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The variable morphological spectrum of penile basaloid carcinomas: differential diagnosis, prognostic factors and outcome report in 27 cases classified as classic and mixed variants

Isabel Alvarado-Cabrero[1], Diego Fernando Sanchez[2,3], Diana Piedras[1], Adriana Rodriguez-Gómez[1], Ingrid M. Rodriguez[2,3], Maria Jose Fernandez-Nestosa[4], Narciso Hernández-Toriz[1] and Antonio L. Cubilla[2,3*]

Abstract

Background: Basaloid carcinomas of the penis, HPV-related tumors, are morphologically less homogenous than originally thought. The study objective was to evaluate the prognostic influence of the basaloid pattern in mixed tumors.

Methods: We studied 154 Mexican patients from the Hospital de Oncología, CMN, Mexico City (2000–2013) and found 27 with basaloid features in at least 20% of the sections classified as classic basaloid (8 cases), warty-basaloid (7), papillary-basaloid (5) and usual-basaloid squamous cell carcinomas (7). We evaluated patients' age, site and size of tumor, histological classification, grade, thickness, anatomical level, vascular and perineural invasion, prognostic index score and node involvement. Penile intraepithelial neoplasia in adjacent epithelia was documented. Follow up ranged from 12–78 months. Statistical methods were Fisher's exact test and Kruskal-Wallis test. Kaplan-Meier method and log-rank test were used for survival analysis. The cutoff for statistical significance was $p < 0.05$.

Results: There were not clinical differences. Microscopically types were distinctive and easy to separate. Usual-basaloid squamous cell carcinomas were smaller, thinner and rarely invaded corpora cavernosa, with a low prognostic index score. Classic basaloid, warty-basaloid and papillary-basaloid carcinomas had higher rates of vascular and perineural invasion and higher prognostic index scores. These findings correlated with the rate of nodal metastasis. The majority of patients with classic and papillary-basaloid neoplasms died from systemic metastasis (87.5 and 80%) whereas only 1 patient with usual-basaloid carcinoma died of the disease (14%).

Conclusions: Basaloid carcinomas are not a single entity but a spectrum of variable histological architectures mixed with those of classic basaloid tumors. Identification of mature squamous cells in a basaloid carcinoma may be important to recognize and report because patients with these tumors may carry a better prognosis.

Keywords: Penile carcinoma, Basaloid carcinoma, Warty carcinoma, Squamous cell carcinoma

* Correspondence: antoniocubillaramos@gmail.com
[2]Instituto de Patología e Investigación, Martin Brizuela 325 casi Ayala Velazquez, Asunción, Paraguay
[3]Facultad de Ciencias Médicas, Universidad Nacional de Asunción, San Lorenzo, Paraguay
Full list of author information is available at the end of the article

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Background

Basaloid carcinomas of the penis are unusual morphologically distinctive, Human papillomavirus (HPV)-related, penile neoplasms [1]. They are morphologically similar to basaloid carcinomas described in other anogenital and non-genital sites (anus, cervix, vulva, head and neck, lung and esophagus) [2–13]. They comprise about 5-10% of penile squamous cell carcinomas (SCC), are frequently associated with HPV genotype 16 [14–16], are positive for p16 by immunohistochemistry [17] and have an aggressive clinical behavior with a high-rate of vascular invasion and inguinal lymph node metastasis [18]. After the pathological examination of a large cohort of about 1.000 penile SCC in an international HPV detection study from 5 continents [19] we found basaloid carcinomas to be morphologically less homogenous than originally thought. In addition to the classic nesting pattern composed of uniform small basophilic cells, we also observed tumors of confluent sheets of larger polyhedral cells as well as others with organoid, neuroendocrine-like and spindle cells. Another observation, more pertinent to this presentation, was that classic basaloid carcinomas may be mixed with features of other variants of squamous cell carcinoma, typically usual, warty and papillary basaloid, and less frequently pseudoglandular and sarcomatoid. Variations in basaloid carcinomas were also noted in head and neck tumors, although at these sites, other anatomical considerations (presence of salivary glands) may add to the confusion [4]. Mixed penile SCC are a poorly understood but not infrequent group of tumors, comprising about a fourth to a third of all penile neoplasms [20]. They are often problematic to classify [21]. The objective of this study was to start expanding the knowledge of penile mixed SCC determining and comparing the prognostic influence of the basaloid pattern in variously identified tumor categories. For this purpose we described and compared clinical, pathological and outcome features of patients with classic basaloid, warty-basaloid, papillary basaloid and usual-basaloid squamous cell carcinomas.

Methods

A retrospective review of clinic-pathological features of 154 patients with invasive penile carcinomas diagnosed and uniformly treated at the Hospital de Oncología, CMN from Mexico City during the period 2000–2013 was performed. Twenty-seven tumors harboring basaloid features in at least 20% of examined tissue sections were selected for the study. All patients were Mexicans and had radical surgical excision, either total (22 cases) or partial (5 cases) penectomies, with bilateral inguinal lymph node dissection in 20 cases. In 7 patients no dissections were performed due to unresectable massive nodal metastasis, supported by physical examination and ultrasonography. Clinical charts and pathology reports were reviewed to obtain clinical, pathological and outcome information. For each case, 8–20 H&E stained slides of the primary specimen were available and included several sections of the primary tumor as well as of adjacent penile tissue for the evaluation of precancerous lesions, the later in the majority of the cases. Clinical data were patients' age, tumor site at presentation and inguinal nodal status at physical examination. Pathological data of primary tumors included gross maximum diameter in cm, histological subtyping of carcinomas, histological grade from 1–3, tumor thickness (in mm), anatomical level of invasion, and perineural and vascular invasion (including lymphatic invasion). Basaloid features were characterized by a confluent or non confluent nesting pattern composed of basophilic cells with uniform small to intermediate size nuclei, frequent central necrosis or abrupt keratinization and numerous mitosis [21]. Tumors composed by at least 80% of cells with basaloid squamous cell carcinoma features [21] were classified as classic basaloid carcinoma, those with mixed features of warty or usual squamous cell carcinoma with at least 20% of basaloid carcinoma were defined as warty-basaloid [22] and usual-basaloid carcinoma respectively. Papillary neoplasms composed of small blue cells were typified as papillary basaloid carcinomas [15]. Grading was performed according to previously reported criteria: extreme differentiation resembling normal squamous epithelium was grade 1, anaplasia, even when focal was grade 3, and the remainder cases were grade 2 [23]. Anatomical levels in the glans were lamina propria (LP), corpus spongiosum (CS) and corpora cavernosa (CC). The tunica albuginea was considered part of CC [24, 25]. Tumor thickness was measured in millimeters from the non-keratinized surface to deepest point of invasion [26]. The Prognostic Index, from 2–7, was determined in each case. It consisted in the addition of numerical values given to histological grades (1, 2 and 3), anatomical levels of invasion (LP = 1; CS/dartos = 2; CC = 3), and perineural invasion (present = 1, absent = 0) [27]. Penile Intraepithelial Neoplasia (PeIN) present in tissues adjacent to invasive carcinomas were classified as differentiated, basaloid, warty, warty-basaloid and mixed [28].

Bilateral inguinal dissections, performed in 20 patients (74%), yielded 338 lymph nodes, 168 in the right and 170 in the left. The number of nodes per patient ranged from 4–14 per side with a median of 8 nodes in each groin. Histologically positive nodes were counted in each side. Follow up ranged from 12–78 months (average 48 months).

Statistical methods to compare features of the 4 variants were Fisher's exact test for categorical variables and Kruskal-Wallis test to compare equality of means among population groups. Kaplan-Meier analysis and

log-rank test were used to evaluate patients' survival. The cutoff for statistical significance was p <0.05. All analysis were made with STATA 11 SE software (Statacorp LP, College Station, TX)

All data generated or analyzed during this study are included in this published article and its supplementary information files (Additional file 1)

Results

Data on clinicopathological features, precancerous lesions, inguinal lymph node metastasis and patients outcome according to histological subtypes are listed in Tables 1, 2 and 3.

Basaloid carcinomas, classic (8 cases)

Patients age ranged from 47–76 years (mean of 66.2). Sites of primary tumors were the glans and coronal sulcus in 7 and 1 patient, respectively. Physical examination of inguinal regions revealed clinically positive palpable nodes in all patients. Surgical treatment consisted of total penectomies in 6 cases, partial penectomy in one case, total penectomy plus emasculation in 1 case with a large neoplasm. Bilateral inguinal lymph node dissections were performed in 5 cases. In 3 patients bilateral unresectable nodes were documented by physical examination and ultrasonography. Radiotherapy to inguinal and pelvic regions was utilized in 5 patients after surgery.

Grossly, tumors were large, firm, irregular, white-gray, beige or dark brown masses replacing the glans measuring 2–8 cm (average 5.7 cm). The cut surface revealed a homogenously beige, deeply infiltrating solid tumor usually affecting the tunica albuginea and erectile tissues of corpora cavernosa. Microscopically there were compact nests of invasive carcinoma with or without central necrosis separated by scant stroma. A clear space surrounding the nests was frequently observed (Fig. 1a). Occasionally, confluence of nests into wider sheets of tumor cells was found (Fig. 1b). Keratinization was scant, however when present it was usually abrupt and located in the center of the nests (Fig. 1c). Cells were basophilic, with uniform round small to intermediate sized nuclei with numerous mitoses. Rarely polyhedrical or spindle cells were noted. Nucleoli were small and inconspicuous. Individual cell necrosis was frequently observed. Mitoses were numerous (about 15–20 per high power fields) (Fig. 1d). In 2 cases the stroma was hyalinized and the tumors appeared organoid. All tumors were poorly differentiated (grade 3) and deeply invasive into corpora cavernosa (8 cases). Tumor thickness ranged from 14–26 mm (average 21.1 mm). Vascular invasion was documented in all cases and perineural invasion in 7 of them. Prognostic Index score were 6 in one case and 7 in 7 cases. Changes of basaloid PeIN were observed in 6 cases and of mixed differentiated-basaloid PeIN in 2 cases.

Inguinal nodal metastases were bilateral in 6 and unilateral in 2 patients. The number of positive nodes varied from 2–3 per groin side and from 2–5 per patient. The majority of nodal metastases were macro metastases with capsular rupture and invasion of peri-nodal adipose tissue. During the follow up (average 4 years) systemic metastases documented by imaging were located in the retroperitoneum (4 cases), lungs (3 cases) and liver (1 case). Seven patients died of systemic metastatic disease and 1 is alive with enlarged retroperitoneal nodes documented 2 years after diagnosis.

Table 1 Pathological features and prognostic factors

	Classic (8 pts)	Papillary (5 pts)	Warty-bas (7 pts)	U-Basaloid (7pts)	p <
Size (cm)	5.7	6.4	4.8	4	0.34
Site Glans (%)	8 (100)	5 (100)	7 (100)	4 (57.1)	-
Thickness (mm)	21.1	19.2	18.4	12.9	0.04
Grade 3 (%)	8 (100)	5 (100)	5 (71.4)	7 (100)	0.15
Inv. CS (%)	0 (0)	1 (20)	3 (42.9)	6 (85.7)	0.004
Inv. CC (%)	8 (100)	4 (80)	4 (57.1)	1 (14.3)	
VI (%)	8 (100)	5 (100)	5 (71.4)	3 (42.9)	0.025
PNI (%)	7 (87.5)	3 (60)	4 (57.1)	2 (28.6)	0.15
PI 7 (%)	7 (87.5)	3 (60)	4 (57.1)	1 (14.3)	0.04

U-basaloid usual-basaloid, *CS* corpus spongiosum, *CC* corpus cavernosum, *VI* vascular invasion, *PNI* perineural invasion, *PI* Prognostic Index

Papillary basaloid carcinomas (5 cases)

Patients age ranged from 52–82 years (mean of 63.8 years). The glans was the preferred primary tumor site (5 cases). In 2 patients the tumor extended to the coronal sulcus. Inguinal nodes were clinically palpable in all cases. The surgical treatment consisted in total penectomies in 5 cases and bilateral inguinal lymph node dissection in 3. In 2 patients there were bilateral unresectable nodes documented by physical examination and ultrasonography. Post operative radiotherapy was administered to 4 patients.

Grossly, tumors sizes ranged from 4–9 cm (average of 6.4 cm) in largest diameter and were exo-endophytic. There was a villous surface with a solid deeply invasive component. Tumor cut surface was white-gray to beige. Microscopically, the papillae had a central fibrovascular core and were composed of homogeneous small basophilic cells similar to those observed in basaloid PeIN, or invasive basaloid carcinomas (Fig. 2). The papillae surface had a thin keratin layer, with parakeratosis and occasional clear koilocytic cells. Whereas the exophytic component of these tumors was non invasive, in all cases there was an invasive component. The morphological features of the invasive tumor were undistinguishable

Table 2 Precancerous lesions (PeIN) according to subtypes

Penile Intraepithelial Neoplasia	Total	Classical Basaloid (8 cases)	Papillary basaloid (5 cases)	Warty-basaloid (7 cases)	Usual basaloid (7 cases)
Differentiated	1	0	0	0	1
Basaloid	12	6	3	1	2
Warty-basaloid	2	0	0	2	0
Warty	1	0	0	1	0
Mixed Diff-basaloid	8	2	1	1	4
Mixed Diff-warty	1	0	0	1	0
Total	25	8	4	6	7

from those described above for classic basaloid carcinomas. Tumor cells in the papillary and non-papillary portions of the neoplasms were poorly differentiated or grade 3 and deeply invaded into corpora cavernosa in 4 cases and corpus spongiosum in another case. Tumor thickness varied from 15–24 mm (average 19.2 mm). Vascular invasion was noted in all cases and perineural invasion in 3 of them. Prognostic Index scores were high, as follow: 5 and 6 (one case each) and 7 (in 3 cases). Associated changes of basaloid PeIN and mixed differentiated-basaloid PeIN were observed in 3 and 1 case respectively.

Inguinal lymph node metastases were present in 3 patients; in two of them they were massive, grossly confluent and bilateral. During follow up, 4 patients developed systemic metastasis as follows: liver and retroperitoneum, retroperitoneum, lung and liver, and lung and retroperitoneum. Follow up varied from 24–60 months (average 42 months). Four patients died of metastatic disease and the remainder was alive with no evidence of disease.

Warty-basaloid carcinomas (7 cases)

Patients ranged from 41–71 years (mean of 55.1 years). Primary tumors were located in the glans in all cases. In two patients tumor extended to the foreskin. Inguinal nodes were clinically palpable in 5 cases. The surgical treatment consisted in total or partial penectomies (5 and 2 cases respectively) and bilateral inguinal lymph node dissections in all cases followed by radiotherapy in 3.

Grossly tumors were in general large, ranging from 1.5 to 8 cm (average of 4.8 cm.) in largest diameter. There was an exophytic papillomatous surface in some cases. Others were irregular white gray tumor masses replacing most of the distal portion of the penis. The cut surface revealed beige to gray solid appearance, with deep invasion into corpus spongiosum or cavernosum. No superficial tumors (invading into lamina propria only) were found. Microscopically, there was either a surface classic warty carcinoma with an underlying deeply invasive carcinoma with basaloid features as described above, or a mixture of warty and basaloid features in the same tumor nests (Fig. 3). Five tumors were poorly differentiated (grade 3) and 4 tumors deeply invaded into CC. Tumor thickness varied from 10–26 mm (average 18.4 mm). Vascular invasion was noted in 5 cases and perineural invasion in 4. Prognostic Index scores varied from 4 (2 cases), 5 (1 case) and 7 (4 cases) The subtypes of PeIN identified in association with the invasive carcinomas were variable: Warty/basaloid (2 cases), basaloid (1 case), warty (1 case), mixed differentiated-basaloid (1 case) and mixed differentiated-warty (1 case).

Inguinal lymph node metastasis was present in 4 patients; in two of them it was bilateral. The range period of follow up was of 3–5 years (average 4.3 years). Four patients developed systemic metastases in the retroperitoneum, liver and lungs. Three patients were dead of metastatic disease at 42 months, one was alive with disease (60 months) and the remainders were alive at 60 (2 cases) and 48 months of follow up (1 case).

Table 3 Regional metastasis, laterality and outcome according to subtypes

	Classical (8 pts)	Papillary (5 pts)	Warty-bas (7 pts)	U-Basaloid (7pts)	$p <$
Positive nodes (%)	8 (100)	3 (60)	4 (57.1)	3 (42,9)	0.08
Bilateral + nodes (%)	6 (75)	2 (40)	2 (28.6)	1 (14,3)	0.2
DOD (%)	7 (87.5)	4 (80)	3 (42.9)	1 (14,3)	0.02 for DOD
NED (%)	0 (0)	1 (20)	3 (42.9)	4 (57,1)	0.043*
AwD (%)	1 (12.5)	0 (0)	1 (14.3)	2 (28,6)	

DOD dead of disease, *NED* no evidence of disease, *AwD* alive with disease. *Combined outcome

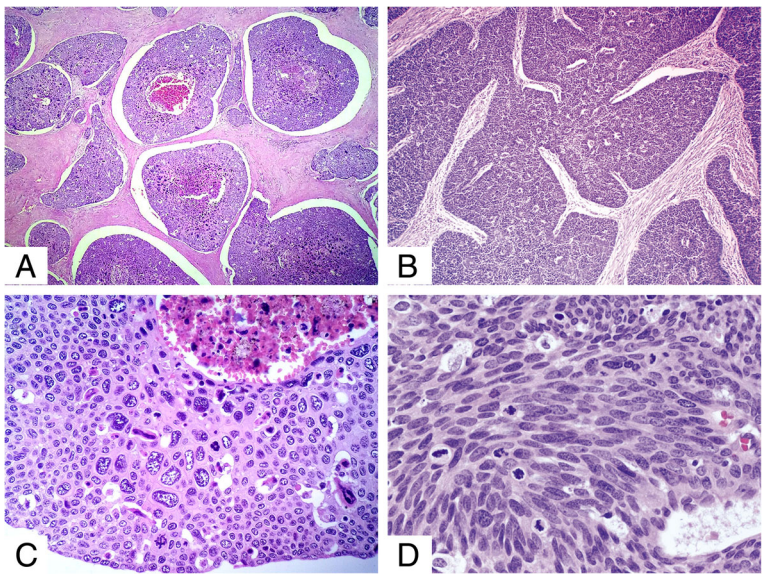

Fig. 1 Classic basaloid squamous cell carcinoma. **a**-Small basaloid cells forming solid nests some of them with comedonecrosis-like foci. **b**- Sheets of basophilic tumor cells with peripheral clear artifacts. **c**- Pleomorphic basaloid cells with numerous mitoses. Note the abrupt keratinization in the upper right corner. **d**- Basaloid carcinoma with spindle cell features. Mitoses are numerous

Mixed usual-basaloid carcinoma (7 cases)

Mean age was 59.3 years ranging from 30–83 years. Tumors were located as follow: glans and coronal sulcus (4 cases), glans-coronal sulcus-foreskin (1 case), foreskin and glans (1 case) and foreskin and coronal sulcus (1 case). Inguinal nodes were palpable in 6 patients. Surgical treatment was total penectomy in 5 cases and partial penectomy in 2 cases. Bilateral inguinal lymph node dissection was performed in 6 patients. Post operatory radiotherapy was administered to 4 patients.

Grossly, tumors were white-gray firm ulcerating and granular masses measuring from 1.5 to 8 cm (average 4 cm). Microscopically, all cases presented mixed features of moderately differentiated grade 2 keratinizing usual squamous cell carcinoma and classic grade 3 basaloid carcinoma. The basaloid component represented

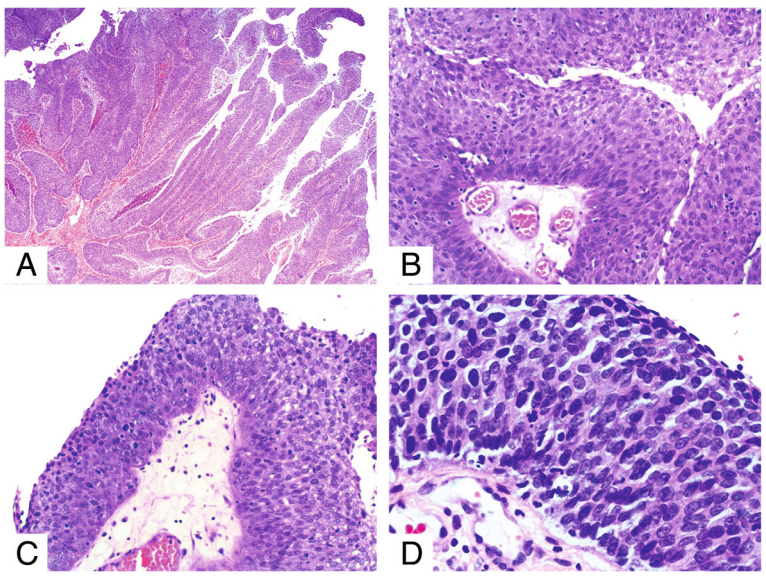

Fig. 2 Papillary-basaloid squamous cell carcinoma. **a**- Low power view showing an exo-endophytic tumor. **b**, **c** Tumor is compose of a central fibrovascular core lined by uniform small basalod cells. **d**- High power view of lining neoplastic epithelium with numerous mitoses

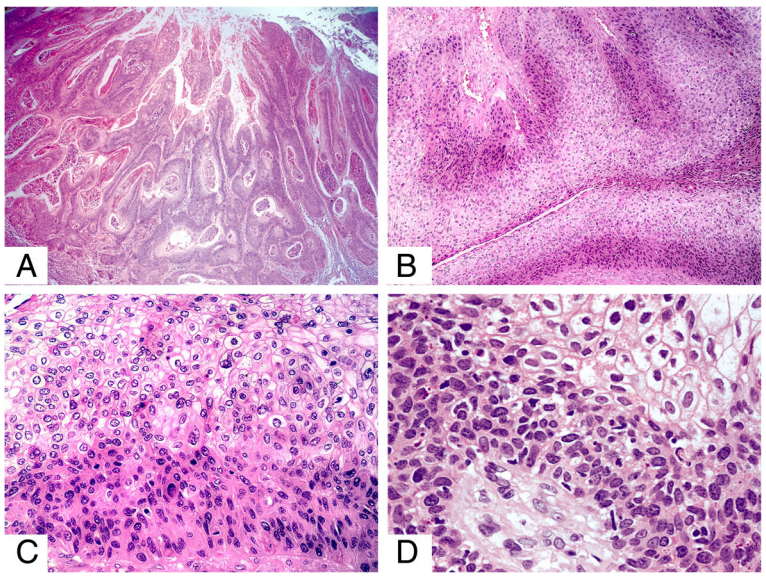

Fig. 3 Warty-basaloid squamous cell carcinoma. **a**- Exophytic warty carcinoma. **b**- Invasive nest composed of small basaloid cells at the periphery and central clear cells. **c**- High power view of **b**. Note the high grade cells in the base contrasting with clear cells with koilocytic changes in the upper half. **d**- Some papillae of **a** show the lower third of its lining composed of small basaloid cells with numerous mitoses and the upper third with koilocytes

20–40% of the neoplasms. Both patterns were in apposition to each other, very rarely mixing their features (Fig. 4). Tumor thickness varied from 6–20 mm. (average 12.9 mm). The majority of the cases invaded into the corpus spongiosum or dartos (6 cases) and only one into corpora cavernosa. Vascular and perineural invasion were documented in 3 and 2 cases, respectively. The majority of the tumor had a prognostic index score of 5. Highest prognostic index score of 7 was present in only 1 case. The subtypes of PeIN identified were variable: mixed differentiated-basaloid (4 cases), basaloid (2 cases), and differentiated (1 case).

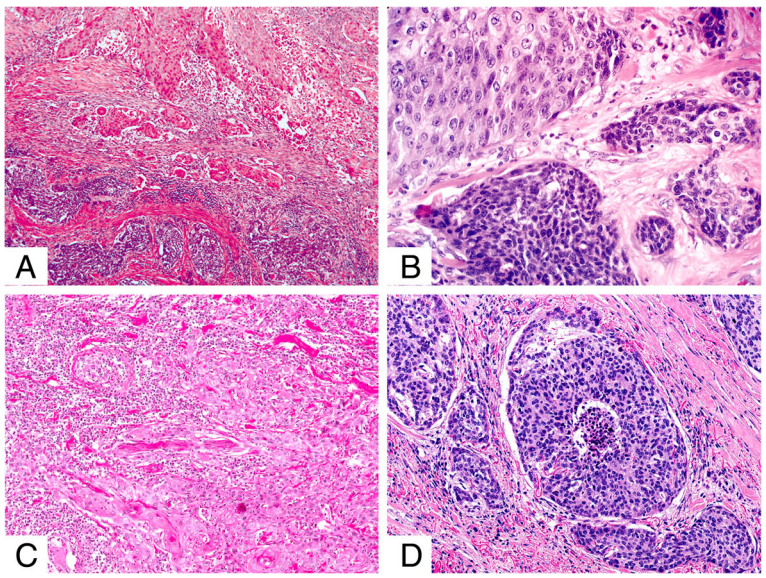

Fig. 4 Usual-basaloid squamous cell carcinoma. **a**- Superiorly, note areas of usual squamous cell carcinoma. The inferior part shows features of classic basaloid carcinoma. **b**- Nests of tumors composed by different cells types. In the upper left, intermediate grade neoplastic cells with ample cytoplasm. In the lower right, nests of small basaloid cells. **c**- Sheets of squamous cell carcinoma of usual type. **d**- Small nests of tumors with characteristic features of basaloid squamous cell carcinoma

Inguinal lymph node metastases were histologically confirmed in 3 patients. Metastases were bilateral in 2 cases and unilateral in 1. After follow up ranging from 36–72 months (average 49 months), 4 patients were alive and with no evidence of disease. One was alive with evidence of retroperitoneal metastasis detected 6 years after diagnosis of the primary tumor. One patient with massive bilateral and confluent nodal metastasis died from pulmonary metastasis 36 months after original surgical resection.

Comparison of classic, warty-basaloid, papillary basaloid and usual-basaloid squamous cell carcinomas

There were no statistically significant differences in the clinical features of patients with the four tumor types. Most tumors originated in the glans, but usual-basaloid carcinomas tended to affect the coronal sulcus or the foreskin more frequently than the other types.

Pathological features of each tumor type were distinctive and easy to separate using diagnostic criteria established for the corresponding tumor. There were some differences in the distribution of pathological prognostic factors (Table 1). Usual-basaloid carcinomas were smaller, thinner, affecting mostly corpus spongiosum and rarely invading corpora cavernosa and perineural spaces. On the contrary, most classic, warty-basaloid and papillary-basaloid carcinomas invaded deeper into corpora cavernosa and demonstrated higher rates of vascular and perineural invasion. Highest scores of the Prognostic Index were found in classic basaloids and lowest in usual-basaloid carcinomas (100% vs. 14%). Penile Intraepithelial Neoplasia was present in 25 patients (92.5%). One lesion subtype was identified in 14 cases (56%) and multiple subtypes in 11 (44%). A difference in the distribution of PeIN subtypes was that whereas classic basaloid carcinomas were mainly associated with basaloid PeIN, the other categories showed an heterogeneous mixtures of differentiated, warty and basaloid PeINs (Table 2).

High rate of nodal metastases was found in the classic, papillary basaloid and warty-basaloid tumor categories. Bilateral metastasis was more frequent in classic and papillary basaloid carcinomas and infrequent in usual-basaloid carcinomas. Massive and confluent bilateral metastases were typical of classic basaloid carcinoma patients at clinical presentation. Lowest rate of inguinal metastases was found in usual-basaloid carcinomas. Systemic metastases were more common in the classic and papillary variants (Table 3). According to the survival curves the majority of patients with classic and papillary basaloid neoplasms died from systemic metastases. Tumor related death was lower in warty-basaloid and usual-basaloid tumors, specially the latter (Fig. 5).

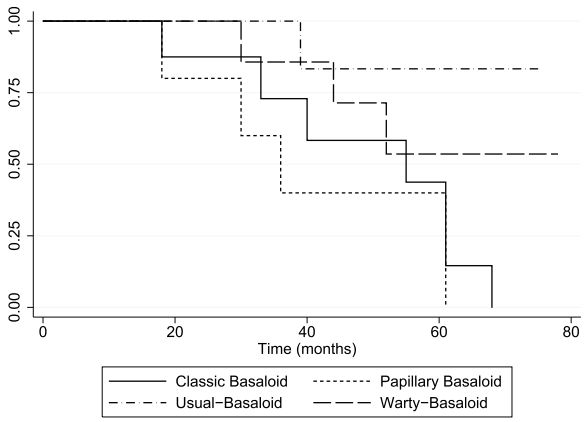

Fig. 5 Kaplan-Meier's survival curve stratified by basaloid carcinoma variants. Log-rank test was $p < 0.0393$

Discussion

Basaloid carcinoma of the penis is not a single entity but part of a heterogeneous spectrum of morphological phenotypes comprising different architectures and cell types. In this study we identified and described pathological features and outcome of basaloid tumors considered classic according to previous reports as well as of other subtypes of penile squamous cell carcinoma harboring in part basaloid features. Estimating the prognostic influence of the basaloid component in these tumors we found that patients with mixed usual SCC and basaloid SCC showed a better prognosis than those with classic, warty-basaloid, or papillary basaloid carcinomas. The later were aggressive tumors with frequent nodal metastasis and high tumor related mortality.

Classic basaloid carcinomas are morphologically distinctive with a uniform morphology but mixtures with the above mentioned categories may occur. In this and in larger series we have observed other tumor features mixed with basaloid carcinomas, such as pseudoglandular, sarcomatoid and adenosquamous, but they were very rare and we could not find sufficient number of cases to justify their inclusion in this report. Considering that the small "basaloid" cell was demonstrated to be the most important tissue marker for HPV in 2 recent studies [29, 30], it is important to identify and delineate clinical, morphological and outcome features of neoplasms containing these cells as part of their histopathological make up. In this study we are also demonstrating this cell to be an indicator of adverse prognosis as compared with the maturing keratining typical squamous cell.

In order to identify classic basaloid carcinomas we excluded tumors which may cause diagnostic confusion such as the small cell neurodocrine and the Merckel carcinomas and the solid variants of conventional squamous cell carcinomas. Neuroendocrine tumors are more organoid and do not usually grow in nests with central

comedo-like necrosis, characteristic of basaloid carcinomas. Keratinization is not a feature of endocrine neoplasms, although it can be rarely seen. In difficult cases immunostains may be necessary. Poorly differentiated squamous cell carcinomas with solid features may simulate basaloid tumors in their nesting pattern of growth but their cells are usually larger, keratinizing eosinophilic and more pleomorphic than the homogeneous small and basophilic cells of the typical basaloid carcinoma. Abrupt keratinization is not a feature of usual squamous cell carcinoma, which characteristically depicts a gradual maturation process, a hallmark for the distinction. In difficult cases, HPV detection and or positive staining with p16 would favor a diagnosis of basaloid carcinoma. Warty-basaloid carcinomas should be distinguished from typical warty or classic basaloid carcinomas. Distinction is based on the recognition of the presence of each tumor diagnostic morphological features in a proportion superior to 20%. The warty component is usually (but not always) present on the surface and the basaloid component is usually endophytic. The differential diagnosis of the papillary variant of basaloid carcinoma was with classic basaloid, urothelial high-grade papillary tumors and with warty-basaloid penile carcinomas. Classic basaloid carcinomas are composed of identical cells but they are not papillary. The separation with urothelial cancers may be problematic in perimeatal neoplasms with a predominant non invasive papillary architecture. Urothelial makers like GATA3 or uroplakin may be necessary as it was in some difficult cases in a previous study [15]. In the present series however, all cases were deeply invasive and the basaloid features of the tumors were easily recognized in the invasive component of the neoplasms. The main difference of papillary basaloid with warty-basaloid carcinoma is in the cell features: whereas the papillae in both tumors are “condylomatous”, that is they are spiky, leaf-like and undulating with prominent central fibro-vascular cores, in warty carcinomas the cells are larger, keratinizing and eosinophilic, with pleomorphic koilocytosis throughout the papillae. These features are not present in the papillary basaloid carcinomas that typically present a uniform small cell pattern with minimal superficial or focal koilocytosis. In the mixed usual-basaloid carcinoma category it was important to determine the boundaries between typical basaloid and usual squamous cell carcinoma. In the later, the tumors were more differentiated, with a gradual maturation and keratinization and the cells were eosinophilic. Although in cases there were transitional areas difficult to evaluate, the morphological contrast between the basaloid and the conventional SCC components was usually striking. We do not know whether the mixed usual-basaloid carcinomas originated as either pure basaloid or typical squamous cell carcinomas and dedifferentiated into the mixed variety. Evaluating the precancerous associated lesions we found that, as expected and reported in other studies [28], most classic basaloid and papillary basaloid carcinomas were associated with basaloid PeIN, and warty-basaloid carcinomas were associated with either warty, basaloid or differentiated PeIN. In the mixed usual-basaloid carcinoma category, associated precancerous lesions were also predominantly mixed differentiated (squamous)-basaloid validating the observation of a significant morphological correspondence of in situ and invasive lesions, even in the mixed tumor categories, as it was shown for the first time in this study. This finding may indicate that these tumors may be pre determined to be mixed from their origin and, as their better outcome appear to show, might represent different biological entities justifying their separation from the other basaloid carcinoma variants which are purely basaloid from the onset. Another difference may be related to the presence of HPV. The majority of classic basaloid carcinomas were reported to be HPV and p16 positive. Although we did not performed HPV studies in this series, in a previous evaluation of classic and mixed tumors among them the usual-basaloid carcinomas we found presence of HPV, using whole tissue sections PCR and p16, in 80% of the former and around in 50% of the later [17, 29].

In this comparative study of 4 morphological variants composed of basaloid cells we demonstrated that the classic and papillary basaloid carcinomas were equally lethal and significantly more aggressive than warty-basaloid carcinomas and especially of basaloid tumors mixed with the usual squamous cell carcinomas. It is known that classic basaloid carcinomas of the penis are associated with higher rates of regional nodal metastasis than other subtypes of penile SCCs except for the sarcomatoid variant [18]. Likewise, the tumors in this series were aggressive, with nodal metastasis present in all cases, usually bilaterally, and with 7 of 8 patients dying from widespread tumor dissemination. Comparing patients outcome in this study with those of 2 published series of American and Brazilian patients, Mexican patients with classic basaloid carcinomas were about 10 years older, their tumors were much larger, invaded deeper into corpora cavernosa and had a higher rate of vascular and perineural invasion. Basaloid carcinomas diagnosed earlier are smaller and thinner and showed better survival [18, 20]. The finding may be an indication of ethnic or cultural (late diagnosis) geographic differences among countries. The differences are more likely restricted to mortality and not to relative frequencies of histological subtypes since in other studies we found no geographical differences in the distribution of histological subtypes of penile SCCs, including the basaloid carcinomas [31, 32]. There is a recent study suggesting a higher frequency of HPV-related carcinomas, among them the basaloid

tumors, in African patients comparing to patients from other continents with the same tumor subtypes [19]. The difference may be due to the higher incidence of HIV infection in Africa and the known tendency for these immuno compromised patients to developed HPV-related genital cancers, including penile cancers [33, 34]. In the large series of 154 mexican patients from which these 27 cases were selected we found one patient with a history of HIV. He had a warty (condylomatous) carcinoma. None of the patients in this basaloid carcinoma series was infected with the HIV.

Clinical data were similar for the 4 tumor types. Prognostic pathological data and outcome of classic, warty-basaloid and papillary basaloid carcinoma patients were similar in accordance to their predominant cell type, but distinct from the usual-basaloid carcinomas. The later patients were younger and their tumor were smaller, more superficial, with lower rates of vascular and perineural invasion and a much higher survival. In addition, site of origin also appeared to differ. Whereas the vast majority of classic, warty-basaloid and papillary basaloid carcinomas originated in the glans, about half of usual-basaloid carcinomas affected the sulcus or foreskin. This finding is consistent with the hypothesis of the preference of HPV-related tumors for the "central" region of the penis, around the meatal area, and of the preference of keratinizing non-HPV-variants of SCCs for the "peripheral" regions (sulcus and foreskin) [35]. Collectively, these findings would indicate the biological and prognostic significance of identifying typical squamous cell features in association with those of classic basaloid carcinomas.

High score prognostic index correlated with patients' outcome in the 4 tumor variants. The lethal combination of adverse factors such as high grade, invasion of corpora cavernosa and perineural invasion [36], which comprises the pathological basis for the highest score category [26, 27], was present in 100, 60, 57 and 14% of the classic, papillary, warty-basaloid and usual-basaloid carcinomas respectively, revealing a significantly distinct behavior potential for the tumors. These figures correlated with the patterns of nodal metastasis. They were frequently bilateral and massive in the classic basaloid, as opposed to the mixed usual-basaloid carcinoma category, but also present in some of the papillary and warty-basaloid types as well. As expected, 6 of 7 patients with grossly massive confluent groin metastasis at clinical presentation in all categories died from disseminated cancer and 1 was alive at last follow up with incurable disease.

Conclusions

In summary, there was a variegated morphological spectrum of penile squamous cell carcinomas harboring at least 20% of basaloid pattern. In accordance with their predominant cell type, the classic, warty-basaloid and the papillary variants showed similar pathological features characterized by large, high grade and deeply invasive tumors resulting in a high rate of nodal metastasis and lethal outcome for most patients. To the contrary, patients with mixed usual-basaloid tumors were smaller, of lower grade, and thinner resulting in a lower incidence of nodal metastasis and a considerably better outcome. Basaloid carcinomas are not a single tumor entity but a spectrum of variable histological architecture and cell types. The identification of areas of typical maturing squamous carcinoma in a basaloid carcinoma is important to recognize and report because these patients may carry a better prognosis.

Additional file

Additional file 1: Spreadsheet with all patients' data codified and tabulated. (XLS 42 kb)

Abbreviations

CC: Corpora cavernosa; CS: Corpus spongiosum; HIV: Human immunodeficience virus; HPV: Human papillomavirus; LP: Lamina propria; PeIN: Penile intraepithelial neoplasia; SCC: Squamous cell carcinoma

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Availability of data and materials

Supporting data of this study are included supplementary information files (Additional file 1)

Authors' contributions

IAC: study conception, data acquisition, and manuscript writing. DFS: Data analysis, interpretation and manuscript writing. DP: Data acquisition and manuscript editing. ARG: Data acquisition and manuscript editing. IMR: Data interpretation and manuscript writing. MJFN: Data analysis and manuscript editing. NHT: Study design and manuscript writing. ALC: Study conception and design, data interpretation, and manuscript writing. All authors approved the final version of the manuscript.

Competing interests

The authors declare that they have no competing interests.

Consent for publication

Not applicable.

Ethics approval and consent to participate

This study was designed to deal with retrospective data and safeguarded the patients' health, well-being and rights. All patients had access to the best available diagnostic methods and treatments according to actual scientific evidence and were not involved in any experimental method. The dignity, privacy, and confidentiality of personal information were guarantied throughout the study by codification and deleting all personal data that could make identifiable individual people.
This study was evaluated and approved by ethics and scientific committee: Comité Científico, Hospital de Oncología, CMN SXXI, IMSS, number: IO-124-2012.

Author details
[1]Hospital de Oncología, Centro Médico Nacional (CMN), IMSS, México D.F., Mexico. [2]Instituto de Patología e Investigación, Martin Brizuela 325 casi Ayala Velazquez, Asunción, Paraguay. [3]Facultad de Ciencias Médicas, Universidad Nacional de Asunción, San Lorenzo, Paraguay. [4]Facultad Politécnica, Universidad Nacional de Asunción, San Lorenzo, Paraguay.

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