Int J Clin Exp Pathol 2013;6(9):1868-1873
www.ijcep.com /ISSN:1936-2625/IJCEP1307030

Original Article

Pigmented squamous intraepithelial neoplasia of the esophagus

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Received July 16, 2013; Accepted August 15, 2013; Epub August 15, 2013; Published September 1, 2013

Abstract: Squamous cell carcinoma (SCC) usually lacks melanocytes within the tumor. A few reports have documented invasive SCC or SCC *in situ* (intraepithelial neoplasia, IEN) with melanocytic hyperplasia within the tumor, referred to as pigmented SCC, in some organs. However, case series of pigmented SCC or IEN of the esophagus have not yet been reported. This is the first study to analyze the incidence and clinicopathological features of pigmented SCC or IEN of the esophagus. We reviewed 18 surgically-resected and 122 endoscopically-resected esophageal specimens, including 79 cases of IEN. Three cases of pigmented IEN were observed in this series, and all of them were located in the middle to lower third of the esophagus. Two of 3 cases had melanocytosis in the non-neoplastic squamous epithelium around the IEN. The incidence of pigmented IEN was 2.5% of all endoscopically resected specimens and 3.8% of IEN cases. No pigmented invasive SCC was detected in both endoscopically-resected and surgically-resected specimens. The mechanism of pigmentation of esophageal IEN is unknown. However, production of melanocyte chemotactic factors by tumor cells has been demonstrated in pigmented SCC of the oral mucosa. Moreover, two of 3 cases of pigmented IEN in the present series had melanocytosis in the non-neoplastic squamous epithelium, and melanocytosis is thought to be associated with chronic esophagitis, therefore, it has been hypothesized that various stimuli can cause pigmentation in squamous epithelium. Additional studies are needed to clarify the mechanism of pigmentation in squamous IEN of the esophagus.

Keywords: Intraepithelial squamous neoplasia, squamous cell carcinoma, esophagus, melanocytes

Introduction

Squamous cell carcinoma (SCC) usually lacks melanocytes within the tumor. A few reports have documented invasive SCC or SCC *in situ* (intraepithelial neoplasia, IEN) with melanocytic hyperplasia within the tumor, referred to as pigmented SCC, in some organs including the skin, oral mucosa, nasal cavity, and uterine cervix [1-4]. In the esophagus, only one case of SCC *in situ* with atypical melanocytic proliferation, which resembled malignant melanoma *in situ*, has been documented [5]. However, case series of pigmented SCC or IEN of the esophagus have not yet been reported. Herein, we were the first to analyze the incidence and histopathological characteristics of pigmented SCC or IEN of the esophagus.

Materials and methods

Patients

The esophageal squamous cell lesions reviewed in this study were collected from the surgical pathology files at our hospital between January 2009 and June 2013. Eighteen surgically-resected and 122 endoscopically-resected specimens (endoscopic mucosal resection or endoscopic submucosal dissection) were retrieved, and the histopathological features of these lesions were reviewed. Histopathological diagnosis was performed according to the World Health Organization Classification of squamous cell carcinoma of the esophagus [6].

Immunohistochemistry

Immunohistochemical analyses were performed using an Autostainer (Benchmark XT system, Ventana Medical System, Tucson, AZ, USA) by the same method as previously reported [7-9]. The following primary antibodies were used: a mouse monoclonal antibody against HMB-45 (HMB-45, Novocastra Laboratories, Ltd., Newcastle upon Tyne, UK), a mouse monoclonal antibody against Melan-A (A103, Novocastra), and a rabbit polyclonal antibody

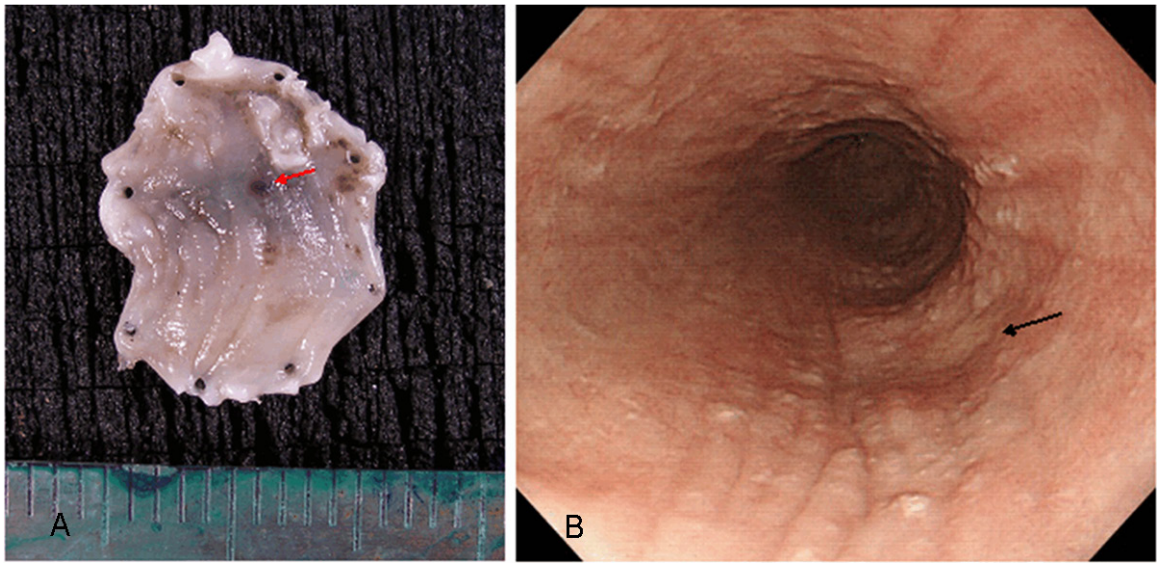

Figure 1. A: Endoscopic mucosal resection specimen of Case 1 showing a slightly depressed lesion with a black spot in the esophagus (arrow). B: Endoscopic findings of Case 2 showing a depressed lesion in the esophagus (arrow).

against S-100 protein (Nichirei Bioscience, Tokyo, Japan).

Results

Endoscopically-resected specimens

One hundred and twenty-two specimens from 102 patients including 79 cases of IEN, 42 cases of SCC invading into the lamina propria or muscularis mucosae (pT1a), and one case of SCC invading into the submucosa (pT1b) were examined. Ten patients had two lesions, 3 patients had 3 lesions, and one patient had 5 lesions.

Three cases of pigmented IEN were identified in this series, and the incidence of pigmented IEN was 2.5% of all endoscopically-resected specimens and 3.8% of IEN cases. No pigmented SCC was observed in pT1 cases. The clinicopathological features of pigmented IEN are described below.

Surgically-resected specimens

All 18 specimens were invasive SCC, and no pigmented SCC was observed.

Clinical features

Case 1: A 69-year-old Japanese male with a past history of lung cancer was found to have a slightly depressed reddish lesion, measuring 5 mm in diameter, in the esophagus 30 cm from incision (**Figure 1A**). A small black spot, measuring approximately 1 x 0.5 mm in diameter, was observed within the lesion (**Figure 1A**).

Case 2: A 70-year-old Japanese male was found to have a depressed reddish lesion, measuring 20 mm in diameter, in the esophagus 38 cm from incision (**Figure 1B**). No macroscopic black spot was noted.

Case 3: A 70-year-old Japanese male was found to have a slightly elevated lesion, measuring 35 x 20 mm, in the esophagus 23 cm from incision. No macroscopic black spot was noted.

Histopathological features

Case 1: Proliferation of atypical squamous cells with slightly enlarged round to oval nuclei was observed in the lower third of the squamous epithelium (**Figure 2A**). Cellular disorganization at the bottom of the squamous epithelium was noted. Dendritic melanocytes without atypia were observed in the lower third of the squamous epithelium (**Figure 2A**). No mitotic figures were present in the melanocytes. Moreover, melanocytic proliferation without atypia was also noted in the basal layer of the non-neoplastic squamous epithelium around the IEN (**Figure 2B**).

Accordingly, a diagnosis of pigmented low-grade IEN was made.

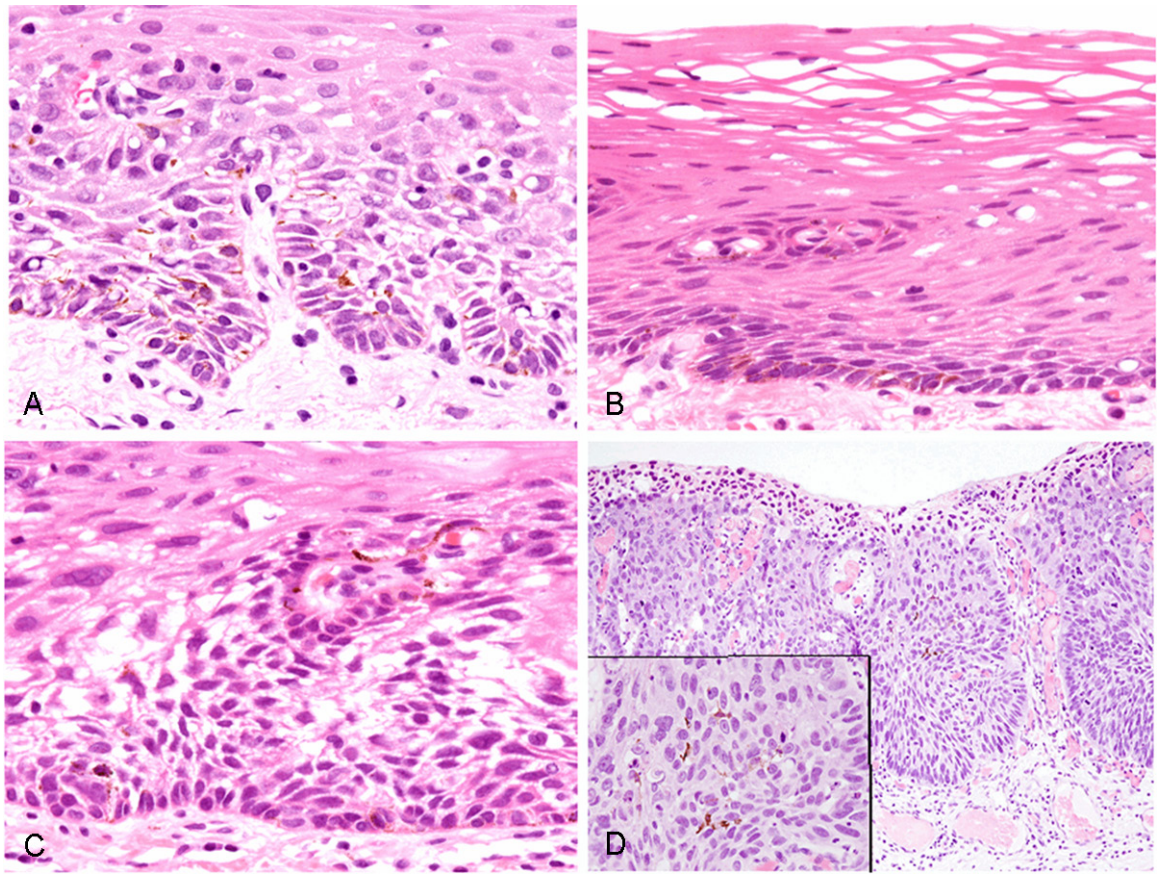

Figure 2. Histopathological findings. A: Case 1. Atypical squamous proliferation is observed in the lower third of the squamous epithelium accompanied by dendritic melanocytes without atypia. HE, x 400. B: Melanocytosis is also present in the non-neoplastic squamous mucosa. HE, x 400. C: Case 2. Atypical squamous proliferation is noted in the lower half of the squamous epithelium with proliferation of dendritic melanocytes without atypia. HE, x 400. D: Case 3. Atypical squamous cell proliferation is observed in the entire layer of the squamous mucosa accompanied by dendritic melanocytes. Dendritic melanocytes are without atypia, and melanin pigment is present within the cytoplasm of the atypical squamous cells (inset). HE, x 200, x 400 (inset).

Case 2: Proliferation of atypical squamous cells with large round to oval nuclei was observed in the lower half of the squamous epithelium accompanied by proliferation of dendritic melanocytes without atypia (**Figure 2C**). Melanin pigment was present within the cytoplasm of some neoplastic squamous cells (**Figure 2C**). Melanocytes were not present in the surrounding non-neoplastic squamous epithelium.

Accordingly, a diagnosis of pigmented high-grade IEN was made.

Case 3: Proliferation of atypical squamous cells with large round to oval nuclei with conspicuous nucleoli was observed in the entire layer of the squamous epithelium (**Figure 2D**). Dendritic melanocytes without atypia were present within the lesion, and melanin pigment was present within the cytoplasm of some neoplastic squamous cells (**Figure 2D**, inset). No invasive growth was noted. Melanocytes were also present in the surrounding non-neoplastic squamous epithelium.

Accordingly, a diagnosis of pigmented high-grade IEN (SCC *in situ*) was made.

Immunohistochemical features

The dendritic melanocytes within IEN were positive for S-100 protein, Melan-A, and HMB-45 (**Figure 3**). Moreover, the melanocytes within the non-neoplastic squamous epithelium in Case 1 and 3 were also positive for S-100 protein, Melan-A, and HMB-45 (**Figure 3**, inset).

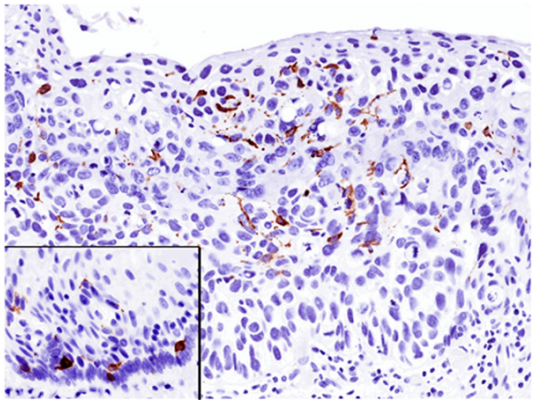

Figure 3. Immunohistochemical features of Case 3. Melan A is expressed in the dendritic melanocytes within the IEN. Melan-A-positive dendritic melanocytes are present in the non-neoplastic squamous epithelium (inset). x 200.

Discussion

The esophagus usually lacks melanocytes, however, malignant melanoma and melanocytosis are rare but well-recognized melanocytic lesions of the esophagus [10]. Esophageal melanocytosis is a rare condition, which is defined as an increase in number of melanocytes without atypia spreading along the basal layer of the squamous epithelium [10]. De la Pava *et al*. first described the presence of melanocytes in the squamous epithelium and lamina propria of the esophagus in 1963 [11]. In their series, 4% of the cases had melanocytes in the esophagus [11]. Furthermore, Ohashi *et al*. reported that 7.7% (5 of 65 cases) of autopsied normal esophageal specimens contained melanocytes [12]. Interestingly, the incidence of esophageal melanocytosis is higher in Japan than in Western countries. Immunohistochemically, these melanocytes in the esophagus are positive for S-100 protein, Melan-A, and HMB-45 [10].

Pigmented SCC is a rare variant of SCC, characterized histopathologically by the presence of non-neoplastic melanocytes within the lesion of SCC. This variant of SCC has been reported in various organs, such as skin, oral mucosa, nasal cavity and uterine cervix [1-4]. Most of the reported cases of pigmented SCC are invasive SCC, however, one case of pigmented IEN of the uterine cervix (cervical intraepithelial neoplasia) has been documented [3]. In the esophagus, Walter *et al*. described a case of multifocal SCC *in situ* adjacent to atypical melanocytic proliferation within the squamous epithelium [5]. These melanocytes had faintly eosinophilic cytoplasm containing focal melanin pigment and medium-sized hyperchromatic nuclei with occasional conspicuous nucleoli as single cells or in small clusters scattered close to the epithelial surface [5]. These features resembled malignant melanoma *in situ*, however, they concluded that these melanocytes were non-neoplastic in nature because the melanocytic lesion had disappeared by follow-up [5].

This report is the first to analyze the incidence and clinicopathological features of pigmented squamous IEN of the esophagus. Our analysis revealed that the incidence of pigmented squamous IEN of the esophagus was 3.8% of all squamous IEN of the esophagus. All patients were elderly males, and all lesions occurred in the middle to lower third of the esophagus. Moreover, no pigmented invasive SCC was present in the present series. All three cases of the present study had positivity for S-100 protein, Melan-A, and HMB-45 in the melanocytes without atypia within the IEN, and 2 of 3 cases had non-neoplastic melanocytes in the non-neoplastic squamous epithelium around the IEN. In the present three cases, melanocytes within the IEN had no nuclear atypia, therefore, a diagnosis of pigmented IEN was straightforward. However, a few cases of pigmented SCC that mimicked malignant melanoma have been documented [13, 14]. Moreover, pigmented SCC sometimes had melanin pigment within the cytoplasm of the neoplastic squamous cells [14], as also seen in 2 of 3 cases in the present series, which resulted in misdiagnosis as malignant melanoma, therefore, differentiation from malignant melanoma is very important for accurate diagnosis and treatment.

The origin and pathogenesis of melanocytosis of the esophagus are not completely understood, however, two hypotheses have been proposed. During embryogenesis, melanocytes arise in the neural crest and migrate to various sites, such as epidermis, uvea, choroid, and leptomeninges, and “aberrant” migration of melanocytes to the esophagus can occur [10, 11, 15]. An alternative possibility is that melanocytes are formed in the esophagus as a result of differentiation from totipotential cells in the basal layer of the squamous epithelium

by various stimuli [15] because melanocytosis of the esophagus is mostly located in the middle to lower third of the esophagus and is frequently associated with chronic esophagitis and reactive squamous hyperplasia [12, 15]. Some authors have hypothesized that gastrointestinal reflux disease can lead to esophageal melanocytosis [12, 15]. In addition, melanocytosis has been identified in 25 to 30% of surgically-resected specimens of malignant melanoma of the esophagus, and some authors have suggested that melanocytosis is a precursor lesion of malignant melanoma [12, 16, 17].

Furthermore, it has been demonstrated that squamous cell carcinoma of the oral mucosa can produce melanocyte chemotactic factors, such as stem cell factor and endothelin-1, resulting in melanocytic colonization within the tumor [18]. The mechanism of pigmentation in squamous IEN of the esophagus is unknown; therefore, additional studies are needed to clarify the precise mechanism of melanocytosis and pigmented IEN of the esophagus.

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References

[1] Morgan MB, Lima-Maribona J, Miller RA, Kilpatrick T, Tannenbaum M. Pigmented squamous cell carcinoma of the skin: morphologic and immunohistochemical study of five cases. J Cutan Pathol 2000; 27: 381-386.

[2] Mathews A, Abraham EK, Amman S, Nair MK. Pigmented squamous cell carcinoma of nasal cavity. Histopathology 1998; 33: 184-185.

[3] Ishida M, Kagotani A, Yoshida K, Okabe H. Pigmented cervical intraepithelial neoplasia. Int J Gynecol Pathol 2013; 32: 146-147.

[4] Mikami T, Furuya I, Kumagai A, Furuuchi H, Hoshi H, Iijima S, Sugiyama Y, Takeda Y. Pigmented squamous cell carcinoma of oral mucosa: clinicopathologic study of 3 cases. J Oral Maxillofac Surg 2012; 70: 1232-1239.

[5] Walter A, van Rees BP, Heijnen BH, van Lanschot JJ, Offerhaus GJ. Atypical melanocytic proliferation associated with squamous cell carcinoma in situ of the esophagus. Virchows Arch 2000; 437: 203-207.

[6] Montgomery E, Field JK, Boffetta P, Daigo Y, Shimizu M, Shimoda T. Squamous cell carcinoma of the oesophagus. In: Bosman FT, Carneiro F, Hruban RH, Theise ND, editors. World Health Organization Classification of Tumours of the Digestive System. Lyon: IARC Press, 2010; pp: 18-24.

[7] Ishida M, Mori T, Umeda T, Kawai Y, Kubota Y, Abe H, Iwai M, Yoshida K, Kagotani A, Tani T, Okabe H. Pleomorphic lobular carcinoma in a male breast: case report with review of the literature. Int J Clin Exp Pathol 2013; 6: 1441-1444.

[8] Ishida M, Iwai M, Yoshida K, Kagotani A, Okabe H. Sarcomatoid carcinoma with small cell carcinoma component of the urinary bladder: A case report with review of the literature. Int J Clin Exp Pathol 2013; 6: 1671-6.

[9] Nakanishi R, Ishida M, Hodohara K, Yoshida T, Yoshii M, Okuno H, Horinouchi A, Iwai M, Yoshida K, Kagotani A, Okabe H. Prominent gelatinous bone marrow transformation presenting prior to myelodysplastic syndrome: A case report with review of the literature. Int J Clin Exp Pathol 2013; 6: 1677-82.

[10] Chang F, Deere H. Esophageal melanocytosis. Morphologic features and review of the literature. Arch Pathol Lab Med 2006; 130: 552-557.

[11] De la Pava S, Nigogosyan G, Pickren JW, Cabrena A. Melanocytosis of the esophagus. Cancer 1963; 16: 48-50.

[12] Ohashi K, Kato Y, Kanno J, Kasuga T. Melanocytes and melanosis of the oesophagus in Japanese subjects-analysis of factors effecting their increase. Virchows Arch A Pathol Anat Histopathol 1990; 417: 137-143.

[13] Sante M, de Giorgi V, Massi D, Chiarugi A, Carli P. Pigmented Bowen's disease mimicking cutaneous melanoma: clinical and dermoscopic aspects. Dermatol Surg 2004; 30: 541-544.

[14] Shields JA, Shields CL, Eagle RC Jr, Singh AD, Demirci H, Wolf MA. Pigmented conjunctival squamous cell carcinoma simulating a conjunctival melanoma. Am J Ophthalmol 2001; 132: 104-106.

[15] Bogomoletz WV, Lecat M, Amoros F. Melanosis of the oesophagus in a Western patient. Histopathology 1997; 30: 498-499.

[16] Kanavaros P, Galian A, Periac P, Dyan S, Licht H, Lavergne A. Primary malignant melanoma of the esophagus arising in melanosis; histological, immunohistochemical and ultrastructural study of a case. Ann Pathol 1989; 9: 57-61.

[17] Piccone VA, Klopsrock R, LeVeen HR, Sika J. Primary malignant melanoma of the esophagus associated with melanosis of the entire esophagus. J Thorac Cardiovasc Surg 1970; 59: 864-870.

[18] Satomura K, Tokuyama R, Yamasaki Y, Yuasa T, Tatehara S, Ishimaru N, Hayashi Y, Nagayama M. Possible involvement of stem cell factor and endothelin-1 in the emergence of pigmented squamous cell carcinoma in oral mucosa. J Oral Pathol Med 2007; 36: 621-624.