Rationale for Indocyanine Green being Formulated as a Sterile Lyophilized Powder for Near-Infrared Fluorescence Imaging

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Indocyanine green (ICG) has experienced significant expansions in its medical applications ever since its initial approval as a near-infrared fluorescence (NIR) imaging agent for cardiac output, hepatic function, and liver blood flow more than 60 years ago. The current ICG products that are approved by the United States Food and Drug Administration (FDA) are sterile lyophilized powder products containing 25 mg of ICG with no more than 5% of sodium iodide. This review article is aimed at providing readers with scientific reasoning on the way the ICG products are designed. Because ICG is unstable in aqueous solutions, it is presented as lyophilized products to increase the shelf-lives of the products. The lyophilized ICG products are sterile and need to be reconstituted using aseptic techniques due to the reconstituted solutions being administered to patients through intravenous injection for determining cardiac output, hepatic function, and liver blood flow, for ophthalmic angiography, and for visualization of vessels, blood flow and tissue perfusion, or by intradermal or interstitial injection for lymphatic mapping. A well-informed knowledge of the biopharmaceutics of ICG can help to maximize its safe and effective usage and stimulate the development of new ICG products and new applications.

Keywords: Indocyanine green, Near-infrared fluorescence, Solubility, Stability, Protein binding, Drug Product, Medical Imaging

Introduction

ICG is a substance that is known for its application as an imaging agent in the medical field. It was developed in World War II by the Eastman Kodak Company for color imaging. On February 9, 1959, the FDA approved the ICG product developed by the late Akorn, Inc. (Lake Forest, Illinois) for medical use to determine cardiac output, liver function, and liver blood flow[1], which still stands today. Later, in 1963, ICG was applied for determining renal blood flow because of its fluorescent properties[2]. Then, in 1965, it was also found to have a use of detecting cardiac murmurs, and finally, evaluating physiological brain perfusion[3]. Several additional indications have since been approved by the FDA[4], and many more indications have been, and continue to be, tested[1].

Although ICG has been used for decades, its significance in determining the function and quality of organs such as the liver or heart and in lymphatic mapping is only ever expanding, which is why it is important for healthcare experts to understand on how this agent works and how it can be applied in the medical field. This paper is intended to provide a review of the biopharmaceutics of ICG. Biopharmaceutics, as explained by the McGraw Hill Access Pharmacy Applied Physical Pharmacy book, is the study of the physical and chemical properties of drugs and their proper dosage as related to the onset, duration, and intensity of drug action[5]; Biopharmaceutics enables the rational design of drug products[6].

Results & Discussion

Physical and Chemical Properties of ICG

ICG is a dark green, odorless, negatively charged polymethine dye with a chemical formula of $C_{43}H_{47}N_2NaO_6S_2$ and a molecular weight of 774.96 grams per mole. Shown in Fig. 1 is the chemical structure of ICG. It has a melting point of 235C, decomposing gradually at temperatures above 200C. The absorbance of ICG in an aqueous solution has a peak of around 750–800 nm and a fluorescence emission peak of about 810 nm[7]. It was reported that a 0.5% (w/w) solution of ICG in water has a pH of six[8]. LogP refers to the logarithm of the partition coefficient of a compound in octanol over water[9]. LogP value is used in pharmaceutical sciences to measure the relative hydrophilicity (water-attractive) and hydrophobicity (non-water-attractive) of a compound[9]; the logP value of ICG is -0.29[10], which means a partition coefficient value of 0.51, indicating that ICG is hydrophilic and relatively more soluble in water than in octanol, less likely to pass through lipid bilayers and more likely to stay in the blood upon intravenous injection[11].

While a solution of ICG can be relatively simple to create, it

is not recommended for it to be used after long-term storage, because the stability of this substance is extremely low; the half-life of an ICG aqueous solution is in the range of 14–17 h in room temperatures [12]. For this reason, it is recommended against using an ICG solution after a long-term storage in research and on patients in the medical field.

ICGs degradation in aqueous solutions can be directly affected by air, light, temperature, the nature of the solvent, additives, and its concentration, and the degradation kinetic follows first order [13]. Li and Smith (2021) described that the combination of light and molecular oxygen promotes three main pathways to chemical degradation; the first one is conducting a reaction with an excited state of singlet oxygen and ICG to produce fragments containing carbonyl that dont absorb or emit NIR light; the second one is conducting a heptamine truncation reaction to create small bits of pentamethine homologue; and finally, the third one is to couple together two molecules of ICG to give an oxidative dimer [14]. Saxena et al. (2023) studied the degradation of ICG in aqueous solutions and reported that the rate constant of the degradation is 0.0412 ± 0.0038 hour^{-1} in dark environments at 22C, 0.0480 ± 0.0052 hour^{-1} in room, lighted environments at 22C, and 0.0344 ± 0.0027 hour^{-1} in dark environments at 8C [13]. For first-order reactions, the rate law is: $\ln[A]_t = \ln[A]_o - kt$, where $[A]_o$ is the initial concentration of the reactants, $[A]_t$ is the concentration of the reactants at time t, and k is the first-order rate constant [15]. Therefore, assuming first-order degradation kinetics, in lighted rooms and at 22C, the half-life of ICG or the time at which 50% of the ICG is degraded in a solution will be around 14.4 ± 2.4 hours (or $-\ln[0.5]/k = -\ln[0.5]/0.048$) [15], and the t_{90} value, or the time at which 10% of the ICG is degraded in a solution, is about 2.2 hours (or $-\ln[0.9]/k = -\ln[0.9]/0.048$) [15], which means that in 2.2 hours, 10% of the ICG will be degraded. Saxena et al. (2023) used 1 μg/L of ICG or ICG-sodium iodide (NaI) in water solution in most of their experiments, and concluded that to slow down ICG degradation in an aqueous solution, a high concentration of ICG solution should be stored at low temperatures and away from light exposure [13].

US FDA approved ICG products

For information on FDA-approved ICG products, one can look up the FDA Approved Drug Products with Therapeutic Equivalence Evaluations, also known as the Orange Book [16], and press the Search the Orange Book Database link. There, one can type in indocyanine green in the search bar; the search results will return a list of FDA-approved ICG products. In the list, only 3 of the 6 results are available on the market and the others are discontinued (Table 1). Two available products are marketed by Renew Pharmaceuticals LTD (now Renew Health Limited), IC-Green and Indocyanine Green, Injectable, 25 mg per vial (Table 1). On Renew Pharmaceuticals webpage (renewhealthlimited.com), ICG is sold under the Diagnostic Green name; in the United States, the product is supplied as IC-Green (indocyanine green for injection, 25 mg/vial); in Canada, the product is named as Indocyanine Green for Injection, USP; in Europe, the Middle East, and Africa (the EMEA countries), the product is sold as Verdye. In its package insert or prescribing information, the stated indications of the Indocyanine Green for Injection, USP are for determining cardiac input, hepatic function and liver blood flow, and for ophthalmic angiography [17]. The stated indications of IC-Green include fluorescence imaging of vessels, blood flow and tissue perfusion, fluorescence imaging of extrahepatic biliary ducts, fluorescence imaging of lymph nodes and lymphatic vessels during lymphatic mapping in adults with cervical and uterine cancer, and ophthalmic angiography [18].

The other product listed in the FDA Orange Book that is available on the market is the ICG product by Novadaq Technologies ULC; the product is named the Spy Agent Green Kit, which contains ICG powder for intravenous or interstitial injection, 25 mg per vial (Table 1). Since 2017, Novadaq Technologies ULC has been acquired by the Stryker Corporation (Kalamazoo, MI). In the Spy Agent Green Kit package insert, there are 4 indications of usage, which are (1) visualization of vessels, blood flow and tissue perfusion, (2) visualization of extrahepatic biliary ducts, (3) lymphatic mapping of cervical and uterine cancer, and (4) lymphatic mapping of breast cancer. For first two indications, the dye should be injected intravenously, for the third indication, the dye should be injected interstitially through the cervix, and for the fourth indication, it should be injected intradermally [19]. For more information about the Spy Agent Green Kit, readers can see the package insert/prescribing information of the product online [19].

Applications of ICG in the Medical Field

Over the past 66 years, the U.S. FDA has approved ICG as an imaging agent in the medical field with a wide range of usages (Table 1). In the next paragraphs, ICGs uses in two of the FDA approved indications will be described.

One of FDA approved indications of ICG is for ophthalmic angiography; angiography is a medical procedure that visualizes the contents of blood vessels using a contrast agent, or in this case, ICG. The technique in ophthalmology helps to visualize blood vessels in the choroid, or a layer of blood vessels and connective tissue between the retina and the sclera (white outer layer) in the eye [20], by injecting ICG into the antecubital vein and imaging the fluorescence of the dye under infrared light. The effectiveness of this method comes from the infrared lights ability to penetrate the retina, which allows for visualizing the deeper layers of the eye when photographed under the infrared camera. Another property of ICG that makes it effective for visualizing the choroid is its ability to bind with blood proteins extensively [21], limiting the leakage of the dye from the vessel

walls. The infrared images visualize any abnormalities in the blood vessels through two contrasting events: hypofluorescence, where the blocked and defective areas appear black and dark in the photograph due to defects blocking ICG from flowing into the defective area; and hyperfluorescence, where an area appears white in the photo due to abnormal blood vessels from cases such as choroidal hemangioma or the thinning and atrophy of retinal pigment epithelium or choriocapillaris[22]. For a more detailed understanding of this technique and its other applications, please refer to an article by Faik Gelisken (2024)[22].

Another one of the FDA approved indications of ICG is to determine liver functions and blood flow; it is routinely used before and after liver surgery and transplantation, as well as in diagnosing liver failure. Though a small molecule, ICG after being injected into the blood will bind with the bloods plasma proteins[21], causing it not be cleared through the kidneys and into urine; instead, the protein bound ICG will be exclusively cleared through the liver to the bile ducts, and then excreted into the fecal matters[21,23]. Therefore, how fast and to what extent the ICG is cleared can be a useful indicator of the quality of liver function and the blood flow[23]. A non-invasive transcutaneous measurement of ICG kinetics is done through a finger clip system. Normally, the transcutaneous measurement of ICG provides liver function data within less than 10 minutes; the measurement provides data such as ICG clearance, ICG half-life, ICG retention ratio after 15 minutes (ICG-R15), and ICG plasma disappearance rate (ICG-PDR)[23]. For example, in a healthy person, the ICG-PDR should be larger than 18%/min, the ICG clearance should be greater than 500 mL/min/m^2, and the ICG-R15 should be 6–12%[23]. For more information on the assessment of liver functions by ICG kinetics, readers can read an article by Samir G. Sakka (2018)[23]. For a description of the other FDA approved indications of ICG, readers can study the package inserts of the Spy Agent Green Kit and Renew Pharmaceuticals IC-Green and Indocyanine Green for Injection, USP[17–19].

Dosage Forms of ICG

Renew Pharmaceuticals LTDs IC-Green (Indocyanine Green for Injection) and Indocyanine Green for Injection, USP, are freeze-dried green powders in amber glass vials (Table 1). Each vial contains 25 mg of ICG, with no more than 5% of sodium iodide. Based on Diagnostic Green webpage, the sodium iodide is added as part of the manufacturing process to lyophilize the ICG, which makes the product more stable and easier to dissolve[24]. Each package of the product contains 6 single-patient-use vials of freeze-dried ICG (25 mg each) and 6 vials of 10 mL of sterile water for injection, USP. The vials should be stored in 20C–25C or 68F–77F, or about room temperatures (Table 1). The reported shelf-life of the ICG product by Renew Pharmaceuticals is 5 years[25]. The shelf-life of a pharmaceutical product is commonly measured using the t_{90} value[15], meaning that after 5 years of storage, around 90% of the original ICG remains in the product. Before use, the ICG powder product should be reconstituted with the sterile water for injection in a sterile environment and used within 6 hours. The reconstituted ICG solution is injected intravenously or interstitially into the patients, which is why the product needs to be sterile, and the reconstitution done using aseptic techniques.

The Spy Agent Green Kit from Novadaq Technologies ULC (now Stryker Corporation) is supplied in a variety of ways; specifically, the Spy Elite Kit or Package, the Spy PHI Kit or Package, Spy Minimally Invasive Surgery (Spy-MIS) Kit or Package, and the Spy Lymphatics Kit or Package (Table 1). Similar to the ICG from Renew Pharmaceutics, the vials should be stored in 20C–25C or 68F–77F, and before use, the product should be reconstituted with the sterile water for injection in a sterile environment and used within 6 hours[19].

Lyophilization or freeze-drying involves subjecting a product to cold temperatures, for example -20C or -40C, and turning all the water in the product into ice, then lowering the pressure to the product which causes the ice to undergo sublimation and turn directly into water vapor without going through the liquid phase[26]. By doing this, the water from the product is extracted while it is in a frozen state, preventing the degradation of the ICG during drying. This process can be explained based on waters phase diagram, which is a graph of the phases water with pressure in the Y-axis and temperature in the X-axis, and there is a triple point where all 3 physical phases of water coexist (0.01C, 0.006 atm)[27]. In pressures above the triple point, increasing the temperature of ice from a lower temperature to a higher one will cause the solid phase to melt into a liquid, then the liquid with vaporize into gas as the temperature continues to rise. But in pressures below the triple point, the water will instead sublime from its solid ice state directly into a gaseous vapor state without melting.

A comparison of the shelf-lives of the freeze-dried ICG products (5 years) and the ICG solutions reconstituted from the freeze-dried ICG products (6 hours) clearly illustrates that freeze-drying makes the ICG much more stable. As mentioned earlier, ICGs degradation in aqueous solutions is affected by air, light, temperature, the nature of the solvent, additives, and its concentration. In a freeze-dried product, water is removed, reducing the overall molecular movement, and the ICG molecules in the powder have reduced access to oxygen and light, all explaining the longer shelf-lives of the lyophilized ICG products. Presenting ICG as a lyophilized product addresses the instability issue of ICG in aqueous solution and increases the shelf-life of the product. However, lyophilized products are not without their disadvantages. The FDA listed several disadvantages of lyophilization of sterile parenterals, including the need for reconstitution with a sterile diluent and cost and complexity of equipment[28]. Reconstitution demands the time of trained

medical personnel. Moreover, errors can occur during reconstitution[29].

Pharmacokinetics and Pharmacodynamics of ICG after Intravenous Injection

Pharmacokinetics (PK) is defined as what the body does to a drug[30], ICG in this case. It describes the movement of drugs in, around, and out the body, or more specifically the absorption, distribution, metabolism, and excretion of a drug[31]. Pharmacodynamics (PD) is defined as what a drug does to the body; it studies the effect of a drug on the body[31]. ICG is used as an imaging agent and thus does not have any FDA-approved indication in treating or preventing any diseases or illness. The PK of ICG is important because of its role as an imaging agent that must properly flow around the body so it can be properly imaged to visualize blood flow. For some of the FDA-approved indications, ICG is intravenously injected into the blood stream. Thus, there is not a need for absorption, which is different from medications taken orally that need to be absorbed through the intestinal wall from the lumen of the gastrointestinal tract into the blood stream. Upon injection into the blood, ICG binds with the plasma proteins in the blood extensively. Cherrick et al. (1960) showed that when ICG was added in a concentration of 0.25 mg/100 mL in 0.5 ml samples of normal human plasma, 95% of the recovered ICG was associated with albumin, 2.4% with α_1-globulin, 2% with α_2-globulin, and 0.6% with β-globulin[21]. Jang et al. (2023) reported that the dissociation constant of ICG and human serum albumin is 0.00792 s^{-1}, and the association constant is 193 $M^{-1}s^{-1}$, with an equilibrium constant of 41 μM[32]. Cherrick et al. (1960) also reported that the initial volume of distribution of ICG in four human volunteers is 3.483 ± 0.724 L[21]. The initial decay rate, half-life, and percent retained 20 min after injection were 18.5 ± 3.1%/min, 3.4 ± 0.7 min, and 3.8 ± 1.0%, respectively[21]. Once the ICG is circulating around the body and the imaging process is done, the ICG does not get filtered from the blood into the urine; Cherrick et al. (1960) reported that urine collected from five patient volunteers during a 6 hour period following the intravenous administration of ICG contained no dye.[21]. Instead, ICG is removed by the liver into the bile and then gets excreted into the fecal matter.

For lymphatic mapping of cervical and uterine cancer, ICG solution is injected interstitially into the cervix to map the lymphatic system around the cervix and determine the extent of cancer spread[33]. For lymphatic mapping of breast cancer, the ICG solution is injected intradermally into the periareolar area to locate the sentinel lymph node and map the lymphatic system around the breast cancer[34]. Based on the prescribing information of Spy Agent Green, following interstitial or intradermal injection, indocyanine green binds to proteins in lymph fluid and the interstitial space, is taken up by the lymphatic vessels, and drain to the lymph nodes.[19]

Binding of ICG to proteins, regardless of whether it is injected intravenously, interstitially, or intradermally, is critical for ICGs clinical applications and its excretion from the body. It is thus beneficial that the FDA-approved ICG lyophilized products only contain ICG with no more than 5% of sodium iodide because when they are reconstituted with sterile water for injection, the ICG dissolves in the solution, making it free to bind to proteins upon injection.

Toxicities

As a chemical meant to be injected into the human body, considerations for its toxicity are critical for ensuring not only effective, but also safe usage. Based on prescribing information of the Spy Agent Green Kit and Renew Pharmaceuticals IC-Green, a clinically significant adverse reaction from ICG is hypersensitivity reactions, including anaphylaxis, urticaria, and death[18,19], therefore, ICG should not be used for people with a history of hypersensitive reactions to ICG. In the package inserts of IC-Green and SPY AGENT GREEN, it is stated that there are no adequate and well-controlled studies of IC-Green (or SPY AGENT GREEN) in pregnant women. Both package inserts also contain risk summary for pregnancy and lactation[17–19]. The safety of ICG in pediatric and geriatric uses has been established[18,19]. Additionally, some studies show that ICG can cause dose-dependent retinal damage, and even lower concentrations of ICG can potentially cause unintentional damage[35]. However, other than the potential risk of allergic reactions, based on the prescribing information of ICG products, it is a mostly safe agent for medical use with proper cautionary measures.

Alternative ICG Formulation Strategies and Future Directions

The current FDA-approved ICG products are sterile lyophilized ICG powder. The lyophilization strategy not only overcomes the instability issue of ICG, but also allows ICG to dissolve in sterile water for injection and freely binds to proteins after injection, which is important for its clinical indications and excretion. Besides lyophilization, other strategies for stabilizing ICG can be found in literature. One such strategy is to store ICG in a frozen environment. Perks et al. reported that ICG reconstituted in sterile water for injection at 2.5 mg/mL did not show any concentration change after 28 days of storage in -20C or -67C[36], suggesting that the ICG solution may not have to be discarded 6 hours after reconstitution if there are frozen. However, the authors did not report the stability of the frozen ICG after more 28 days of storage, and the exact shelf-life of the ICG frozen solution is not known. Another reported strategy to increase the storage stability of ICG without lyophilization is to include additional excipients. Laurent, Sayah, and Lopez reported that ICG solutions containing ascorbic acid and salts

did not show any ICG degradation after 26 weeks, or half a year, of storage at room temperature away from light[37]. Another tested strategy is to incorporate ICG in various micelles, nanoparticles, and nanobubbles[12,38–44]. For example, Schwartz et al. showed that ICG is more stable in water containing 0.1% of Tween 20, with less than 5% reduction in emission intensity over 4 months[45]. Kirchherr et al. reported that ICG in Solutol HS 15 and Cremophor RH40 appeared stable after 30 days of storage at 25C[39]. Other researchers reported that ICG showed better stability in nanoparticles[12,40], but overall, the stability ICG in those formulations remain unsatisfactory for long-term storage. Still, another reported strategy is to chemically modify ICG to deuterated ICG[14], which increases the aqueous half-life of ICG by a factor of 3.1. Clearly, the FDA-approved sterile lyophilized ICG powder products represent the best formulation for long-term storage of ICG products. Other strategies introduce various additional excipients without extending the shelf-life of ICG to a level equal to that of the FDA-approved ICG products. Importantly, including more excipients or incorporating ICG in various nanoparticles or micelles could make it impossible to use ICG to perform the indications of the current FDA-approved products, although the new ICG formulations may allow new medical indications of ICG. For example, some ICG nanoparticles have reported applications in fluorescence imaging-guided photothermal therapy of tumors[41,42,44]. Finally, besides the indications listed in the package inserts of the two FDA-approved ICG products, scientists and clinicians will likely find new applications for the sterile lyophilized ICG products. An example of ICG in action are cases of anastomotic leakage, a frequent surgical issue where the contents of the guts leak out during or after surgical anastomosis, which can be prevented through the use of ICG to properly visualize and evaluate the colons of the patient to make more precise and accurate operations[46]. Another application of ICG is imaging for lung tumors through inhaling the substance into the lung[47]; the inhalation method provides a far more detailed and clearer image of the lung functions as opposed to injection and serves its purpose as an imaging agent with incredible efficiency.

Conclusions

Ever since the FDAs initial approval of using ICG as an imaging agent to determine cardiac output, hepatic function, and liver blood flow about 66 years ago, many additional indications have been approved. However, ICG remains mainly administered by intravenous injection, and the FDA approved products remain sterile lyophilized ICG powders to be dissolved or reconstituted in sterile water for injection in a sterile environment. Due to its peculiar poor stability in aqueous solution, ICG is freeze-dried into a dry powder in an amber glass vial, which drastically increases the shelf-life of the ICG products to 5 years at 20-25oC; however, upon reconstitution in water, the ICG solution must be used within 6 hours. Upon reconstitution, ICG dissolves in the solution and is free to bind to proteins after injection, which is critical for its clinical applications and its excretion from the body. Alternative ICG formulations strategies can be found in literature, although they do not necessarily have longer shelf-lives than sterile lyophilized ICG powder, but they could potentially allow ICG new clinical indications in the future.

Methods

The literature searching strategy to find information for this paper was to refer to the PubMed website, the FDA website of Approved Drug Products with Therapeutic Equivalence Evaluations[16], Google and Google Scholar, then searching up topics such as indocyanine green biopharmaceutics, Indocyanine green solubility, Indocyanine green stability, Indocyanine green chemical degradation, Indocyanine green pharmacokinetics, etc. Some information was from the webpages of the two FDA-approved ICG product Applicant Holders, Renew Pharmaceuticals LTD (now Renew Health Limited) and NOVADAQ Technologies ULC (now Stryker Corporation).

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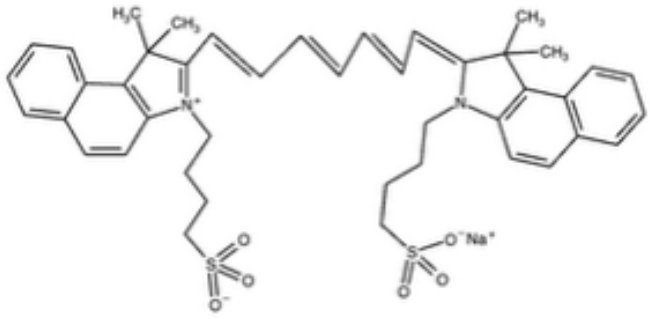

Fig. 1 Chemical structure of ICG (drawn based on SPY AGENT GREEN prescribing information using the RCSB PDB Chemical Sketch Tool, https://www.rcsb.org/chemical-sketch).

References

1 P. University, *Center for Research Innovation in Biotechnology (CRIB), College of Pharmacy Indocyanine green, Clinical Drug Experience Knowledgebase*, https://cdek.pharmacy.purdue.edu/api/39278.

2 J. Walker, H. Silva, T. Lawson, J. Ryder and S. Shaldon, *Renal blood flow in acute renal failure measured by renal arterial infusion of indocyanine green*, https://doi.org/10.3181/00379727-112-28214.

3 T. Huffman, R. Goodwin, R. Leighton, J. Ryan and C. Wooley, *Intracardiac phonocardiography in the differential diagnosis of continuous murmurs*.

4 L. Mulugeta, *Memorandum, Review of revised label and labeling*, https://www.accessdata.fda.gov/drugsatfda_docs/nda/2018/211580Orig1s000OtherR.pdf.

5 W. Mobley, *Chapter 1. Introduction to Biopharmaceutics*.

6 L. Shargel, S. Wu-Pong and A. Yu, *Chapter 14, Biopharmaceutic considerations in drug product design and in vitro drug product performance*.

7 L. Cui, A. Enochs, R. Lurie and A. Dylla, *Characterization and photochemical exploration of indocyanine green and its aggregation properties*.

8 E. McCorquodale and C. Colyer, *Indocyanine green as a noncovalent, pseudofluorogenic label for protein determination by capillary electrophoresis*.

9 L. Shargel, S. Wu-Pong and A. Yu, *Chapter 10. Physiologic drug distribution and protein binding*.

10 J. Sangster, *LOGKOW; editing status 2024-12-13; re3data.org - Registry of Research Data Repositories*, http://doi.org/10.17616/R36C73.

11 A. Mansoor and N. StatPearls, https://www.ncbi.nlm.nih.gov/books/NBK545280/.

12 V. Saxena, M. Sadoqi and J. Shao, *Enhanced photo-stability, thermal-stability and aqueous-stability of indocyanine green in polymeric nanoparticulate systems*, https://doi.org/10.1016/j.jphotobiol.2004.01.002.

13 V. Saxena, M. Sadoqi and J. Shao, *Degradation kinetics of indocyanine green in aqueous solution*, https://doi.org/10.1002/jps.10470.

14 D. Li and B. Smith, *Deuterated indocyanine green (ICG) with extended aqueous storage shelf-life: Chemical and clinical implications*, https://doi.org/10.1002/chem.202102816.

15 D. Bhangare, N. Rajput, T. Jadav, A. Sahu, R. Tekade and P. Sengupta, *Systematic strategies for degradation kinetic study of pharmaceuticals: an issue of utmost importance concerning current stability analysis practices*, https://doi.org/10.1186/s40543-022-00317-6.

16 U. Food and D. Administration, *Approved drug products with therapeutic equivalence evaluations, Orange book*, https://www.fda.gov/drugs/drug-approvals-and-databases/approved-drug-products-therapeutic-equivalence-evaluations-orange-book.

17 *17 INDOCYANINE GREEN for injection prescribing information*, https://diagnosticgreen.com/am/package-insert/.

18 *18 IC-Green prescribing information*, https://www.accessdata.fda.gov/drugsatfda_docs/label/2024/011525s039lbl.pdf.

19 *19 SPY AGENT GREEN prescribing information*, https://www.accessdata.fda.gov/drugsatfda_docs/label/2023/211580s006lbl.pdf.

20 *20 NCI Dictionary of Cancer Terms, Choroid*, https://www.cancer.gov/publications/dictionaries/cancer-terms/def/choroid.

21 G. Cherrick, S. Stein, C. Leevy and C. Davidson, *Indocyanine green: observations on its physical properties, plasma decay, and hepatic extraction*, https://doi.org/10.1172/JCI104072.

22 F. Gelisken, *Indocyanine green angiography*, https://doi.org/10.4274/tjo.galenos.2023.89735.

23 S. Sakka, *Assessment of liver perfusion and function by indocyanine green in the perioperative setting and in critically ill patients*, https://doi.org/10.1007/s10877-017-0073-4.

24 R. Hehrmann, *Indocyanine green and iodine/iodide allergies*, https://diagnosticgreen.com/row/news/indocyanine-green-and-iodine-iodide-allergies/.

25 *25 Diagnostic Green, Use of Verdye (ICG) in minimally invasive surgery (MIS*, https://diagnosticgreen.com/row/use-of-verdye-icg-in-minimally-invasive-surgery-mis/#:~:text=Before%20administration%20the%20powder%20must%20be%20reconstituted,should%20be%20used%20immediately%2C%20protected%20from%20light.

26 S. Nail, S. Jiang, S. Chongprasert and S. Knopp, *Fundamentals of freeze-drying*, https://doi.org/10.1007/978-1-4615-0549-5_6.

27 *27 Chapter 13.20. Phase diagram for water*, https://chem.libretexts.org/Bookshelves/Introductory_Chemistry/Introductory_Chemistry_(CK-12)/13%3A_States_of_Matter/13.20%3A_Phase_Diagram_for_Water, in LibreTexts Chemistry (CK-12 Foundation, 2022/8/8/).

28 U.S., *Food and Drug Administration*.

29 *29 Institute of Safe Medication Practices. Reconstitution errors again: This time using the wrong diluent*.

30 P. Glassman and V. Muzykantov, *Pharmacokinetic and pharmacodynamic properties of drug delivery systems*, https://doi.org/10.1124/jpet.119.257113.

31 K. Ernstmeyer and E. Christman, *Chapter 1. Pharmacokinetics Pharmacodynamics*, Ernstmeyer, E. Christman).

32 H. Jang, M. Song, C. Park, J. Youn, Y. Lee, G. Cheon and K. Kang, *Imaging of indocyanine green-human serum albumin (ICG-HSA) complex in secreted protein acidic and rich in cysteine (SPARC)-expressing glioblastoma*, https://doi.org/10.3390/ijms24010850.

33 M. Frumovitz, M. Plante, P. Lee, S. Sandadi, J. Lilja, P. Escobar, L. Gien, D. Urbauer and N. Abu-Rustum, *Near-infrared fluorescence for detection of sentinel lymph nodes in women with cervical and uterine cancers (FILM): a randomised, phase 3, multicentre, non-inferiority trial*, https://doi.org/10.1016/S1470-2045(18)30448-0.

34 E.-M. Grischke, C. Rohm, M. Hahn, G. Helms, S. Brucker and D. Wallwiener, *ICG fluorescence technique for the detection of sentinel lymph nodes in breast cancer: Results of a prospective open-label clinical trial*.

35 E. Rodrigues, C. Meyer, S. Mennel and M. Farah, *Mechanisms of intravitreal toxicity of indocyanine green dye: implications for chromovitrectomy*, https://doi.org/10.1097/01.iae.0000253051.01194.ab.

36 W. Perks, S. Law, R. Marchesano and S. Walker, *Stability of 2.5 mg/mL indocyanine green (ICG) solutions stored in syringes at 25oC*, https://www.metrodis.org/uploads/document_library/53fbdc39933b6d09f8bdb650a0f01207.pdf.

37 M. Laurent, B. Sayah and N. Lopez, *Stable formulations of indocyanine green*, https://patents.google.com/patent/WO2022194731A1/en.

38 Y. Ding, C. Wang, B. Lu and Y. Yao, *Enhancing the stability and photothermal conversion efficiency of ICG by pillar[5]arene-based host-guest interaction*, https://doi.org/10.3389/fchem.2021.775436.

39 A. Kirchherr, A. Briel and K. Mader, *Stabilization of indocyanine green by encapsulation within micellar systems*, https://doi.org/10.1021/mp8001649.

40 E. Lee, M. K. and S. Lee, *Enhanced stability of indocyanine green by encapsulation in zein-phosphatidylcholine hybrid nanoparticles for use in the phototherapy of cancer*, https://doi.org/10.3390/pharmaceutics13030305.

41 C. Ting, Y. Chou, S. Huang and W. H, *Chiang Indocyanine green-carrying polymeric nanoparticles with acid-triggered detachable PEG coating and drug release for boosting cancer photothermal therapy*, https://doi.org/10.1016/j.colsurfb.2021.112048.

42 L. Yan and L. Qiu, *Indocyanine green targeted micelles with improved stability for near-infrared image-guided photothermal tumor therapy*, https://doi.org/10.2217/nnm.14.118.

43 L. Yang, B. Huang, F. Chen, J. Jin, Z. Qin, F. Yang, Y. Li and N. Gu, *Indocyanine green assembled nanobubbles with enhanced fluorescence and photostability*, https://doi.org/10.1021/acs.langmuir.0c02288.

44 M. Yaseen, J. Yu, M. Wong and B. Anvari, *Stability assessment of indocyanine green within dextran-coated mesocapsules by absorbance spectroscopy*, https://doi.org/10.1117/1.2821423.

45 A. Schwartz, L. Joline, E. Snow, S. francke, C. Marques and R. bernardes, *Stabilizing indocyanin green for use as quantitative standards*, https://iovs.arvojournals.org/article.aspx?articleid=2377551.

46 P. Fransvea, M. Chiarello, V. Fico, M. Cariati and G. Brisinda, *Indocyanine green: The guide to safer and more effective surgery*, https://doi.org/10.4240/wjgs.v16.i3.641.

47 Y. Quan, C. Oh, D. Jung, J. Lim, B. Choi, J. Rho, Y. Choi, K. Han, B. Kim, C. Kim, J. Park and J. Kim, *Evaluation of intraoperative near-infrared fluorescence visualization of the lung tumor margin with indocyanine green inhalation*, https://doi.org/10.1001/jamasurg.2020.1314.

Table 1. FDA approved ICG products currently on the US market. Information is based on FDA Orange Book and package inserts or prescribing information.

Applicant holder	Product name	Dosage form	Route of administration	Indications	Storage and shelf-life
Renew Pharmaceuticals LTD	IC-Green (indocyanine green for injection), for intravenous or interstitial use. Package configuration: 6 25 mL single-patient-use vial of IC-Green and 6 single-dose vials of Sterile Water for Injection (10 mL)	Single-patient-use vial of IC-Green (25 mg each) as sterile, lyophilized green powder, with single dose vial of sterile water for injection, USP (10 mL). pH of 5.5-7.5 when reconstituted, use within 6 h	Intravenous injection	For ophthalmic angiography, visualization of extrahepatic biliary ducts, or visualization of vessels, blood flow, and tissue perfusion	20-25°C, 5 years
			Interstitial injection	For lymphatic mapping of cervical and uterine cancer	
Renew Pharmaceuticals LTD	Indocyanine Green for Injection, USP	Sterile, lyophilized green powder (25 mg/vial, with ≤5% of NaI). Packaged with sterile water for injection (WFI), USP pH ~6.5 upon reconstitution, use within 6 h	Intravenous injection	For determining cardiac output, hepatic function, and liver blood flow; for ophthalmic angiography	20-25°C, 5 years
Novadaq Technologies ULC (now Stryker Co)	Spy Elite Kit or Pack (of 6 kits) Each kit contains one 25 mg SPY AGENT GREEN vial, one 10 ml sterile WFI plastic vial, one ND8000 sterile drape	Spy Agent Green (indocyanine green for injection, USP) is a sterile, lyophilized, green powder containing 25 mg of ICG with not more than 5% of NaI, and HCl or NaOH may have been used to adjust the pH before lyophilization pH ~5.5-7.5 upon reconstitution, use within 6 h	Intravenous injection	For use with SPY Elite System For visualization of vessels, blood flow and tissue perfusion	20-25°C, Shelf-life is unknown but likely 5 years
	SPY-PHI Kit or Pack (of 6 kits) Each kit contains one 25 mg SPY AGENT GREEN vial, one 10 ml sterile WFI plastic vial, one SPY-PHI (HH2000) sterile drape		Intravenous injection Intradermal injection for use with SPY PHI System for lymphatic mapping of breast cancer	For use with SPY-PHI System For visualization of vessels, blood flow and tissue perfusion For lymphatic mapping	
	SPY-MIS Kit or Pack (of 6 kits) Each kit contains one 25 mg SPY AGENT GREEN vial, one 10 ml Sterile WFI plastic vial, two 3 mL syringes, two 10 ml syringes, two 18 G, 1-inch needles, labels for syringes		Intravenous injection	For use with Advanced Imaging Modality (AIM) and PINPOINT Systems For visualization of vessels, blood flow and tissue perfusion For visualization of extrahepatic biliary ducts	
	SPY Lymphatics Kit or Pack (of 6 kits) Each kit contains one 25 mg SPY AGENT GREEN vial, two 10 ml sterile WFI plastic vial, one 10 mL syringe, two 3 ml syringes, one luer-lock 10 ml syringe with controlled handle, two spinal needles (22G, 3.5 inch), labels for syringes		Lymphatic mapping of cervical and uterine cancer after interstitial injection	For use with the AIM L10 and L11 Light Sources and 1688 Camera System, and the PINPOINT System For lymphatic mapping	