largest series), and is more common in blacks than whites [3]. Currently, about 35 cases of co-existing ACC and invasive squamous cell carcinoma (SCC) of the uterine cervix have been documented in the literature [4]. ACC has been thought to behave more aggressively than SCC, but a standard treatment method has not been established due to its rarity. Therefore, patients with ACC still received the same treatment as SCC patients [5].

The relation of the cervical tumor to ACC of the salivary gland and upper respiratory tract neoplasms containing myoepithelial cells is still unknown. Unlike ACC of other sites, cervical ACC contained few myoepithelial cells as detected by electron microscopy or S-100 and actin immunohistochemistry. Some researchers have suggested that "adenoid cystic" carcinoma is a more appropriate designation for the

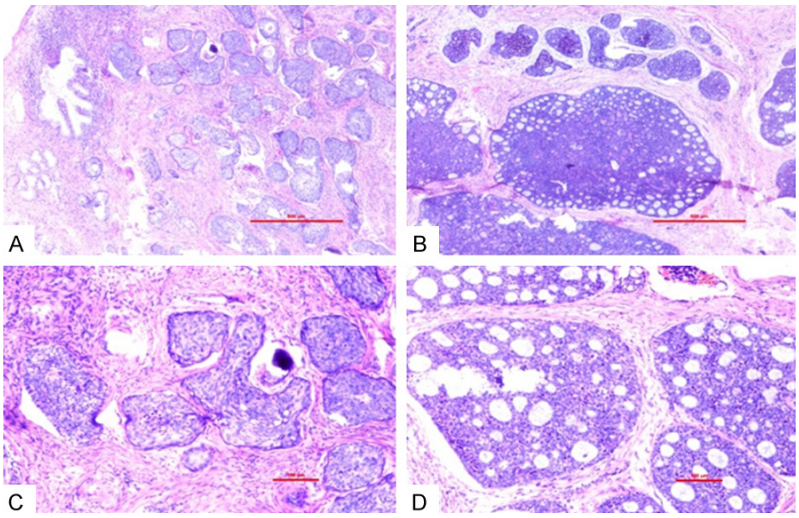

Figure 1. Hematoxylin and eosin (HE) of the SCC and ACC components in cervical tumor. A and C. The invasive SCC component of the tumor. B and D. The ACC component of the tumor with a cribriform growth pattern. Note: A and B (HE, ×4 objective lens); C and D (HE, ×10 objective lens).

cervical tumor, because it lacks consistency with the better-known tumor of salivary gland origin.

Integration of high-risk HPV DNA, including type 16, has been implicated in the pathogenesis of ACC [6]. Grayson proposed the "reserve cell origin "for the tumor, and claimed that ACC and the basaloid cystic carcinoma of the cervix derive from a common progenitor cell [7].

ACC behaved aggressively with frequent local recurrences or metastatic spread. In a review of 43 cases, overall survival for ACC patients with stage I was about 56% at 3-5 years [8]. Although some studies had reported a higher survival rate, it still appeared to be considerably lower than other types of stage I cervical cancer. ACC of the cervix should be differentiated form adenoid basal carcinoma of the cervix [9-13].

Here, a rare case of cervical ACC coexistence with invasive SCC is reported, and clinical and histological features are described, as well as the distinct immunostaining pattern. A literature review was also conducted for comparison.

Methods

Patient's information

A 71-year-old Chinese woman had abnormal vaginal bleeding for more than 20 days, and watery vaginal discharge for more than half a year. She had been post-menopausal for 20 years without a history of any other gynecologic problems. She experienced sterilization using an intrauterine device 40 years ago. No routine cervical cancer screening was conducted previously.

Histological features of the tumor

Cervical cytology screening for the patient indicated highly suspicion for high-grade squamous intraepithelial lesion (ASC-H). An exophilic and polypoid nodule with a size of 15 mm * 10 mm * 10 mm on the inferior lip of the cervix was found according to cervical examination. The fragile polyp would bleed a lot when touched.

Microscopic observation of cervical colposcopy biopsy samples showed nesting of basaloid tumor cells. Some foci showed acinar or tubular formation with abundant luminal mucin. Adequate evaluation of post-surgical specimens was needed to confirm whether it was invasive basaloid squamous carcinoma or ACC, or poorly differentiated adenocarcinoma.

The patient underwent extensive radical hysterectomy plus bilateral salpingectomy and ovariectomy, as well as bilateral pelvic lymph node dissection. Histological examination revealed that the tumor was composed of two distinct morphological types. One was an adenocarcinoma with a cribriform growth pattern and abundant luminal mucin, which was consistent with ACC (**Figure 1B**, **1D**). The other was an invasive SCC without obvious keratinization and necrosis (**Figure 1A**, **1C**). The ACC component was surrounded by the SCC component. The circumjacent SCC component had a solid basaloid pattern without keratinization and central necrosis. The ACC component was more prominent (65%) than the SCC component (35%). Both the ACC and SCC components infiltrated greater than 2/3 of the full thickness of the cervix.

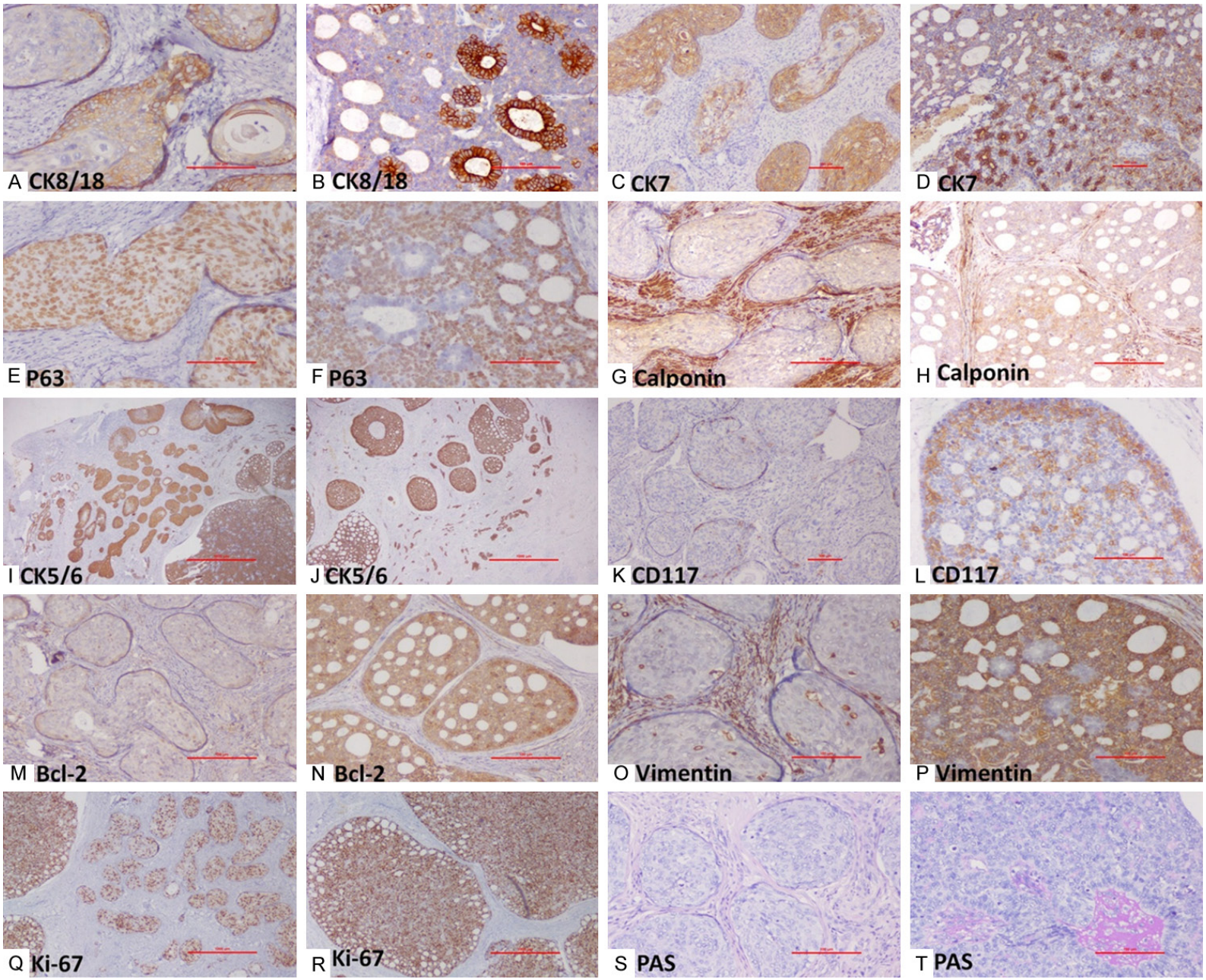
A CK8/18
B CK8/18
C CK7
D CK7
E P63
F P63
G Calponin
H Calponin
I CK5/6
J CK5/6
K CD117
L CD117
M Bcl-2
N Bcl-2
O Vimentin
P Vimentin
Q Ki-67
R Ki-67
S PAS
T PAS

Figure 2. Immunostaining of SCC and ACC components in cervical tumor. A. CK8/18 was weakly positive in the SCC component. B. CK8/18 was partially strongly positive in the tumor cells located duct in the ACC component. C. CK7 was partially positive in the SCC component. D. CK7 was positive in the ACC component similar to that of CK8/18. E. P63 was diffuse positive in the SCC component. F. P63 was mostly positive in myoepithelial cells in the pseudocysts of the ACC component. G. Calponin was negative in the SCC compartment. H. Calponin was locally positive in myoepithelial cells of ACC compartment. I. CK5/6 exhibited strong and diffuse plasma positive staining in the SCC component. J. CK5/6 was mostly positive in the ACC component. K. CD117 was negative in the SCC component. L. CD117 was locally positive in the luminal cell layers in ACC component while the myoepithelial cells do not express C-kit. M. Bcl-2 was patchy positive in the SCC component. N. Bcl-2 was strongly and diffusely membrane and cytoplasmic positive in ACC component. O. Vimentin was negative in the SCC component. P. Vimentin was strongly and diffusely positive in the SCC component. Q. Ki-67 was almost 40-60% positive in the SCC component. R. Ki-67 was nearly 70% positive in the ACC component. S. PAS staining was negative in the SCC component. T. Duct-like structures that contain extracellular matrices filled with eosinophilic periodic acid-Schiff (PAS) positive material were detected in the ACC component. Note: A, B, E-H, K-P, S and T (HE, ×20 objective lens); C and D (HE, ×10 objective lens); I, J, Q and R (HE, ×2 objective lens).

Table 1. Summary of the immunostaining pattern of ACC and SCC

	CK8/18	CK7	P63	Calponin	CK5/6	CD117	Bcl-2	Vim	Ki-67	PAS
SCC	Weakly +	Partial +	Diffuse +	-	Diffuse +	-	Patchy +	-	Almost 40-60% +	-
ACC	Partial +	+	Mostly +	Locally +	Mostly +	Locally	Diffuse +	Diffuse +	Almost 70%	+

Immunohistochemical features of the tumor

CK8/18 and CK7 were two key adeno-epithelial markers. In the SCC component, weakly positive expressed CK8/18 (**Figure 2A**) and partially positive expressed CK7 (**Figure 2C**) was found. However, the ACC component exhibited partially strongly positive CK8/18 (**Figure 2B**) and positive expressed CK7 (**Figure 2D**).

There were also differences in expression of myoepithelial markers (including P63, Calponin and CK5/6) in the SCC component and the ACC component. For the SCC component, it presented strong and diffuse nuclear positive staining of P63 (**Figure 2E**), negative Calponin (**Figure 2G**), as well as strong and diffuse cytoplasmic positive staining of CK5/6 (**Figure 2I**). At the same time, mostly positive P63 (**Figure 2F**), locally positive Calponin (**Figure 2H**), as well as mostly positive CK5/6 (**Figure 2J**) was observed in the ACC component.

ACC was usually strongly immunoreactive for CD117, which was a helpful adjunct to distinguish between the ACC and SCC components. In this case, negative CD117 (**Figure 2K**), patchy positive Bcl-2 (**Figure 2M**), as well as negative Vimentin (**Figure 2O**) occurred in the SCC component. However, the ACC component showed locally positive CD117 (**Figure 2L**), strong and diffuse expression of Bcl-2 (**Figure 2N**), and strongly and diffusely positive Vimentin (**Figure 2P**).

In addition, Ki-67 was almost 40-60% positive in the SCC component (**Figure 2Q**), while nearly 70% in the ACC component (**Figure 2R**). Periodic acid-schiff (PAS) staining was negative in the SCC component (**Figure 2S**), whereas PAS positive expression was detected in the ACC component (**Figure 2T**).

More detailed information on expression of the above proteins is shown in **Table 1**.

Treatment

According to the NCCN Guidelines Version 1.2016 Cervical Cancer, this case belonged to IB1. Before surgery, the patient's pelvic cavity computed tomography demonstrated a well enhanced mass (red arrows) in the uterine cervix (**Figure 3**). She underwent radical hysterectomy with bilateral pelvic lymphadenectomy, and bilateral salpingectomy and ovariectomy due to lack of fertility. After surgery, she was treated with radiation (200cGy). Two months after treatment, no abnormal reaction was occurred and the patient was with blood routine test results and good mental state.

Discussion

ACC (also called adenocystic carcinoma, cylindroma or cylindromatous adenocarcinoma) is a malignant epithelial neoplasm derived from the salivary glands, which might occur in variety of other sites, such as minor and major glands,

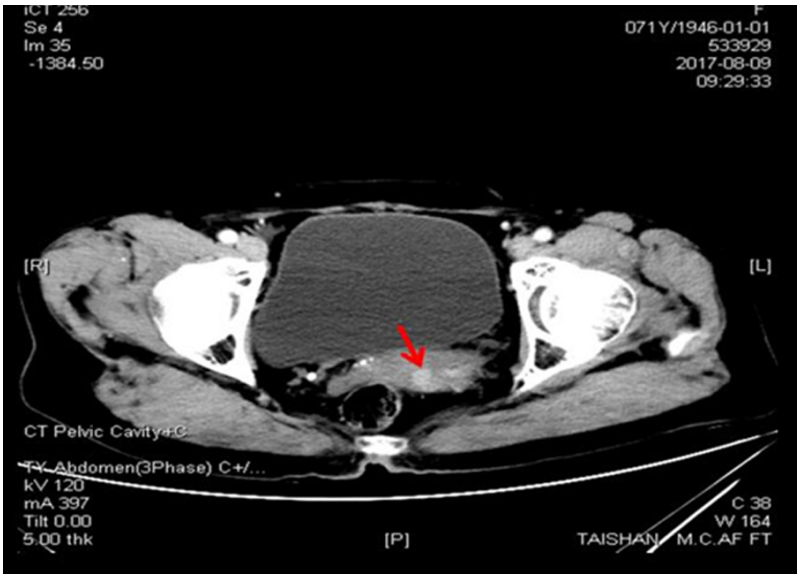

Figure 3. Pelvic cavity computed tomography demonstrated a well-demarcated mass (red arrows) in the uterine cervix.

lacrymal glands, mucous glands of the aerodigestive tract, skin, breast and lung [14, 15].

ACC of the uterine cervix is the least common of the cervical reserve cell carcinomas but their histological patterns were distinctive and easily recognized. The majority of reported cases of ACC had more specific architectural features with the typical cribriform pattern. Cribriform pattern was the most characteristic pattern in ACC, and was composed of polygonal to spindled cells forming numerous duct-like structures that contained extracellular matrices filled with homogenous eosinophilic PAS positive material or granular basophilic material [16, 17]. In the tubular type, small ductal structures formed by one or two layers of polygonal tumor cells were intermingled with solid strands of tumor cells. The solid type was characterized by solid sheets with mitotic activity and frequently with central necrosis. Small clusters of larger polygonal cells were occasionally present. This pattern was morphologically least differentiated and was not diagnostic by itself. Perzin et al. [18, 19] described a grading system for ACC based on distinctive histologic patterns: tubular, cribriform and solid. They believed that the three patterns reflected a progression of cellular proliferation and aggressiveness of biologic behavior. The tumor was assigned to three histological grades: grade I, a well-differentiated tumor composed of tubular and cribriform patterns without solid components; grade II, a tumor with a pure cribriform pattern or mixed with less than 30% of solid area; grade III, a tumor with marked predominance of the solid pattern.

ACC was generally a locally aggressive tumor and had a high propensity for local recurrence and distant metastasis [20]. Thus, it needed to be differentiated from other less aggressive tumors. Immunohistochemically, ACC cells stained positively with broad spectrum cytokeratin, CAM 5.2, CK7, CD117, and EMA. CEA might also be expressed. Nevertheless, cervical ACC contained few or no myoepithelial cells, which could distinguish this tumor from ACC of other sites. In fact, S-100 protein reactivity occurred in some cases while others were unreactive or just focally and weakly reactive [7, 21, 22]. Also, ACC has rarely been reported to be associated with intraepithelial squamous neoplasia, invasive SCC, adenocarcinoma, and sarcoma in the uterine cervix [22, 23].

The differential diagnosis included small cell carcinoma, adenoid basal carcinoma and nonkeratinizing SCC.

A review of cases from the literature are also summarized. The patients' age at diagnosis, the tumor size, the cancer stage, high-risk HPV detection results, the microscopic appearance, the treatment methods and the duration of survival are summarized in **Table 2** according to the publication year. It could be seen that ACC usually occurred in older women in their sixth and seventh decades. The vast majority of patients developed their tumors several years after menopause, although the tumor could occur in younger women. Very rare cases have been reported in women less than 40 years old [24], and it was reported that these cases were related to human papilloma virus (HPV) DNA (types 16, 18, and 31) [24-26]. So, ACC of the cervix in younger individuals also took on aspects of a sexually transmitted disease [27]. In our case, human papilloma virus (HPV) DNA

Table 2. Review of previous reported cases of cervical ACC associated with invasive SCC

Author	Publication Year	Age at diagnosis	Tumor size	Cancer stage	HPV result	Microscopic appearance	Treatment method	Duration of survival
Paalman et al. [1]	1949	67	NA	NA	NA	ACC + CIS	Surgery	NA
Tchertkoff et al. [34]	1962	63	NA	NA	NA	ACC + invasive SCC	Surgery	NA
Moss et al. [35]	1964	56	NA	NA	NA	ACC + invasive SCC + CIS	Surgery	NA
Gallager et al. [36]	1971	NA	NA	NA	NA	2 of 4 had ACC + CIS	NA	NA
Miles et al [37]	1971	NA	NA	NA	NA	3 of 12 had ACC + invasive SCC, 1 of 12 had ACC + CIS	Surgery	NA
Ramzy et al. [38]	1975	68	NA	NA	NA	ACC + invasive SCC	Surgery + R	DOD, 27 months
Shingleton et al. [39]	1977	78	NA	FIGO III	NA	ACC + focal SCC	R	DOD, 3 months
		65	NA	FIGO IIB	NA	ACC + focal SCC	Surgery	NED
		64	NA	NA	NA	ACC + poorly differentiated SCC	Surgery	DOD, 5 months
Fowler et al. [40]	1978	NA	NA	NA	NA	1 of 9 had ACC + invasive SCC	NA	NA
Gupta et al. [41]	1979	47	NA	FIGO III	NA	ACC + CIS	NA	DOD, 12 months
Hoskins et al. [42]	1979	54	NA	FIGO IB	NA	Adenosquamous carcinoma and the adeno component is ACC	Surgery + R	NED, 12 months
Bittencourt et al. [43]	1979	60	7 cm	FIGO IB	NA	ACC + invasive SCC	Surgery + R	Surgery + R
		72	NA	FIGO IB	NA	ACC + invasive SCC	R	NED, 27 months
Mazur et al. [44]	1982	76	NA	NA	NA	ACC + invasive SCC + CIS	Surgery + R	NA
Van Dinh et al. [45]	1985	NA	NA	NA	NA	4 of 9 had ACC + CIS	NA	NA
Berchuck et al. [29]	1985	72	9.5 cm	FIGO IIIB	NA	ACC + large cell squamous components	Sugery + R	NED, 6 months
Musa et al. [46]	1985	NA	NA	NA	NA	2 of 17 had ACC + invasive SCC, 2 of 17 had ACC + CIS	NA	NA
Ferry et al. [21]	1988	NA	NA	NA	NA	4 of 14 had ACC + CIS, 1 had ACC + SCC + CIS + ABC	NA	NA
King et al. [23]	1989	27	No gross nodule	FIGO IB	NA	ACC + CIS	Surgery	NED, 12 months
Albores et al. [7]	1992	NA	NA	NA	NA	3 of 7 had ACC + focal invasive SCC, 1 had ACC + CIS chemotherapy	NA	NA
Dixit et al. [33]	1994	30	NA	FIGO IIIB	NA	ACC + CIS	R	NED, 50 months
Manhoff et al. [47]	1995	80	NA	NA	NA	ACC + focally invasive SCC + malignant stroma	Surgery + R	NED, 6 months
Vuong et al. [2]	1996	60	2cm	NA	NA	ACC + CIS	Surgery	NED, 60 months
Grayson et al. [25]	1996	NA	NA	NA	HPV16 pos	ACC + superficially invasive SCC	NA	NA
Mathoulin et al. [48]	1998	73	5cm	NA	NA	ACC + a rare focus of SCC + sarcoma	Sugery + R	NED, 6 months
Grayson et al. [3]	1999	NA	NA	NA	NA	6 of 18 had ACC + invasive SCC, 1 had ACC + invasive SCC + ABC	NA	NA
Yang et al. [26]	1999	36	No gross nodule	NA	HPV16, 18 pos in SCC, neg in ACC component	ACC + SCC + CIS + condyloma	Surgery	NA
Grayson et al. [49]	2001	68	NA	NA	HPV16 pos	ACC + SCC + CIS	NA	NA
		55	NA	NA	HPV16 pos	ACC + SCC + CIS + focal ABC	NA	NA
		36	NA	NA	Neg	ACC + SCC	NA	NA
Ishiko et al. [50]	2001	76	NA	FIGO IB	NA	ACC + CIS	NA	NED, 12 months
Seth et al. [51]	2009	24	3 cm	NA	NA	ACC + focal SCC	Surgery	NA
Xiaohua Shi [4]	2014	68	0.5 cm	FIGO I	HPV16, 18 pos	ACC and SCC	NA	NA
Xiaohua Shi [22]	2015	64	5 cm	FIGO I	HPV, pos	ACC and SCC	Sugery + R + C	NED, 20 months
		63	10 cm	FIGO I	HPV, pos	ACC and SCC	Sugery + R + C	NED, 25 months
		77	15 cm	FIGO I	HPV, pos	ACC and SCC	Sugery + R + C	NED, 20 months

(types 16) was positive in both the ACC component and the SCC component.

The tumor size varied from 0.5 cm to 15 cm. The cancer stage of most cases was diagnosed in early stages, as were most cervical carcinomas. ACC of cervix was characterized by highly aggressive behavior. Therefore, outcomes in advanced disease (stage III and IV) were invariably poorly, and the shortest period was 3 months after diagnosis as reported.

ACC of cervix was a rare entity and it was considered as a radiosensitive tumor, but at the same time, highly aggressive biological behavior of the tumor led to early recurrences and metastases. Management of cervical ACCs comprised surgical intervention as the initial strategy for early stages. Radiation therapy was usually recommended as adjuvant treatment. The role of chemotherapy was undefined as adjuvant or primary treatment, although in some cases, chemotherapy had been used for recurrent or metastatic disease [28, 29]. ACC of cervix was considered as radiosensitive tumor and better results had been seen in reported cases as compared to those seen in cases where surgery had been done alone, in early stages [8, 30]. Distant metastases were the main determinant of the survival. Therefore, majority of patients required aggressive local and systemic therapy, considering high local and distant relapses.

Ethassani et al. reported 13 cases of stage IIIB ACC cervix in 2008 and they claimed that the first case was successfully managed with concurrent chemo-radiation [31]. Nishida et al. treated a case of stage IIIB with radiotherapy alone. The patient showed no evidence of recurrent tumor at 5 years after radiotherapy [32]. Dixit et al. reported eight patients of stage III who had taken radiotherapy treatment. Only one patient remained disease free at 11 months and none remained disease free for more than two years, with NED rate of 12.5% (l/8) [33]. Cervical ACC was much more aggressive than SCC. Generally, treatment for ACC was the same as that for SCC of the cervix. A combination of surgery, chemotherapy and radiation therapy was recommended. However, because of the rarity of this tumor, there has been no universal consent in the standard treatment of this devastating malignancy. When there was coexistence of these two malignant tumors in the uterine cervix, a close follow up was highly recommended.

Conclusion

ACC of the cervix is a rare, particularly aggressive neoplasm. It requires enhancement of postoperative treatment regimens and careful follow-up and thus needed to be distinguished from other tumors with similar histological aspects. Association of ACC with SCC suggests a common origin, and further studies are required to explore the histogenesis of ACC in the uterine cervix.

Disclosure of conflict of interest

None.

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