Basaloid squamous cell carcinoma of esophagus expressing KIT: A case report with immunohistochemical analysis

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Abstract Basaloid squamous cell carcinoma of esophagus (BSCC-E) is rare. This case report is the first demonstrating KIT protein expression of BSCC-E. A 74-year-old man presented with dysphagia. Endoscopy revealed a polypoid tumor (2 × 2 × 2.5 cm) with a stalk in cervical esophagus. Biopsy showed squamous cell carcinoma with undifferentiated areas. An endoscopic submucosal dissection (ESD) was performed. Grossly, it was solid tumor with white cut surface. Histologically, the tumor was hypercellular carcinoma consisting of solid areas of island. The tumor cells were composed of basaloid malignant cells with hyperchromatic nuclei, scant cytoplasm, and basophilic cytoplasm. Many mitotic figures were recognized. Foci of comedonecrosis were scattered. Areas of squamous and glandular differentiations were scattered. Immunohistochemically, the tumor cells were positive for pancytokeratin (PCK) CAM5.2, PCK AE1/3, cytokeratin (CK) 7, KIT, CEA, CA19-9, EMA, Ki-67 (labeling index = 80%). The tumor cells were negative for CK20, PDGFRA, NSE, vimentin, estrogen receptor, p53 protein, chromogranin, synaptophysin, CD56, and TTF-1.

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1. Introduction

Basaloid squamous cell carcinoma (BSCC) of esophagus (BSCC-E) is a rare malignant neoplasm. It is morphologically characterized by basal cell carcinoma-like squamous cell carcinoma frequently showing glandular differentiation and comedonecrosis [1]. The cells of BSCC have hyperchromatic nuclei increased nucleo-cytoplasmic ratio, and basophilic cytoplasm, thus resembling basal cell carcinoma of the skin [1]. A review of English literature by PubMed search revealed about 50 case reports or case series of BSCC-E [2,3] and about 10 case reports or case series with immunohistochemical study [1,4,5]. However, there are no reports of the protein expression and gene mutational status of KIT (CD117) and platelet-derived growth factor-α (PDGFRA) in BSCC-E.

KIT and PDGFRA genes, both mapped to 4q12, encode receptor tyrosine kinase oncoproteins called KIT (CD117) and PDGFRA, respectively [6–11]. Both molecules are transmembranous oncoproteins involved in tumorigenesis of some neoplasms including gastrointestinal stromal tumor (GIST), acute myeloid leukemia, mast cell neoplasms, germ

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cell tumors, melanoma, neuroendocrine carcinomas (NEC), large cell neuroendocrine carcinoma (LCNEC), adenoid cystic carcinoma (ACC), small cell lung carcinoma (SCLC), and extra-pulmonary small cell carcinoma (SmCC) [12–27]. The hot spots of gene mutations are exons 9, 11, 13, and 17 of KIT gene and exons 12 and 18 of PDGFRA gene [6–11].

Herein, reported is a case of BSCC-E with protein expression of KIT but without protein expression of PDGFRA. A genetic analysis revealed no mutations of the KIT and PDGFRA genes.

2. Case report

A 74-year-old man presented with dysphagia and anemia. Blood test revealed anemia (red blood cells $282 \times 10^4/\mu l$, normal $450–550 \times 10^4/\mu l$), high creatinine (1.32 mg/dl, normal 0.4–1.2), high C-reactive protein (5.72 mg/dl, normal 0–0.3), and low Fe (17 μg/dl, normal 54–200). The serum tumor markers showed elevated SCC (2.0 ng/ml, normal 0–1.5). Serum CEA was within normal ranges. Upper gastrointestinal endoscopy revealed a polypoid tumor (2 × 2 × 2.5 cm) with a stalk in the cervical esophagus (Fig. 1). The biopsy showed squamous cell carcinoma with undifferentiated areas. Imaging modalities (CT, PET, PET-CT, and MRI) showed no tumors other than the esophageal tumor. Because the tumor was polypoid and the biopsy showed no apparent invasion, an endoscopic submucosal dissection (ESD) of the polypoid tumor was performed successfully.

Gross pathological examination showed that the tumor is solid tumor with white cut surface (Fig. 2). Histologically, the tumor is hypercellular carcinoma consisting of solid areas of island (Fig. 3A). The tumor was continuous with surface esophageal squamous epithelium which showed carcinomatous changes. The tumor cells were composed of basoloid malignant cells with hyperchromatic nuclei, scant cytoplasm, and basophilic cytoplasm (Fig. 3A, B, C and D). Many mitotic figures were recognized. Characteristically, foci of comedonecrosis were scattered (Fig. 3A and B). Areas of squamous differentiation (Fig. 3C) and glandular differentiation (Fig. 3D) were also scattered. The pathological diagnosis was BSCC of the esophagus. According to WHO blue book [1], it was typical BSCC-E.

An immunohistochemical analysis was performed by the Dako Envision method (Dako Corp, Glostrup, Denmark), as previously reported [28,29]. Immunohistochemically, the tumor cells are positive for pancytokeratin (PCK) CAM5.2 (Fig. 4A), PCK AE1/3, cytokeratin (CK) 7, KIT (Fig. 4B), CEA, CA19-9 (Fig. 4C), EMA, Ki-67 (labeling index = 80%) (Fig. 4D). The tumor cells were negative for CK20, PDGFRA, neuron-specific enolase (NSE), vimentin, estrogen receptor, p53 protein, chromogranin, synaptophysin, CD56, and TTF-1.

A molecular genetic analysis of KIT gene (exons 9, 11, 13, and 17) and PDGFRA (exons 12 and 18) gene was performed by the PCR direct sequencing method, as previously reported [12–27]. The exons of both genes were selected because they are frequent mutation sites [6–27]. In brief, genomic DNA was extracted from paraffin blocks with proteinase K digestion and phenol/chloroform extraction, and subjected to PCR for 40 cycles (94 °C for one minute, 52 °C for one minute, 72 °C for one minute), using a thermal cycler (GeneAmp PCR system 9700, Applied Biosystems, ABI, CA). The annealing temperature was 53 °C. PCR products were extracted, and subjected to a computed automatic DNA sequeencer (ABI PRIZM 3100 Genetic Analyzer, Applied Biosystems, ABI, CA).

The retrospective genetic analysis using PCR-direct sequencing method in paraffin sections identified no mutations of KIT (exons 9, 11, 13 and 17) and PDGFRA (exons 12 and 18) genes in the present tumor.

After the ESD, re-endoscopy was performed, and it did not reveal tumor. However, biopsy showed a few carcinoma...
cells; the lesion was burned. The third biopsy showed no residual tumor. Now, the patient is free from tumor by endoscopy and various imaging techniques 9 months after the first ESD, and is followed-up.

3. Discussion

The present case is the first of esophageal BSCC with examination of immunoreactive KIT (CD117) and...
PDGFRA proteins and with a genetic examination of KIT and PDGFRA genes. The present case showed that the tumor cells of esophageal BSCC expressed KIT protein but not PDGFRA protein. No mutations of KIT (exons 9, 11, 13, and 17) and PDGFRA (exons 12 and 18) genes were recognized. Although the genetic analysis is narrow, these seem new findings.

In the head and neck region, adenoid cystic carcinoma (ACC) is famous for positive KIT protein [6,8,9]. From the findings of the current case, it can be stressed that BSCC of esophagus is included into KIT-positive neoplasms in addition to ACC. In KIT-positive ACC, no mutations of KIT is seen [6,8,9], similar to the current case. To the best of author’s knowledge, there have been no studies of PDGFRA protein and gene mutational status in ACC. The present case showed that BSCC of the esophagus did not show PDGFRA protein expression and PDGFRA gene mutation.

In the esophagus, small cell carcinoma (SmCC) is known to express KIT and PDGFRA protein, but no mutations of KIT and PDGFRA genes are seen in esophageal small cell carcinoma [8,9]. Therefore, esophageal SmCC and BSCC-E are included into KIT-positive neoplasms of the esophagus, in addition to ACC.

Positive immunoreaction of KIT and PDGFRA protein does not necessarily imply gene mutations of KIT and PDGFRA. In general, KIT protein-positive tumors include GIST, acute myeloid leukemia, mast cell neoplasms, germ cell tumors, melanoma, neuroendocrine carcinomas (NEC), large cell neuroendocrine carcinoma (LCNEC), ACC, small cell lung carcinoma (SCLC), and extrapulmonary SmCC [12–27]. Of these neoplasms expressing KIT, KIT mutations are shown to be present in GIST [6–11,20,22,24,30], germ cell tumors [18,21], and malignant melanoma [19]. PDGFRA mutations are seen mainly in GIST [6–11,22]. In contrast, neuroendocrine carcinomas [13], large cell neuroendocrine carcinomas [11,12], ACC [6,8,9], SCLC [13,31,32], and extrapulmonary SmCC [12,14–20,24–27] are shown to express KIT and PDGFRA proteins, but no mutations are seen in these genes. Sihto et al. [31] insisted that KIT expression in SCLC is due not to KIT gene mutations but to KIT gene amplification. Other factors of KIT protein positivity and no KIT gene mutations include enhanced transcription, as distinct from increased tumor copy number, as well as mRNA isoform stability changes which can be encountered in various tumors. Such a status may be operative in KIT protein-positive but KIT gene mutation-negative neoplasms such as SCLC, extrapulmonary SmCC, ACC, LC, and LCNEC. In the present BSCC of the esophagus also, KIT expression may be due to KIT gene amplification or other factors.

Recently, the phosphorylation (activation) status of KIT and PDGFRA has been studies [30,33]. This is particularly important in KIT mutation-negative tumors as in the present case. KIT kinase activation and downstream signaling proteins leading to tumorigenesis have been studied, but little is known as yet. Protein kinase C-theta and PI3-kinase/AKT are activated in imatinib-resistant GIST [30], and analyses of these KIT signaling molecules may be important in the treatment of GIST. Such studies are not performed in SCC. In the present case, the author could not investigate these molecules, because no relevant antibodies were available. KIT tyrosine kinase activity and KIT signaling abnormalities in SCC remain to be studied.

In the current case, the first biopsy was pathologically diagnosed as squamous cell carcinoma with undifferentiated areas. Therefore, it seems that the biopsy diagnosis of BSCC of the esophagus is difficult. The present tumor showed characteristic gross appearance, i.e., polyp with a stalk. Fortunately, this feature made it easy for endoscopists to remove the tumor. The patient finally underwent complete resection of the esophageal BSCC. The patient is healthy without any tumors in the body.

The pathological diagnosis of the current tumor seems definite and typical, according to WHO [1]. The basaloïd malignant cells, focal squamous and glandular differentiation, many mitotic figures, characteristic basaloïd nature of tumor cells, and spotty comedonecrosis are all characteristic features of BSCC of esophagus [1]. The present case is not pure SCC because it showed broad areas of basaloïd features.

An immunohistochemical study was performed in the current case. The tumor cells are positive for PCK CAM5.2 and AE1/3, CK7, KIT, CEA, CA19-9, EMA, Ki-67 (labeling index = 80%). In contrast, the tumor cells were negative for CK20, PDGFRA, NSE, vimentin, estrogen receptor, p53 protein, chromogranin, synaptophysin, CD56, and TTF-1. The positive reaction of PCK and CK7 shows epithelial nature of the tumor. The positive reaction of CEA and CA19-9 may imply that BSCC has glandular phenotypes although they are thought to be irrelevant in immunohistochemically; especially CEA and CA19-9 were accentuated in the glandular differentiation areas of the current tumor. Positive EMA may imply epithelial nature. The high Ki-67 labeling (labeling index = 80%) may imply high cell proliferative activity and relatively malignant nature of the present tumor. The CK7+/CK20− pattern of the present tumor is compatible with primary BSCC of esophagus. The negative reaction of NSE, chromogranin, synaptophysin, and CD56 demonstrates that the present BSCC of the esophagus is not NEC or that the current tumor shows no neuroendocrine differentiation. The negative reaction of estrogen receptor and TTF-1 imply that the current tumor has no relationship to hormone producing tumors or lung adenocarcinoma. The negative vimentin may indicate that the tumor is not mesenchymal tumor. The present tumor was negative for p53 protein, suggesting no p53 gene mutations.
In summary, the author reported the first case of esophageal BSCC expressing KIT. PDGFRA was not expressed. A genetic analysis showed no mutations in the KIT and PDGFRA genes. An immunohistochemical study was performed.

The author has no conflict of interest.

This work was approved by ethics committee.

The work was performed by only the author and by only author’s money.

References


