

## Review

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# Utilization of indocyanine green fluorescent imaging (ICG-FI) for the assessment of microperfusion in vascular medicine

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**Abstract:** Intraoperative valuation of organ and tissue microperfusion is always a current topic in different surgical situations. Although indocyanine green fluorescent imaging (ICG-FI) has turned to be a more and more common technique to evaluate organ perfusion, only few studies tried to quantitatively validate the technique for microperfusion assessment. The aim of the following manuscript is to present the results of our interdisciplinary research confirming additional quantitative assessment tools in different surgical conditions. Thus, we are implementing the background-subtracted peak fluorescent intensity (BSFI), the slope of fluorescence intensity (SFI), and the time to slope (TTS) using ICG-FI in several regions of interest (ROI).

**Keywords:** fluorescent imaging; ICG; microperfusion; quantitative assessment; validation.

## Intraoperative imaging

### ICG visualization

Due to its biological characteristics following intravenous injection, indocyanine green (ICG) is a suitable contrast agent for fluorescent imaging (FI) using the near-infrared

laser technology [1]. The technology is based on the fluorescence of ICG, a non-toxic, water-soluble fluorescent dye that has been used extensively for >40 years for the examination of liver function, microcirculation, and ophthalmic angiography. The dye immediately binds to plasma proteins and remains intravascular after systemic intravascular injection. Because of its rapid elimination exclusively by the liver (half-life, 2.4 min), repeated injections of ICG are possible. Adverse reactions related to its clinical use are rare, and the side effects other than iodine allergy have not been reported [1, 2]. In plasma, it displays an absorption maximum at 805 nm and an emission maximum at 830 nm [1].

For ICG-FI, the fluorescent dye ICG is intravenously applied through a peripheral venous line (0.01 mg/kg body weight) [3–8] and the tissue in the region of interest is illuminated with near-infrared light at a wavelength of 785 nm with a total output of 80 mW in a field of view of 10 cm in diameter (1 mW/cm<sup>2</sup>) operating at a distance of approximately 20 cm above tissue. The fluorescence emission of the excited dye is detected by an infrared-sensitive charge-coupled device (CCD) camera system (dynamic range 54 dB). In our studies, the camera was equipped with a band pass filter for the selective transmission of light at the emission maximum of ICG (830 nm) [9–11] (Figure 1). The fluorescent cardiac imaging (FCI) images are displayed and recorded in real-time on a computer monitor. In our studies, we used a digital image processing system with a temporal resolution of 40 ms and spatial resolution of about 0.2 mm at a penetration depth of 4 mm for quantitative assessment of tissue perfusion (LLS GmbH, Ulm, Germany) [9].

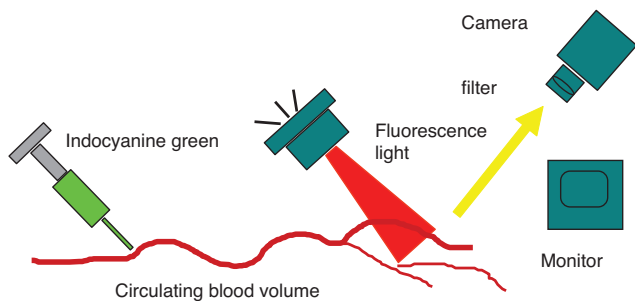
The laser has an excellent safety profile for both patient and operating staff. Because of the low power density of the emitted laser energy, there is no tissue warming. Further, eye protection is not required because the laser light energy is dispersed and remitted light has no hazardous potential ( $I = 1 \text{ mW/cm}^2$ ).

Although ICG-FI has turned to be a more and more common technique to evaluate intraoperative organ perfusion, only few studies tried to quantitatively validate the technique for visceral perfusion assessment.

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**Figure 1:** Indocyanine green fluorescent imaging (IDG-FI): After intravenous injection, the region of interest is illuminated with near-infrared light (785 nm).

The fluorescence emission of the excited dye is detected by an infrared-sensitive charge-coupled device camera system (dynamic range 54 dB) equipped with a band pass filter for the selective transmission of light at the emission maximum of ICG (830 nm).

In our studies, we confirmed additional methods for quantitative assessment using ICG-FI [9–11]:

First, *background-subtracted peak fluorescent intensity (BSFI)* in the time-dependent fluorescence intensity curves is a useful parameter for quantitative assessment during repeated injections of ICG. By subtraction of residual fluorescent intensity of the previous injection, misinterpretation of false high fluorescence intensities can be avoided. Thus, several injections in short-time intervals can be performed with viable results, while solely visual interpretation would mislead to false high perfusion levels. Nevertheless, capillary leakage must be taken into consideration due to increased fluorescence intensity in the extravascular space.

Second, the maximal *slope of fluorescence intensity (SFI)* during the increase of the time-dependent fluorescent intensity can be assessed. This allows the quantitative determination of the increase of fluorescent signal in the region of interest (ROI), independent of the background fluorescent signal. Nevertheless, the SFI is dependent on the blood flow and therefore dependent on the cardiac output.

### Cardiac surgery

Intraoperative assessment of graft patency and detection of significant coronary artery bypass stenosis is of major importance for optimal early and long-term outcome following coronary artery bypass grafting (CABG).

However, no routine quality control is performed by most cardiac surgeons because the techniques used suffered from time-consuming, unreliable, invasiveness, or lack of visualization.

FCI using the dye ICG is a non-invasive imaging technology for intraoperative visualization of coronary vessels

and myocardial perfusion and can detect patency of the bypass graft as well as graft occlusion [3–7]. Several studies demonstrated that the FCI technique was clinically useful for evaluation of graft patency using a similar ICG-based imaging device (SPY; Novadaq Technologies; Toronto, Canada) for quality control after CABG [3–6]. Solely visual assessment of non-occlusive graft-flow abnormalities could not be identified reliably [3, 4, 8]. Therefore, additional quantitative assessment of myocardial perfusion by FCI is a useful tool for intraoperative quality control.

Quantitative assessment of myocardial perfusion during coronary stenosis of variable severity was validated in an experimental setting [9–11]. For FCI, ICG (0.01 mg/kg body weight) is intravenously applied through a peripheral venous line and the heart is illuminated with near-infrared light at a wavelength of 785 nm with a total output of 80 mW in a field of view of 10 cm in diameter (1 mW/cm<sup>2</sup>) operating at a distance of 20 cm above the left ventricle. The fluorescence emission of the excited dye was detected by an infrared-sensitive charge-coupled device camera system (dynamic range 54 dB) equipped with a band pass filter for the selective transmission of light at the emission maximum of ICG (830 nm; Figure 2). The FCI images were recorded in real-time and analyzed using a digital image processing system as previously described (LLS GmbH, Ulm, Germany) [9–11].

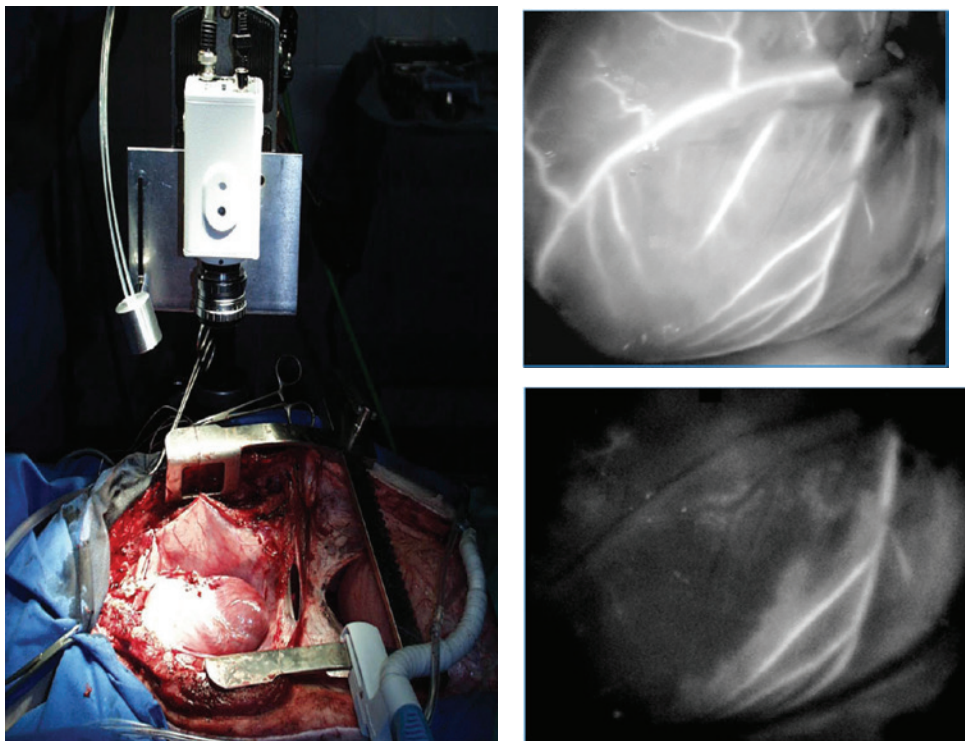
The experimental study was able to show that quantitative assessment of myocardial perfusion during flow-limiting coronary stenosis of high severity and even during non-flow limiting stenosis is possible by FCI (Figure 2).

Nevertheless, the experimental studies were evaluated under perfectly controlled hemodynamic conditions in open-chest pigs. Direct transfer of the experimental findings into a clinical setting in humans might be limited: rotation of the heart is necessary for proper imaging of the lateral or posterior wall. Changes in hemodynamic parameters such as heart rate, blood pressure, and cardiac output as well as optical parameters like scattering due to overlying fat tissue and movement of the beating heart may influence absolute values. Furthermore, in a clinical setting, competitive coronary flow and collateral flow may affect perfusion measurements, which have to be proved in future studies.

Finally, FCI might be a useful tool for intraoperative quality control during CABG.

### Vascular surgery

Peripheral arterial disease (PAD) is a major health problem affecting over 200 million people worldwide [12]. Demographic changes inevitably lead to increasing numbers



**Figure 2:** Left: experimental setting of ICG-FI.

The tissue in the region of interest is illuminated with near-infrared light at a wavelength of 785 nm with a total output of 80 mW in a field of view of 10 cm in diameter ( $1 \text{ mW/cm}^2$ ) operating at a distance of approximately 20 cm above tissue. The fluorescence emission of the excited dye is detected by an infrared-sensitive charge-coupled device (CCD) camera system (dynamic range 54 dB). Right: ICG-FI before stenosis (above) and myocardial perfusion during flow-limiting coronary stenosis of high severity (lower).

subsequently requiring an optimal best medical or invasive treatment. As the gold standard Ankle-brachial index (ABI) measurement is certainly the oldest method for diagnosis of PAD and assessment of lower limb macroperfusion [13]. However, its utilization is limited in patients with extensive calcification of the medial vessel layer causing incompressibility of peripheral arteries during measurement leading to falsely elevated pressure values. Other measuring tools, such as toe-pressure brachial index measurements (TBI), photoplethysmography, and transcutaneous oxygen pressure measurements ( $\text{TcPO}_2$ ), are used for assessing microperfusion of the foot [14, 15]. However, their application requires sensors or probes being placed on the foot to conduct microperfusion measurements. Hence, usability may be limited in patients with foot ulcers and infected tissue. Furthermore, diabetes mellitus poses a significant risk factor for disturbing the micro-circulation of the lower extremity leading to diabetic foot ulcers with increasing incidence and prevalence that may contribute to increasing numbers of amputations [16]. As described above, ICG-FI serves as a tool for assessing the microperfusion of various tissues without necessary probes that have to be placed on the area of interest. For that purpose, it has been widely

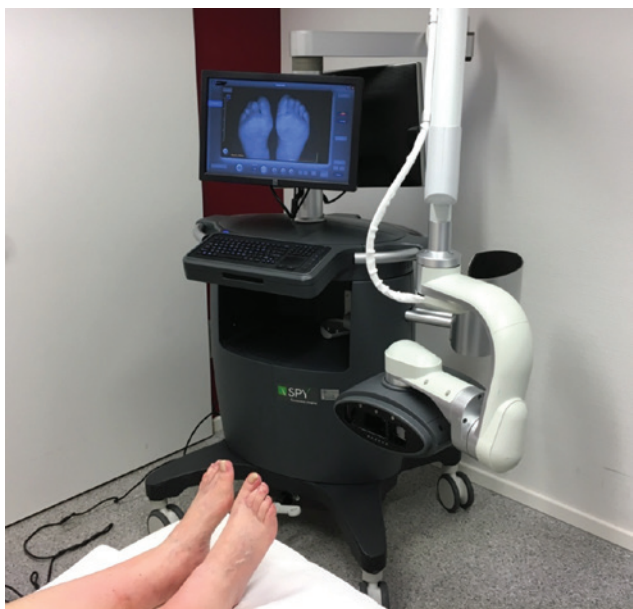
used in various medical disciplines over the 30 years with excellent results in terms of validity and reproducibility [9, 17–21]. Considering the advantages of this technique, it might be surprising that its utilization for the assessment of micro-circulation in PAD is relatively new. Braun et al. and Colvard et al. successfully tested ICG-FI (SPY system, NOVADAQ, Bonita Springs, USA) as a tool for quality control by evaluating the perfusion of the foot in patients with PAD prior and after a revascularization procedure [22, 23]. Quantitative evaluation of data was performed via the SPY software-package assessing time-intensity curves of fluorescent intensity by ingress, ingress rate, egress, and egress rate. Results were successfully correlated to ABI and Toe-pressure measurements (TP). Igari et al. correlated results of pre- and postoperative ICG-FI to ABI, TBI, and TP [24]. They were the first in assessing different regions of interest (ROIs) of the foot and described “time from ICG onset to maximum intensity” ( $T_{\text{max}}$ ), time elapsed from fluorescence onset to half the maximum intensity” ( $T_{1/2}$ ) and “fluorescence intensity measured 10 seconds after the onset of fluorescence” ( $\text{PDE}_{10}$ ) as new parameters used for the assessment of curves obtained from ICG-FI. They found  $T_{\text{max}}$  and  $T_{1/2}$  more significantly correlated with ABI, TBI,



and TP [24]. In another study, Venermo et al. showed repeatability of time-intensity curves obtained from ICG-FI using T1/2 and PDE10 in patients undergoing a revascularization procedure. Interestingly a strong correlation of TcPO<sub>2</sub> and PDE10 for diabetic patients was observed [25]. Although the process of ICG-FI measurements of the foot may vary between the different studies, the main principle stays the same. Our protocol and setting for ICG-FI measurements can be obtained from Table 1 and Figure 3. In a recently published study, we introduced a new way of assessing time-intensity curves obtained from ICG-FI in patients that underwent open-surgical revascularization procedure for PAD [26]. Time-intensity curves were analyzed offline using our own software package (Institute for Laser Technology, University Ulm, GmbH) that enables manually fitting the slope of fluorescence intensity (SFI) to the maximum incline of the curve (Figure 4) [26]. Since results obtained

**Table 1:** Protocol for ICG-FI measurement.

Clear the patient of contraindications for ICGA
Eliminate all light sources and remove any clothes or wound dressings
Wait for 15 min and measure systolic blood pressure
Position the camera head perpendicular to the plantar region of the foot with the help of two overlapping laser beams
Administer a bolus of dissolved indocyanine green at a dose of 0.1 mg/kg and immediately inject 10 mL normal saline to flush intravenous line
Initiate capturing for a sequence of 272 s without moving the camera

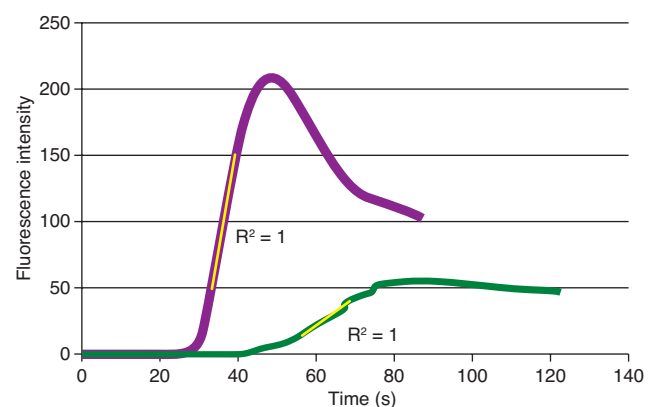


**Figure 3:** Setting for ICG-FI measurements. The camera is positioned perpendicular to area of interest. ICG-FI is captured when all light sources are removed.

from maximum fluorescent intensity measurements, which are analyzed by many previous studies, can be influenced by skin color, light in the room, distance to the camera, peripheral edema, and previous injections of ICG causing a residual fluorescent signal may in our opinion over or underestimate perfusion. We successfully correlated results to perfusion measurements obtained from ABI and photoplethysmography and thereby proved the practical feasibility of this new technique of analyzing data [26]. ICG-FI can be utilized for various purposes in vascular medicine. Rother et al. recently used ICG-FI for assessing indirect and direct revascularized angiosomes after tibial bypass surgery [27]. Lepow et al. showed first attempts in using the ICG-FI to visually assess a chronic diabetic ulcer and intraoperatively evaluated wound margins during debridement in the operating room [28]. They concluded that ICG-FI may serve as a tool for predicting wound healing and help in determining resection margins during surgical treatment of chronic wounds. Figure 5 presents a patient suffering from microembolization of digitus I of the left foot, which was caused by atrial fibrillation without sufficient anticoagulation (Figure 5). Visual assessment of the bare foot and the image obtained from ICG-FI consistently proved full recovery after medical treatment with heparin and Alprostadil (Prostvasin) for 10 days.

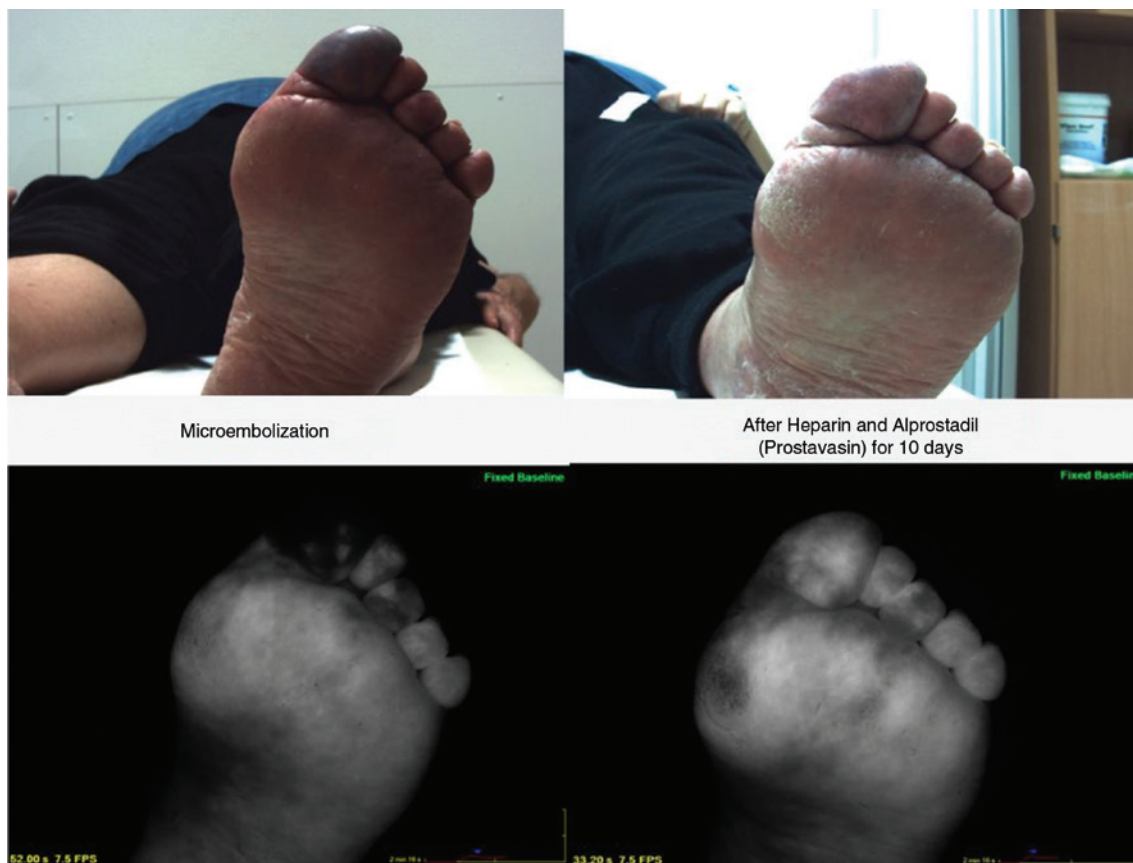
### Visceral surgery

Intestinal microperfusion and viability of intestinal organs is usually estimated from the color of serosal surface,



**Figure 4:** Green time-intensity curve corresponding to preoperative ICG-FI measurement.

Purple time-intensity curve corresponding to postoperative ICG-FI measurement. Yellow line (linear regression trendline) corresponding to SFI manually fitted to the maximum incline of the curves. Reprinted from Publication *Vasa*, 46/5, H. C. Rieß et al., A new tool for fluorescent imaging, pp. 383–388. Copyright Hogrefe Publishing Corp. Reproduced with permission.



**Figure 5:** Microembolization of digitus I of the left foot. Before (left side) and after 10 days of treatment with Heparin and Alprostadil (Prostavasin).

presence of peristalsis, pulsation, and bleeding from marginal arteries. However, clinical assessment remains subjective and still depends on the personal experience of the surgeon [29]. ICG-FI is getting more and more common to evaluate tissue perfusion because of its impact in anastomotic healing [30–32]. However, the method still remains subjective caused by solely visual interpretation of fluorescence intensity as a parameter of perfusion. Our study group recently evaluated the role of FI for quantitative assessment of intestinal perfusion first in a gastric tube model as a one-vessel model and second in an experimental setting of mesenteric ischemia.

Although numerous objective quantitative techniques of intraoperative bowel viability assessment are available, fluorescence based methods seem to be most promising [29, 33]. Several studies describe clinical use of ICG-FI in esophageal [32, 34–37] and colorectal surgery [31, 38–46].

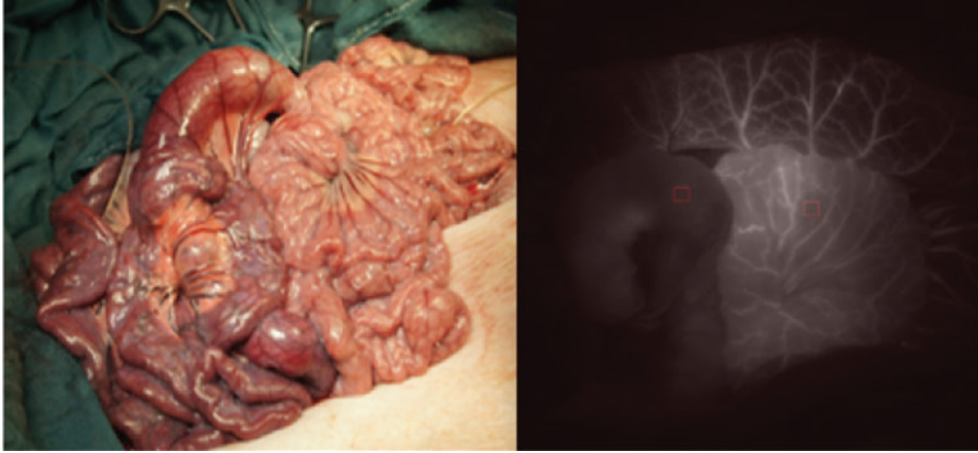
Zehetner et al. concluded in their study on 150 patients undergoing esophagectomy, that intraoperative real-time assessment of perfusion correlated with the likelihood of an anastomotic leak and confirmed the critical relationship between good perfusion and anastomotic healing

[35]. Same observation has been recently published by Karampinis et al. presenting the results in 55 patients [32]. However, only visual and no quantitative assessment of the ICG-FI data was performed.

Toens et al. conducted an experimental study on mesenteric ischemia in a rabbit model comparing ICG-FI to radioactive microspheres. Validation of ICG-FI was performed by the increment of fluorescence in the ischemic ROIs compared to a control loop as reference region. ICG-FI was concluded to be a feasible, reliable, and valid technique for mesenteric blood flow assessment [47].

Diana et al. showed in a porcine study the real-time assessment of intestinal viability using ICG-FI. Quantitative assessment of ICG-FI was reached by calculating the time to peak in ischemic and non-ischemic areas of the bowel. They concluded that ICG-FI perfusion assessment may effectively detect bowel perfusion [33]. They concluded in a following study that 25% less perfusion certainly impairs anastomotic healing without generating surgical complications [48].

In our studies (manuscript under submission), ICG-FI correlated perfectly with the predicted perfusion changes



**Figure 6:** Induced mesenteric ischemia in a porcine model on the left. On the right side, the corresponding ICG-FI picture with total loss of fluorescence in the left loop.

using the explained evaluation tool SFI, BSFI, and TTS. In our one-vessel model, we found a strong correlation between our evaluation technique, and the fluorescent microspheres as the gold standard in experimental perfusion assessment.

The aim of our second study was to establish and validate our quantitative assessment tools for intraoperative perfusion measurement in ischemic and post-ischemic bowel (Figure 6). Acute mesenteric ischemia remains a highly morbid condition with poor outcome [49]. It is mandatory to confirm diagnosis, assess bowel viability, perform revascularization if useful, and resect non-viable bowel as early as possible. Evaluation of bowel vitality is still subjective, and may lead to resection of segments that potentially can recover [49]. Thus, objective non-invasive methods to assess intestine viability are needed to determine correctly the extent of ischemia and to evaluate anastomosis in several visceral procedures.

The use of ICG-FI in evaluation of ischemic bowel has been described in small case series so far [46, 50–52]. All authors conclude that ICG-FI is easy to handle and little time-consuming even in emergency situations [46]. Some changed their intraoperative decision after assessing bowel perfusion via ICG-FI, still with the limitation of only subjective visual evaluation [46, 52]. Behrendt et al. investigated bowel perfusion after simple manual manipulation using ICG-FI in a rat model using a perfusion index between region of interest and AMS as reference region. They proved the system capable on quantifying gastrointestinal transit and bowel perfusion [53].

Matsui et al. performed similar experiments in a porcine model combined with a survivor study in rats. He used 30 min of ischemia to induce ischemia in 2–12 cm of bowel. ICG-FI was quantified by absolute fluorescence intensity and contrast-to-background ratio. They assumed

that clinical assessment alone had high sensitivity, but relatively low specificity, and therefore accuracy remained low in predicting animal survival. Surprisingly, the quantitative metrics like maximum fluorescence intensity to control were only marginally better than clinical assessment [54].

Our study evaluated the use of ICG-FI in the assessment of bowel viability after different ischemic periods in a porcine model. In contrast to existing studies, ischemic period was adjusted to realistic clinical setting. ICG-FI showed significant difference in tissue viability comparing 6 and 10 h of ischemia. While macroscopic damage of bowel tissue after 10 h of ischemia was obvious even by solely visual inspection, detection of tissue damage after 6 h of ischemia remained difficult. Thus, quantitative ICG-FI can predict tissue damage even if qualitative macroscopic evaluation fails (Figure 5).

In connection with this work, we observed two major points, which affect the interpretation of the ICG-FI technique in daily life visceral surgery.

Hyper intensity due to capillary leakage and thus the pooling of ICG in the tissue may lead to misinterpretation. In the case of visual assessment alone, there is a risk that previously damaged areas will be assessed as adequately perfused because of the ongoing fluorescence of the tissue. Normally, this pooling fluorescence lasts longer due to the loss of epithelial integrity. A well-perfused tissue shows a quickly influx of ICG, and a slower, but visible, outflow.

Hemodynamic alterations during operation or in between the patients causes different ICG-FI. In our studies we observed that higher cardiac causes elevated ICG-FI levels. Hemodynamics, such as cardiac output, blood pressure, and vascular resistance, volume load, and catecholamine substitution can influence ICG-FI results. In the clinical setting, those parameters have to be taken



into consideration for valid measurement. If a stable setting is not possible, either a ratio to a baseline value, or comparison to a non-ischemic area during the same measurement is mandatory. So far, this important influencing factor seems to be underestimated in the literature.

SFI and BSFI measurements are valid quantitative assessment tools using ICG-FI for intraoperative quality evaluation of tissue perfusion. The translation to clinical use is therefore much easier and will be performed in the future. First observational studies in humans are mandatory to evaluate predictive value of ICG-FI assessment.

## Conclusion

ICG-FI is a promising tool for intraoperative evaluation of microperfusion in different interdisciplinary settings. As discussed above solely, visual assessment might be misleading; therefore, quantitative determination of microperfusion should be implemented into every days clinical evaluation. Thus, in vascular surgery, ICG-FI can be successfully used for perioperative quality control and postoperative follow-up. In cardiac and visceral surgery, ICG-FI is useful intraoperative assessment tool to evaluate tissue perfusion after ischemia and anastomotic sides enabling immediate revision or decision making. Further clinical studies are needed to prove the findings and integrate the technique into daily routine.

### Author Statement

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### Author Contributions

Anna Duprée: conceptualization; investigation; writing – original draft; writing – review and editing. Henrik Rieß: conceptualization; data curation; writing – original draft. Christian Detter: conceptualization; writing – review and editing. Eike Sebastian Debus: conceptualization; writing – review and editing. Sabine Helena Wipper: conceptualization; data curation; investigation; methodology; writing – original draft; writing – review and editing.

## References

- [1] Cherrick GR, Stein SW, Leevy CM, Davidson CS. Indocyanine green: observations on its physical properties, plasma decay, and hepatic extraction. *J Clin Invest* 1960;39:592–600.
- [2] Hope-Ross M, Yannuzzi LA, Gragoudas ES, Guyer DR, Slakter JS, Sorenson JA, et al. Adverse reactions due to indocyanine green. *Ophthalmology* 1994;101:529–33.
- [3] Reuthebuch O, Haussler A, Genoni M, Tavakoli R, Odavic D, Kadner A, et al. Novadaq SPY: intraoperative quality assessment in off-pump coronary artery bypass grafting. *Chest* 2004;125:418–24.
- [4] Taggart DP, Choudhary B, Anastasiadis K, Abu-Omar Y, Balacumaraswami L, Pigott DW. Preliminary experience with a novel intraoperative fluorescence imaging technique to evaluate the patency of bypass grafts in total arterial revascularization. *Ann Thorac Surg* 2003;75:870–3.
- [5] Desai ND, Miwa S, Kodama D, Cohen G, Christakis GT, Goldman BS, et al. Improving the quality of coronary bypass surgery with intraoperative angiography: validation of a new technique. *J Am Coll Cardiol* 2005;46:1521–5.
- [6] Desai ND, Miwa S, Kodama D, Koyama T, Cohen G, Pelletier MP, et al. A randomized comparison of intraoperative indocyanine green angiography and transit-time flow measurement to detect technical errors in coronary bypass grafts. *J Thorac Cardiovasc Surg* 2006;132:585–94.
- [7] Detter C, Russ D, Iffland A, Wipper S, Schurr MO, Reichenspurner H, et al. Near-infrared fluorescence coronary angiography: a new noninvasive technology for intraoperative graft patency control. *Heart Surg Forum* 2002;5:364–9.
- [8] Balacumaraswami L, Abu-Omar Y, Choudhary B, Pigott D, Taggart DP. A comparison of transit-time flowmetry and intraoperative fluorescence imaging for assessing coronary artery bypass graft patency. *J Thorac Cardiovasc Surg* 2005;130:315–20.
- [9] Detter C, Wipper S, Russ D, Iffland A, Burdorf L, Thein E, et al. Fluorescent cardiac imaging: a novel intraoperative method for quantitative assessment of myocardial perfusion during graded coronary artery stenosis. *Circulation* 2007;116:1007–14.
- [10] Wipper S, Reiter B, Russ D, Hahnel F, Kersten JF, Kolbel T, et al. Distinction of non-ischemia inducing versus ischemia inducing coronary stenosis by fluorescent cardiac imaging. *Int J Cardiovasc Imaging* 2016;32:363–71.
- [11] Detter C, Russ D, Kersten JF, Reichenspurner H, Wipper S. Qualitative angiographic and quantitative myocardial perfusion assessment using fluorescent cardiac imaging during graded coronary artery bypass stenosis. *Int J Cardiovasc Imaging* 2018;34:159–67.
- [12] Fowkes FG, Rudan D, Rudan I, Aboyans V, Denenberg JO, McDermott MM, et al. Comparison of global estimates of prevalence and risk factors for peripheral artery disease in 2000 and 2010: a systematic review and analysis. *Lancet* 2013;382:1329–40.
- [13] Aboyans V, Criqui MH, Abraham P, Allison MA, Creager MA, Diehm C, et al. Measurement and interpretation of the ankle-brachial index: a scientific statement from the American Heart Association. *Circulation* 2012;126:2890–909.
- [14] Allen J. Photoplethysmography and its application in clinical physiological measurement. *Physiol Meas* 2007;28:R1–39.
- [15] Lusiani L, Visona A, Nicolin P, Papesso B, Pagnan A. Transcutaneous oxygen tension (TcPO<sub>2</sub>) measurement as a diagnostic

- tool in patients with peripheral vascular disease. *Angiology* 1988;39:873–80.
- [16] Sharma M, Nazareth I, Petersen I. Trends in incidence, prevalence and prescribing in type 2 diabetes mellitus between 2000 and 2013 in primary care: a retrospective cohort study. *BMJ Open* 2016;6:e010210.
- [17] Brown N, Strong R. Infrared fundus angiography. *Br J Ophthalmol* 1973;57:797–802.
- [18] Craandijk A, Van Beek CA. Indocyanine green fluorescence angiography of the choroid. *Br J Ophthalmol* 1976;60:377–85.
- [19] Yeoh MS, Kim DD, Ghali GE. Fluorescence angiography in the assessment of flap perfusion and vitality. *Oral Maxillofac Surg Clin North Am* 2013;25:61–6, vi.
- [20] Ashitate Y, Stockdale A, Choi HS, Laurence RG, Frangioni JV. Real-time simultaneous near-infrared fluorescence imaging of bile duct and arterial anatomy. *J Surg Res* 2012;176:7–13.
- [21] Carus T, Dammer R. Laparoscopic fluorescence angiography with indocyanine green to control the perfusion of gastrointestinal anastomoses intraoperatively. *Surg Technol Int* 2012;22:27–32.
- [22] Braun JD, Trinidad-Hernandez M, Perry D, Armstrong DG, Mills JL, Sr. Early quantitative evaluation of indocyanine green angiography in patients with critical limb ischemia. *J Vasc Surg* 2013;57:1213–8.
- [23] Colvard B, Itoga NK, Hitchner E, Sun Q, Long B, Lee G, et al. SPY technology as an adjunctive measure for lower extremity perfusion. *J Vasc Surg* 2016;64:195–201.
- [24] Igari K, Kudo T, Toyofuku T, Jibiki M, Inoue Y, Kawano T. Quantitative evaluation of the outcomes of revascularization procedures for peripheral arterial disease using indocyanine green angiography. *Eur J Vasc Endovasc Surg* 2013;46:460–5.
- [25] Venermo M, Settembre N, Alback A, Vikatmaa P, Aho PS, Lepantalo M, et al. Pilot assessment of the repeatability of indocyanine green fluorescence imaging and correlation with traditional foot perfusion assessments. *Eur J Vasc Endovasc Surg* 2016;52:527–33.
- [26] Riess HC, Dupree A, Behrendt CA, Kolbel T, Debus ES, Larena-Avellaneda A, et al. Initial experience with a new quantitative assessment tool for fluorescent imaging in peripheral artery disease. *Vasa* 2017;46:383–8.
- [27] Rother U, Lang W, Horch RE, Ludolph I, Meyer A, Gefeller O, et al. Pilot assessment of the angiosome concept by intraoperative fluorescence angiography after tibial bypass surgery. *Eur J Vasc Endovasc Surg* 2018;55:215–21.
- [28] Lepow BD. The use of SPY intra-operative vascular angiography as a predictor of wound healing. *Podiatry Management* 2011:141–8.
- [29] Urbanavicius L, Pattyn P, de Putte DV, Venskutonis D. How to assess intestinal viability during surgery: a review of techniques. *World J Gastrointest Surg* 2011;3:59–69.
- [30] Thompson SK, Chang EY, Jobe BA. Clinical review: healing in gastrointestinal anastomoses, part I. *Microsurgery* 2006;26:131–6.
- [31] Degett TH, Andersen HS, Gogenur I. Indocyanine green fluorescence angiography for intraoperative assessment of gastrointestinal anastomotic perfusion: a systematic review of clinical trials. *Langenbecks Arch Surg* 2016;401:767–75.
- [32] Karampinis I, Ronellenfitsch U, Mertens C, Gerken A, Hetjens S, Post S, et al. Indocyanine green tissue angiography affects anastomotic leakage after esophagectomy. A retrospective, case-control study. *Int J Surg* 2017;48:210–4.
- [33] Diana M, Noll E, Diemunsch P, Dallemagne B, Benahmed MA, Agnus V, et al. Enhanced-reality video fluorescence: a real-time assessment of intestinal viability. *Ann Surg* 2014;259:700–7.
- [34] Shimada Y, Okumura T, Nagata T, Sawada S, Matsui K, Hori R, et al. Usefulness of blood supply visualization by indocyanine green fluorescence for reconstruction during esophagectomy. *Esophagus* 2011;8:259–66.
- [35] Zehetner J, DeMeester SR, Alicuben ET, Oh DS, Lipham JC, Hagen JA, et al. Intraoperative assessment of perfusion of the gastric graft and correlation with anastomotic leaks after esophagectomy. *Ann Surg* 2015;262:74–8.
- [36] Murawa D, Hunerbein M, Spychala A, Nowaczyk P, Polom K, Murawa P. Indocyanine green angiography for evaluation of gastric conduit perfusion during esophagectomy – first experience. *Acta Chir Belg* 2012;112:275–80.
- [37] Pacheco PE, Hill SM, Henriques SM, Paulsen JK, Anderson RC. The novel use of intraoperative laser-induced fluorescence of indocyanine green tissue angiography for evaluation of the gastric conduit in esophageal reconstructive surgery. *Am J Surg* 2013;205:349–52; Discussion 52–3.
- [38] Kudzus S, Roesel C, Schachtrupp A, Hoer JJ. Intraoperative laser fluorescence angiography in colorectal surgery: a non-invasive analysis to reduce the rate of anastomotic leakage. *Langenbecks Arch Surg* 2010;395:1025–30.
- [39] Kin C, Vo H, Welton L, Welton M. Equivocal effect of intraoperative fluorescence angiography on colorectal anastomotic leaks. *Dis Colon Rectum* 2015;58:582–7.
- [40] Jafari MD, Wexner SD, Martz JE, McLemore EC, Margolin DA, Sherwinter DA, et al. Perfusion assessment in laparoscopic left-sided/anterior resection (PILLAR II): a multi-institutional study. *J Am Coll Surg* 2015;220:82–92 e1.
- [41] Sherwinter DA. Transanal near-infrared imaging of colorectal anastomotic perfusion. *Surg Laparosc Endosc Percutan Techn* 2012;22:433–6.
- [42] Sherwinter DA, Gallagher J, Donkar T. Intra-operative transanal near infrared imaging of colorectal anastomotic perfusion: a feasibility study. *Colorectal Disease* 2013; 15:91–6.
- [43] Hellan M, Spinoglio G, Pigazzi A, Lagares-Garcia JA. The influence of fluorescence imaging on the location of bowel transection during robotic left-sided colorectal surgery. *Surg Endosc* 2014;28:1695–702.
- [44] Protyniak B, Dinallo AM, Boyan WP, Jr., Dressner RM, Arvanitis ML. Intraoperative indocyanine green fluorescence angiography – an objective evaluation of anastomotic perfusion in colorectal surgery. *Am Surg* 2015;81:580–4.
- [45] Boni L, David G, Mangano A, Dionigi G, Rausei S, Spampatti S, et al. Clinical applications of indocyanine green (ICG) enhanced fluorescence in laparoscopic surgery. *Surg Endosc* 2015;29:2046–55.
- [46] Nowak K, Sandra-Petrescu F, Post S, Horisberger K. Ischemic and injured bowel evaluation by Fluorescence imaging. *Colorectal Dis* 2015;17 Suppl 3:12–5.
- [47] Toens C, Krones CJ, Blum U, Fernandez V, Grommes J, Hoelzl F, et al. Validation of IC-VIEW fluorescence videography in a rabbit model of mesenteric ischaemia and reperfusion. *Int J Colorectal Dis* 2006;21:332–8.
- [48] Diana M, Halvax P, Dallemagne B, Nagao Y, Diemunsch P, Charles AL, et al. Real-time navigation by fluorescence-based



- enhanced reality for precise estimation of future anastomotic site in digestive surgery. *Surg Endosc* 2014;28:3108–18.
- [49] Meng X, Liu L, Jiang H. Indications and procedures for second-look surgery in acute mesenteric ischemia. *Surg Today* 2010;40:700–5.
- [50] Iinuma Y, Hirayama Y, Yokoyama N, Otani T, Nitta K, Hashidate H, et al. Intraoperative near-infrared indocyanine green fluorescence angiography (NIR-ICG AG) can predict delayed small bowel stricture after ischemic intestinal injury: report of a case. *J Pediatr Surg* 2013;48:1123–8.
- [51] Ryu S, Yoshida M, Ohdaira H, Tsutsui N, Suzuki N, Ito E, et al. Intestinal blood flow assessment by indocyanine green fluorescence imaging in a patient with the incarcerated umbilical hernia: report of a case. *Ann Med Surg* 2016;8:40–2.
- [52] Nitori N, Deguchi T, Kubota K, Yoshida M, Kato A, Kojima M, et al. Successful treatment of non-occlusive mesenteric ischemia (NOMI) using the HyperEye Medical System for intraoperative visualization of the mesenteric and bowel circulation: report of a case. *Surg Today* 2014;44:359–62.
- [53] Behrendt FF, Tolba RH, Overhaus M, Hirner A, Minor T, Kalff JC. Indocyanine green fluorescence measurement of intestinal transit and gut perfusion after intestinal manipulation. *Eur Surg Res* 2004;36:210–8.
- [54] Matsui A, Winer JH, Laurence RG, Frangioni JV. Predicting the survival of experimental ischaemic small bowel using intraoperative near-infrared fluorescence angiography. *Br J Surg* 2011;98:1725–34.

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**Supplementary Material:** The article (<https://doi.org/10.1515/iss-2018-0014>) offers reviewer assessments as supplementary material.



## Reviewer Assessment

Anna Duprée<sup>a,\*</sup>, Henrik Rieß<sup>a</sup>, Christian Detter, Eike S. Debus and Sabine H. Wipper

# Utilization of indocyanine green fluorescent imaging (ICG-FI) for the assessment of microperfusion in vascular medicine

<https://doi.org/10.1515/10.1515/iss-2018-0014>

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## Reviewers' Comments to Original Submission

### Reviewer 1: anonymous

Apr 22, 2018

**Reviewer Recommendation Term:** Accept with Minor Revision  
**Overall Reviewer Manuscript Rating:** 70

#### Custom Review Questions

Custom Review Questions	Response
Is the subject area appropriate for you?	4
Does the title clearly reflect the paper's content?	4
Does the abstract clearly reflect the paper's content?	3
Do the keywords clearly reflect the paper's content?	4
Does the introduction present the problem clearly?	2
Are the results/conclusions justified?	3
How comprehensive and up-to-date is the subject matter presented?	4
How adequate is the data presentation?	3
Are units and terminology used correctly?	4
Is the number of cases adequate?	3
Are the experimental methods/clinical studies adequate?	N/A
Is the length appropriate in relation to the content?	4
Does the reader get new insights from the article?	4
Please rate the practical significance.	4
Please rate the accuracy of methods.	N/A
Please rate the statistical evaluation and quality control.	N/A
Please rate the appropriateness of the figures and tables.	3
Please rate the appropriateness of the references.	3
Please evaluate the writing style and use of language.	3
Please judge the overall scientific quality of the manuscript.	3
Are you willing to review the revision of this manuscript?	Yes

**Comments to Authors:****A. Overall evaluation and general comments**

Your paper proposed a nice overview on indocyanine green fluorescent imaging (ICG-FI) for the assessment of microperfusion in vascular medicine. This is a very interesting topic that will probably have a growing interest in clinical practice.

There are some general points I would like to comment:

- A1: I would suggest putting in perspective the different technique that are proposed to evaluate tissue perfusion as perfusion angiography to help in demonstrating the particular role of ICG-FI.

**B. Detailed evaluation, specific deficiencies with suggestions for improvement****Abstract:**

Give the meaning for ICG-FI line 2 since it appears for the first time and write only ICG-FI at the end of the section.

**Intraoperative imaging:**

You discussed the difficulties to evaluate all the surfaces of the heart and the necessity of a 20 cm distance between the camera and the tissue. Is there a risk of partial distorsion of the results in non-perfectly plane tissue?

Page 3 line 3: define CABG since cited for the first time

Page 6, line 7: use only FI since already defined

Page 7 line 3, you should cite references corresponding to your studies

Page 7, line 16: use past tenses

Page 7, line 23: ad “et al” after Matsui

**Figures:**

In your legends you have 2 figures 4

**Reviewer 2: anonymous**

May 14, 2018

**Reviewer Recommendation Term:**

Revise with Major Modifications

**Overall Reviewer Manuscript Rating:**

N/A

**Custom Review Questions****Response**

Is the subject area appropriate for you?	5 - High/Yes
Does the title clearly reflect the paper's content?	4
Does the abstract clearly reflect the paper's content?	3
Do the keywords clearly reflect the paper's content?	4
Does the introduction present the problem clearly?	3
Are the results/conclusions justified?	3
How comprehensive and up-to-date is the subject matter presented?	5 - High/Yes
How adequate is the data presentation?	3
Are units and terminology used correctly?	2
Is the number of cases adequate?	2
Are the experimental methods/clinical studies adequate?	2
Is the length appropriate in relation to the content?	3
Does the reader get new insights from the article?	3
Please rate the practical significance.	4
Please rate the accuracy of methods.	2
Please rate the statistical evaluation and quality control.	1 - Low/No
Please rate the appropriateness of the figures and tables.	2
Please rate the appropriateness of the references.	3
Please evaluate the writing style and use of language.	2
Please judge the overall scientific quality of the manuscript.	3
Are you willing to review the revision of this manuscript?	Yes

**Comments to Authors:**

Dear Sir/ Madam,

Thank you for sending your article „Utilization of Indocyanine green Fluorescent Imaging (ICG-FI) for the assessment of microperfusion in vascular medicine“. This is a review paper that provides new aspects of the use of ICG-FI different specialties.

Here are my comments:

1. The draft is not properly formatted yet for Innovative Surgical Sciences
2. Looks like the initial study of BSFI was conducted long time ago. Any follow up on that? Any updated results? More information on methodology is needed as well as statistical analysis.
3. Is there a control group?
4. Your discussion is incomplete. Need to add the limitations of your investigation.
5. Conclusion. Share the future direction and applicability of your results in the usage of ICG-FI for the assessment of microperfusion in vascular medicine.

Major concerns:

What is new in your current manuscript and what is the new message? Is the new message worthy of a new manuscript or just a letter to the editor?

Many Thanks

As to the current draft you sent me, it is not adequate for me to provide the opinion for you to submit the manuscript as such.

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## Authors' Response to Reviewer Comments

May 30, 2018

Reply to reviewers comments

**Reviewer 1**

A1: the different techniques for tissue perfusion evaluation are mentioned in the subsections for each application area.

B: Because of the depth of field there is no distortion of results in non-plane tissue. For cardiac surgery a heart beat tracking is being planned.

**Reviewer 2**

1. The manuscript is formatted according to the author instructions

2-5. This work is a summary of our previous work. The method has been proofed in several experimental settings and different fields.

We performed several independently studies on that. To describe the detailed study protocol of each study in each subsection would go beyond the scope of this paper. We refer to the individual publication for this purpose, although some results still remain in the submission process.

## Reviewers' Comments to Revision

### Reviewer 2: anonymous

Jun 25, 2018

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**Reviewer Recommendation Term:**

Accept

**Overall Reviewer Manuscript Rating:**

75

**Custom Review Questions**

Is the subject area appropriate for you?  
 Does the title clearly reflect the paper's content?  
 Does the abstract clearly reflect the paper's content?  
 Do the keywords clearly reflect the paper's content?  
 Does the introduction present the problem clearly?

**Response**

5 - High/Yes  
 5 - High/Yes  
 4  
 5 - High/Yes  
 5 - High/Yes



Are the results/conclusions justified?	5 - High/Yes
How comprehensive and up-to-date is the subject matter presented?	5 - High/Yes
How adequate is the data presentation?	5 - High/Yes
Are units and terminology used correctly?	4
Is the number of cases adequate?	4
Are the experimental methods/clinical studies adequate?	4
Is the length appropriate in relation to the content?	4
Does the reader get new insights from the article?	5 - High/Yes
Please rate the practical significance.	5 - High/Yes
Please rate the accuracy of methods.	5 - High/Yes
Please rate the statistical evaluation and quality control.	4
Please rate the appropriateness of the figures and tables.	5 - High/Yes
Please rate the appropriateness of the references.	5 - High/Yes
Please evaluate the writing style and use of language.	4
Please judge the overall scientific quality of the manuscript.	5 - High/Yes
Are you willing to review the revision of this manuscript?	Yes

**Comments to Authors:**

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