



Case Series

Applications of indocyanine green in surgery: A single center case series

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ABSTRACT

Background: Fluorescence imaging using indocyanine green (ICG) has revolutionized commonly performed general surgical procedures by providing superior anatomic imaging and enhancing safety for patients. ICG, when injected, shows a bright green fluorescence when subjected to the near infra-red (NIR) spectrum.

Materials and methods: We employed the use of ICG in Laparoscopic cholecystectomy, Intestinal Colorectal Anastomosis and Hernia to assess vascularity of resected ends and bowel viability, Sentinel Lymph node mapping, Vascular surgery to assess amputation stump success and in assessing Flap Vascularity and healing.

Results: ICG when administered had successfully shown bright green fluorescence in different cases thereby aiding in surgical procedures.

Conclusion: Routine intraoperative use of ICG could pave the way for a more objective assessment of different surgical circumstances and thereby reduce personalized barriers to surgery. ICG fluorescence therefore seems to be a promising apparatus in standard general surgical procedures minimizing untoward errors and improving patient conformance.

1. Introduction

Indocyanine Green (ICG), used for fluorescence imaging, came into being and was approved for clinical usage during mid to late 1950s. However, its utilization for the purpose of angiography was not until after over 10 years of its formulation by the Kodak Research Laboratories [1].

The principle behind ICG angiography (ICGA), lies in the ability of the dye molecules to become fluorescent at an excitation wavelength of approximately 750–800 nm in the near infra-red (NIR) spectrum which can then be observed at an emission wavelength of over 800 nm [1–3]. ICG can be administered via the intravenous route and is considered relatively safe, with no known toxic metabolites and rapid excretion via the liver [2,4].

The compound was first brought into play in the early 1970s in the field of ophthalmology for the assessment of retinal vessels [1]. Since its advent, there have been several advancements in the applications of ICG, especially in the surgical field, such as intraoperative mapping and biopsy of sentinel lymph node biopsy, measurement of hepatic function prior to resection, neurosurgical cases to detect vascular anomalies, cardiovascular cases for patency and assessment of vascular

abnormalities, predicting healing following amputations, visualization of hepatobiliary anatomy and blood vessels, reconstructive surgery, to assess flap viability and for the evaluation of tissue perfusion following major trauma and burns [1,3].

ICG has been approved by the FDA and is safe for use in humans. It can be administered intravenously or intra-arterially. Indocyanine green (ICG) is a tricarboyanine compound which is soluble in water and has a high safety profile. It absorbs Near infrared light at a wavelength of approximately 750–800 nm and re-emits it as fluorescence at an approximate wavelength of 830 nm. The rapid and extensive binding of this compound to the plasma proteins ensures its confinement to the intravascular compartment with little or leakage to the interstitium thereby making it an ideal fluorophore for use in angiography. Since it is nontoxic and nonionizing, it has a good patient safety profile and can be administered for clinical use with a lower risk of side effects. It is metabolized by the liver within 3–5 min and excreted in bile with no known metabolites. ICG is safe to be administered repeatedly due to its short half-life. Documented allergy to iodides or iodinated imaging agents are absolute contraindications to ICG as sodium iodide contributes 5% of its total organic composition. Anaphylactic reaction is a life-threatening side effect of ICG although very few reported cases are

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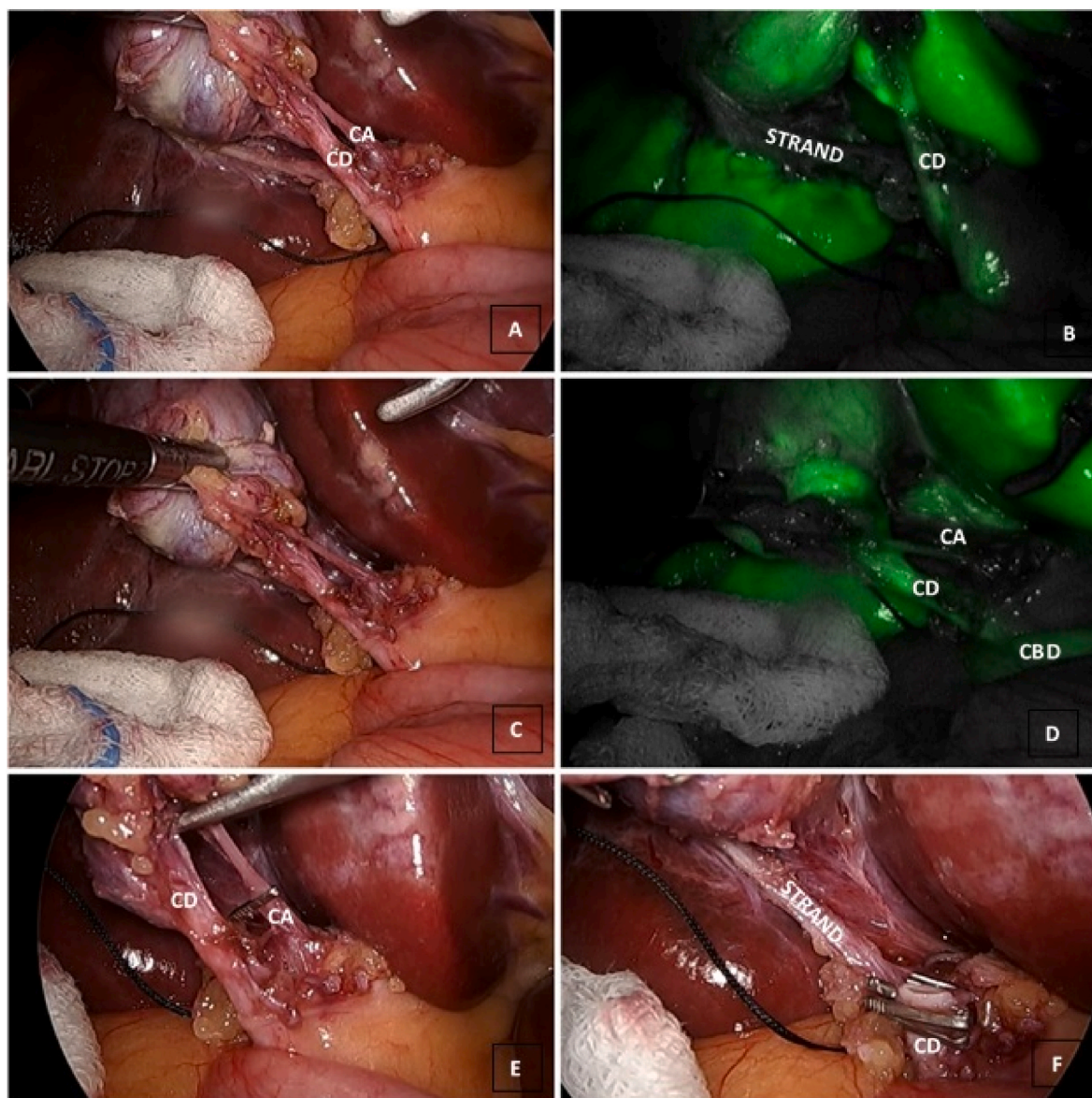


Fig. 1. A case of cholelithiasis with biliary colic undergoing Laparoscopic cholecystectomy with the aid of NIV mode. (A) Showing the Calot's triangle with a tubular structure noted in the posterior aspect, which would be easily confused as the cystic duct. (B) NIV mode with green fluorescence differentiating the cystic duct from the strand. (C) Showing the Calot's triangle. (D) NIV mode identifying the green fluorescence in the cystic duct further visualizing the CBD, with no uptake seen in the cystic artery. (E) White light mode showing the clipping of the cystic artery. (F) Post clipping and cutting of the cystic duct. CD – Cystic duct, CA – Cystic artery, CBD – Common Bile duct (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

documented [1,5,6].

The laparoscopy system used was STRYKER 1588 AIM with Near Infrared Visualization (NIV mode) with STRYKER precision LED light source (Stryker Endoscopy, San Jose, CA). The laparoscopic camera comes with a dual inbuilt white light and near infrared mode which has the ability to detect light at a wavelength of 823 nm. Structures which take up the ICG dye fluoresce bright green when subjected to NIV mode against a black backdrop.

In this case series, we aim to evaluate the application and efficacy of indocyanine green (ICG) for fluorescence imaging in different commonly performed procedures in surgery.

2. Materials and Methods

2.1. Population and study design

Patients admitted for both elective and emergency cases were

considered and ICG was administered in individualized doses to assess the potential applications. This study is a single tertiary center case series carried out between 2019 and 2022 and all the procedures were performed by senior consultant surgeons of our institution. This work has been reported in line with the PROCESS guidelines [7] and ethical approval was received from our institutional ethical committee bearing number IEC KMC MLR 09/2021/287. The procedures were recorded and the video reviewed by the consultant surgeons.

2.2. Inclusion criteria

All patients aged 18 years and above presenting with a newly diagnosed surgical problem with indications falling within the objectives of the study were considered. Written informed consent was obtained from all the participants. Preoperative risk factors such as the age and sex of a patient, presence of any comorbidities, prior treatment history (steroids, radiation, transfusion) were recorded. The American Society of

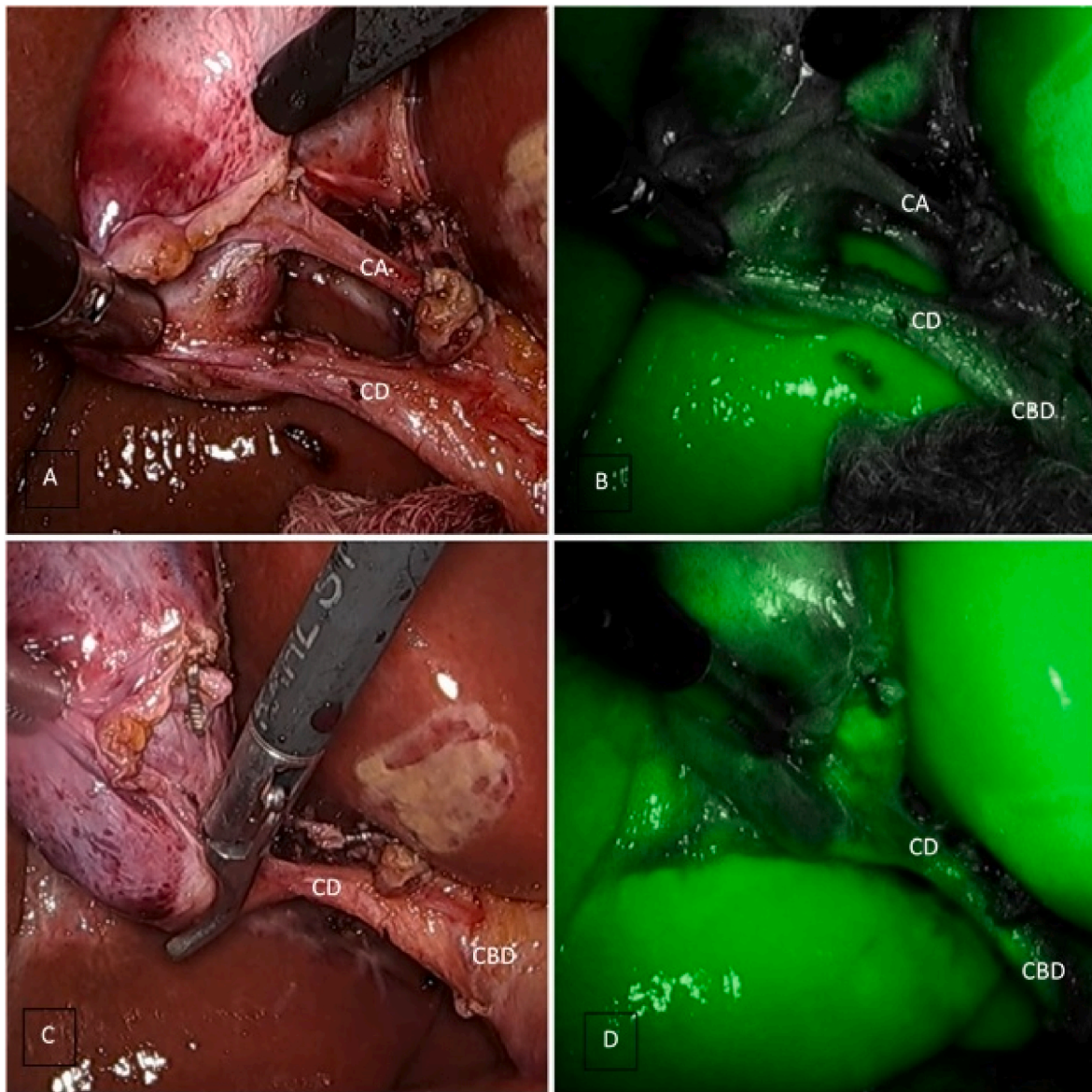


Fig. 2. Case of Chronic Cholecystitis with Cholelithiasis and Choledocholithiasis status quo ERCP and Stenting undergoing interval cholecystectomy. (A) Calot's triangle with Critical View of Safety. (B) NIV mode showing green fluorescence confirming the cystic duct from the cystic artery. (C) Clipping of the cystic duct safeguarding the CBD. (E) Green fluorescence taken up by the gall bladder, cystic duct and CBD as seen in NIV mode. CD – Cystic duct, CA – Cystic artery, CBD – Common Bile duct, ERCP – Endoscopic retrograde cholangiopancreatography. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

Anesthesiologists (ASA) score was also recorded.

2.3. Exclusion criteria

History about hypersensitivity reaction to ICG dye and/or compounds containing iodine. Pregnant and lactating mothers were excluded.

2.4. Description of procedure

ICG dye (Aurogreen, Aurolab, Madurai, India) consists of 25 mg of dye in a powdered form, with 5 ml distilled water for reconstitution. The dye was reconstituted and different doses were instilled for various indications, after appropriate anesthesia. Laparoscopy system used was STRYKER 1588 AIM with Near Infrared Visualization (NIV mode) to visualize the fluorescence.

1. To visualize the biliary tree, 5 mg (1 ml) of reconstituted dye was given to patients intravenously 2 h prior to the incision time of Laparoscopic cholecystectomy and critical view of safety was identified.
2. Patients undergoing colorectal surgeries with intestinal resection and anastomosis, 7.5 mg (1.5 ml) of ICG dye was injected intravenously after intestinal resection and the perfusion of the anastomotic ends was assessed.
3. Patients presenting with complicated (incarcerated/strangulated) hernia to assess the bowel viability, 7.5 mg (1.5 mL) of ICG dye was administered IV.
4. Oncological cases presenting for adequate clearance, 5 mg (1 ml) of ICG dye was injected in the peri tumor region/peri areolar region for sentinel lymph node mapping. 10 min post injection, the local lymph node basin was explored to identify the sentinel node.
5. Patients with peripheral arterial occlusive disease presenting with gangrene to assess the vascularity of the limb, level of amputation

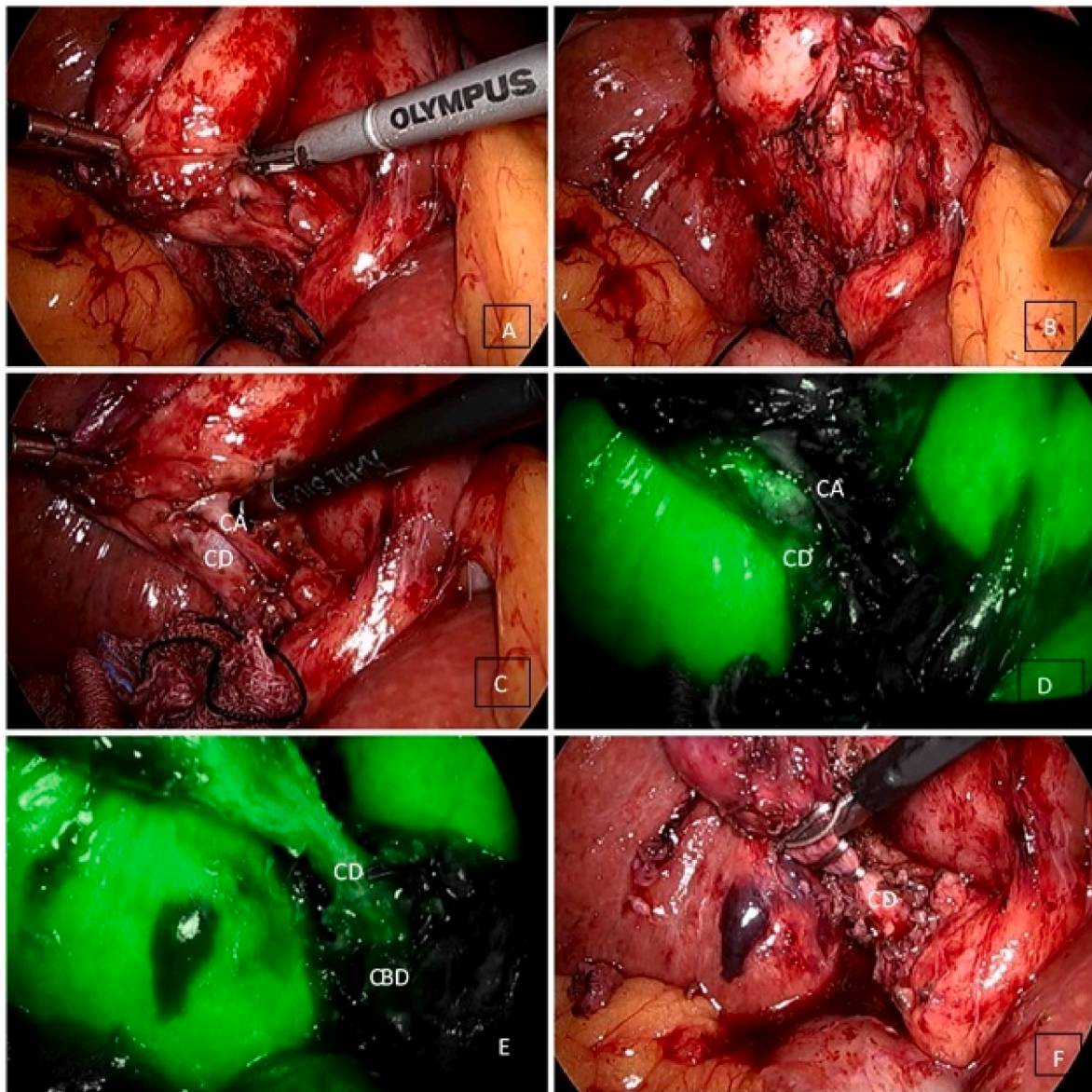


Fig. 3. A case of Acute cholecystitis undergoing Laparoscopic cholecystectomy. (A) & (B) showing the dense adhesions and inflamed gall bladder with no identifiable anatomy. (C) After dissection, two tubular structures identified near the base of the gall bladder. (D) and (E) NIV mode identifying the cystic duct from the cystic artery, which shows bright green fluorescence, which can be further traced down to identify the CBD. (F) Clipping of the cystic duct after confirming its anatomical location.

CD – Cystic duct, CA – Cystic artery, CBD – Common Bile duct. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

and healing of stump coupled with vascular bypass interventions, 7.5 mg (1.5 ml) of ICG was administered.

- Patients undergoing major reconstructive surgeries and local flap procedures following plastic surgical principles were subjected to 7.5 mg (1.5 ml) of ICG to predict flap uptake.

3. Results

3.1. Application of ICG in laparoscopic cholecystectomy

Symptomatic cholelithiasis, Acute and Chronic cholecystitis including complications like perforation, empyema, gangrene was included. Patients with Common bile duct (CBD) calculus were evaluated in the form of Magnetic Resonance Pancreatography (MRCP) and posted for Endoscopic Retrograde Cholangiopancreatography (ERCP) and stenting. These patients were then taken up for Interval

Cholecystectomy after 6 weeks.

A total of 70 laparoscopic cholecystectomies were performed in our hospital over a span of 1 year. 42 of them were females and 28 of them were males. Symptomatic Cholelithiasis was the indication for performing Laparoscopic cholecystectomy in 35 of these patients, 12 of them had Acute cholecystitis with inflamed and edematous gall bladder, 8 with Chronic cholecystitis. 15 of them presented with Cholelithiasis with obstructive jaundice who subsequently underwent ERCP and stenting to achieve biliary clearance and then taken up for interval cholecystectomy 6 weeks later.

All patients were intravenously injected with 5 mg (1 mL) of reconstituted ICG dye 2 h prior to the incision time of the laparoscopic procedure. The procedure was then performed using standard laparoscopic cholecystectomy principles. Calot's triangle was dissected to attain Critical View of Safety before proceeding with the clipping of the Cystic Artery and Cystic Duct. In cases where dissection was difficult

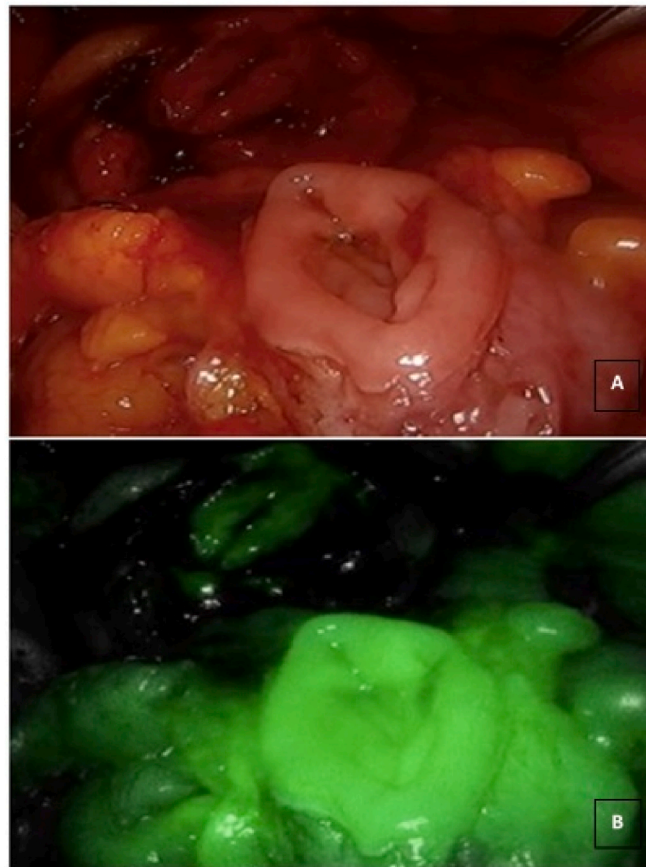


Fig. 4. Case of Carcinoma sigmoid colon who underwent left radical hemicolectomy with colorectal anastomoses. (A) Distal end of resected bowel. (B) Good uptake seen in NIV mode after ICG injection.

with dense adhesions, fundus first approach was employed to achieve a secure dissection.

Using the NIV mode of the laparoscope, bright green fluorescence was seen delineating the gall bladder and biliary tract confirming the location of the Cystic Duct and CBD, however no fluorescence was seen in any of the non-biliary structures, thereby aiding in the accurate anatomical location of these structures (Fig. 1).

Critical view of safety was achieved to identify the two structures entering the gall bladder. Cystic duct was confirmed with the aid of the NIV mode of the laparoscope which had a bright green fluorescence, whereas cystic artery had no uptake (Fig. 2). This was performed in every case of laparoscopic cholecystectomy prior to clipping and cutting of the cystic artery and duct. The CBD was safeguarded in each of these cases by tracing the fluorescence beyond the Rouvier's sulcus of the liver and any clip deployed was placed above it to prevent CBD injury.

We report accurate localization of the biliary tree in 100% of the patients surveyed regardless of the complication status (Fig. 3). ICG had also precisely identified the CBD thereby preventing untoward CBD injury. ICG had also aided the surgeon in identifying any variable biliary anatomy further assuring the surgeon in preventing any unpredictable injury.

3.2. Application of ICG in colorectal surgeries for assessing vascular perfusion of anastomotic ends

A case series was done to evaluate if the incidence of anastomotic leak after colorectal surgeries is altered following routine use of ICG-FA.

Patients planned for elective procedures involving resection and anastomosis of large bowel were included in the study and were

preoperatively evaluated for the resectability of the disease (involving a CECT abdomen and chest and colonoscopy with biopsy).

6 cases of resection and anastomosis were documented (including 3 males and 3 females, with an average age of 60.1 years), out of which, resection for malignancy was the indication in 5 patients and 1 patient presented with a colo-vaginal fistula and underwent sigmoid resection and anastomosis for the same.

All these patients, undergoing resection and anastomosis were subjected to ICG-enhanced fluorescence, wherein 7.5 mg (1.5 ml) of ICG dye was injected intravenously after intestinal resection and the time of the injection was recorded, followed by introduction of a laparoscope in NIV mode for visualization of perfusion after 1 min of the injection and the perfusion of the bowel was assessed subjectively depending on the intensity of the fluorescence (green color) (Fig. 4). Since the intensity of the fluorescence cannot be determined objectively, a subjective assessment of bowel perfusion under standard white light by the surgical team is done and indicated as 'adequate' (uniform to that of proximal colon) or 'insufficient'. A leak test was performed, and vascularity of the bowel re-evaluated in the NIV mode, following the completion of anastomosis.

Postoperative evaluation of anastomotic leak was done clinically by assessing the presence of features of infection (temperature, tachycardia, tachypnoea, hypotension), severe abdominal pain, abdominal distension, which was then followed up and co-related with laboratory and ultrasound findings.

In one patient, perfusion was judged to be inadequate, and a re-resection of the bowel was performed followed by a re-assessment of perfusion before anastomosing (Fig. 5). All the cases (100%) showed ICG-enhanced fluorescence and any changes in the surgical plan were considered only after performing fluorescence angiography. None of

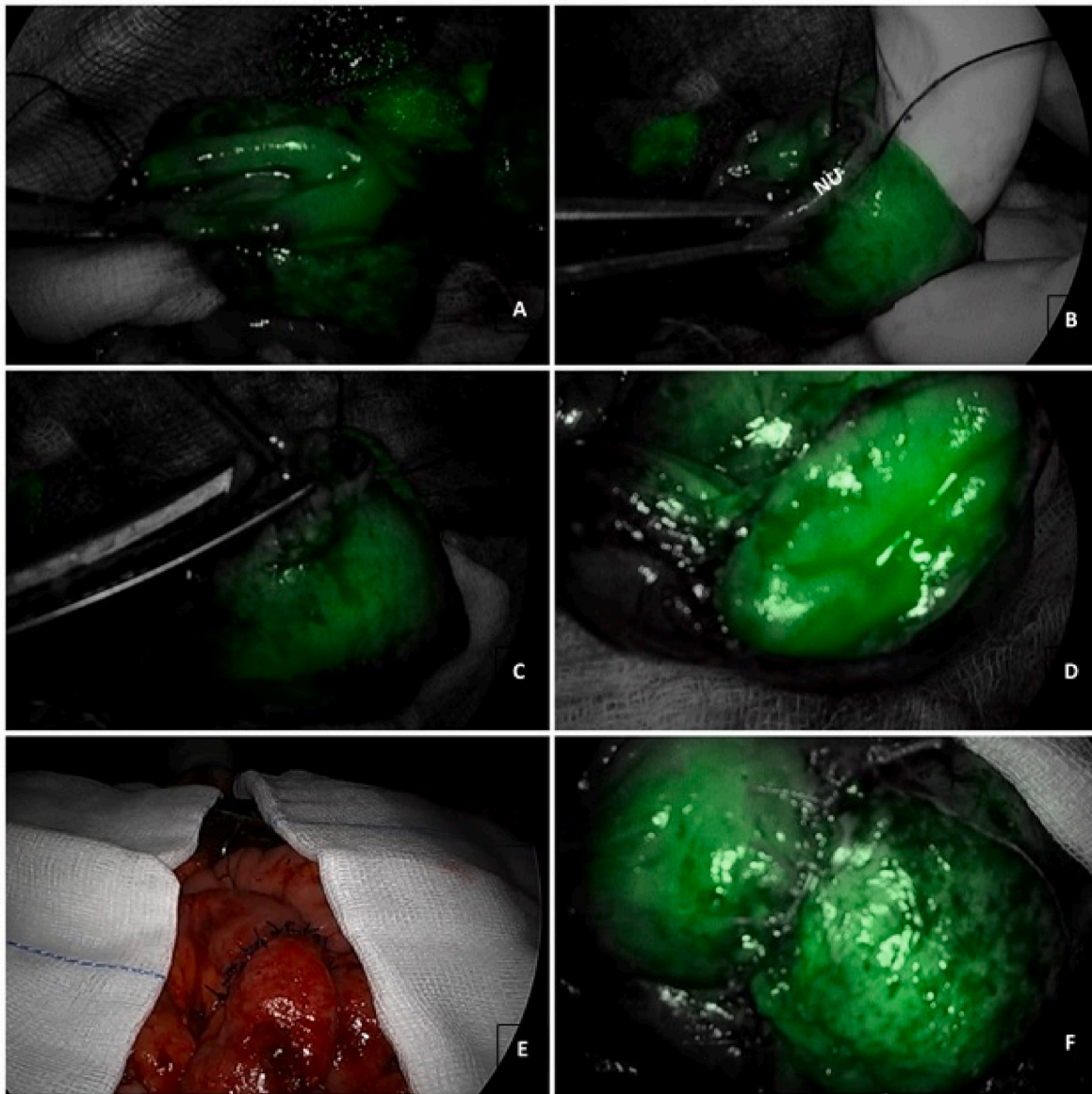


Fig. 5. Case of Carcinoma caecum undergoing Radical Right Hemicolectomy. (A) NIV mode showing the distal resected end of transverse colon. (B) No uptake on the antimesenteric border of the resected bowel segment. (C) Excision of the bowel edge showing poor uptake. (D) Post excision of the bowel edges, showing good uptake and vascularity on the entire circumference of the resected bowel. (E) Ileo-transverse anastomoses done. (F) NIV mode showing good uptake on either ends of the anastomotic suture line.

NU – No uptake.

these patients had clinical anastomotic leaks in the postoperative period.

3.3. Application of ICG in cases of hernia to assess bowel viability

A case series was done to evaluate the intraoperative usage of ICG dye in the determination of bowel viability in cases of incarcerated hernia.

Five patients presenting with incarcerated hernias were studied. All of these patients underwent objective assessment of the bowel to look for viability. 7.5 mg (1.5 mL) of ICG dye followed by a 10 ml normal saline flush were administered IV. Post injection of ICG, fluorescence was observed within 60sec, with areas of good uptake showing bright green fluorescence and poor uptake being visualized as dark areas.

In two of the patients, visual inspection following the reduction of the incarcerated bowel showed the presence of an edematous bowel with a deep red color and the peristalsis could not be assessed. Following the administration of ICG, good fluorescence was observed under NIV

mode, and hence resection and anastomosis was avoided (Fig. 6).

In one of the cases, patient had presented with irreducible complete inguinal hernia. On exploration, small bowel loops were found to be densely adherent to each other which couldn't be separated individually. Hence decision was taken to proceed with resection and anastomoses. Post resection, distal and proximal resected end of the bowel appeared viable on visual inspection. However, on instillation of ICG, distal resected segment of bowel showed no uptake on NIV mode, thus leading to a decision of performing re-resection. Prior to proceeding with anastomoses, viability was reconfirmed with ICG which showed good uptake in both the resected ends, which was rechecked post anastomoses (Figs. 7 and 8).

3.4. Application of ICG for the mapping of sentinel lymph node

A case series was done to assess the use of ICG for the mapping of sentinel lymph nodes.

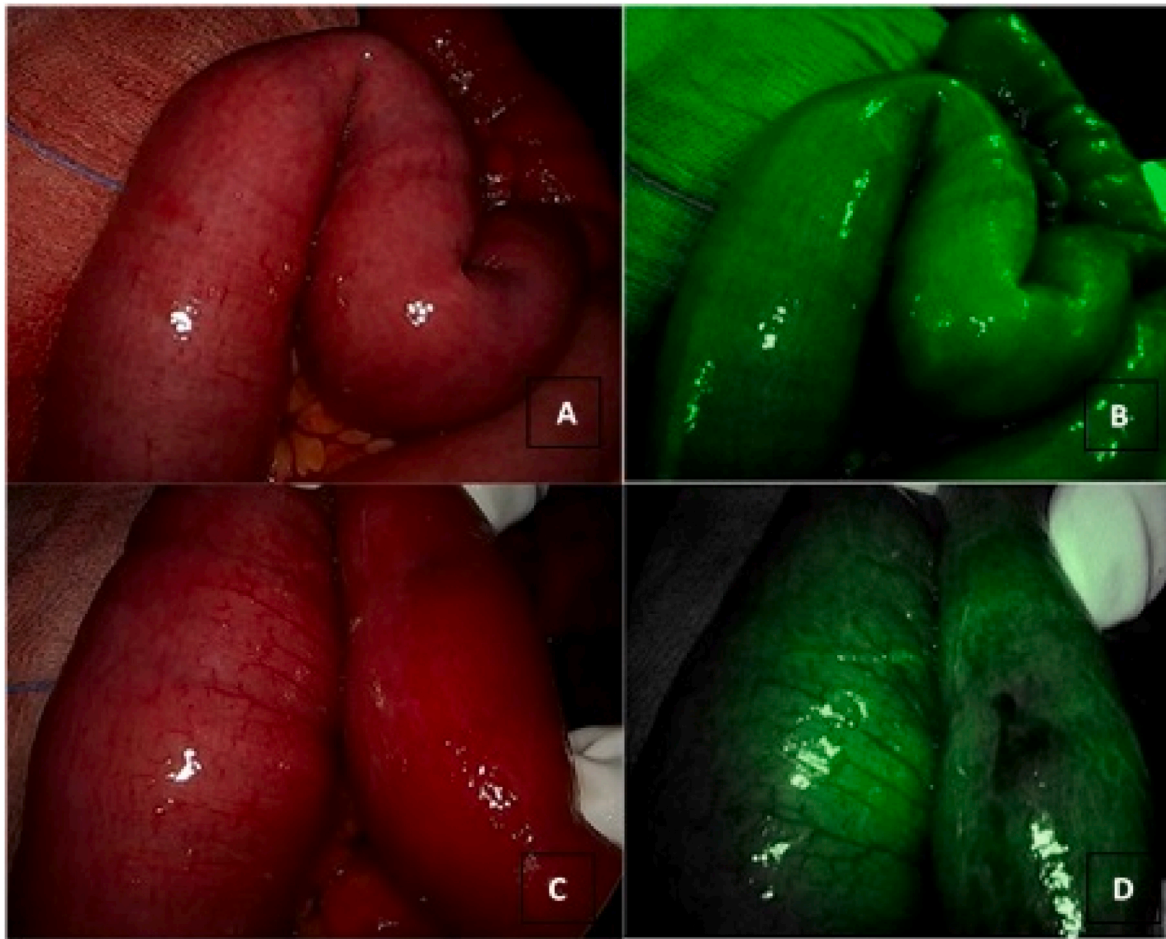


Fig. 6. Case of Right complete irreducible inguinal hernia who underwent inguino-scrotal exploration with small bowel loops as content. (A) & (C) Dilated, edematous and congested ileal loops. (B) & (D) Good uptake seen on the bowel wall using ICG in all the contents of the hernial sac, thereby avoiding resection.

We had employed the applications of ICG in 6 cases of biopsy proven Breast Cancer, 5 cases of Oral Malignancies and 2 cases of skin malignancy of the extremities.

All these patients, undergoing cancer clearance were subjected to ICG-enhanced fluorescence, wherein 5 mg (1 ml) of ICG dye was injected in the peri tumor region/peri areolar region. 10 min post injection, the local lymph node basin was explored to identify the sentinel node. The procedure of clearance of the primary tumor was done following standard oncologic clearance protocols. The regional nodal basin was then subjected to the introduction of a laparoscope in NIV mode for visualization of the Sentinel node, which was identified as a bright green fluorescence. The regional lymph node dissection was then undertaken following the general oncological principles, with the echelon node as a guide for optimal lymphatic clearance.

Among the 6 cases with Biopsy proven Breast cancer, all of them had undergone a Breast Conservation Surgery/Quadrantectomy with the identification of the sentinel axillary lymph node. ICG was injected and the echelon node was identified as a bright green fluorescence (Figs. 9 and 10). Using this fluorescence as a guide, these women underwent Axillary Lymph node dissection, and all the echelon nodes were cleared and sent for histopathological examination. Drains were placed in the axilla which was removed on POD 3. None of the patients had any wound complications and there were no reported cases of seroma formation. The histopathological examination revealed all the 6 cases to have no evidence of Lymph node involvement with all the women graded as an N0 axilla.

A case series of 5 patients with biopsy proven oral malignancies involving tongue, buccal mucosa and lip were studied. These patients clinically had no palpable cervical neck lymph nodes. The procedure for the primary was carried out using standard oncological principles and ICG was given to recognize the sentinel lymph node. The sentinel nodes were identified and subjected to biopsy (Fig. 11). The HPE report of all the 3 cases revealed no lymph node involvement, thus avoiding a formal neck dissection in these patients. However, in 2 of the cases of oral malignancy (tongue), the sentinel lymph node biopsy was positive, with these patients undergoing a formal supraomohyoid neck dissection to clear off the draining lymph node basin.

We also studied 2 patients with skin malignancy, squamous cell carcinoma of the left thumb and a malignant melanoma of the right index finger. Both patients had no clinically palpable axillary lymph nodes and were taken up for amputation of the digits with sentinel lymph node biopsy. ICG was injected and the sentinel nodes were identified in the axilla (Fig. 12), which were subjected to a biopsy. None showed evidence of metastatic deposits.

3.5. Application of ICG in cases of amputation to assess stump vascularity and level of amputation

Case of a 57-year male who is a chronic smoker with chronic limb ischemia with dry gangrene of the left 4th toe. The patient is a diabetic for 10 years, which is well controlled. He had undergone Rays amputation of the 5th toe in view of dry gangrene at a local hospital before

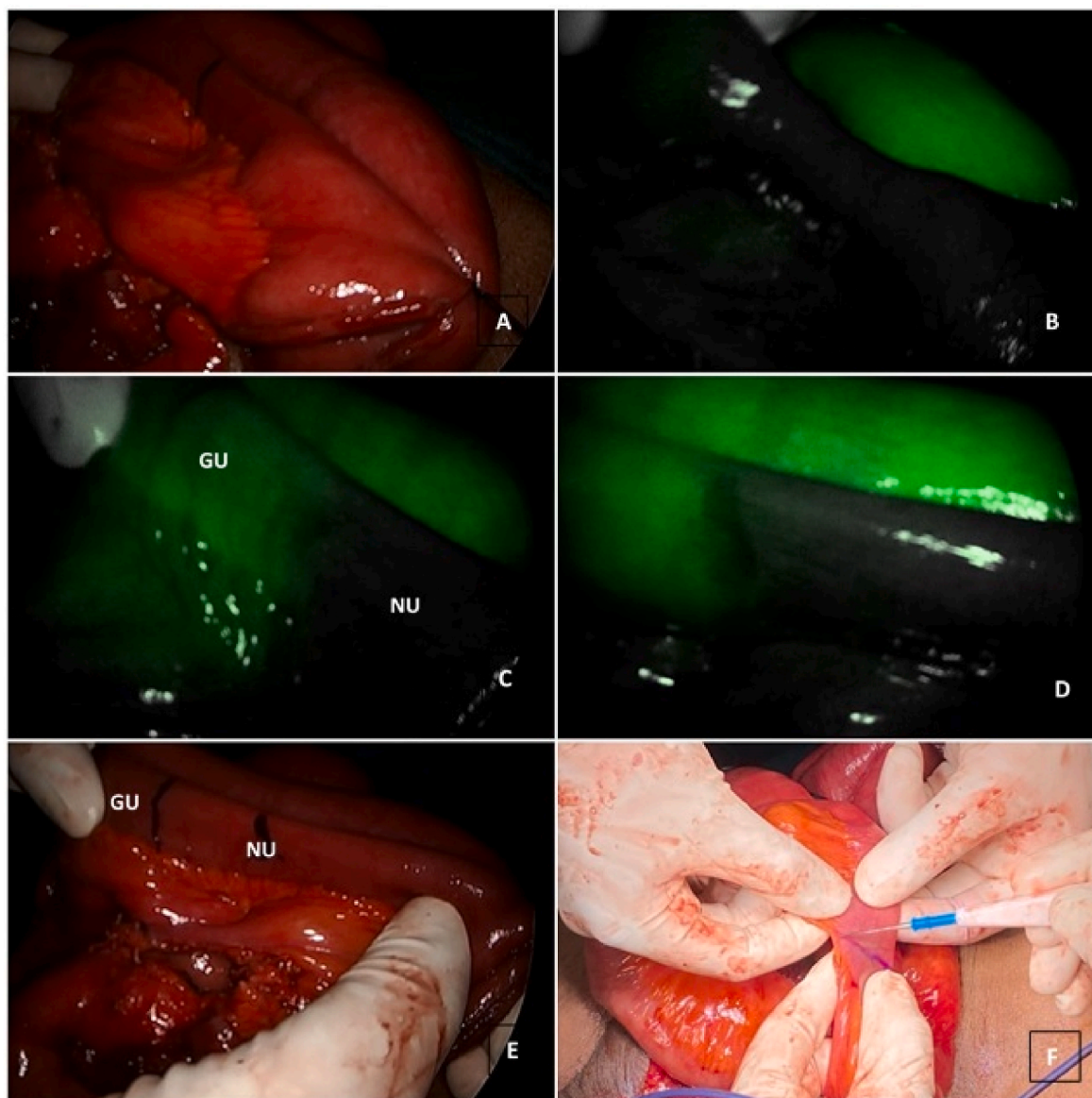


Fig. 7. Case of Right Complete irreducible inguinal hernia with features of obstruction who underwent inguino-scrotal exploration. 15 cm of Ileum with dense fibrous adhesions with clumped bowel were the contents of the sac which could not be separated individually, approximately 10 cm from the ileocaecal junction. Patient had to undergo resection of the adherent ileal loops with end to side anastomoses. (A) Proximal and Distal end of the ileum on white light. (B), (C) & (D) NIV showing no uptake in the distal 7 cm of the resected ileum, with clear line of demarcation which has been marked. (E) White light showing the good vs no uptake segments, which look morphologically normal on naked eye examination. (F) Resection of the segment along the line marked. GU – Good uptake, NU – No uptake.

being referred to our center. Clinically there were absent Popliteal, ATA, PTA, and DPA pulses, and the limb was cold till mid leg level. The patient was evaluated by a CT aortogram with bilateral lower limb angiogram which showed stenosis of the left proximal and mid superficial SFA causing 50% narrowing with normal distal flow and mildly reduced calibre of the distal left SFA, Popliteal artery, ATA, PTA, PA and DPA with normal contrast opacification. The aorta visualized was normal.

The patient was planned for a left Femoro-Popliteal bypass using a synthetic Dacron graft. 7.5 mg of ICG was injected to visualize vascularity in the ischemic limb prior to the bypass, thereby serving as a baseline. The areas of demarcation between the well and poorly vascularized segments proximal to the gangrenous toes were also identified to serve as a marker of level of amputation.

After a successful left femoro-popliteal bypass, ICG was re administered to identify the improved vascularity in the left limb. The amputation of the 4th toe was done at a level identified by the demarcation

line and the stump was sutured. The results obtained were tabulated.

PRE-BYPASS		
LOCATION	TIME WHEN ICG DETECTED	TIME WHEN MAXIMALLY DETECTED
Dorsum	49 sec	1min 32 sec
Thigh	4min 20sec	6min 30sec
POST BYPASS		
Dorsum	40sec	1min 03sec
Thigh	2min 16sec	4 min

As shown above, there was a significant improvement in the vascularity of the limb with faster perfusion of the ICG to the re vascularized limb. Post operatively, the limb was monitored clinically with good capillary perfusion, warmth and absence of rest pain. The amputation stump healed well with no evidence of flap necrosis (Fig. 13). The patient is on regular follow up and has no wound site complications.



Fig. 8. (G) & (H) Distal resected end and end to side anastomoses performed. (I) NIV showing good uptake across the suture line and on either end of the anastomoses. (J) Resected specimen showing the dense adhesions and clumping.

3.6. Application of ICG in assessing flap vascularity and uptake

A case series was done to assess the vascularity of flaps and its success. 9 cases of biopsy proven Breast Cancer and 3 cases of Oral Malignancy were taken into the study. 7.5 mg (1.5 ml) of ICG followed by 10 ml of normal saline flush was done to visualize the green fluorescence on the edges of the skin margins using NIV mode.

Females with both Early breast cancer and Locally advanced disease post neo adjuvant therapy were taken up for radical mastectomy and reconstruction of the chest defect using local advancement flaps. All these females were subjected to a lengthy oblique incision extending from the left infraclavicular region to the left anterior axillary line across the breast. After adequate oncologic clearance, patients were planned for primary closure of the wound, to approximate the defect. Keeping in mind the extensive nature of resection, the bulkiness of the tumor and the need for future post-operative radiotherapy to the chest wall and the breast bed, the patient was subjected to ICG to assess the vascularity of the flap edges.

A case series of 3 patients with biopsy proven oral buccal mucosa

malignancy were taken up for composite resection with radical neck dissection along with reconstruction of the facial defect using pectoralis major myocutaneous flap (PMMC). ICG was given to assess the vascularity of the pectoralis muscle and the its cutaneous component.

In both these series, areas showing absent/poor fluorescence were marked, excised and freshened, till the area of bright perfusion with brisk bleeding. The wound edges were approximated using 2-0 nylon mattress sutures after confirming all the edges of the wound, circumferentially, showed good fluorescence (Fig. 14). All the flaps were monitored regularly in the post-operative period for any evidence of flap necrosis, failure and wound dehiscence (Fig. 15) (Fig. 16).

4. Discussion

4.1. Application of ICG in laparoscopic cholecystectomy

Laparoscopic cholecystectomy is considered to be the gold standard and most commonly used surgical technique in cases of cholelithiasis, with injury to bile duct being one of its complications, with an overall

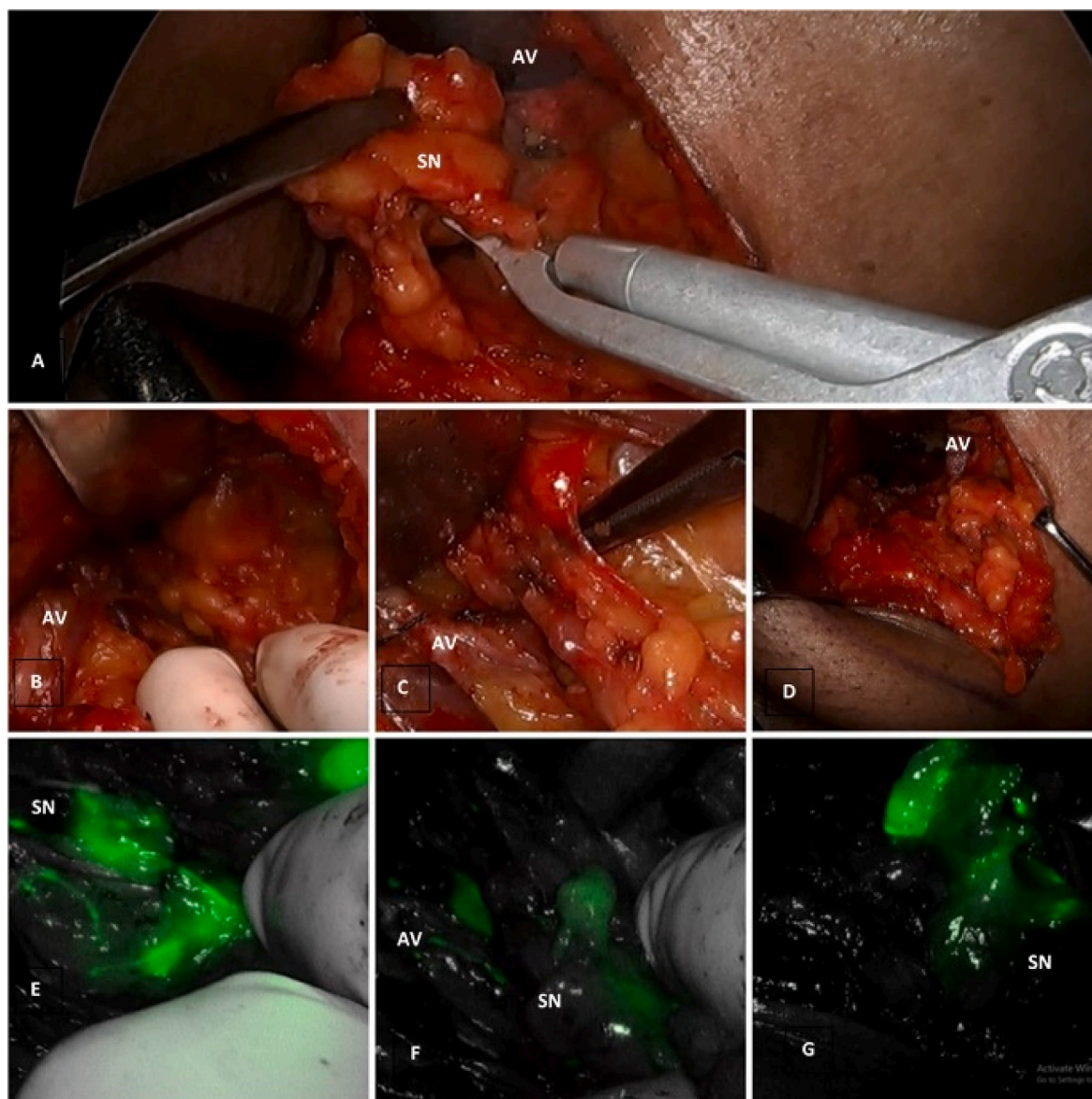


Fig. 9. Case of biopsy proven Infiltrating ductal carcinoma of the left breast T2N0M0 planned for left Breast conservation surgery with Sentinal lymph node biopsy using ICG. (B), (C) & (D) White light image showing Anterior and Central group of lymph nodes along the axillary vein which clinically appeared normal and unenlarged. However, when subjected to NIV mode, they showed bright green fluorescence suggestive of reactivity in those nodes as seen in (E), (F) & (G). These echelon nodes were then cleared as shown in (A) and sent for histopathological examination.

AV – Axillary vein, SN – Sentinal node. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

prevalence of 0.4%, which can increase up to 4% in the presence of acute cholecystitis [8,9]. Biliary anatomy is highly variable with a number of variations in the biliary tree, which serve as a challenge in performing safe cholecystectomy.

While intraoperative cholangiography is the standard procedure for biliary imaging [10], it is a time-consuming procedure with additional risks of injury to bile duct during catheterization and radiation exposure to patient and OT staff. Moreover, presence of dense adhesions in the cysto-hepatic triangle may prevent the delineation of the cystic duct, which is a prerequisite to dye injection, thus leading to failure of the procedure.

ICG with the NIV mode had helped in performing a safe cholecystectomy and significantly reduced the chances of biliary injury by accurately identifying biliary structures. It had also assisted in evaluating aberrant biliary anatomy and preventing any unforeseen complications.

4.2. Application of ICG in colorectal surgeries for assessing vascular perfusion of anastomotic ends

The most significant complication following a colorectal resection and anastomosis procedure is anastomotic leak, with a reported incidence of 1–30% and accounting for 1/3rd of deaths following a low anterior resection, with a further increase in cases of intraperitoneal leak [11,12].

Several iatrogenic and patient-related factors are associated with an increased risk of anastomotic leak; however, the single most important and modifiable factor is the inadequate flow of blood to the bowel, thus necessitating the need for evaluation of bowel perfusion, viability and anastomotic integrity to potentially improve the rate of anastomotic leaks [13].

Conventionally, evaluation of bowel is based on intraoperative clinical assessment by the surgeon via visual inspection of gross findings such as color, pulsations, peristalsis, bleeding from cut edges; however

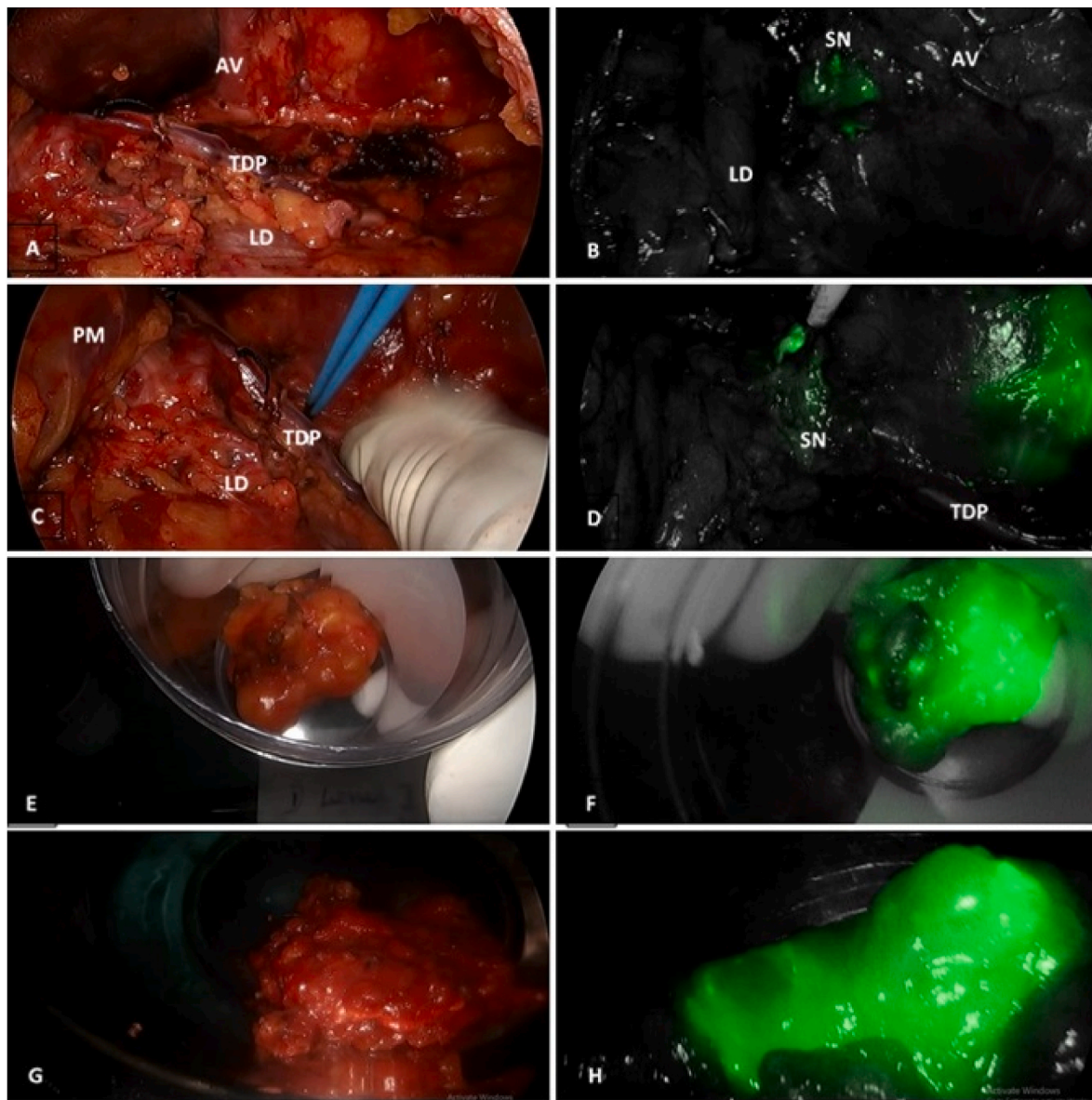


Fig. 10. Case of biopsy proven Infiltrating Ductal carcinoma of right breast T1N0M0 was taken up for a right Breast Conservation surgery with Sentinal lymph node biopsy using periareolar injection of ICG. (A) Axillary anatomy showing no gross abnormally enlarged nodes. (B) NIV mode showing central group of nodes along the axillary vein with green fluorescence suggestive of reactivity. (C) and (D) showing the removal of these reactive nodes as seen on white light and ENC mode. (E) & (G) showing the removed nodes after dissection. (F) & (H) Green fluorescence seen in the nodes post excision depicting the echelon nodal status and then subjected for HPE.

AV – Axillary vein, SN – Sentinal node, TDP – Thoracodorsal pedicle, LD – Latissimus dorsi, PM – Pectoralis major. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

prospective studies show that such a method has a low predictive accuracy and lacks objectivity and value for the development of an anastomotic leak [14], thereby leading to an increase in the use of ICG dye for the assessment of perfusion of anastomotic ends in colorectal surgeries.

ICG had successfully evaluated the perfusion of the anastomotic ends of bowel by subjectively showing bright green fluorescence and hence being an indirect indicator of the vascularity. This has significantly reduced the rates of anastomotic leaks and improved the anastomotic success in colorectal anastomoses.

4.3. Application of ICG in cases of hernia to assess bowel viability

Incarcerated hernia commonly presents with features of intestinal obstruction; with a mortality rate of around 3%, which may rise up to

20% in cases of bowel resection, especially in elderly patients, and thus requires urgent intervention in the form of an exploratory laparotomy procedure for the release of the constriction ring and for determining the viability of the bowel [15,16].

As with all surgeries involving the bowel, visual inspection under standard white light is the traditional method to assess perfusion and viability based on color, pulsations, peristalsis, bleeding from cut edges, however, it is an unreliable method due to low accuracy of predicting ischemia and the decisions of different surgeons are vastly subjective [14, 15]. Other methods which have been used for evaluating perfusion and have subsequently been found unsatisfactory include pulse oximetry, doppler ultrasound, radioisotopes, hydrogen gas clearance etc. [16].

Incarcerated hernia serves as a challenge for the operating surgeon as the decision to resect the oedematous and inflamed hernial contents always is in dilemma, as this is highly objective and not truly

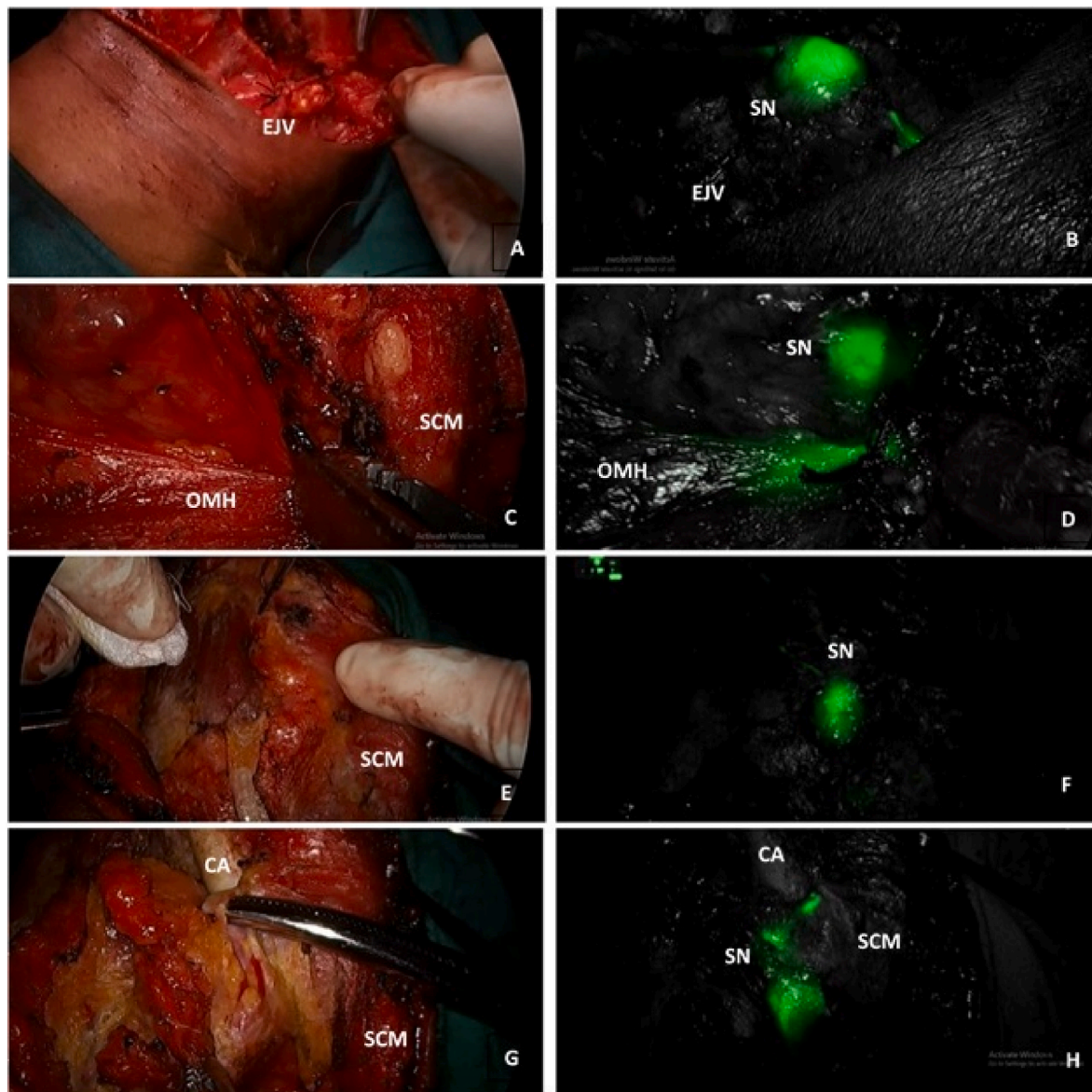


Fig. 11. A case of Squamous cell carcinoma of Left lateral border of tongue T1N0M0 who underwent wide local excision of the primary and subjected for left sentinel node biopsy of cervical node using peritumoral injection of ICG. (A), (C), (F) & (G) White light image showing the left anterior triangle of the neck in the region of the level III node close to the carotid sheath underneath the ligated EJV and deep to the SCM. These nodes appeared morphologically normal and unenlarged. (B), (D), (F) & (H) NIV mode identifying the echelon node of the level III nodal basin and subjected for histopathological examination.

EJV – External jugular vein, SCM – Sternocleidomastoid, CA – Carotid artery, SN – Sentinel node, OMH – Omohyoid.

representative of the vascularity. The application of ICG has therefore avoided unnecessary resections in such patients by accurately predicting the vascularity of the hernial contents and thereby avoiding an extensive intervention, as shown in our series of cases. ICG had therefore aided in demonstrating perfusion of the segments and reducing the risks of a missed ischemia.

4.4. Application of ICG for the mapping of sentinel lymph node

As the lymphatic system is one of the major routes via which a malignancy spreads, most cancers disseminate first to the regional nodal basin. Sentinel node is the first point of entry to a nodal basin. Therefore, for a cancer to metastasize to the nodal basin, it must first pass through the sentinel node; hence leading to its importance in assessing the nodal status, which in turn aids in proper staging of the disease, along with predicting outcome and need for adjuvant treatment [1,17]. Initially developed for assessing nodal metastasis in cases of parotid carcinoma

[18,19]; sentinel lymph node biopsy has now gained widespread usage as a minimally invasive method for the assessment of lymph node status in various cancers such as penile cancer, breast cancer, melanoma, head and neck tumors, as well as gastrointestinal and endocrine tumors [18].

Currently, the methods commonly employed for the mapping of the sentinel lymph node include-

1. Blue dye technique (methylene blue is more commonly used as compared to isosulfan blue and patent blue due to lesser restrictions as compared to the latter two, such as life-threatening anaphylaxis).
2. Radionuclide method involving the injection of technetium 99 tagged sulfur colloid [17].

However, there are several drawbacks associated with both the methods. The blue dye technique carries the risk of anaphylaxis, skin tattooing, skin necrosis and bluish discoloration of urine. Moreover, the rate of detection of the sentinel node with the blue dye method alone are



Fig. 12. A case of Squamous cell carcinoma of the left thumb with no palpable axillary nodes planned for carpometacarpal joint dislocation of the thumb along with axillary sentinel lymph node biopsy. (A) Peritumoral injection of ICG. (B) Line of proposed amputation site. (C) & (D) Axillary exploration being done to identify the sentinel node which was then excised and subjected to histopathology. (E) & (G) White light image of the pre and post amputation of the thumb. (F) & (H) NIV mode showing good fluorescence suggestive of the vascular status of the thumb pre and post amputation.
SN – Sentinel node.

relatively lower, which may have a considerable impact on the prognosis of the patient [20]. Similarly, the need for expensive equipment, handling and disposal of radioactive substance and the risk of radiation exposure to healthcare workers and patients are a few of the deterrents to the radionuclide method [17,21,22].

ICG had accurately identified the most reactive node in the nodal basin, which on further HPE revealed the presence/absence of metastases thereby confirming the utility of ICG in detecting such echelon nodes. The absence of metastases in the sentinel node, as described above, provided us with evidence of probable absence of metastases in the other lymph nodes and thereby avoided a formal lymphatic clearance. This avoided majority of the post-operative complications associated with an extensive lymph node dissection such as seroma formation, hematoma, neurovascular injury, lymphedema, flap necrosis.

4.5. Application of ICG in cases of amputation to assess stump vascularity and level of amputation

Peripheral arterial occlusive disease is commonly encountered in the surgical OPD, with majority of the patients presenting with gangrene, ischemic ulcer and pain. In view of the chronic arterial occlusion, there is reduced vascularity and healing of such an ulcer and gangrene is always in doubt. This becomes pronounced especially when an amputation is planned for such patients and the healing of the stump is hampered due to the poor perfusion, thereby subjecting the patient towards a re-amputation. The vascularity of any given tissue is assessed clinically with good bleeding from the wound edges, skin color along with the warmth of the skin [23–25]. But clinical judgment need not always be accurate and is highly subjective, therefore not always reliable [24,25,27,28]. The level of amputation is always in doubt with the surgeon pushed into a dilemma of not cutting too much versus cutting too little and comprising the healing of the stump. Predicting the healing



Fig. 13. (A) & (B) Chronic limb ischemia showing dry gangrene of the left 4th toe and 5th toe non-healing stump along with ischemic changes on the dorsum of the foot. (C) Post Femoropopliteal bypass showing the incision made for the harvesting of the saphenous vein. (D) Sutured stump site following amputation of the 4th toe and improvement in the ischemic changes post bypass with no evidence of stump necrosis.

of such ischemic limbs is currently the focus on recent ongoing studies. Recently, ICG has been used to visualize tissue perfusion intra-operatively in various surgical fields, including cardiac surgery, transplantation and flap surgery. ICG can also help in determining the extent of debridement required in a necrotic limb [25,26]. We thus employed the application of ICG in detecting regional tissue vascularity in an ischemic limb by providing us a clear visual demarcation between the viable and non-viable tissue and thus predict the future healing. Although CT/MR angiography remains the gold standard to assess the vascular system, ICG can be employed intra operatively to provide a real time visual confirmation on the vascularity and thereby guiding the operating surgeon. It is also economical, nontoxic contrast which can be used in patients with renal conditions rather than CT/MR angiography.

We have evidence showing the successful application of ICG in a patient with Chronic limb ischemia with dry gangrene. It has accurately predicted the regional tissue perfusion and thus can be used as a guide for successful wound healing. Thus, ICG can be used as to assess the vascular supply of an ischemic limb and is relatively safe and easy to perform.

4.6. Application of ICG in assessing flap vascularity and uptake

Reconstruction following major resection surgery is one of the most crucial events for both closures of defects as well as cosmetic purposes. The vascular supply of the flap is the single most important factor determining the healing and uptake of the transferred tissue. A flap is a segment of tissue with its own parent blood supply, the orientation of which has categorized flaps into random, axial or perforator based. Different methods like USG, MRI, CTA have been employed to predict the vascularity of the flap and thereby foresee flap uptake and healing. However, they do not accurately represent the degree of perfusion of the flap and are unreliable in accurately assessing the success of flap surgery [29–31]. As mentioned earlier, the vascularity of any given tissue is assessed clinically with good bleeding from the wound edges, skin color along with the warmth of the skin [23–25].

The use of near-infrared fluorescence imaging has revolutionized the extensive applications of ICG in vascular surgery to study the perfusion of the flap edges, its micro vascularity and thereby to predict the uptake and minimize the possibility of flap necrosis, failure and congestion.

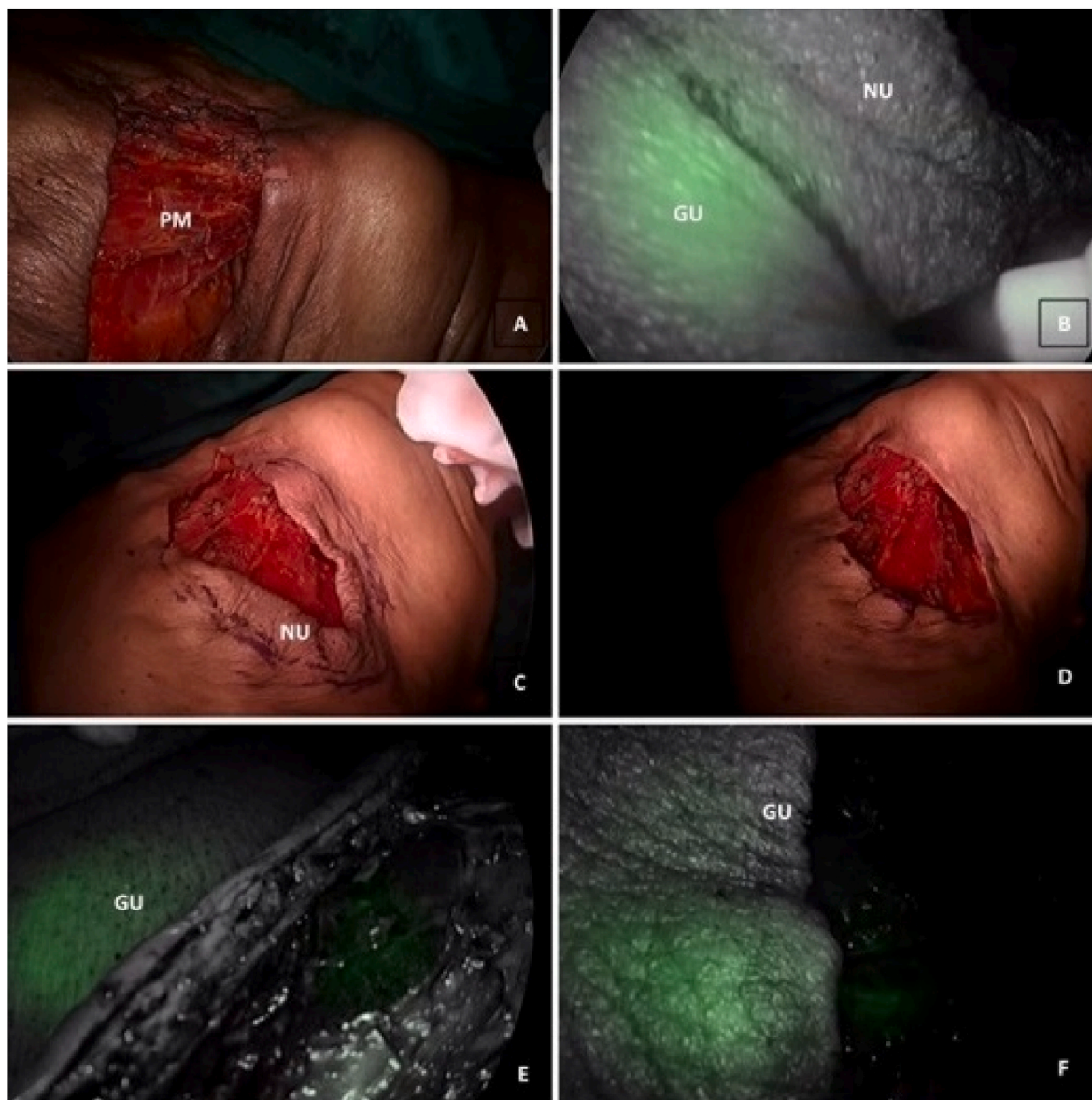


Fig. 14. ICG being utilized to examine the vascularity of skin flap in a case of Carcinoma left breast T3N1M0 who underwent Left modified radical mastectomy. (A) White light image showing the skin edges post mastectomy before primary closure. (B) NIV mode depicting area of no uptake of ICG on the skin edge with the lune of demarcation being marked. (C) & (D) Marked skin area depicting the region of no uptake and post excision of the area. (E) & (F) NIV mode confirming the vascularity of the skin edge before primary suturing.

PM – Pectoralis major, GU – Good uptake, NU – No uptake.

As evidenced by our study, the administration of ICG fluorescence imaging, is a useful and successful accompaniment to wound healing. ICG had accurately predicted the micro vascular supply of the flap and thereby beneficial in successful uptake of the skin flap and thereby cost effective. ICG therefore can be used to assess the flap vascularity, design the flap, evaluate the micro vascular network, to predict the flap uptake, monitor for post-operative complications and to check the lymphatic system function.

5. Conclusion

Fluorescence imaging using ICG-NIV camera has revolutionized common and routinely performed surgical procedures to minimize post-operative complications and providing a superior anatomic visualization of different structures. ICG is easily available, noninvasive and due to its wide margin of safety, can be employed in different surgical scenarios. It remains the only fluorophore to be used in humans. Due to its

short half-life it can safely be used in patients with renal dysfunction and can be employed for multiple administrations. Routine intraoperative use of ICG could pave the way for a more objective assessment of different surgical circumstances and thereby reduce personalized barriers to aciurgy. ICG fluorescence therefore seems to be a promising apparatus in standard general surgical procedures minimizing untoward errors and improving patient conformance.

Ethical approval

Ethical approval was received from the Institutional Ethics Committee of Kasturba Medical College, Mangalore (Reg. no. ECR/541/Inst/K A/2014/RR-20) bearing number IEC KMC MLR 09/2021/287.

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Fig. 15. Post-operative flap inspection of the same patient after the utilization of ICG. (A), (B), (C), (D), (E) & (F) Depicting the flap status on POD 1, 3, 7, 13, 30, 35 respectively.

Author contribution

Dr. Talha Ahmed – Study concept, Study design, Data collection, Writing of original draft, Surgical Operation. Dr. Manohar Pai – Reviewing and final approval of the version to be submitted, Surgical Operation. Dr. Esha Mallik – Study concept, writing and editing of article. Dr. George Varghese - Study concept, writing and editing of article. Dr. Sharad Ashish – Study concept, Data collect and analysis. Dr. Abhijith Acharya – Writing and editing of article, Surgical Operation. Dr. Avinash Krishna – Proof reading the manuscript, editing of article.

Trial registry number

1. Name of the registry: Research Registry
2. Unique Identifying number or registration ID: researchregistry7655
3. Hyperlink to your specific registration (must be publicly accessible and will be checked): <https://www.researchregistry.com/browse-the-registry/#home/registrationdetails/620e5213fd1e4d001e6edbb1/>

Guarantor

Dr. Talha Ahmed and Dr. Manohar Pai.

Informed consent

Written informed consent was obtained from all the patients for publication of this case series and accompanying images. A copy of the written consent is available for review by the Editor-in-Chief of this journal on request.

Provenance and peer review

Not commissioned, externally peer-reviewed.

Declaration of competing interest

The authors have NO affiliations with or involvement in any organization or entity with any financial interest (such as honoraria;



Fig. 16. Case of Squamous cell carcinoma of the left buccal mucosa T2N1M0 who underwent Commando Operation with Left modified radical neck dissection along with pectoralis major myocutaneous flap. (A) PMMC flap being raised to cover the defect on the neck and face. (B) & (C) NIV mode showing good uptake in the flap throughout the muscle and the cutaneous component. (D), (E) & (F) Post-operative day 30 showing excellent uptake of the PMMC flap with good healing and no evidence of skin necrosis. PMMC - Pectoralis major myocutaneous flap, GU - Good uptake

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Appendix A. Supplementary data

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