

## **Imaging Spectroscopy, A New Non-Destructive Method for Materials Analysis**

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*Verbatim transcript (from audio tape) of presentation made by Dr. Warren Grundfest on September 6, 1998 at the Shroud meeting in Dallas, Texas. Note that Dr. Grundfest projected a number of slides to illustrate his presentation. Wherever possible, these have been included in the text of this transcript.*

### Introduction by Barrie Schwartz:

For those of you who have known me for a long time, I think you've all heard me, over the years, talk about my friend and mentor, Dr. Warren Grundfest. Dr. Grundfest is the Director of the Laser Research and Technology Development Laboratory at Cedars-Sinai Medical Center, and there are many other credits that I could give him, but the biggest credit I want to give him is that he has supported me in my work, both on the Internet (Shroud of Turin Website at <http://www.shroud.com>) and the Shroud of Turin CD-ROM for the last three years. So I am very proud and pleased to present Dr. Warren Grundfest.

### Dr. Warren Grundfest:

Thank you Barrie. It's been my pleasure to work with Barrie and, through him, I've got a much greater and better understanding of the Shroud of Turin and its implications. I think what you may not know is that I've known Barrie since 1987 and since that time, from the very first time I walked into his studio, he began showing me some of the images he had taken back in 1978. So that's where this comes from. Over the years we've talked about can we better preserve the Shroud of Turin.

Recently, in our laboratory we've helped design some very exciting and interesting technology for optical imaging. And so what I'm going to talk you about today is something called imaging spectroscopy. This is a new, non-destructive method for materials analysis.

I will show you some of the medical applications which will hopefully give you an idea of how we can apply this to the Shroud of Turin, which may yield some very fascinating and, I believe, important information.

This is actually a joint effort between Barrie and myself. Barrie is an imaging expert in his own right and has had a lot to do with some of the concepts I'm going to present today.

Just for everybody in the room, for the non-scientists, everybody knows if you see light coming out of a flashlight, it's white. But in fact that white light is composed of the entire spectrum, from red to orange to yellow to green to blue to purple and into the infrared or up into the ultra-violet, depending upon which direction you're going. But your eye doesn't perceive it as such. We perceive it as one color at one point in space, red or green or blue or white or yellow or gold.

Optical spectroscopy is a non-destructive, quantifiable analysis of the optical signals from an object by light wavelengths or light colors. So that we can determine how much of a particular color is at each point in an image. Now optical imaging, I think you're all familiar with, is the use of light to record the structural characteristics of an object. What is its shape, what is its form? And we do this on everything from microscope slides to standard camera images.

What really has become available only very recently and only through a lot of work by NASA, are the types of devices they put up in spacecraft that really aren't adaptable to something we use in a room. We've developed something that allows us to obtain wavelength intensity information. That means how much color from each type of light at each point in space from every point in an optical image.

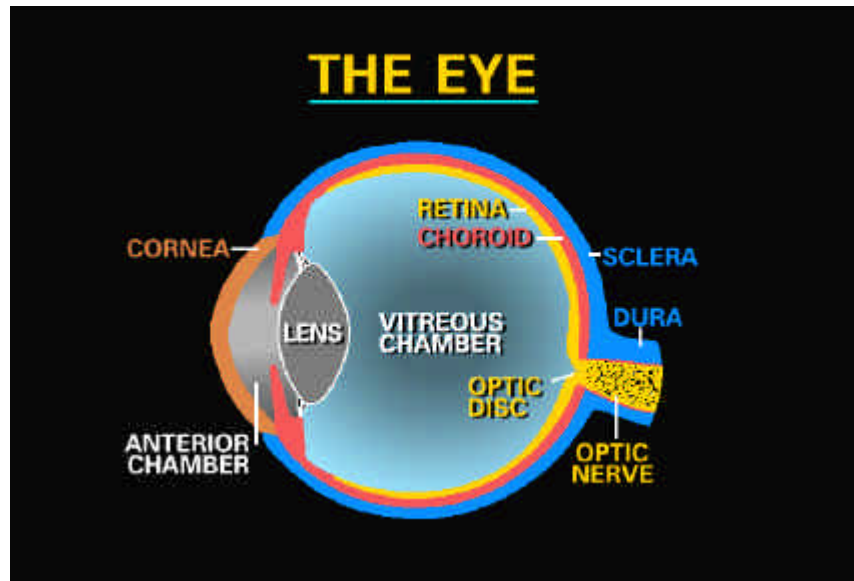
Now in the past you could do this point by point by point, and you'd be there for a week trying to get one image. However, we can now do this all at once. I think you are all familiar with the so-called CCD or "chip" cameras that we all use in our camcorders and other things. Well, however many pixels there are, however many little squares there are (on the chip) in that camera, we can get that many number of pixels to give us a complete image. So for every point in that image, we can now get an optical spectrum at that point. This is a tremendous amount of information; it is 90 megabytes of data per image. So part of the problem is how do you analyze all that data and what does it mean?

The human eye perceives light reflected into it from objects in the environment. This optical signal is composed of the various wavelengths of light: red, green, blue, yellow. The eye and the brain process this information into a perceived color. They unify it. They take it all together and give you one color, which is a sum of all the wavelengths for a given point in the image.

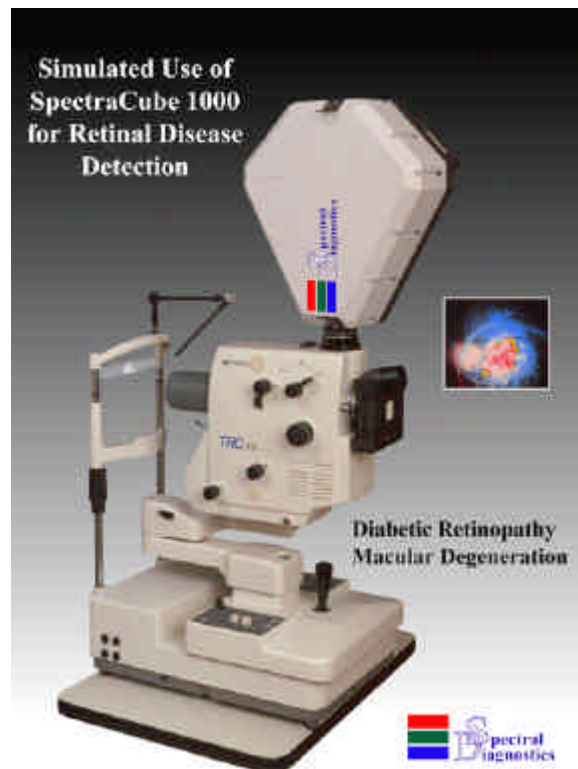


With our device, we do the reverse. Spectral imaging separates out all the colors at each point in the image. The various substances that absorb and reflect light at a specific wavelength can be determined, allowing precise, non-destructive analysis of the tissue. So if we have a protein such as hemoglobin or albumen, or if we have materials like cellulose, we can in fact understand those spectral patterns.

In direct reference to the Shroud, what we see in medical imaging will not be the same, because these materials have been exposed to oxygen and to heat. So it's not quite so simple as taking something out of a textbook. But in fact, we believe very strongly that this technique can be used with any optical image, specifically looking at the Shroud of Turin. And by comparing known spectral patterns that we create in the laboratory, we can then obtain information from the imaging spectrometer and develop a map of chemical species. So we can see things that you can't see with the naked eye. That are *impossible* to see with the naked eye. So we can see if the blood spatters drip in a particular direction. We can see how the albumen is leached out. We can see patterns in the cellulose or the fibers that are not visible, even to polarized light microscopy, although we can use these techniques and they are additive. They may give us different results and then we have to reconcile them, but they can be all put together to give us much more information.

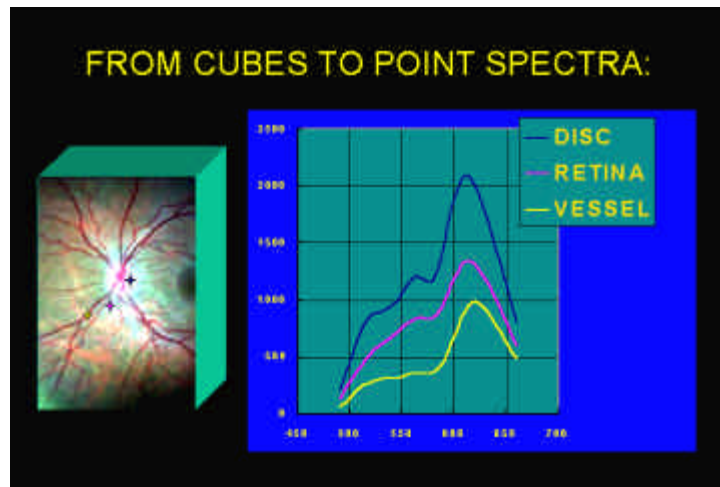
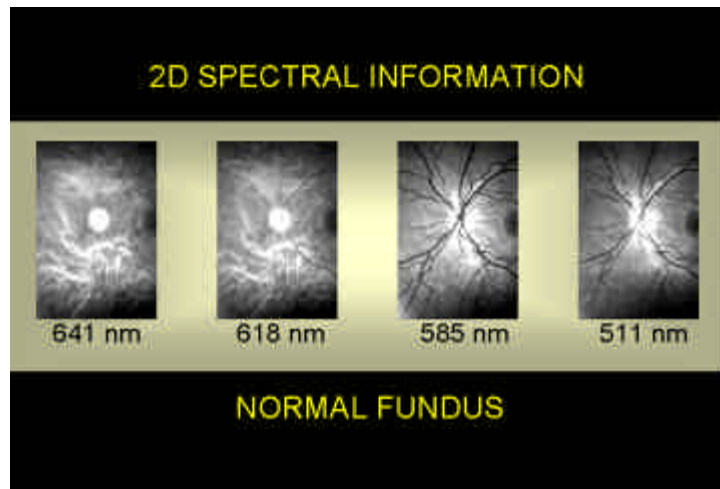


I just want to show you a few of the medical applications to illustrate this. I think you are all familiar with the eye. The human eye is a marvelous device. The light comes in through the lens, the cornea and its focused on the retina. How does the retina exist? It has no blood vessels of its own. It has a layer behind it called the choroid. And that's where a lot of diseases take place. But imaging that choroid, imaging behind the retina is a problem, because you've got to see through the retina to get there. And remember, the purpose of the retina is to absorb light, so seeing through something that absorbs light is not necessarily easy.

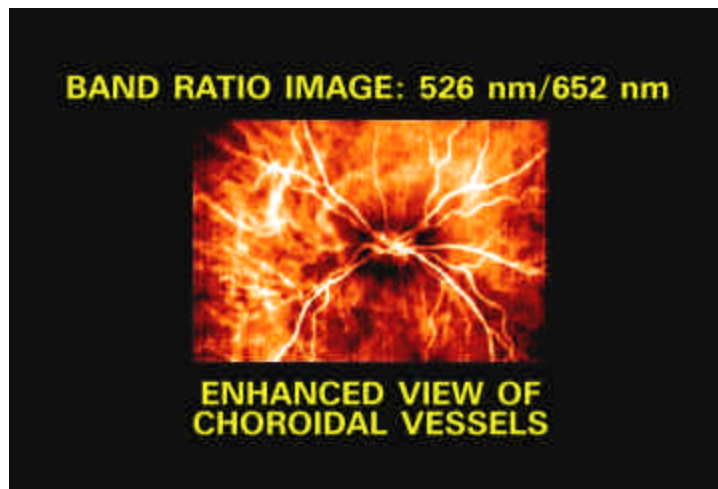
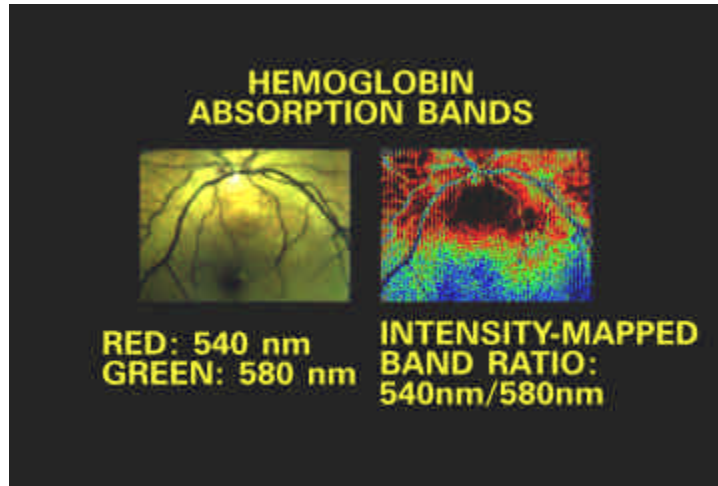


We came up with this technology which is a box like this (see above illustration). This is the device we use to take standard images of the eye called a retinal camera. We use it for diagnosing various diseases that occur with diabetes or with macular degeneration, which is a leading cause of blindness in the elderly. What we get is an image that is quite spectacular. Quite different than any image obtained so far.

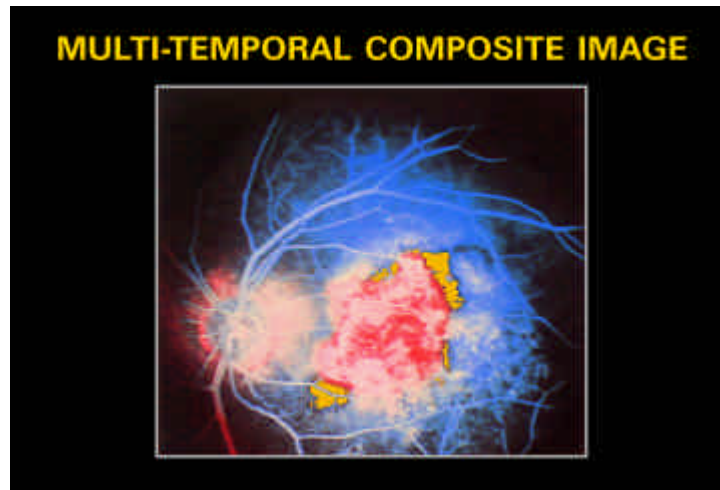
To give you an idea of how this works, this is your standard image (see illustrations below). This is what the retina looks like when we look through that camera at high resolution. You can see the arteries and veins.



But at every point in that image we can get a spectra, whether it is similar to hemoglobin or similar to the proteins, I won't go into the details. But we can, with very high resolution now, map in the retina the spectral components.



And what does that allow us to do? We can, in fact, record the veins, (the arteries here have disappeared) and the choroidal vessels, this white hazy material from all the veins in the back. Normally, we would need to give you an injection of dye and flash some bright xenon lights in your eye. The dye may make you get nauseous and throw up in the process, so this new technique is a little more pleasant for the patient.



But perhaps more spectacularly, after we process the image by doing some mathematics and ratioing the entire image between two different wavelengths or two different colors, these vessels stand out. When we first showed this to our ophthalmologists they couldn't believe it. They said, "It shouldn't work. There should be lots of reasons it doesn't work." But it does.

I won't go into the mathematics of why. But the fact is we can now see things that you cannot see with the naked eye. Similarly, we can map hemoglobin. I just want to show you one example. This is intensity mapped ratios of oxygenated and deoxygenated hemoglobin in the eye. You can see very spectacular, very specific patterns in this patient who we know had certain areas, namely this area here (in reference to projected 35mm slide illustration), that had laser treatment. So there wasn't any hemoglobin. We can be very precise. We know where he was treated.

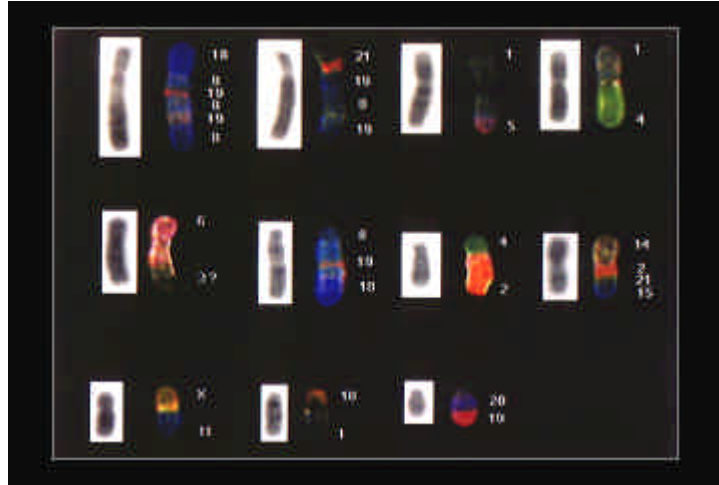
Using the same techniques, we can obtain unique spectral patterns for various proteins, such as blood and saliva in the laboratory. Once we create a library of spectra, we can use them as a reference to what is on the actual cloth.

Comment from the audience interjected here by Dr. Alan Adler:

Also, the spectral state. Hemoglobin exists in lots of states and it's a real problem on the Shroud to know what some of those states are.

Dr. Grundfest:

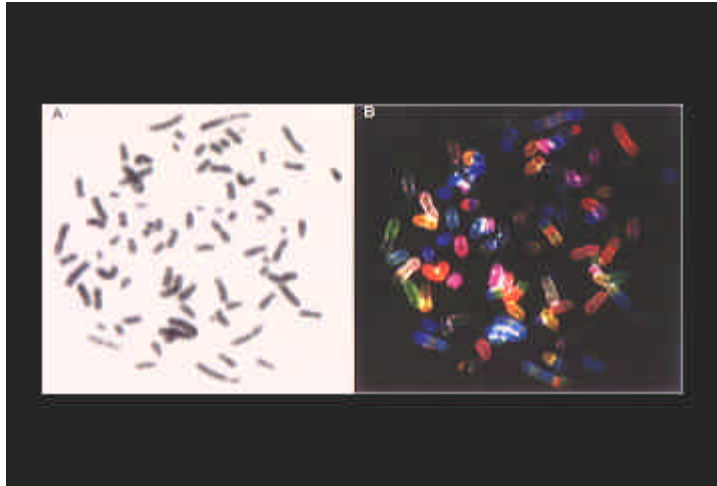
This would allow us to do that in fact. He's talking about how much oxygen is in the hemoglobin, what the electronic state is and that determines its color and its absorption. So in fact, it would allow us to do that, not only for the hemoglobins, but also for the albumens and some of the other compounds, because this works in the infrared.



Now to give you an idea of how this works with dyes. You can say fine, that works for hemoglobin but what about for other things? This is a very nice application of this technology (see above illustration). After we developed it we went to Thomas Reid of the NIH (National Institute of Health) who is head of their genetics laboratory. People are always trying to look at genes. They are always trying to understand birth defects. What he was able to do by using five specific dyes and hooking this device up to a microscope was, for the first time in history, get spectral banding patterns in human chromosomes.

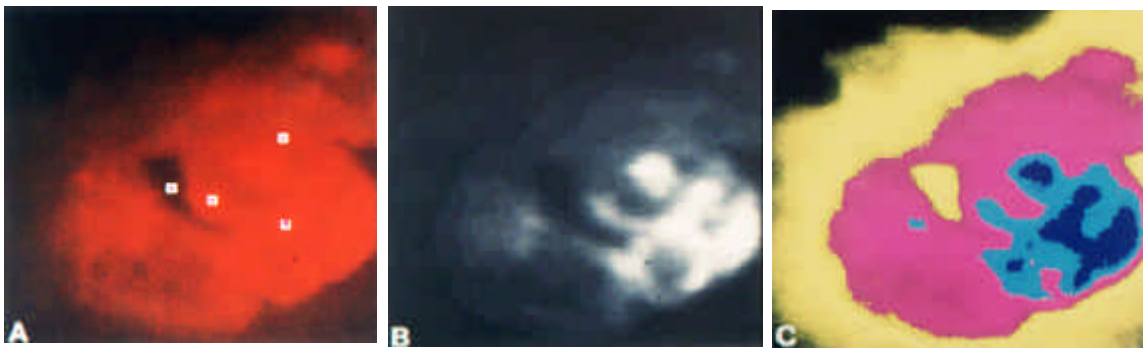
That means we can sort every chromosome by its spectral color or composition. So here's the swatch that you get when you take these out of the body and you grow some cells (it takes about 24 hours). We can go all the way from one through twenty-two and x and y and they all have their own unique characteristics. In fact, this was part of a publication in *Science* showing some translocations or genetic abnormalities. This is very important in diagnosing and treating children.





By looking at the color analysis if you will, this same mapping (was done). We were able to see little tiny bits of things at very high resolution. This is the old way of doing it (A, above left), the so-called black and white karyotype staining. And that's the new way (B, above right). I think that just from looking with your own eyes you can get the impression there's a lot more information here (above right) than there is there.

That's not to say this doesn't work. It's to say that we've gone (to) the next step. Now, this actually got published in *Science* in July of 1996, and so we think this technique is quite valid. We've gone on at the start to apply this to studying cancers. And cancers themselves have their own particular spectra. Now there's no dye here. This is the dye that is endogenous to the tumors. This is the image under red light illumination (A, left below). This is the image mapped (B, below center). And this is a false color image (C, below right) and at each point there is a different spectra. We can actually find germinal centers of the tumor, the areas where the most active proliferation is. This also can help us find the borders so when we take out the tumor, we know we've got it all.



We can in fact, do anatomic mapping using this technology. Well how would we apply this to Shroud of Turin? How would we tell what's different? Whether its blood or other stains, we can take the area that we know and say we want to find all the areas that are spectrally identical and map it that way. We don't need apriori knowledge, because this machine has the ability to say well, I see this bump, let me find all the bumps that are similar in the whole image. And you can do that in either high resolution or looking just at a small area, or across the entire cloth. That is in fact what we would propose to do is take both high resolution and low resolution images (low meaning larger images, not necessarily a lower number of pixels).

By using carefully calibrated light sources, a pattern of reflected light can be used to map specific compounds on the Shroud as well. So we can use this in two ways. We can use it to say, let's find the areas that are similar and also let's see if we can identify some of these chemical species. Now this allows us to image the Shroud and simultaneously map the various substances present. And we should be able to sort out those that are overlapping, which I think to date has been very difficult to do.

This includes the images areas, the bloodstains, the water marks, the scorches, the burns and the cloth itself. As we get more knowledge, we might be able to do this in real time. We can in fact begin to see where the overlaps are and which came first.

The technology, when combined with high resolution imaging and appropriate light sources, will allow us to characterize the properties of the image on the Shroud, versus the background. I think that perhaps may be the greatest value of this technology. From this data we can determine the rate of image degradation. I think the science is nice, the hypotheses are nice, but one of the things we want to do is really preserve this magnificent and quite special cloth. Over time, this technique should allow us to quantify the rate and causes of image degradation. The data can then guide future conservation and preservation efforts and hopefully, measure their effectiveness.

I don't want to stand here and say "we know" we can do all this all at once. This will take some time, and it may be that we can examine the Shroud once, and then have the opportunity to do that (again) at some later date. But we can gain an awful lot of information just from one image. From several images we can gain tremendous amounts. Remember, 90 megabytes of spectral data from one image.

This work can be supported by my laboratory at Cedars-Sinai Medical Center. And since we have the spectrometer, we have the light sources and we have Barrie's expertise, we look forward to hopefully examining the Shroud with this technology. Thank you.

*A series of questions was then asked by members of the audience which led to further discussion of the technology . Unfortunately, there was no microphone set up to record the audience, so some of the questions were impossible to transcribe completely. In some cases the questions were summarized or paraphrased and are not completely verbatim. In all cases they are accurate as to general content. They are included here with that understanding.*

Question from Dr. Alan Adler:

Warren, as Gus said, my job is to rain on everybody's parade. I think it's no secret here that I'm on the Conservation Committee (referring to the committee of scientists and scholars appointed by the Archbishop of Turin to guide the future preservation and conservation efforts on the Shroud). And I've been suggesting to the Cardinal that we do some of these things for a number of years. (continues for approximately one minute but unintelligible). I have to ask you, how far into the infrared can you go?

Dr. Grundfest:

The instrument we have only goes down to about 1.1 microns. We have access to an instrument, but we don't own it, that works all the way down to 10 microns.

Question from Dr. Alan Adler:

What is the resolution?

Dr. Grundfest:

In the blue, its about three nanometers. When you get down to the IR at about one micron its about fifteen nanometers, so it's pretty good. But by the time you're at ten (microns), it will probably be 100 nanometers, maybe 200.

Question from Kevin Moran:

What type of interferometer is used in your spectrometer?

Dr. Grundfest:

It's a Fourier Transform Signac Interferometer.

Question from Kevin Moran:

And what was the resolution time for example, in the retinal study where you were looking in the eye?

Dr. Grundfest:

The spatial resolution of the spectrometer depends upon the attached optics. When attached to a standard retinal imaging camera (fundus camera), the resolution is approximately 20 microns. Those images were acquired in eleven seconds, but newer versions of the instrument have reduced the time to a few seconds.

Question from Dr. Alan Adler:

...There's another problem on the Shroud. It may be a little naïve to say we're going to identify compounds. We can do that with the blood but I'm afraid that... with the images, you are really looking at a mixed structure to begin with...

Dr. Grundfest:

We've actually given a fair amount of thought to this.

Question from Dr. Alan Adler:

You could look at a peak in the UV and compare it with a peak in the near IR, that would tell us everywhere what the ratio of the final form was... even though you couldn't say how you got there.

Dr. Grundfest:

Exactly. In fact you would examine this using UV, visible and IR portions of the spectrum. This would need a lot of thought and a lot of planning. It's not, we show up and take pictures. One of the things that this would involve using the facilities not only in my laboratory but others, to postulate what the compounds are ahead of time and get a reference library of spectra. What that means is we take albumen, hemoglobin, sweat and a whole lot of other things on linen itself and age them and then expose them and then get reference spectra. And that doesn't mean we will be successful. I'm not going to stand here and tell you we can do this. I'm going to tell you we can get the information.

Question from Dr. Alan Adler:

...Recognizing the fact once you've got it into the computer, you can do all kinds of things that we never normally do... you don't have to understand everything.

Dr. Grundfest:

That's correct.