

correlated with the histopathologic findings (by a histopathologist and an endoscopist independently, and in phase two by a histopathologist and an endoscopist together). The aim of the pilot study was to obtain basic microscopic images of a healthy esophagus, esophagitis, BE and EAC.

## Results

A total of 14 patients were enrolled in the study and underwent an upper endoscopy with pCLE, of which 10 patients were male and 4 were female. The average age was 45.8 years and the mean BMI was 26.2. The following diagnoses were endoscopically identified and later confirmed by histopathology results: 3 patients with reflux esophagitis, 4 with BE (3 patients with intestinal metaplasia (IM), 1 patient with LGD), 3 patients with EAC and 4 persons were included in the healthy cohort. Half of the patients were current users of proton pump inhibitors, 3 patients were smokers, 3 patients were ex-smokers, and in 4 patients hiatal hernia was present among the endoscopic findings. The baseline characteristics of the patients can be seen in table 1.

The upper endoscopy examination was followed by pCLE. No adverse effects were observed after the application of the contrast agent. Videos were recorded during all of the pCLE procedures. Every video record was evaluated during the endoscopy, and then re-evaluated with a final histopathology result according to each diagnosis of reflux esophagitis, BE (IM, LGD), EAC, and also in the healthy cohort.

In a healthy esophagus we observed these pCLE images: normal squamous epithelium which appears as typical scale-like cells (fig. 3a), without the presence of inflammatory cells, sporadically intrapapillary capillary loops were also observed (fig. 3 b).

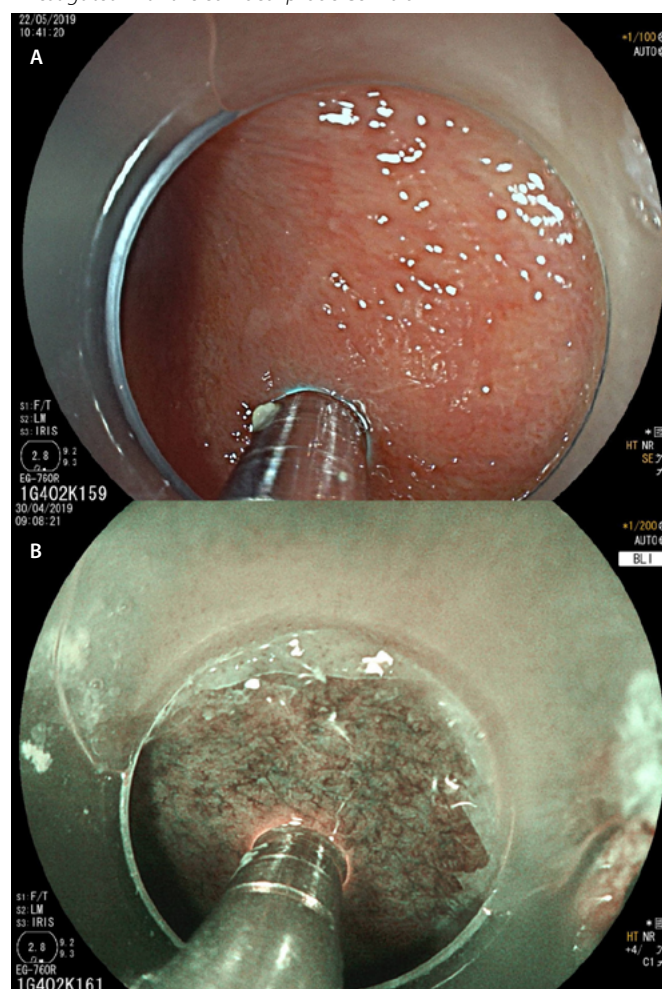
In patients with reflux esophagitis we saw columnar cells, hyperemia and inflammatory cells in the area of the gastroesophageal junction (fig. 4a), and in the squamous epithelium, stromal papillae with hyperemia in the distal esophagus (fig. 4 b).

The characteristic pCLE figures which we saw (and recorded) during the procedure in nondysplastic BE were: columnar cells and uniform villiform architecture and with dark goblet cells (fig. 5a, 5 b). In the patient with both BE and LGD we observed: dark non-round glands, lack of goblet cells, a sharp cutoff of darkness and variable cell size (fig. 6a, 6 b). No patient with HGD participated in our pilot study. In patients with EAC the following signs were seen: disorganization or a loss of structure, dark columnar cells with severe nuclear atypia and dilated irregular vessels (fig. 7a, 7 b).

The comparison of the pCLE images and the final histopathology figures from the biopsies taken can be seen in Fig. 3c–7c.

The correlations between the pCLE images captured by endoscopists and the definitive histopathology results are summarized in table 2. The correct diagnoses based on real-time pCLE were evaluated by an endoscopist in 11 of the 14 cases (78.6 %). The average time of pCLE examination needed to obtain a diagnosis based on pCLE images was 8 minutes.

**Fig. 2.** Endoscopic view in high-definition white-light endoscopy (HD-WLE) (a) and blue light imaging (BLI) (b) of nondysplastic Barrett's esophagus investigated with the confocal probe Cellvizio®



**Tab. 2.** Correlation between pCLE images and definitive histopathology samples

Variable	Total	Healthy cohort (n = 4)	Reflux esophagitis (n = 3)	Barrett's esophagus (n = 4)	Esophageal adenocarcinoma (n = 3)
<b>Histology</b>	14	Esophageal squamous epithelium: 4	Reflux esophagitis (grade 1): 2, (grade 2): 1	IM:3, LGD:1	Esophageal adenocarcinoma: 3
<b>Correct pCLE established diagnoses/definitive histopathology diagnoses</b>	11/14	4/4	2/3	3/4	2/3
<b>Average time of pCLE procedure needed to establish diagnoses based on pCLE images (minutes)</b>	8.0	6.0	7.5	9.6	9.0

pCLE – probe-based confocal laser endomicroscopy, IM – intestinal metaplasia, LGD – low grade dysplasia