A lower dose of fluorescein sodium is more suitable for confocal laser endomicroscopy: a feasibility study

Ya-Lin Zhang, MD,[1,2] Liang Bai,[3] Zhen Li, MD, PhD,[1] Rui Ji, MD, PhD,[1] Chang-Qing Li, MD, PhD,[1] Xiu-Li Zuo, MD, PhD,[1] Ya-Fei Yin,[1] Jia-Xin Du,[1] Zhen-Zhen Zhai, MD,[4] Xiao-Zhong Gao,[2] Yan-Qing Li, MD, PhD[1]

Jinan, Shandong, Weihai, Dezhou, China

Background and Aims: Image quality can be guaranteed with the conventional dosage of fluorescein sodium in probe-based confocal laser endomicroscopy (pCLE). However, yellow discoloration of the skin seriously affects daily life and simultaneously increases the risk of adverse events such as allergic reactions. The aim of this study was to test whether a lower dosage of fluorescein sodium can provide satisfactory image quality and to compare the diagnostic accuracy of gastric intestinal metaplasia (GIM) through a randomized blind controlled trial.

Methods: Consecutive patients were randomly assigned to different doses of fluorescein sodium. Image quality was determined by the endoscopists' subjective assessments and signal-to-noise ratio (SNR) assessment systems. Skin discoloration was tested using a neonatal transcutaneous jaundice detector. In addition, consecutive patients with a known or suspected diagnosis of GIM were examined by pCLE with the lower dose and the traditional dose.

Results: Only 0.01 mL/kg dose of 10% fluorescein sodium led to a significant decrease in image quality ($P < .05$), and a dose of 0.02 mL/kg had the highest SNR value ($P < .05$). There were no significant differences in skin discoloration between the 0.01 mL/kg and 0.02 mL/kg doses ($P = .148$) and no statistical difference in the diagnostic accuracy of pCLE for GIM between the 0.02 mL/kg and 0.10 mL/kg doses ($P > .05$). The kappa values for the correlation between pCLE and histopathology were 0.867 (95% confidence interval, 0.782–0.952) and 0.891 (95% confidence interval, 0.811–0.971).

Conclusions: The 0.02 mL/kg dose of 10% fluorescein sodium seems to be the best dose for pCLE in the upper GI tract, with comparable image quality with the conventional dose and insignificant skin discoloration. This dose is also very efficient for the diagnosis of GIM. (Gastrointest Endosc 2016;84:917-23.)

INTRODUCTION

Probe-based confocal laser endomicroscopy (pCLE) is an endoscopic technique for obtaining in vivo real-time images of the GI tract.[1-4] It facilitates the recognition of macroscopically invisible histology of the GI tract, such as cellular and subcellular structures, capillaries, and red blood cells,[5] and can even capture video images of the blood flow.[6] This noninvasive technique has the potential to realize an in vivo virtual biopsy,[7] provide precise real-time guidance for endoscopic therapy, and reduce the need for random blind biopsy.[8] Contrast agents with reflection properties such as fluorescein sodium, acriflavine, and cresyl violet are needed.[9] Acriflavine and

Abbreviations: CI, confidence interval; CLE, confocal laser endomicroscopy; GIM, gastric intestinal metaplasia; pCLE, probe-based confocal laser endomicroscopy; SNR, signal-to-noise ratio; TB, total bilirubin.

DISCLOSURE: *All authors disclosed no financial relationships relevant to this publication.*

This study was funded by the National Natural Scientific Funding of China (NSFC81330012).

0016-5107/$36.00
http://dx.doi.org/10.1016/j.gie.2016.05.011

Received December 9, 2015. Accepted May 4, 2016.

Current affiliations: Department of Gastroenterology, Laboratory of Translational Gastroenterology, Qilu Hospital, Shandong University, Jinan, Shandong (1), Department of Gastroenterology, Weihai Municipal Hospital, Weihai, Shandong (2), Department of Hepatobiliary Surgery, Weihai Municipal Hospital, Weihai, Shandong (3), Department of Gastroenterology, Dezhou People's Hospital, Dezhou, Shandong, China (4).

Reprint requests: Yan-Qing Li, Department of Gastroenterology, Qilu Hospital, Shandong University, 107 Wenhuaxi Road, Jinan 250012, China.

If you would like to chat with an author of this article, you may contact Dr Yan-Qing Li at liyanqing@sdu.edu.cn.

cresyl violet are specific nuclei staining agents and are only administered topically on the surface of the mucosa. They are used for the diagnosis of dysplasia, which requires assessment of the nuclei.[10,11] Fluorescein sodium is administered intravenously to show tissue, cellular, and microvessel structures.[12] Due to its ease of use, excellent fluorescent properties, nonmutagenic activity, low cost, and rare adverse events, fluorescein sodium has been safely used for several decades in ophthalmology[15] and is now the most applied contrast agent used in pCLE.[13,14] Furthermore, it is also a small molecule (molecular weight 376) and has been widely used as a hydrophilic marker molecule for paracellular permeability studies.[16-19] Fluorescein sodium is predominantly excreted by the kidneys and its plasma half-life is 24 hours.[6,20] The conventional intravenous dose is in the range of 5 to 10 mL of 10% fluorescein sodium in the United States and other countries around the world.[8,21,22] This dose of fluorescein sodium is almost completely excreted within 48 hours after intravenous administration. Although the image quality can be guaranteed using this dose range of fluorescein sodium,[23] the color of the skin and urine becomes obviously yellow after fluorescein sodium injection,[8] which can seriously affect daily life and work. In our pilot trials, we found that a lower dose of fluorescein sodium can also produce clear images in many cases; for example, 1 or 2 mL of 10% fluorescein sodium can produce clear images. Thus, the aim of this study was to optimize the appropriate dose of fluorescein sodium that can provide a high-quality image to ensure a satisfactory endoscopy examination, while avoiding yellow discoloration of the skin, through a randomized blind controlled trial. pCLE is an efficient and potentially important method for the diagnosis of gastric intestinal metaplasia (GIM) in vivo.[24,25] We also compared the accuracy of the optimized dose and the conventional dose of fluorescein sodium for the diagnosis of GIM.

METHODS

Derivative study

Patients. Patients who underwent upper GI endoscopy from August to September 2015 were enrolled in this study. Patients with normal mucosa and minor gastritis aged from 18 to 80 years were included in the study. Minor gastritis is defined as endoscopic edematous mucosa without erosion or bleeding. According to our experience, minor gastritis does not seriously affect the image quality compared with erosion or bleeding, which do compromise the image quality by inducing excessive leakage of fluorescein sodium. Exclusion criteria were as follows: patients with serious coagulopathy dysfunction, severe cardiopulmonary disease, bronchial asthma, liver or kidney dysfunction, allergy to fluorescein sodium, and pregnant or breastfeeding women. Those with a peptic ulcer, gastric cancer, a remnant stomach, or acute serious gastritis were also excluded. Participants were randomly divided into 4 groups based on the dose of fluorescein sodium according to computer-generated randomized numbers. All participants were informed about the aim of this study and gave their written informed consent.

Fluorescein sodium. All participants were given 1 mL of 1% fluorescein sodium intravenously (Baiyunshan Co., Guangzhou, China) to test for allergy 20 minutes before the endoscopy. Patients with allergic reactions were excluded. The candidate doses of fluorescein sodium were as follows: standard dose, 0.10 mL/kg and 3 lower dosages (0.05 mL/kg, 0.02 mL/kg, and 0.01 mL/kg). Once the gastric antrum was clearly visible, the assistant (Y.-L.Z.) promptly administered fluorescein sodium intravenously in accordance with the patient's randomized group, which was blind to the endoscopist.

Endoscopic procedure. Endoscopists with extensive experience (T.-Y., X.-M.G.) were responsible for the procedures. Both endoscopists were blinded to the patient group. Preparation for pCLE was the same as for conventional GI endoscopy. Participants fasted for at least 8 hours and were given chymotrypsin orally to prevent interference from mucus and bubbles in the stomach before the examination. Scopolamine (10 mg) was administered conventionally to reduce gastric peristalsis except for those patients with urinary retention, glaucoma, or other contraindications. All patients underwent general anesthesia with propofol (Fresenius Kabi Austria GmbH, Graz, Austria) and electrocardiographic monitoring. All procedures were performed by standard white-light endoscopy (Pentax i10, Tokyo, Japan). If a biopsy was necessary, the patient was excluded from this study.

pCLE. The probe (Cellvizio Gastro Flex UHD; Mauna Kea Technologies, Paris, France) was inserted through the biopsy channel of the endoscope and gently positioned in direct contact with the mucosa. The greater curvature of the gastric antrum in a range of 3 to 4 cm to the pylorus was selected as the best observation spot for its good stability and convenience for touching the gastric mucosa with the pCLE. The pCLE has a 2.5-mm diameter flexible probe with a confocal view of 240 μm, a scanning speed of 12 frames/s, and a scanning depth of 60 μm, producing a real-time video. After injecting fluorescein sodium, we recorded real-time videos for 1 minutes, 5 minutes, 10 minutes, 15 minutes, and 20 minutes. Every pCLE recording was done under fixed brightness, and the laser volume was located in the control panel on the pCLE display and operation screen, in order to avoid the effects of the laser and brightness. The pCLE videos for each patient were stored in a specific folder for later analysis.

Yellow skin. A transient yellowing of the skin and urine is a common adverse event after injection of fluorescein sodium, although fluorescein sodium can be metabolized completely after enough water intake for about 24 to 48 hours. Therefore, the patients' jaundice values on the