tutorial

Fundus photography and fluorescein angiography

Fluorescein angiography

Introduction

Fluorescein angiography is a clinical diagnostic test procedure in which rapid-sequence photography is performed after intravenous injection of sodium fluorescein. The procedure allows the documentation of the retinal circulation. Of equal importance is the ability of the angiogram to outline the entire retinal vascular bed. Previous clinical methods of examination of the fundus enabled the physician to effectively see only a portion of the retinal vasculature, namely the larger blood vessels. Due to the miniscule caliber of the retinal capillaries, much of the more important structures could not he visualized. Now, with the use of fluorescein dye, combined with retinal fluorescence photography techniques, these mi crovascular structures may he clearly demonstrated.

The angiogram is helpful in the evaluation and demonstration of systemic diseases such as diabetes and hypertension through its ability to show the changes that may take place in the microvascular structures. Differential diagnoses of a number of eye conditions such as circulatory and inflammatory disorders are made possible due to time differences in the appearance of the dye in certain areas of the retina and to dye flow or leak patterns which are unique to certain disease entities.

The use of fluorescein dye in ophthalmology is not a recent development but dates back as far as 1881, ten years after the substance was first synthesized by Von Baeyer (Maumenee, 1968). At that time Erhlich observed that after dye was injected into the blood stream, the dye appeared in the anterior chamber. This disclosure led others to make use of the substance as a "marker" in the study of the circulation of the aqueous humor, and by 1900 many investigators had done extensive work with the dye. Burke (1910) also used fluorescein dye to study fundus pathology by administering five grams of the dye to adults in coffee and two to three grams to children. With these concentrations, he was able to observe fluorescence in retinal and choroidal lesions with ordinary illumination. The conditions of inflammatory lesions, chorioretinitis and optic neuritis were reported as appearing heavily stained; healthy retinas did not stain. Fisher (1929) reported his observations of dynamic blood flow through the iris vessels of a rabbit after injection of fluorescein dye. Maumenee (1955) gave an intravenous injection of fluorescein to study a choroidal hemangioma by ophthalmoscopy, and later he and MacLean (1960) applied the same technique of fluorescein angioscopy in the differential diagnosis of malignant melanomas and choroidal hemangiomas.

In 1961, David, Saito and Heyman were able to calculate arm-to-retina circulation time by intravenously injecting fluorescein dye into the arm and watching for its appearance in the central retinal arteries. This determination was added to those standard values of circulation times for arm-to-lip time, arm-to-leg time, etc., which had already been established by investigators in other areas of medicine.

Novotney and Alvis' work (1961.) was a significant contribution in clinical and investigative ophthalmology in that they formulated the photographic technique for fluorescein angiography. This technique has remained essentially unchanged over the years.

Fluorescence

if certain substances are exposed to any form of short wave electromagnetic radiation, energy is absorbed by that substance, which changes the internal condition of the molecules by raising the electrons to a higher energy level. As the molecules return to their original state, the substance will emit radiation of a longer wavelength. This phenomenon is known as luminescence, of which there are two types: Fluorescence occurs during exposure to the exciting radiation and ceases within a very short time (10—s seconds) after removal of the stimulus. In other substances, the molecules disturbed by the absorption of energy do not return immediately to their original state; emission continues after the exciting source is cut off. This type of delayed light emission is called phosphorescence.

Fluorescence photography

Two filters are required when photographing fluorescence (Anon., 1974), an excitation filter and a barrier filter. The excitation filter is placed over the irradiation source and its function is to transmit only that radiation that will stimulate fluorescence and to absorb all other wavelengths. Since the efficiency of filters is less than optimum, it is inevitable

that the excitation filter will transmit some amount of unwanted or undesirable light to the subject along with the exciting radiation. This would certainly compromise the image quality of the fluorescing body, since the unwanted light is usually of considerably greater intensity than that of fluorescence and would cause more exposure of the film than would the fluorescence. The barrier filter, therefore, must absorb all of this unwanted, reflected light and transmit only the emitted fluorescence. The perfect filter set would have high transmittance by both filters in the respective wavelength bands for excitation and emission. The barrier filter would absorb all wavelengths except that of the fluorescence. The resultant photograph would then have maximum contrast between the fluorescent body and its nonfluorescing surroundings.

After performing spectrofluorometric evaluations of blood-fluorescein mixtures, Novotney and Alvis learned that peak excitation occurred at 490 nm, and peak emission of the fluorescence was found to he at 520 nm. The initial selection of filters by these men was probably based on the premise that the source of radiation must be converted to the appropriate (color) portion of the visible spectrum, and that the barrier filter should be selected to transmit according to the color of fluorescence. Accordingly, the Kodak Wratten filter, no. 47, was chosen as the excitation filter, and the Wratten filter, no. 58, as the barrier filter. Ansco super Hypan film was used and force developed in Ethol UFG developer for 10 minutes at 70°F.

Since this early work, other investigators have utilized a number of filter combinations for angiography. See the listing in Figure 28. The early sets, chosen on the basis of availability were destined to be replaced by more efficient combinations. Haining and Lancaster (1968) in an attempt to improve angiographic results through better filtration, did a series of front-face spectrofluorometric evaluations using different concentrations of fluorescein in heperanized whole blood taken from patients ten to fifteen minutes after being injected with fluorescein. They observed that peak excitation ranged between 460 nm and 480 nm, depending upon the concentration of fluorescein, blood hemoglobin and pH. Peak emission of fluorescence remained constant, however, at 520 nm, which is consistent with

Figure 28—Excitation and barrier filters used by various authors*

Author	Publication Date	Excitation Filter	Barrier Filter
Novotney and Alvis	1961	KW47 (Blue)†	KW58 (Green)†
Dollery and Hodge	1962	KW47B (Deep blue)†	KW58 (Green)†
Hart, et al	1963	KW47 (Blue)†	KW56 (Light green)†
Smith, Gass, and Justice	1964	KW47 (Blue)†	KW56 (Light green)†
Oosterhuis and Lammens	1965	BG12 (Light blue)††	GG14 (Dark yellow)††
Shimizu	1965	FG18 (Blue)§	FG17 (Green)§
Allen, et al	1966	KW47A (Light blue)†	KW15 (Deep yellow)†
Shimizu	1966	FG18 (Blue)§	FG12 (Yellow)§
Tani	1966	FG18 (Blue)§	FG17 (Green)§
Haining and Lancaster	1968	Baird Atomic B4‖	Baird Atomic B5‖
Frazier and Allen	1974	KP490¶ and BG38††	KW15 (Deep yellow)†

*A full bibliographic listing for each author is contained in the Reference section of this article.
†Kodak Wratten filters are available from photo dealers and described in Kodak Publication No. B-3, *KODAK Filters for Scientific and Technical Uses.*
††Available from Schott Optical Glass, Inc., Duryea, Pennsylvania.
§ Available from Fuji Photo Film Co. Ltd., Tokyo, Japan.
‖ Available from Baird Atomic Inc., Bedford, Massachusetts.
¶Manufactured by E. Leitz, Inc., Rochleigh, New Jersey.

Novotney and Alvis' finding. It became obvious that the absorption filters used up to that time were poor for angiography due to their poor transmission properties (the Kodak Wratten filter, no. 47, transmits only 42 percent at 440 nm, and the no. 58 filter only 52 percent at 520 nm) in the desired wavelengths. Figure 29A shows the no. 47 filter with its companion barrier filter, the Wratten no. 15. Additionally, the transmission curves for these filters have curved rather than linear gradients leading up to these peaks, with small "toes" near both ends of the transmission bands. These "toes" are responsible for a cross-over point common to both filters which permits unwanted light to be reflected from the subject and to strike the film. Another feature of the "perfect" filter set would be an optical matching of the excitation and barrier filters so that there would be minimal overlapping of their transmission curves; if there is any overlapping, the amount of energy transmitted should be below the threshold of film sensitivity.

Haining & Lancaster recommended the use of interference filters in order to take advantage of their high transmission peaks at the required wavelengths (Figure 29B). The Baird Atomic B4 excitation filter and B5 barrier filter permitted a 90 percent transmission at 520 nm, with a minimal crossover of the two curves at 500 nm. Designed for monochromatic transmission, these narrowband filters could be made to pass light very selectively and indeed proved to be more efficient for angiography.

Frazier and Allen (1974) worked on the assumption that different blood-fluorescein mixtures might require different wavelengths to excite peak fluorescence. A basis was needed to explain some of the clinical experiences in which an otherwise routine angiographic procedure was carried out on a cooperative patient but which resulted in angiographic negatives which lacked good, strong fluorescence. In other words, the wavelengths required for peak excitation in those patients may have been different than that of the exitation filter used, especially if the filter was a narrowband, high peak interference filter. Accordingly, a search for a filter which might have a high peak of transmission in all the observed wavelengths (460 nm through 490 nm) disclosed a filter made by Leitz, the KP 490. This filter transmits a broad band between 350 nm and 510 am, with 80 to 90 percent transmission of most wavelengths between 400 nm and 500 nm, with an extremely sharp cut-off at 500 nm (Figure 29C). Very little transmission takes place between 500 Inn and 51.0 nm, but a second transmittance band. is present from 650 nm to about 700 nm, with a peak of 60 percent transmission at about 685 nm. The filter combination of Frazier and Allen consists of three filters, the KP 490 (excitation) a Wratten no. 15 (barrier) and a BG38, which absorbs most of the red light transmitted through the KP 490.

The circulation

Since the injection site for fluorescein angiography is generally at the antecubital vein in the arm, there is a delay-time between the injection and the appearance of dye in the retinal vessels. This period is referred to as the arm-to-retina circulation time, the average time being 1.0 to 12 seconds in a normal patient. About one second prior to the filling of the central retinal artery (Figure 30), the choroidal vessels fill, thus causing a brightening of the field. This phenomenon is commonly referred to as "background fluorescence" or "choroidal flush." The dye enters the retinal vasculature through the central retinal artery (early arterial phase), fans out into the superior and inferior arteries along the temporal vascular arcades as well as nasally. The venous system drains the deoxygenated blood out of their respective arterial counterparts, and since the circulation around the disc centrally is made up of shorter arterial-to-venous loops, the venous filling is seen there, even as the arterial filling is progressing outward to the periphery. The phenomenon of "laminar flow," (ie, the white stripe outlining the borders of the vein) is seen simultaneously with the filling of many of the peripheral arterioles. The "full arterio-venous phase" occurs when both arterial and venous systems are equally filled with dye and are fluorescing equally. This is followed by the dye draining from the arteries, thus leaving the veins more fluorescent than the arteries (venous phase).

An important factor in obtaining good angiographic negatives is that of strong photographic contrast between the vasculature and the background. This contrast is proportional to the concentration of the dye, and may be a variable factor, depending upon the rate of injection as well as the pH of the blood. It is therefore desirable to give a rapid injection in order to get a concentrated bolus of dye into the blood stream, rather than a long, drawn out injection, which will allow the dye to he diluted by the blood, thus diminishing contrast. Some institutions utilize an intravenous saline flush immediately after the injection. This is not done in our laboratory since it adds to the complexity of the procedure.

Sequencing

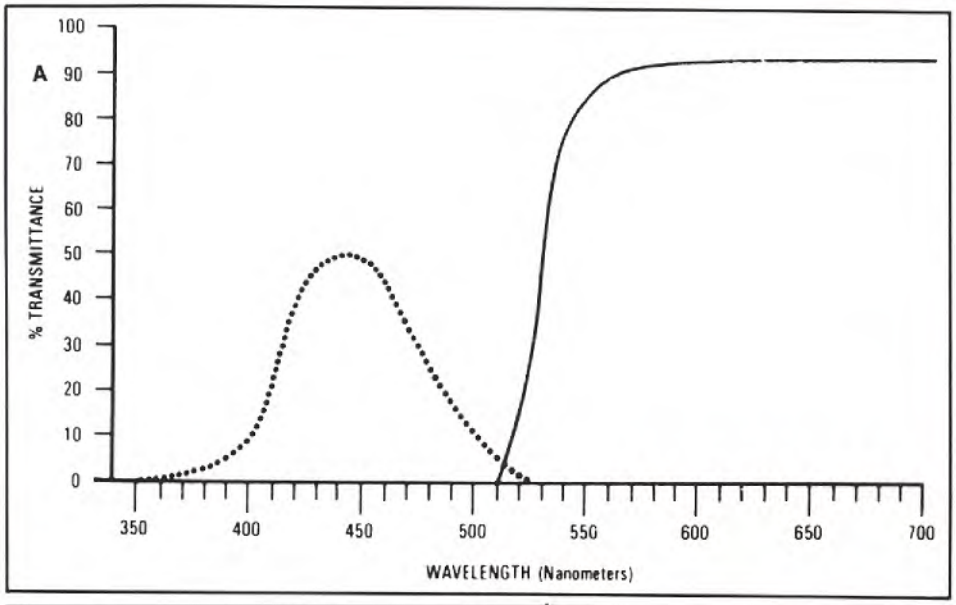
Since the evaluation and interpretation methods of angiographic data may vary from one physician to another, the protocol for angiography is established by the individual doing the interpretation. Protocols in most laboratories has been established to record the actual flow of dye through the retinal vasculature. It is important to establish standard techniques that are universally accepted in order to render consistency to all data.

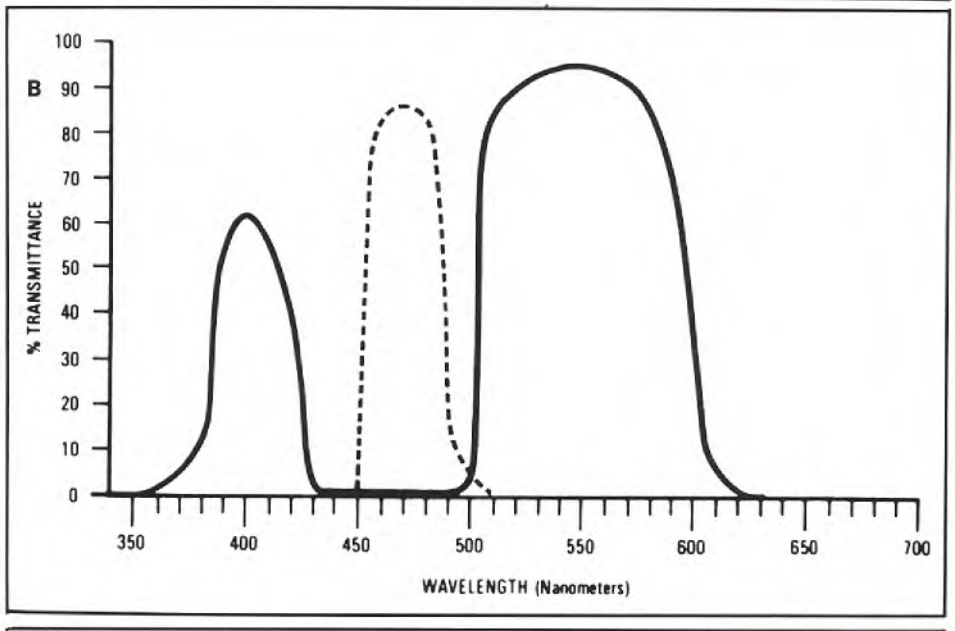
It is not necessary to exceed a rate greater than one picture per second in rapid-sequence photography for clinical studies. This rate of photography will undoubtedly cover any clinical diagnostic situation. A faster rate might be required in clinical research in order to obtain usable data in the very early portion of the dye-flow cycle. Since the average arm-to-retina circulation time is in the order of 10 to 12 seconds, in order to conserve film, delay 5 to 7 seconds after the beginning of injection before commencing with photography. Take 25 to 30 pictures to document the fluorescent pattern of the retinal circulation from the choroidal flush stage through to the full venous phase. Once the full venous phase is seen, little else of diagnostic value takes place until a period of time passes.

Differential diagnosis of many conditions is made by comparison of a picture made more than 15 minutes after injection (commonly referred to as a "late" photograph) to either a control picture taken before the injection ("blank" photograph) or an early arterial picture. It is therefore imperative to obtain good control photographs and good late photographs as well as early arterial-phase pictures.

The procedure

Though the basic technical procedures for angiography are identical to those of single-frame fundus photography, the two techniques require quite different approaches on the part of the photographer. As was discussed pre-





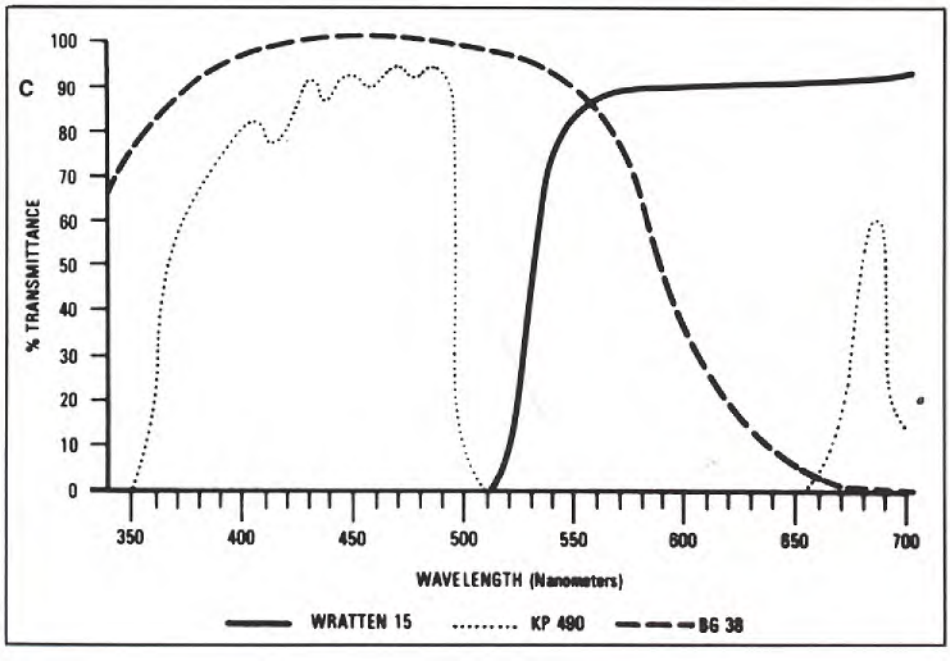
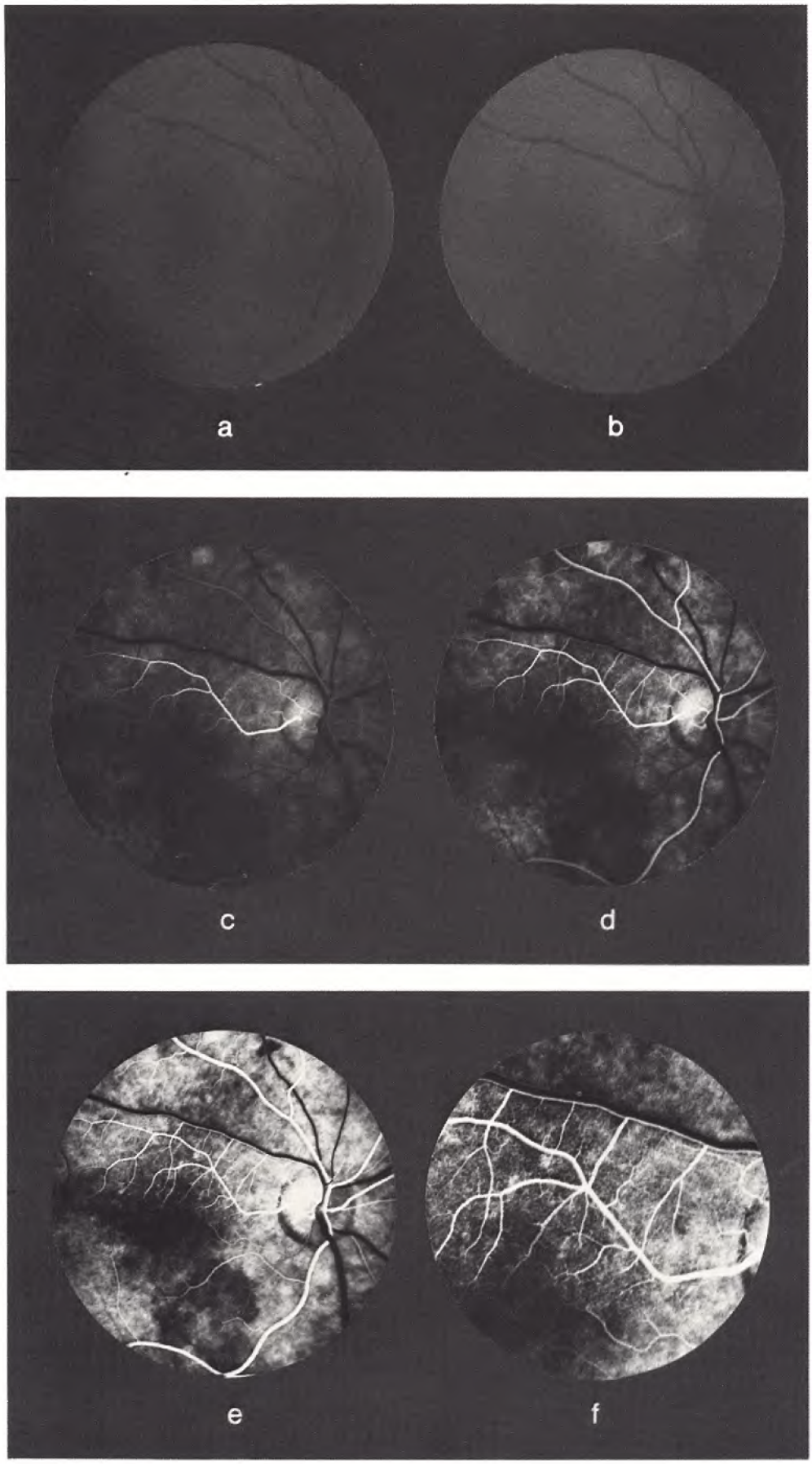


Figure 29—Transmittance curves for various filter combinations used in fluorescein angiography. A—The Kodak Wratten filter, no. 47 (blue), dotted line, and the Kodak Wratten filter, no. 15 (deep yellow), solid line. B—The interference filters, Baird Atomic B4 (solid line) and Baird Atomic B5 (dotted line), used for black-and-white fluorescein angiography. C—A recent filter combination for fluorescein angiography. The exciter filter, the Leitz KP490, is shown by the dotted line. The solid line shows the characteristics of the Kodak Wratten filter, no. 15 (yellow), which is used as the barrier filter. The Schott BG38 filter, dashed line, also serves as a barrier filter in absorbing the unwanted red transmittance of the KP490 filter (660 to 700 nm).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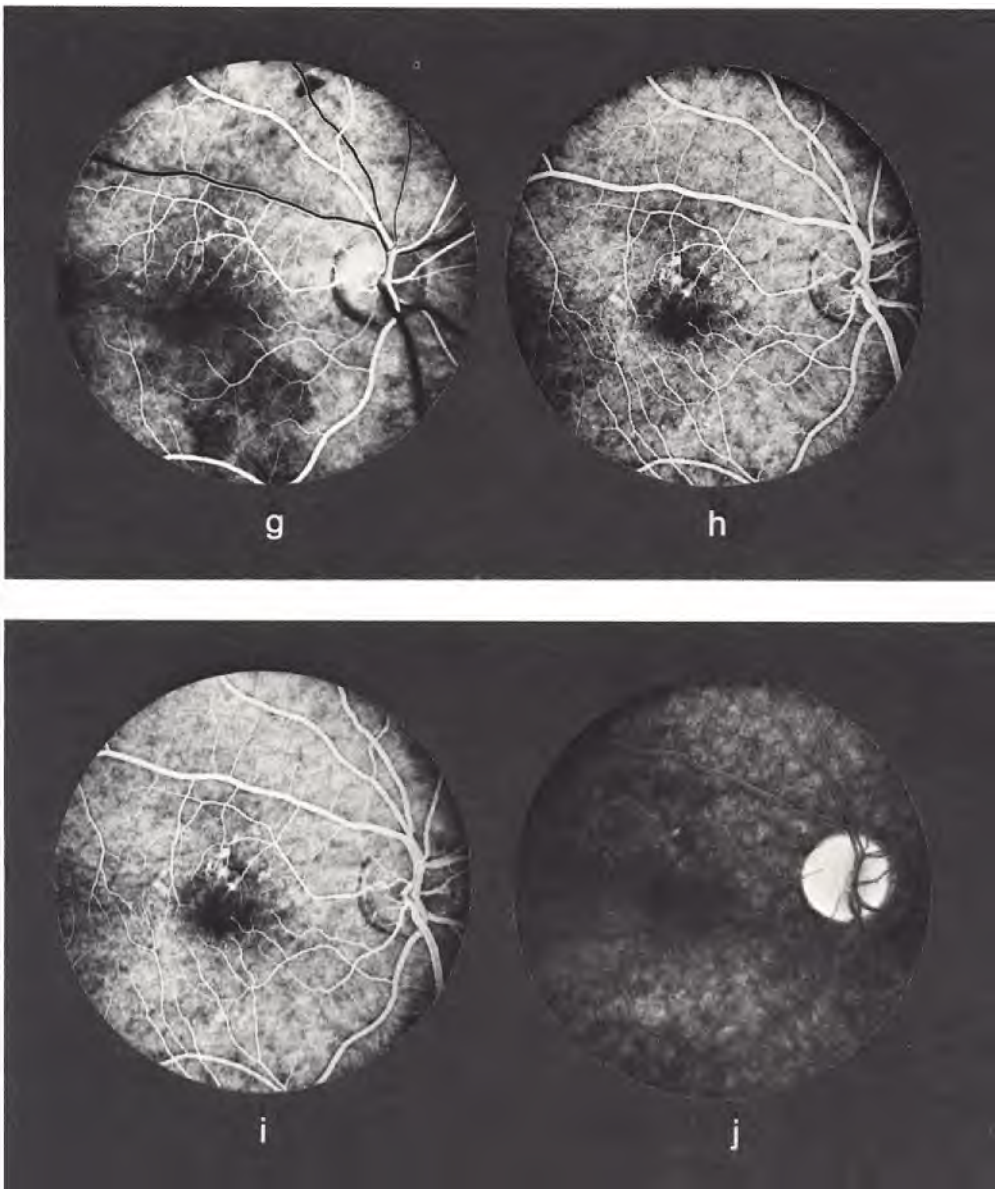

Figure 30—Fluorescein angiography is the only effective method which permits the detailed study of retinal circulation. A—The control, or pre-injection picture, is relatively dark since *the* dye has not yet reached the vasculature. **B-As** the dye approaches the retinal vessels, the brightness of the background increases as the choroid fills with dye. If there is a cilio-retinal artery present, that is the first to fill. **C---**Further filling of the cilio-retinal artery. The arterial phase has begun. D—The early arterial phase. E—Continuation of arterial filling and the venous phase. Venous filling is characterized by "laminar flow", the white stripe lining of the vein. F—A close-up view of the central retinal vein as seen in E. Laminar flow is shown. G, H, I—Continuation of circulation through the arterial-venous phase. J—The "late" photograph taken 20 minutes after the injection of dye.

Figure 31—Patient positioned at the fundus camera table. The injection is given by the physician in the antecubital vein.

viously, the patient in our laboratory receives a fairly detailed orientation to the procedure to be undertaken. In this way, the patient knows in advance what he will experience and what is expected of him. The following is a brief summary our procedures:

1. The patient is positioned at the camera as usual, and extends an arm forward on an arm board (Figure 31).
2. The field designated for study is visualized and critical focus is achieved under white light. The excitation filter is rotated into place and a control picture (pre-injection) is taken.
3. The physician inserts the hypodermic needle into the antecubital vein.
4. The patient's position is rechecked, appropriate changes are made if necessary, to regain the original position and a signal is given to the physician to begin the injection.
5. The time is marked at the beginning of the injection.
6. The photographer delays 5 to 7 seconds, then begins photography, taking pictures at one-second intervals.
7. If the study concerns only the posterior pole or one specifically designated field, 30 pictures are taken in rapid sequence. If the patient has diabetes or hypertension, 20 pictures of the posterior pole are taken in rapid sequence, followed by survey photographs of the seven standard fields as described previously.
8. The late photographs are made 20 minutes after the injection.
9. Upon completion of the angiogram, we inform the patient that, due to the presence of dye in the system, the urine will be deep in color, which is a normal phenomenon. Additionally, the complexion may change to a pale yellow color, which may last for a few hours.

The ability of the photographer to establish a good working rapport and to be able to manage the patient during angiography is probably the most important factor in obtaining consistently good results. Orientation of the patient is only the first step in this process. In spite of this orientation, many patients must be repeatedly reminded to maintain their original position as the angiography progresses. Many patients will draw increasingly away from the headrest as the rapid-sequence photography is taking place. This comes either as a result of the injection or the effect of the repeated flashes of light.

The photographer should not assume a position directly behind the instrument, but sit at the left corner of the fundus camera table. This enables him to extend his left arm out to reach the patient's eyelids. From this position it is possible to reach over the barrel of the fundus camera and to retract the patient's left upper eyelid without any difficulty. The photographer's arm can be supported by gripping the top of the headrest bar with the left index finger while retracting the lid, if necessary, with the thumb. The other fingers can rest lightly on the patient's head. If, as time passes, the photographer feels a lessening of tension on his fingertips, he is immediately aware that the patient's position is changing. Promptly compensate by manipulation of the joy stick. This technique is not only useful to determine the patient's movement during serial angiography, but it has also been useful to let the photographer know that the patient might be feeling uncomfortable. If the movements are numerous or are quick and jerky, immediately check the patient to determine what is happening. During the orientation of the patient, stress the fact that he will be able to blink and breath in a normal fashion. Many patients are relieved to hear this, and they advise us later that this was an important piece of information to have beforehand. Also ask that patients keep their teeth together, lest they rest too lightly on the chinrest and move during angiography as they breath.

Some physicians remove the needle from the arm immediately after the injection. Many times the practice is to bend the arm upwards to apply pressure to the site of injection to prevent the formation of sub-epidermal hematomas. Unfortunately, the time sequences for these actions usually coincide with the arrival of the dye in the retinal vasculature, and often these movements cause the patient's head to move about. This movement results in flares and haze in the pictures. It is a good practice to leave the needle in place until after the rapid-sequence photography is completed.

It is good general practice to have the patient's clothing around the neck loosened prior to angiography. This may be helpful in allowing the patient to breath comfortably during the procedure, but more importantly, it will be necessary if the patient becomes ill.

Complications of fluorescein angiography

It is generally recognized that fluorescein dye is a relatively harmless substance to the human system. Nevertheless, it must also be acknowledged that patients can and sometimes do react to the *procedure* just as much as to the presence of dye in the system. Thus, it is important for the physician and photographer to be aware of some of the reactions that may occur, and to be prepared

to deal with them.

Most reactions encountered are probably psychogenically triggered mechanisms due to anxiety and nervousness. Most common of these reactions is transient nausea, which may occur within the first minute after the injection. The ability of patients to cope with this reaction is varied, but when they are fully oriented to the procedure, and are aware of the importance of maintaining a stable position during that first minute, many have been able to remain in this position for serial angiography, sitting back away from the camera only if the discomfort is too severe. If the nausea persists, have the patient take a few deep breaths through an open mouth. This will help the nausea to pass. It is wise to keep an emesis basin nearby in the event that the patient has to vomit. Other reactions that have been encountered have been a flush of warmth, a metallic taste on the tongue, drying of the mouth (or the reverse condition, sudden salivation), sneezing fits, lightheadedness and even fainting.

Occasionally, the physician may have a little difficulty in maintaining proper position of the needle after placing it into the vein. Upon beginning the injection, he discovers that the dye is not being injected into the blood stream, but rather into the soft tissue around the vein. Such extravasation of dye into the tissues is very painful to patients. They may experience a throbbing pain in the arm or a tingling in the fingers, or both. Local massage and ice-packs will help relieve the pain.

There is also a chance that the patient will experience an allergic reaction to the fluorescein dye, which may be manifested in the form of a rash or hives (urticaria). See Figure 32. These reactions generally occur within the 3- to 10-minute period after the injection of dye. The reaction usually involves the face and trunk, and varies in degree of severity. The use of an antihistamine, either by means of an intramuscular injection or a tablet taken orally, effectively counteracts the reaction and brings quick relief. The physician will want to detain the patient for some length of time in order to be certain that the medication has been completely effective. Though these ministrations of medications are the responsibility of the physician, in all probability the reactions will occur with only the photographer in attendance.

In many practical working situations, the physician often leaves the angiographic room once the injection of dye has been given and serial angiography is under way. It is therefore imperative that each angiographer be aware of the reactions that may occur, be able to recognize signs of distress and be trained in the proper course of action. *(Only a few courses are actually available to the photographer).* The first action to be taken is to look after the immediate welfare of the patient. In the event that the patient gets light-headed or even faints while still seated at the camera, be sure that the clothing around the neck is loose. Get the patient into a stable position so that he does not hit his head against any part of the apparatus or nearby furniture. The hest and quickest course in stablizing the patient is to lay the patient down on the floor, The second action to be taken is to notify the physician that the patient is in distress. After this has been done, circumstances dictate what the photographer should do. It is usually not wise to leave the patient alone at these times unless absolutely necessary.

A most important, general responsibility that falls upon the photographer's shoulders is to be certain that the angiographic room is equipped for any of these emergency situations. The room should be large enough to allow the patient to be stretched out on the floor. The photographer, therefore, should not allow the room to be cluttered with furniture or unnecessary items. A two-way intercom system should he installed to enable the photographer to contact the office secretary or the physician without having to leave the room. A stretcher should be kept in the room along with an "emergency kit" of materials and medications. This kit should contain instruments and medications which should not be used except for emergency situations. Do not allow anyone to borrow a tourniquet or syringe from this kit, at the risk of forgetting to replace it.

The emergency kit in our laboratory contains the following:

1. Tourniquets, adults and children's sizes
2. Plastic airways for adults and children
3. Ambu bag
4. Oxygen
5. Syringes
6. Ammonia ampules or smelling salts
7. Emesis basin
8. Adhesive tape
9. Blood pressure cuff and stethoscope
10. Epinephrine hydrochloric solution, 1/1000
11. Diphenylhydramine hydrochloride (Ben edryl)

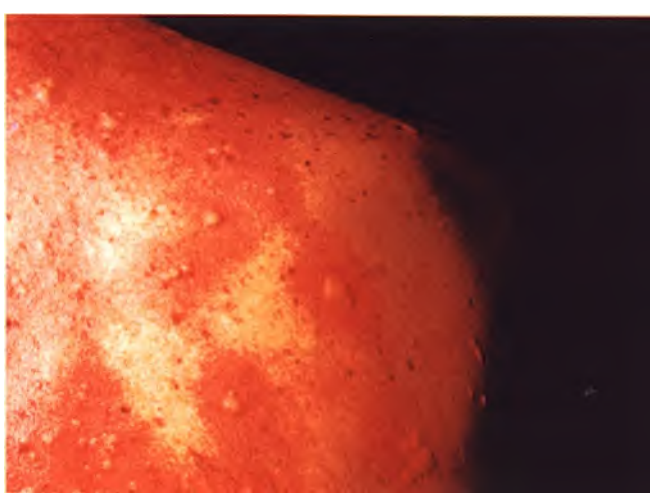

Figure 32—An allergic reaction to fluorescein dye, These are often treated with an antihistamine.

Color fluorescein angiography

Shikano and Shimizu (1968) were the first to experiment with the concept of obtaining angiographic studies in color. Using a Fuji TV-B (purple) filter for excitation and a Fuji 12 (yellow) barrier filter, color transparencies were obtained with Fuji color reversal film (ASA 100). This film and filter combination rendered many of the fundus details in their natural colors. Matsui (1969) improved upon the

color rendition of these fundus details with the use of the Kodak Wratten filter no. 32 (magenta) for excitation, and the Wratten no. 58 for the barrier filter. This ability to record fundus details in natural colors and the fluorescein dye passing through the vasculature as greenish-yellow was very appealing in the early days of angiography, hut the technique never gained any degree of popularity.

The interpretive advantage of color angiography over black-and-white recordings is subject to wide differences of opinion, and whether or not a laboratory will include this as part of their regular angiographic program will be dependent upon various factors. Generally speaking, color angiography's greatest advantage is its ability to record the fundus details in their natural colors, so that as the dye passes through the retinal vasculature, it is possible to separate true fluorescence from pseudofluorescence. This is often difficult for a beginner to determine in the evaluation of the angiographic data. A serious disadvantage, however, is the lack of photographic contrast between the fluorescing bodies and their surround; this can be a severe handicap to the interpreter who is interested in seeing possible changes in microvascular structures. Additionally, the image quality of the color angiogram suffers greatly in the presence of the least amount of intraocular haze, whereas the black-and-white angiogram may still yield useful diagnostic data.

Darkroom procedures

Standard darkroom procedures prevail in the processing and printing of the angiographic negatives. However, due to the low level of intensity of the emitted fluorescence, the films must be forced developed in order to obtain a negative with good density and contrast. Various factors such as the intensity of the illuminating source, the filter combinations and film type used are determining factors. The table in Figure 33 shows some of the developer and development time combinations used by various angiographic labs.

As discussed previously, the biomedical photographer may find himself on the periphery of an angiographic program rather than being totally involved. This position is often very difficult in that these individuals must provide a photographic service either on short notice or with little opportunity to become familiar with the procedures. Many institutional photographic departments are simply the photofinishing lab for the ophthalmology department. Their sole function is to process and print angiographic films turned over to them by the eye department. In many such cases, the angiography is performed by a resident, who might be assigned to that task in addition to his other functions. It is a common, unfortunate occurrance that if the films are not good, the physician immediately blames the darkroom technician; additionally, little thought is given to the necessity of giving the technician any instruction in the subject matter being dealt with. It would he of value to the biomedical photographer or to the darkroom technician working with these films to have some background knowledge and to be able to evaluate the negatives in the event of problems.

A number of factors may contribute to thin (underexposed) angiographic negatives. Attempting angiography through a poorly dilated pupil without compensation in the output of the electronic flash will certainly result in thin negatives. It is therefore helpful to increase the output of the exposing source. Hazy intraocular media may cause some loss of exposure, but generally, it affects only the photographic contrast between the fluorescent and nonfluorescent bodies. Thin negatives can also be caused by a sudden loss of power due to voltage drop or to a malfunction of the power generator, permitting the electronic flash tube to ignite but at less than peak intensity for that output setting. Misalignment of certain parts of the optical system may also cause difficulty in this regard.

The insertion of the wrong filters into the optical pathway may cause thin negatives. This point is illustrated by recent experience in the angiographic program at a doctor's private office. He installed a Zeiss fundus camera in his office and started the program by doing the angiography himself. His knowledge of the

Figure 33—Photographic considerations with different power generators

Power Generator	Filters (Exciter/Barrier)	Kodak Film	Flash Setting	Developer	Development Time/Temperature
Topcon TRC-F	BA-15*	Tri-X	100 watt-seconds	Kodak D-11	7 mins 70°F
Topcon TRC-F	E4-B5†	Tri-X	100 watt-seconds	Kodak D-11	6 mins 70°F
Topcon TRC-F3	BA-15*	Tri-X	100 watt-seconds	Kodak D-11	8½ mins 68°F
Zeiss Siemens	E4-B5†	Plus-X	3	Kodak D-11	7 mins 68°F
Zeiss Dyonics	E4-B5†	Tri-X	200 watt-seconds	Kodak HC-110,1:7	10 mins 70°F
Zeiss Dyonics	E40-B50†	Tri-X	200 watt-seconds	Kodak HC-110,1:7	6 mins 70°F
Zeiss FF II	E4-B5†	Tri-X	High	Kodak D-11	6 mins 72°F
Zeiss FF II	KW47A-W15††	Tri-X	Medium	Acufine	8½ mins 68°F
Zeiss PP 260	E4-B5†	Plus-X	3	Kodak D-11	7 mins 68°F
Zeiss FF III	E4-B5†	Tri-X	2	Kodak D-11	7 mins 72°F
Zeiss FF III	E40-B50†	Tri-X	2	Kodak D-11	5 mins 72°F

*Matched interference filters by Topcon Corporation of America, Paramus, New Jersey.
†Interference filters by Spectrotech Company, Lincoln, Massachusetts.
††Kodak Wratten filters by Eastman Kodak Company, Rochester, New York.

Figure 33— Careful control of all the photographic parameters, power generator, barrier and exciter filters, film, electronic flash setting, developer, and development time and temperature is required to produce good angiographic negatives. The various combinations listed here are from an informal survey of angiographers.

technical procedures was fairly good, and he began to obtain satisfactory results. A short time later, he decided to train one of his office staff to do this work, in addition to her other functions. This assistant learned the basic procedures well enough to get good results. She turned the exposed films over to a biomedical photographer for processing. A satisfactory working arrangement evolved, and in time, a good angiographic program routinely was being conducted. Suddenly, several groups of negatives were extremely thin and of no diagnostic value. The physician questioned the processing technique, but the photographer claimed that all darkroom procedures were correct. Technical representatives from the camera manufacturer were called, and after numerous visits, no explanation was found. All electronic and optical parts were found to be in good working order. Repeated calls to the main office were made to complain about the camera equipment and service. I was asked to consult on this problem. A lengthy conference with the angiographer did not indicate anything faulty in her procedures that may have been the cause of the problem. The electronic flash tube, when measured with an electronic flash meter placed over the front lens, delivered the correct amount of light at each flash setting.

Later, in setting the camera up for angiography, I noticed that a green filter was in the barrier filter's position. It came to light that the physician had done a color angiogram on a patient some time ago and had replaced the interference barrier filter with a Wratten no. 58 filter. Upon completion of that study the physician did not re-install the barrier filter normally used for black-and-white angiography. Thus, the loss of negative density was due to a wrong barrier filter.

Conversely, forgetting to correctly position either the excitation or barrier filter will result in dense negatives on which no dye may be seen. Standardization of camera technique will minimize these errors.

Summary

The preceding discussions were intended to provide a comprehensive guide for beginners so that they will understand and be able to perform the tasks of fundus photography and fluorescein angiography. In order to achieve this, it was felt that certain background information in related areas would help to make the job easier. In reviewing the material presented, a few important factors come to mind which bear repetition:

1. **Determine your eyepiece setting** and do all photography without accommodation of the eyes.
2. For critical focus, you must **see a sharp image of the cross-hairs superimposed over a sharp image of the fundus.**
3. For critical illumination (and optimum camera position), **look for the deepest tone of color.** This point is vital in angiography if the details of the fundus cannot he visualized at all times after the excitation filter is rotated into place. It is not necessary to **see** details; simply look for the deepest tone of blue.
4. **Establish a good working rapport with the patient.** This is of equal importance with technical expertise. The ability of the photographer to calm the nervous patient and to manage that patient during angiography may make the difference between success and failure.

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