[Resubmission manuscript-clean copy]

Metachronous multiple esophageal squamous cell carcinomas and Lugol-voiding lesions after endoscopic mucosal resection

Y. Urabe[1], T. Hiyama[2], S. Tanaka[3], S. Oka[3], M. Yoshihara[2], K. Arihiro[4], K. Chayama[1]

[1]Department of Medicine and Molecular Science, Division of Frontier Medical Science, Programs for Biomedical Research, Graduate School of Biomedical Sciences, Hiroshima University, Hiroshima, Japan
[2]Health Service Center, Hiroshima University, Hiashihiroshima, Japan
[3]Department of Endoscopy, Hiroshima University Hospital, Hiroshima, Japan
[4]Department of Anatomical Pathology, Hiroshima University Hospital, Hiroshima, Japan

Correspondence: Toru Hiyama, M.D., Ph.D., Health Service Center, Hiroshima University, 1-7-1 Kagamiyama, Higashihiroshima 739-8521, Japan
Phone: +81-82-424-6191
Fax: +81-82-422-7156
E-mail: tohiyama@hiroshima-u.ac.jp

Abstract

Background and Study Aims: Endoscopic mucosal resection (EMR) has been applied to the treatment of superficial esophageal squamous cell carcinoma (SCC). The incidence and characteristics of metachronous multiple esophageal SCCs and Lugpl-voiding lesions (LVLs) were investigated in a retrospective study in patients with superficial esophageal SCC after EMR treatment.

Patients and Methods: Ninety-six esophageal SCC patients treated by EMR who were followed up by endoscopy for 12 months or longer were included. Clinicopathologic parameters such as tumor size and location and presence of LVLs were examined.

Results: Twelve (13%) patients had synchronous multiple SCCs, and 12 (13%) developed metachronous multiple SCCs. The mean annual incidence of newly diagnosed tumor was 4.4%. The incidence of speckled pattern of LVLs was 20/74 (27%) in solitary SCC, 5/10 (50%) in synchronous multiple SCCs, and 10/12 (83%) in metachronous multiple SCCs. The incidence of presence of speckled pattern of LVLs was significantly higher in patients with multiple SCCs than in patients with solitary SCC (68% vs 27%, $P = 0.0004$).

Conclusions: Patients who have undergone EMR for esophageal SCC, especially those with metachronous multiple LVLs in the background mucosa, should undergo follow-up with close endoscopic observation with Lugol staining.

Keywords: multiple esophageal squamous cell carcinoma; Lugpl-voiding lesions; endoscopic mucosal resection; incidence; characteristics

Introduction

Esophageal cancer is the seventh most common cancer worldwide, and its incidence has increased rapidly over the past 3 decades [1]. Marked geographic variation exists in the incidence of this cancer, and regional differences exist in the prevalence of adenocarcinoma and squamous cell carcinoma (SCC). Although most tumors are adenocarcinomas in Western societies, SCCs constitute 80% of esophageal cancers in the world. This tumor type is especially prevalent in Asia, in countries such as China, India and Japan [2].

Chromoendoscopy with Lugol solution is highly sensitive for identifying dysplasia and superficial SCC [3,4]. According to the Lugol staining pattern, completely 'voiding' areas were found in approximately 90% of high-grade dysplasia and carcinoma, whereas approximately 90% of staining areas where were less intensely stained than normally stained epithelium were non-dysplastic lesions and the remaining 10% were dysplasia [5]. Therefore, Lugol-voiding lesions (LVLs) are detectable not only in dysplasia and carcinomas but also in non-dysplastic areas such as esophagitis.

Superficial esophageal SCC is defined as a lesion that is confined to the esophageal mucosa or submucosa, regardless of the presence or absence of regional lymph node metastases [6]. The rate of diagnosis of superficial esophageal SCC has increased due to improved diagnostic procedures such as endoscopic examination, including the chromoendoscopy with Lugol solution [7,8].

Recently, the minimally invasive technique of endoscopic mucosal resection (EMR) has been applied to the treatment of superficial esophageal SCC [9,10]. The disease can recur in the esophagus in two patterns: local recurrence and metachronous multiple SCCs. Because EMR spares a larger area of the esophageal mucosa, metachronous multiple SCCs may occur more frequently after EMR than after surgical treatment. One of the advantages of EMR is the high quality of life resulting from preservation of the entire esophagus; however, the preserved esophagus might still require surgical resection later if metachronous multiple SCCs develop. Incidence and characteristics of superficial metachronous esophageal SCCs and the association of LVLs with metachronous esophageal SCCs are still unclear, and analysis of these factors would be useful for deciding how best to follow the preserved whole esophagus following EMR. We therefore carried out a retrospective investigation to clarify the incidence and characteristics of metachronous multiple esophageal SCCs that developed after EMR treatment in patients with superficial esophageal SCC, and examined between presence

of LVLs and metachronous multiple esophageal SCCs.

Patients and methods

From December 1990 to March 2007, 140 esophageal SCC patients treated with EMR at Hiroshima University Hospital (Hiroshima, Japan) were enrolled in this study. Patients with lesions meeting all of the following criteria were considered suitable candidates for EMR at the institution: clinical findings of SCC confined within intraepitherial (EP) and lamina propria muscularis (LPM) without lymph node metastases and distant metastases. SCCs confined within EP and LPM are early stages, and are known to have no lymph node and distant metastases. EMR was performed using the strip biopsy method. The description of the SCCs and histologic evaluation of the resected specimens were in accordance with the Japanese Classification of Esophageal Carcinoma [11]. For tumor location, the esophagus was divided into 5 regions: the cervical esophagus (Ce), upper thoracic esophagus (Ut), middle thoracic esophagus (Mt), lower thoracic esophagus (Lt), and the abdominal esophagus (Ae). Periodic endoscopic examinations were recommended at least annually to look for recurrences of esophageal SCC.

Synchronous multiple esophageal SCCs were diagnosed in accordance with the criteria of Warren and Gates [12]. Lesions detected within 1 year of the initial EMR were regarded as synchronous multiple lesions because microcancers might have been missed at the time of EMR. Metachronous multiple lesions were defined as new esophageal SCCs in different areas from the initial cancer and occurring at least 1 year after the initial EMR. Patients who had both synchronous and metachronous SCC were assigned to the metachronous SCC group. For patients with synchronous and metachronous multiple lesions, data for the largest or initial lesion were analyzed.

The endoscopic examinations were carried out with the spraying of Lugol's solution by using catheter to detect recurrent lesions. Clinicopathologic characteristics, including patient age at diagnosis, sex, macroscopic type, location of lesion, tumor size, and presence of LVLs in background esophageal mucosae, were evaluated. Speckled pattern of LVLs were defined as the cases with much more than 10 small LVLs or numerous irregular-shaped multiform LVLs in the esophageal mucosae (Figure **1**) [13].

For statistical analysis, chi-square test, and Fisher’s exact probability test were used. $P < 0.05$ indicated statistical significance.

Results

Of the 140 superficial esophageal SCC patients who were initially treated with EMR, 96 were followed up by endoscopy for 12 months or longer and were included in this analysis. The characteristics of these 96 patients are listed in Table **1**. Median age at diagnosis was 64.5 years (range, 47-88 years), the median follow-up period by endoscopy was 62.7 months (range, 12-154 months), and the median follow-up number of endoscopic examination was 9.74 times (range, 2-31 times). None of the patients had local recurrence and distant metastasis, and died of esophageal SCC, and there were no treatment-related deaths. Six patients died of malignant diseases in other organs (2 lung cancers, one gastric cancer, one tongue cancer, one hepatic cancer, and one malignant lymphoma). Four patients died of pneumonia, 2 died of hepatic failure, 2 died of renal failure, and 3 died suddenly of an unknown cause. None of the patients had serious adverse reactions associated with Lugol staining.

Twelve (13%) of the 96 patients had synchronous multiple superficial esophageal SCCs (9 with double, 2 with triple and one with quadruple lesions) (Figure **2**). Two of these patients subsequently developed metachronous SCCs. In total, 12 (13%) patients developed metachronous multiple esophageal SCCs, and 2 of these patients subsequently developed a third SCC. The median interval between the diagnosis of the primary esophageal SCC and the diagnosis of the first metachronous multiple SCC was 27.3 months (range, 13-45 months). All of these patients had intramucosal SCCs. The mean annual incidence of metachronous cancer was 4.4% (Figure **3**).

The characteristics of the patients with solitary, synchronous multiple, and metachronous multiple SCCs are shown in Table **1**. There were no significant differences between solitary, synchronous, and metachronous SCCs in regard to patient sex, macroscopic type, tumor size. The age at diagnosis of initial SCC in multiple metachronous SCC patients tended to be younger than the age at diagnosis in solitary SCC and multiple synchronous SCC patients. The incidence of speckled pattern of LVLs was 20/74 (27%) in solitary SCC, 5/10 (50%) in synchronous multiple SCCs, and 10/12 (83%) in metachronous multiple SCCs. The incidence of the presence of speckled pattern of LVL was significantly higher in synchronous and metachronous multiple SCCs than in solitary SCC ($P = 0.0005$ by Fisher's exact probability test).

The pattern of primary and secondary lesions in multiple SCCs is shown in Table **2**. Interestingly, 36% of secondary lesions were located in the same segment of esophagus as the primary lesion.

Discussion

In the present study, we showed that metachronous multiple esophageal SCCs develop frequently in patients who have undergone EMR treatment for esophageal SCC and that careful endoscopic observation is required for these patients, especially with LVLs in the background esophageal mucosa.

An increased rate of metachronous multiple esophageal SCCs was expected in patients treated by EMR for esophageal SCC because the largest area of esophageal mucosa remains in these patients. Several researchers have reported that the rates of metachronous multiple esophageal SCCs after EMR for superficial esophageal SCC were 7.8%-20.0% [7,14-16]. In the present study, the mean annual incidence of metachronous cancer was 4.4%, which was comparable with previous reports. In addition, 20% of the patients in the present study developed metachronous lesions within 3 years after initial EMR. Therefore, careful annual follow-up endoscopic examinations for at least for 3 years after initial EMR may be needed for patients who have been treated with EMR for esophageal SCC.

In the present study, synchronous multiple esophageal SCCs were detected relatively frequently (13%), as were metachronous lesions. Kuwano et al. [17] detected synchronous multiple esophageal lesions in Japanese patients more frequently, in 11 of 43 (26%) patients. The difference in rates between Kuwano et al's study and the present study may reflect different populations. Because synchronous multiple esophageal lesions may be observed relatively frequently, other areas in the esophagus must be checked carefully so as not to miss the synchronous lesion(s).

To detect synchronous or metachronous esophageal SCCs, the Lugol spraying method has been recommended. Because squamous dysplasia and SCC lack glycogen, these lesions are revealed as LVLs. Researchers have reported the usefulness of this method to diagnosis esophageal SCC. For example, Shimizu et al. [18] examined 326 consecutive patients with head and neck cancer, patients who are at high risk of esophageal SCC, and reported that 45% of high-grade intraepithelial neoplasias were missed without Lugol staining. Dubuc et al. [19] examined the advantages of systemic Lugol staining in a total of 1095 patients in 62 endoscopy centers. Of the 35 cancers detected, 20% were detected only after Lugol staining. Lugol staining improves the diagnostic accuracy of endoscopy.

In addition to aiding in the diagnosis of esophageal SCC, use of the Lugol spraying

method can evaluate the risk states of developing SCC. Speckled pattern of LVLs is the endoscopic characteristics. It has been reported that metachronous multiple esophageal SCC developed in 38% of patients treated with EMR for esophageal SCC with speckled pattern of LVLs in the background mucosa and in only 9% of those without LVLs [13]. The cumulative proportion of metachronous multiple SCCs was significantly higher in patients with speckled pattern of LVLs than in those without LVLs. Presence of multiple LVLs was the independent risk factor for multiple cancers (relative risk, 67; 95% confidence interval (CI), 15-310) [20]. In the present study, the incidence of the presence of speckled pattern of LVLs was significantly higher in synchronous and metachronous multiple SCCs than in solitary SCCs. These results may truly imply that the existence of speckled pattern of LVLs may indicate that the esophageal SCC is at high risk of progressing to metachronous multiple SCCs.

Characteristics of synchronous and metachronous multiple superficial esophageal SCCs were not significantly different in the present study, and these lesions arose in an area of the esophagus similar to that of the primary lesion. It is well established that multiple cancers can occur in the esophagus. This phenomenon has been explained by the concept of "field cancerization," in which repeated exposure to carcinogens leads to an accumulation of genetic alterations, resulting ultimately in the development of multiple and independent cancers [21]. Multiple LVLs in the esophageal mucosa are significantly associated with heavy consumption of alcohol. Additionally, *p53* mutations and the pattern of DNA methylation in intraepithelial neoplasia (IEN) and SCC occurring in the same patients were examined [22]. The pattern of methylation in individual loci indicated a progressive accumulation of methylations from non-neoplastic epithelium to IEN to SCC in individual patients. The authors suggested a field of methylation may precede and has some causative relation with the development of IEN and SCC. However, they also found that the specific mutations in *p53* differed between IEN and SCC from the same patients. Mutations in *p53* may occur independently in multiple sites and may contribute to carcinogenesis as well as DNA methylation. This findings on the *p53* mutations may be contradictory to the concept of "field cancerization."

Risk factors of esophageal SCC such as genetic factors, including genetic polymorphisms, and environmental factors, such as tobacco smoking and alcohol consumption, have been reported to be important [23]. As for genetic polymorphisms, the *1 and *2 genotypes of *ALDH2*, a gene encoding an alcohol metabolic enzyme, were reported to increase the risk of esophageal SCC by meta-analysis (odds ratio (OR), 3.19; 95% CI, 1.86-5.47) [24]. The Val allele of *CYP1A1* (codon 462), a gene encoding a carcinogen metabolic enzyme, was also reported to increase the risk of esophageal

SCC by meta-analysis (OR, 1.37; 95% CI, 1.09-1.71 for *1*2 vs *1*1 and OR, 1.44; 95% CI, 1.17-1.78 for *2*2 vs *1*1) [25]. However, the influences of genetic polymorphisms may be relatively less than those of environmental factors such as tobacco smoking and alcohol consumption. For tobacco smoking, a dose-response trend has been reported. Relative risks of developing esophageal cancers are 1.4 for persons who smoke ≤ 30 cigarettes per day and 3.1 for persons who smoke > 30 cigarettes per day compared with non-smokers [26]. Alcohol consumption is also linked to increased risk of esophageal cancers. For persons who consume more than 4 drinks (= 47.5 g of pure ethanol) per day, the relative risk of developing such cancers is 4.2, compared with non-drinkers. A synergistic effect was observed in persons who both smoke tobacco and drink alcohol. The relative risks of developing the cancers are 12.7 among those who smoke > 30 cigarettes a day and consume > 4 drinks per day. In the present study, the age at diagnosis of initial SCC in multiple metachronous SCC patients tended to be younger than the age at diagnosis in solitary SCC and multiple synchronous SCC patients. The findings might represent an inherited genetic susceptibility to cancer and/or patients' habits of tobacco smoking and alcohol consumption.

To prevent death due to esophageal SCC, screening programs that use endoscopy or other modalities are crucial. As described previously, Lugol staining is useful to detect early and small lesions of the esophagus. However, Lugol staining often causes mucosal irritation leading to retrosternal discomfort [27]. Complications of Lugol staining, including acute esophageal and gastric mucosal damage, have also been reported [28]. Recently, the usefulness of a narrow-band imaging system with magnifying endoscopy has been reported [29-32]. This method requires no chemical agents and is without complications. It has been applied clinically to the detection of esophageal SCC and also head and neck cancers and may overtake Lugol staining in efficacy [33]. Further research is needed to clarify the significance of the narrow-band imaging system in screening programs.

In addition to screening programs, educational programs warning of the risks of tobacco smoking and alcohol consumption are crucial. People at high risk, such as heavy smokers and drinkers and those with a family history of upper aerodigestive tract cancer, have an 8-fold greater risk of developing a second cancer compared with those without a family history of any cancer [34]. Such at-risk people need to acquire this knowledge and participate in a screening program.

In conclusion, esophageal SCCs are often newly detected after initial EMR for esophageal SCC. Patients who have undergone EMR for esophageal SCC, especially those with multiple LVLs in the background mucosa, should be followed with close

endoscopic observation with Lugol staining. Our recommendation is that such patients undergo annual endoscopic examination.

References

1. Holmes RS, Vaughan TL. Epidemiology and pathogenesis of esophageal cancer. Semin Radiat Oncol 2007; 17: 2-9
2. Xing D, Tan W, Lin D. Genetic polymorphisms and susceptibility to esophageal cancer among Chinese population (review). Oncol Rep 2003; 10: 1615-1623
3. Meyer V, Burtin P, Bour B et al. Endoscopic detection of early esophageal cancer in a high-risk population: does Lugol staining improve videoendoscopy? Gastrointest Endosc 1997; 45: 480-484
4. Dawsey SM, Fleischer DE, Wang GQ et al. Mucosal iodine staining improves endoscopic visualization of squamous dysplasia and squamous cell carcinoma of the esophagus in Linxian, China. Cancer 1998; 83: 220-231
5. Mori M, Adachi Y, Matsushima T et al. Lugol staining pattern and histology of esophjageal lesions. Am J Gastroenterol 1993; 88: 701-705
6. Tachibana M, Kinugasa S, Shibakita M et al. Surgical treatment of superficial esophageal cancer. Langenbecks Arch Surg 2006; 391: 304-321
7. Kanamoto A, Yamaguchi H, Nakanishi Y et al. Clinicopathological study of multiple superficial oesophageal carcinoma. Br J Surg 2000; 87: 1712-1715
8. Katagiri A, Kaneko K, Konishi K et al. Lugol staining pattern in background epithelium of patients with esophageal squamous cell carcinoma. Hepatogastroenterology 2004; 51: 713-717
9. Yokoyama A, Muramatsu T, Ohmori T et al. Multiple primary esophageal and concurrent upper aerodigestive tract cancer and aldehyde dehydrogenase-2 genotype of Japanese alcoholics. Cancer 1996; 77: 1986-1990
10. Katada C, Muto M, Manabe T et al. Local recurrence of squamous-cell carcinoma of the esophagus after EMR. Gastrointest Endosc 2005; 61: 219-225
11. Japanese Society for Esophageal Disease. Guidelines for Clinical and Pathological studies on Carcinoma of the Esophagus, 10th edn. Tokyo: Kanehara; 2007
12. Warren S, Gates O. Multiple primary malignalnt tumors. A survey of the literature and a statistical study. Am J Cancer 1932; 16: 1358-1414
13. Muto M, Hirohata S, Nakane M et al. Association of multiple Lugol-voiding lesions with synchronous and metachronous esophageal squmous cell carcinoma in patients with head and neck cancer. Gastrointest Endosc 2002; 56:517-521
14. Shimizu Y, Tukagoshi H, Fujita M et al. Metachronous squamous cell carcinoma of the esophagus arising after endoscopic mucosal resection. Gastrointest Endosc 2001; 54: 190-194

15. Katada C, Muto M, Manabe T et al. Local recurrence of squamous-cell carcinoma of the esophagus after EMR. Gastrointest Endosc 2005; 61: 219-225
16. Muto M, Hitomi Y, Ohtsu A et al. Association of *aldehyde dehydrogenase 2* gene polymorphism with multiple oesophageal dysplasia in head and neck cancer patients. Gut 2000; 47: 256-61
17. Kuwano H, Ohno S, Matsuda H et al. Serial histologic evaluation of multiple primary squamous cell carcinomas of the esophagus. Cancer 1988; 61: 1635–1638.
18. Shimizu Y, Tukagoshi H, Fujita M et al. Endoscopic screening for early esophageal cancer by iodine staining in patients with other current or prior primary cancers. Gastrointest Endosc 2001; 53: 1–5.
19. Dubuc J, Legoux JL, Winnock M et al. Endoscopic screening for esophageal squamous-cell carcinoma in high-risk patients: a prospective study conducted in 62 French endoscopy centers. Endoscopy 2006; 38: 690-695.
20. Muto M, Takahashi M, Ohtsu A et al. Risk of multiple squamous cell carcinomas both in the esophagus and the head and neck region. Carcinogenesis 2005; 26: 1008-1012
21. Slaughter DP, Southwick HW, Smejkal W. Field cancerization in oral stratified squamous epithelium; clinical implications of multicentric origin. Cancer 1998; 82; 1454-1459
22. Ishii T, Murakami J, Natohara K et al. Oesophageal squamous cell carcinoma may develop within a background of accumulating DNA methylation in normal and dysplastic mucosa. Gut 2007; 56: 13-19
23. Hiyama T, Yoshihara M, Tanaka S et al. Genetic polymorphisms and esophageal cancer risk. Int J Cancer 2007; 121: 1643-1658
24. Lewis SJ, Smith GD. Alcohol, ALDH2, and esophageal cancer: a meta-analysis which illustrates the potentials and limitations of a Mendelian randomization approach. Cancer Epidemiol Biomarkers Prev 2005; 14: 1967-1971
25. Yang CX, Matsuo K, Wang ZM et al. Phase I/II enzyme gene polymorphisms and esophageal cancer risk: a meta-analysis of the literature. World J Gastroenterol 2005; 11: 2531-2538
26. Zeka A, Gore R, Kriebel D. Effects of alcohol and tobacco on aerodigestive cancer risks: a meta-regression analysis. Cancer Causes Control 2003; 14: 897-906
27. Kondo H, Fukuda H, Ono H, Gotoda T, Saito D, Takahiro K, et al. Sodium thiosulfate solution spray for relief of irritation caused by Lugol's stain in chromoendoscopy. Gastrointest Endosc 2001;53:199-202.
28. Park JM, Lee IS, Kang JY et al. Acute esophageal and gastric injury: complication

of Lugol's solution. Scand J Gastroentrol 2007; 42: 135-137

29. Inoue H, Sasajima K, Kaga M et al. Endoscopic in vivo evaluation of tissue atypia in the esophagus using a newly designed integrated endocytoscope: a pilot trial. Endoscopy 2006; 38: 891-895
30. Yoshida T, Inoue H, Usui S et al. Narrow-band imaging system with magnifying endoscopy for superficial esophageal lesions. Gastrointest Endosc 2004; 59: 288-295
31. Hirata M, Tanaka S, Oka S et al. Magnifying endoscopy with narrow band imaging for diagnosis of colorectal tumors. Gastrointest Endosc 2007; 65:988-995
32. Hirata M, Tanaka S, Oka S et al. Evaluation of microvessels in colorectal tumors by narrow band imaging magnification. Gastrointest Endosc 2007 ; 66: 945-952
33. Katada C, Nakayama M, Tanabe S et al. Narrow band imaging for detecting superficial oral squamous cell carcinoma: a report of two case. Laryngoscope 2007; 117: 1596-1599
34. Morita M, Kuwano H, Ohno S et al. Multiple occurrence of carcinoma in the upper aerodigestive tract associated with esophageal cancer: reference to smoking, drinking and family history. Int J Cancer 1994; 58: 207-210

Legends

Table **1** Characteristics of patients with early esophageal cancer and their initial lesions

Table **2** Clinicopathologic similarities of multiple esophageal Squamous cell carcinomas.

Figure **1** Representative examples of speckled pattern of Lugol-voiling lesions (LVLs). A; much more than 10 small LVLs. B; numerous irregular-shaped multiform LVLs.

Figure **2** Endoscopic images from a representative patient with synchronous multiple superficial esophageal SCCs. A, B, C: Endoscopic findings before Lugol staining. D, E, F: Endoscopic findings after Lugol staining. G, H, I: Schematic images.

Figure **3** Incidences of esophageal SCC development in esophageal SCC patients treated by EMR and in controls during the observation period.

Table 1

	Solitary (n = 74)	Synchronous (n = 10)	*P*-value*	Metachronous (n = 12)	*P*-value*
Age at diagnosis (yrs)			0.93		0.21
Median	64.7	65.9		61.5	
Range	47-88	50-78		50-75	
Sex			0.10		0.07
Male	58	10		12	
Female	16	0		0	
Macroscopic type			0.60		0.58
0-IIa or 0-I	2	0		0	
0-IIc	72	10		11	
Location of lesion					
Cervical esophagus (Ce)	1	0		1	
Upper thoracic esophagus (Ut)	7	0		1	
Middle thoracic esophagus (Mt)	49	5		8	
Lower thoracic esophagus (Lt)	15	4		2	
Abdominal esophagus (Ae)	2	1		0	
Tumor size (mm)					
-10	24	3		6	
11-20	25	1		3	
21-50	19	5		2	
51-	6	1		1	
Endoscopic follow-up period (months)					
Median	44	42.9	0.21	67.4	0.59
Range	12-154	12-99		14-110	
Presence of speckled pattern of LVLs	20 (27%)	5 (50%)	0.14	10 (83%)	0.0001

Two patients had both synchronous and metachronous cancers. In this analysis, they are grouped with the metachronous cancer subgroup. For patients with synchronous multiple lesions, data for the initial or the largest lesion were analyzed. LVLs, Lugol-voiding lesions.

**P-value* was calculated compared to solitary.

Table **2**

Primary lesion→ Secondary lesion	Synchronous (10 lesions)	Metachronous (12 lesions)	Total (22 lesions)
Location			
Ce→Ce/Ut/Mt/Lt/Ae	0/0/0/0/0	0/0/0/1/0	0/0/0/1/0
Ut→Ce/Ut/Mt/Lt/Ae	0/0/0/0/0	0/0/1/0/0	0/0/1/0/0
Mt→Ce/Ut/Mt/Lt/Ae	0/0/1/3/0	0/0/3/4/1	0/0/4/7/1
Lt→Ce/Ut/Mt/Lt/Ae	0/0/3/2/0	0/0/0/1/1	0/0/3/3/1
Ae→Ce/Ut/Mt/Lt/Ae	0/0/0/0/1	0/0/0/0/0	0/0/0/0/1
Same location	5 (40%)	4 (33%)	8 (36%)
Tumor Size			
-10→-10/11-20/21-50/51-	3/0/0/0	1/3/2/0	4/3/2/0
11-20→-10/11-20/21-50/51-	1/0/0/0	0/3/0/0	1/3/0/0
21-50→-10/11-20/21-50/51-	2/2/1/0	0/0/2/0	2/2/3/0
51-→-10/11-20/21-50/51-	0/1/0/0	0/0/1/0	0/1/1/0

Figure 1

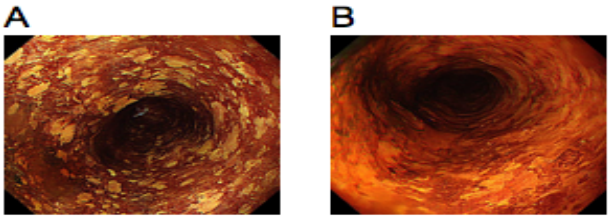


Figure 2

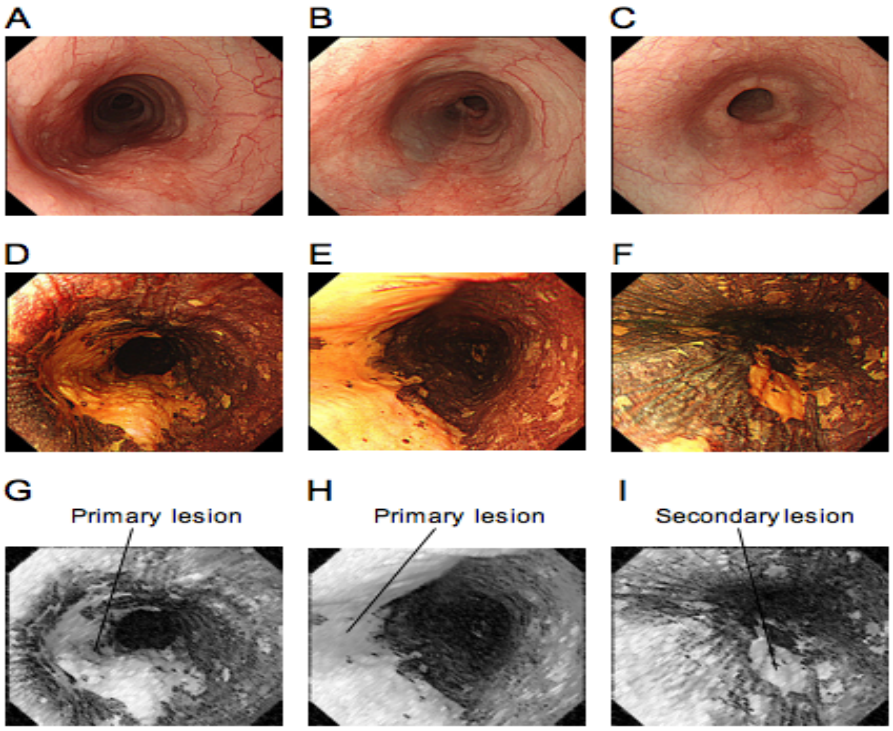
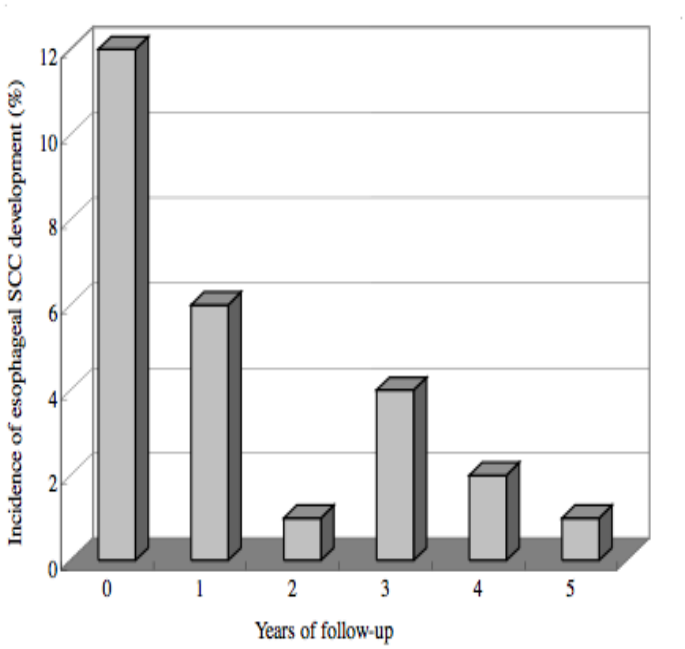

A
B
C
D
E
F
G
Primary lesion
H
Primary lesion
I
Secondary lesion

Figure 3



Number of esophageal SCC patients	96	96	80	48	32	25
Number of SCCs developed	12	6	1	4	2	1
(%)	12.5	6.3	1.3	8.3	6.3	4.0